Different patterns of stage-specific horizontal distribution between two sympatric oligohaline copepods along a macrotidal estuary (Chikugo River, Japan): implications for life-history strategies

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Stage-specific horizontal distribution was compared between two sympatric oligohaline copepods to elucidate their life-history strategies related to reproduction: free-spawning Sinocalanus sinensis and egg-carrying Pseudodiaptomus inopinus. Copepods were sampled by vertical hauls of a 100-μm mesh plankton net along the macrotidal Chikugo River estuary in southwestern Japan from 2005 to 2006. Sinocalanus sinensis dominated throughout the year except for the warm season, when P. inopinus outnumbered S. sinensis. The spatiotemporal correspondence of peak densities with the estuarine turbidity maximum was evident for S. sinensis but ambiguous for P. inopinus. Nauplii of S. sinensis were always distributed on average downstream from copepodids and adults, whereas P. inopinus usually showed the opposite pattern of stage-specific horizontal distribution. During large freshwater discharges, S. sinensis was more subject to washout from the estuary. In addition, the potential vulnerability of S. sinensis to droughts was suggested by low tolerance for high salinities. Consequently, egg-carrying P. inopinus has higher flexibility to live under varying estuarine conditions especially during the warm season. In contrast, ecological specializations, such as tidal vertical migration and detritivorous feeding, are considered to underlie the dominance of the free-spawning S. sinensis under the conditions prevailing in the macrotidal environment.

KEYWORDS: copepod; estuarine turbidity maximum; Pseudodiaptomus inopinus salinity; Sinocalanus sinensis
INTRODUCTION

Position maintenance is essential for estuarine zooplankton species to maintain their populations against the net seaward flow. Tidal vertical migration has been observed in many species from diverse taxa (Kimmerer et al., 1998, 2002; Ueda et al., 2010). Since tidal currents are generally stronger in the upper water column, zooplankton swim up into the water column during the flood tide and stay close to the bottom during the ebb tide. Given their strong preference for low salinities, oligohaline species are considered to have particularly efficient mechanisms for position maintenance (Devreker et al., 2008, 2010; Suzuki et al., 2009a). For example, reproduction in the upper estuary cuts the risk of washout for offspring. Egg-carrying species have the advantage of minimizing the washout of eggs over free-spawning species. Fast growth during early life stages is a possible mechanism to shorten the critical period for position maintenance. The efficiency of position maintenance, combined with life-history strategies, determines the population stability of each oligohaline species in estuaries.

In Japan, the oligohaline copepod Sinocalanus sinensis occurs only in macrotidal estuaries in the innermost part of the Ariake Sea (Hiromi and Ueda, 1987; Ohtsuka et al., 1995; Ueda, 2005). In contrast, the copepod Pseudodiaptomus inopinus is widely distributed in brackish waters of East Asia (Ohtsuka et al., 1995; Sakaguchi et al., 2011). In the macrotidal Chikugo River estuary, the largest estuary flowing into the Ariake Sea, S. sinensis overwhelmingly dominates the oligohaline copepod community except in the warm season when P. inopinus outnumbers S. sinensis (Suzuki et al., submitted). Both species display tidal vertical migration to facilitate their position maintenance within the estuary under usual freshwater discharge levels (Ueda et al., 2010). In conditions of large floods, adults of both species are able to stay in tidal channels, whereas nauplii and copepodids are more subject to washout (Ueda et al., 2004). Besides the apparent behavioral similarities between the two species, S. sinensis displays more consistent patterns of tidal vertical migration, especially at night (Ueda et al., 2010). As for reproduction, S. sinensis spawns free eggs, whereas P. inopinus carries egg sacs. In addition, slightly different preferences for environmental variables are suggested: S. sinensis occurs at lower salinities and higher turbidities and over a wider range of temperature than P. inopinus (Suzuki et al., submitted). Although close comparisons between the two sympatric copepods will provide key information about the dominance of S. sinensis in the macrotidal environment, such a study has not yet been conducted.

To reveal information about the different life-history strategies between the two sympatric oligohaline copepods S. sinensis and P. inopinus, the present study investigates their horizontal distribution along the macrotidal Chikugo River estuary. In addition to monthly sampling through the year, more intensive sampling was conducted to examine the influence of the fortnightly tidal cycle and discharge fluctuations. Copepods were identified to the species level and classified into life-cycle stages. To test vulnerability to high salinities, the mortality of S. sinensis was examined empirically by transferring individuals to various salinities. Finally, the dynamics of stage-specific horizontal distribution were compared between the two species in relation to varying conditions in the estuary.

METHOD

Study area

The Chikugo River estuary is the largest estuary in the Ariake Sea in terms of both catchment area (2860 km²) and freshwater discharge (annual median of daily averages: 60 m³ s⁻¹). The tidal reach extends to the Chikugo Weir 23 km upstream from the river mouth (Fig. 1). Strong tidal currents completely mix the water column during spring tides, whereas partial stratification occurs during neap tides. An estuarine turbidity maximum (ETM) develops at low salinities especially during spring tides under usual freshwater discharge levels (Suzuki et al., 2007). The ETM fundamentally differs from simple loading of allochthonous sediments in that the ETM autochthonously develops through the hydrodynamic function of tidal pumping and estuarine circulation (Allen et al., 1980; Unclews et al., 2002). The ETM is usually located 10–20 km upstream from the river mouth at spring high tide, although it is transported back and forth over a 20-km range along the estuary with the semidiurnal tidal cycle between high and low tides. Detritus of phytoplankton and terrestrial plants accumulates into the ETM throughout the year, whereas phytoplankton occurs abundantly during the warm season only (Suzuki et al., 2012). Large floods occasionally affect the estuary and wash away the ETM especially during the warm season (Suzuki et al., 2009b).

Seven regular sampling stations (R1–R7; Fig. 1) were set up at intervals of 1.5–5.5 km along the lower reaches of the Chikugo River, ranging from the river mouth to the upper limit of the tidal reach (23 km upstream). Three regular stations (E1–E3; Fig. 1) were set up along the main tidal channel of the river so that E1 was near the mouth of the river and E3 was at the...
edge of the tidal flat (9 km offshore). The freshwater discharge was continuously monitored 26 km upstream and the data were uploaded to the website by the Chikugogawa River Office. For tidal ranges, water level differences were calculated between low and high tides in the morning using values that had been predicted for the location of Ohmuta (16 km offshore) by Japan Meteorological Agency (Japan Meteorological Agency, 2004, 2005).

Field sampling

Monthly sampling was conducted at the 10 regular stations from May 2005 to December 2006. All sampling dates coincided approximately with spring tides. A plankton net (45-cm mouth diameter, 100-cm long, 0.1-mm mesh aperture) equipped with a digital flow meter was towed vertically from close to the bottom to the surface at \( \sim 50 \text{ cm s}^{-1} \). Catches were preserved in 5\% formalin seawater solution. The volume of water filtered was estimated using the flow meter. Plankton-net hauls and environmental surveys were started at the uppermost station (R7) and finished at the lowermost station (E3) within 4–5 h around high tide in the morning. Temperature, salinity and turbidity were measured from the bottom to the surface at depth intervals of 1 or 2 m using environmental monitoring systems (6920 Sonde and 650 MDS Display, YSI, USA; Compact-CTD, Alec Electronics, Japan).

To examine the influence of the fortnightly tidal cycle on the stage-specific horizontal distribution, a series of frequent sampling was conducted at the same 10 stations in March and April 2005. Furthermore, the influence of discharge fluctuations was assessed by another series of frequent sampling from June to August 2005. During the latter sampling campaign, three extra stations (R2.5, R3.5 and R6.5; Fig. 1) were added to focus on the ETM and its surrounding waters, whereas sampling was not conducted at stations far downstream from the ETM. The procedures for collecting biological and physical data were the same for those of the monthly sampling. Turbidity was not measured on 19
and 24 April 2005, 24 July 2005 and 14 February 2006, due to mechanical faults with the turbidity sensor.

**Laboratory analysis**

For each station on each sampling date, copepod species were identified under a stereo microscope (×20–50 magnification) or an optical microscope (×200–400 magnification), before being counted quantitatively. Approximately 100 individuals were classified into four life-cycle stages (i.e. nauplius, copepodid, adult male and adult female) separately for *S. sinensis* and *P. inopinus*. For adult females of *P. inopinus*, egg-carrying individuals (gravid females) were separated from non-gravid females. The center of horizontal distribution (CHD) was calculated for each life-cycle stage on each sampling date using the equation:

\[
\text{CHD} = \frac{\sum n_i d_i}{\sum n_i}
\]

where \(n_i\) is the density of each life-cycle stage (ind. L\(^{-1}\)) at station \(i\), and \(d_i\) is the distance (km) between station \(i\) and the river mouth. Upstream and offshore distances are represented by positive and negative values, respectively. As density was highly variable between sampling dates, relative abundance \(\left[n_i/\sum n_i\right]\) is used to improve the visibility of stage-specific horizontal distribution in some figures.

Temperature, salinity and turbidity were averaged through the water column, since copepods were sampled by vertical hauls. Detailed information concerning environmental conditions during the sampling campaigns are published elsewhere (Suzuki et al., 2007, 2009b, 2012). There is a possibility of underestimates for smallest nauplii of *S. sinensis* and *P. inopinus* in the present study, as it is uncertain whether the 0.1-mm mesh net is fine enough (cf. Kuwatani, 2006). The vertical migration of all stages of the two copepod species has already been illustrated by depth-stratified sampling under varying estuarine conditions (Ueda et al., 2004, 2010).

**Tests for the salinity tolerance of *S. sinensis***

The salinity tolerance of *S. sinensis* was examined during 72 h at various salinities as a function of survival rates. Live copepods were collected by horizontal hauls of a plankton net (45-cm mouth diameter, 100-cm long, 0.3-mm mesh aperture) in the surface water at R2.5 on 5 May 2006. Using a 20-L bucket filled with ambient water (19.5°C, salinity 0.9), copepods were transported to the laboratory within 30 min. The bucket was placed in a thermostatic water bath (20.0°C) and then kept undisturbed for 30 min. For a direct transfer experiment, seven salinities (i.e. 0, 1, 5, 10, 15, 20 and 30) were produced by diluting artificial sea water with dechlorinated tap water. After macroscopic identification, *S. sinensis* adults were directly introduced into each salinity using a wide-mouthed pipette. For a stepwise transfer experiment, *S. sinensis* adults were first introduced into dechlorinated tap water. To obtain three target salinities (i.e. 15, 20 and 25), the experimental water was replaced partially with artificial sea water four times within the first 12 h of the experiment (Table I). All treatments were duplicated and conducted simultaneously using 1-L plastic beakers arranged in the water bath. Approximately 100 individuals were introduced into each beaker. Food and aeration were not provided during the experiments.

Survival was examined 4, 8, 12, 18, 24, 30, 36, 48, 60 and 72 h after the introduction of *S. sinensis* into the experimental water. Dead individuals were defined as those that did not respond to tactile stimulation by a fine pipette. These individuals were removed and preserved in 5% formalin water solution separately for each beaker. In addition, survivors were counted at the end of the experiments to confirm the number of individuals that had been introduced into each beaker. Survival rates were averaged between duplicates in each treatment. Microscopic examination confirmed the overwhelming dominance of adult copepods in the introduced *S. sinensis*. Although representatives of other copepod species than *S. sinensis* were rarely found, nine individuals of *Tortanus derjugini* were found in one of duplicates for the stepwise transfer experiment (target salinity 20).

Because of the possibility of predation by carnivorous *T. derjugini* on *S. sinensis* during the experiment, this beaker was not included in the calculation of survival rates.

**RESULTS**

**Year-round patterns**

The daily freshwater discharge changed drastically between June and September, whereas it usually

<table>
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<th>Target salinity</th>
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<td>25</td>
<td>9.6</td>
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Values represent means between duplicates.
remained below 100 m$^3$ s$^{-1}$ in the other months (Fig. 2A). Large flood events (>500 m$^3$ s$^{-1}$) occurred twice in 2005 and four times in 2006. Heavy rainfall maintained freshwater discharge above 100 m$^3$ s$^{-1}$ almost all the time from late June to September in 2006. The mean temperature for the 10 stations reached up to 30°C in summer (June–August) and dropped to 10°C in winter (December–February). The ETM was observed at salinity 1 through the year except summer 2006, when large freshwater discharges decreased salinity and turbidity through the estuary (Fig. 2B). Turbidity usually exceeded 200 NTU at low salinities (<10), whereas it remained below 100 NTU close to the upper limit of the estuary.

Spatiotemporal changes in the density of *S. sinensis* exhibited close relationships with those in salinity and turbidity (Fig. 2C and D). Adults/copepodids and nauplii of *S. sinensis* maintained higher densities (>1 ind. L$^{-1}$) at low salinities (<10) through the year. However, *S. sinensis* almost disappeared from the estuary in July and August 2006 under repetitive floods. The density of *S. sinensis* usually remained below 0.01 ind. L$^{-1}$ close to the upper limit of the estuary as well as in the lower estuary (salinity >20). In contrast, the density of *P. inopinus* showed clear seasonal changes with a minimum between January and March (Fig. 2E and F). In the other months, adults/copepodids and nauplii of *P. inopinus* occurred at greater numbers of stations (i.e. wider ranges of salinity) than *S. sinensis* ($P < 0.05$, the Wilcoxon signed-rank test). Higher densities (>1 ind. L$^{-1}$) were observed in adults/copepodids and nauplii of *P. inopinus* over a 10-km range along the estuary especially during the warm season.

*Sinocalanus sinensis* showed a consistent pattern of stage-specific horizontal distribution along the estuary: nauplii occurred on average downstream from adults and copepodids irrespective of the season (Fig. 3). The CHD of naupliar *S. sinensis* was always located downstream from the CHD of adult/copepodid *S. sinensis* ($P < 0.05$, the Wilcoxon signed-rank test). Although *P. inopinus* had the same pattern of stage-specific horizontal distribution in winter, it displayed the opposite pattern in the other seasons: nauplii occurred on average upstream from adults and copepodids (Fig. 4). The CHD of gravid females was usually located upstream from the CHD of the other life-cycle stages (excluding nauplii, $P < 0.05$, the Wilcoxon signed-rank test).

Responses to the fortnightly tidal cycle

The tidal range changed between ca. 50 and 500 cm in March and April 2005 (Fig. 5A). For clarity, tidal ranges were classified into three phases of the fortnightly tidal cycle: neap (<200 cm), medium (200–400 cm) and spring (>400 cm) tides. The daily freshwater discharge always remained around 70 m$^3$ s$^{-1}$ with the exception of two small peaks in late March (up to 160 m$^3$ s$^{-1}$). Temperature increased gradually from 9 to 18°C during the sampling period (not shown). The ETM developed markedly at salinity 1 during spring tides, although it almost disappeared during neap tides (Fig. 5B). Higher turbidities (>200 NTU) were observed only at low salinities (<10) during spring tides.

Temporal changes in the density of *S. sinensis* corresponded closely with those in the ETM (Fig. 5C and D). Adults/copepodids of *S. sinensis*, in particular, showed higher densities during spring tides. The density of naupliar *S. sinensis* showed almost the same fortnightly pattern during the sampling period, except during the spring tide in late March. The density of *S. sinensis* always exceeded 1 ind. L$^{-1}$ at salinities 1–10. In contrast, the density of *P. inopinus* showed no clear temporal agreement with the ETM (Fig. 5E and F). Adults/copepodids and nauplii of *P. inopinus* remained at low densities in March (<0.1 ind. L$^{-1}$) before increasing to relatively high densities in April (>0.3 ind. L$^{-1}$).

Within the distributional range of *S. sinensis*, naupli occurred on average downstream from adults and copepodids irrespective of the fortnightly tidal cycle (Fig. 6A). The CHD of naupliar *S. sinensis* was always located downstream of the CHD of adult/copepodid *S. sinensis* ($P < 0.05$, the Wilcoxon signed-rank test). There was no clear pattern of stage-specific horizontal distribution for *P. inopinus* during the sampling period (Fig. 6B).

Responses to discharge fluctuations

In summer 2005, the daily freshwater discharge remained below 100 m$^3$ s$^{-1}$ until late June with a minimum (<20 m$^3$ s$^{-1}$) from 17 to 25 June (Fig. 7A). The discharge increased in early July and caused a large flood from 9 to 14 July with a peak of 1768 m$^3$ s$^{-1}$ on 11 July. Following this flood, the discharge settled down to the previous level by late July. The tidal range changed between ca. 150 and 400 cm during the sampling period. Temperature increased from 24 to 28°C before decreasing to <24°C during the flood (not shown). After the flood, temperature increased again and remained above 26°C. The ETM developed at low salinities (<10) before the flood, whereas during the flood both salinity and turbidity decreased markedly (Fig. 7B). The well-developed ETM was observed again at the spring tide of early August.

The density of *S. sinensis* varied drastically in response to discharge fluctuations as well as the fortnightly tidal cycle (Fig. 7C and D). Although the density peak of
Fig. 2. Seasonal changes in the densities of *S. sinensis* and *P. inopinus* with relation to environmental parameters observed in the Chikugo River estuary during 2005 and 2006. Environmental parameters include freshwater discharge and temperature (A) and turbidity and salinity (B). Adult/copepodid and naupliar stages are shown separately for *S. sinensis* (C and D) and *P. inopinus* (E and F). The isopleth charts are overlaid with salinity contour lines. Filled triangles represent the locations of the observation stations.
Fig. 3. Bar charts showing the stage-specific horizontal distribution of *S. sinensis* along the Chikugo River estuary from May 2005 to December 2006. Four life-cycle stages (nauplius, copepodid, adult male and adult female) are represented by different patterns. The density of *S. sinensis* was converted into relative abundance on each sampling date (see the “Method” section). Absolute values of maximum density (ind. L⁻¹) are also shown on the charts. Vertical lines represent locations of salinities 1 and 10 (average through the water column).
Fig. 4. Bar charts showing the stage-specific horizontal distribution of *P. inopinus* along the Chikugo River estuary from May 2005 to December 2006. Five life-cycle stages (nauplius, copepodid, adult male, non-gravid female and gravid female) are represented by different patterns. Maximum densities (ind. L⁻¹) are shown on the charts for each sampling date. See Fig. 3 for details.
S. sinensis always corresponded to salinity 1, the location moved upstream under small discharges and at spring tides. Adult/copepodid S. sinensis decreased in density gradually under minimum discharge levels in June and disappeared during the flood in middle July. The density of adult/copepodid S. sinensis increased to > 20 ind. L\(^{-1}\) within 2 weeks after the flood. The density of naupliar S. sinensis was lower than 1 ind. L\(^{-1}\) before and during the flood, but reached > 20 ind. L\(^{-1}\) one week after the flood. In relation to the fortnightly tidal cycle, the density of naupliar S. sinensis showed maxima during spring tides and minima during neap tides. In contrast, the density of P. inopinus responded primarily to the flood (Fig. 7E and F). Before the flood,
adult/copepodid *P. inopinus* remained at 10 ind. L$^{-1}$, whereas naupliar *P. inopinus* concentrated (up to 40 ind. L$^{-1}$) particularly close to the upper limit of the estuary. The density of adult/copepodid *P. inopinus* decreased to <1 ind. L$^{-1}$ during the flood and increased dramatically to >40 ind. L$^{-1}$ after the flood.

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**Fig. 6.** Bar charts showing the stage-specific horizontal distribution of *S. sinensis* (A) and *P. inopinus* (B) along the Chikugo River estuary in spring 2005. Neap, medium and spring tides correspond to <200, 200–400 and >400 cm in the tidal range, respectively. Maximum densities (ind. L$^{-1}$) are shown on the charts for each sampling date. See Fig. 3 for details.
Similar temporal changes in density were observed in naupliar *P. inopinus* during and after the flood. Before the flood, the *S. sinensis* population comprised primarily adults, whereas nauplii and copepodids dominated after the flood (Fig. 8A). During the sampling period in summer 2005, there was no consistent pattern of stage-specific horizontal distribution of *S. sinensis*. As for *P. inopinus*, nauplii and copepodids always dominated (Fig. 8B). Nauplii of *P. inopinus* generally occurred upstream from adults and copepodids before the flood, whereas the opposite pattern of stage-specific horizontal distribution was observed under large freshwater discharges from 4 to 19 July. *Pseudodiaptomus inopinus*, on the other hand, did not show a consistent pattern of stage-specific horizontal distribution afterward.

**Salinity tolerance of *S. sinensis***

In the direct transfer experiment, the survival rate of *S. sinensis* dropped to 12% in salinity 30 within the first 4 h, whereas it remained above 70% in the other treatments until 36 h after transfer (Fig. 9A). A gradual decrease in the survival rate was observed in salinity 20 beyond 36 h after transfer. At the end of the experiment (i.e. 72 h after transfer), the final survival rate was generally higher at lower salinities: 82, 86, 61, 67, 74, 49 and 1% in salinity 0, 1, 5, 10, 15, 20 and 30, respectively. In the stepwise transfer experiment, the survival rate remained above 90% in the three treatments within the first 12 h, when salinity was increased gradually to the three target values (i.e. 15, 20 and 25). The survival rate in salinity 15, 20 and 25 decreased slowly to 92, 71 and 57% within the first 36 h and to 73, 43 and 16% at the end of the experiment, respectively.

**DISCUSSION**

**Free-spawning and egg-carrying strategies**

The different patterns of stage-specific horizontal distribution between *S. sinensis* and *P. inopinus* are primarily attributable to their respective spawning strategies. Nauplii of free-spawning *S. sinensis* were observed consistently downstream from copepodids and adults throughout the year. The same pattern of stage-specific horizontal distribution was observed in the *S. sinensis* population irrespective of the fortnightly and semi-diurnal tidal cycles (cf. Ueda *et al.*, 2010). This consistency could be explained by the vulnerability of *S. sinensis* eggs to downstream dispersal until hatching. Although no detailed information is available for eggs of *S. sinensis*, the congener *S. tenellus* spawns free eggs that require 1.5–5.0 days for hatching at 10–20°C (Kimoto *et al.*, 1986). In the Chikugo River estuary, eggs of *S. sinensis* are likely suspended by strong tidal currents and dispersed downstream at least for 1 day. The residual current flows seaward at 9.6 km day$^{-1}$ (11.1 cm s$^{-1}$) along the estuary, as the estuary is ≈300 m in width and 3 m in depth with a freshwater discharge of 100 m$^3$ s$^{-1}$ on average. Given that the tidal reach stretches over 23 km, eggs of *S. sinensis* would be vulnerable to washout from the estuary especially during the cold season, when eggs develop slowly.
Fig. 8. Bar charts showing the stage-specific horizontal distribution of *S. sinensis* (A) and *P. inopinus* (B) along the Chikugo River estuary in summer 2005. Crosses represent stations where sampling was not conducted. Maximum densities (ind. L$^{-1}$) are shown on the charts for each sampling date. See Fig. 3 for details.
Moreover, the naupliar stages of *S. sinensis* continue for 5 days at 25°C (Kuwatani, 2006). Even though tidal vertical migration can reduce the downstream transport of *S. sinensis* during the naupliar stages (Ueda et al., 2010), nauplii of *S. sinensis* are considered to be very subject to washout from the estuary due to their low swimming ability and their inevitable downstream dispersal before hatching.

Gravid females and nauplii of egg-carrying *P. inopinus* were usually distributed upstream from the other life-cycle stages, indicating that gravid females release nauplii upstream. Although the egg-carrying strategy of *P. inopinus* minimizes the washout of eggs from the estuary, nauplii of *P. inopinus* occurred more downstream than copepods and adults in winter. Given the low swimming ability of nauplii, this finding could be explained by downstream dispersal during the extended duration of the naupliar stages at low temperatures. As the duration of the naupliar stages of *P. inopinus* is 3 days at 25°C (Kuwatani, 2006), the critical period for the position maintenance of *P. inopinus* likely continues for longer than 3 days in winter. Without tidal vertical migration during the naupliar stages, the residual current would transport nauplii of *P. inopinus* to the sea within 3 days. Consequently, the egg-carrying strategy and tidal vertical migration are together considered to be the central mechanisms for the position maintenance of *P. inopinus* in estuaries.

### Specificity for the macrotidal environment

During the frequent sampling of spring 2005, the influence of the fortnightly tidal cycle was more evident on the density of *S. sinensis* than on that of *P. inopinus*. Higher densities of *S. sinensis* observed during spring tides could be explained by the dense concentration of *S. sinensis* in the well-developed ETM, where plant detritus is especially abundant (Suzuki et al., 2007). Given that plant detritus is an energy source for coastal and estuarine copepods (Heinle and Flemer, 1975; Heinle et al., 1977; Roman, 1984), the well-developed ETM potentially provides *S. sinensis* with abundant food (Suzuki et al., 2012). During spring tides, *S. sinensis* may concentrate in the well-developed ETM and swim up into the water column to feed on plant detritus. In contrast, *S. sinensis* may be distributed closer to the bottom along the estuary during neap tides. The anticipated changes in distribution could be caused by not only active (i.e. tidal vertical migration; Ueda et al., 2010) but also passive processes (i.e. particle trapping; Castel and Veiga, 1990; Roman et al., 2001). Aside from changes in distribution, higher densities of naupliar *S. sinensis* possibly also translate into increases in the number of nauplii in the population, since the naupliar stages continue for only a few days (Kuwatani, 2006). During spring tides, better particle trapping combined with greater food availability has the high potential to increase the survival rate of naupliar *S. sinensis* in the well-developed ETM. Though the spatial distribution of

![Fig. 9. Survival rates of *S. sinensis* at different salinities during the direct (A) and stepwise (B) transfer experiments. Symbols represent mean survival rates between duplicates.](https://academic.oup.com/plankt/article-abstract/34/12/1042/1418945)
S. sinensis remains to be studied (cf. Shoji et al., 2006), the observed fortnightly cycle of density strongly suggests the ecological specificity of S. sinensis for the macrotidal environment.

The timing of the seasonal increase in the density of P. inopinus corresponded to water warming and the phytoplankton bloom in the Chikugo River estuary (i.e. April–May; Suzuki et al., 2007, 2012). Higher water temperature and higher food availability probably accelerated the reproduction and survival rates of P. inopinus. In contrast to S. sinensis, lack of ecological specificity for the macrotidal environment was indicated by less spatiotemporal correspondence between P. inopinus distribution and the ETM. Irrespective of the tidal cycles, P. inopinus ceases tidal vertical migration at night to stay in the upper water column (Ueda et al., 2010). As the risk of visual predation decreases at night, P. inopinus possibly gives the highest priority to feeding on phytoplankton close to the water surface. These ecological characteristics suggest that P. inopinus depends on less plant detritus but more on phytoplankton especially during the warm season. As P. inopinus often dominates oligotrophic estuaries where both plant detritus and phytoplankton seem scarce (Sakaguchi et al., 2011), this species might feed on other food items such as attached algae as well. Consequently, P. inopinus is considered not specialized to live in the macrotidal environment but generalized to live in various estuarine conditions.

**Vulnerability to discharge fluctuations**

The high vulnerability of S. sinensis to discharge fluctuations was revealed by the frequent sampling in summer 2005. The density of S. sinensis, nauplii in particular, decreased in June under the smallest monthly discharge during the last decade (1999–2005). A gradual increase in salinity possibly reduced growth and survival rates of S. sinensis during the drought period. This scenario is partly supported by lower survival rates of S. sinensis observed at higher experimental salinities. Although long-term effects of salinity remain to be studied especially in the early life-cycle stages, S. sinensis possibly requires lower salinities for its optimum growth and survival than other estuarine copepods (cf. Roddie et al., 1984; Kimoto et al., 1986). In addition, the drought may have driven the S. sinensis population into the upper limit of the estuary, leading to significant cannibalism by adults on nauplii (cf. Hada and Uye, 1991). After the drought period, S. sinensis disappeared from the estuary owing to a large flood. The free-spawning strategy of S. sinensis is disadvantageous to successful reproduction under elevated freshwater discharge levels. During large floods, naupliar and copepodid S. sinensis are very subject to washout from the estuary, although adult S. sinensis is able to stay in the bottom of tidal channels (Ueda et al., 2004). Although S. sinensis does not necessarily undergo population loss in the case of a single flood (Ueda et al., 2004), the delayed recovery of population observed in summer 2006 indicates the high vulnerability of S. sinensis to repetitive floods. In summary, the population stability of S. sinensis is likely to be threatened by both droughts and floods.

Egg-carrying P. inopinus showed relatively low vulnerability to discharge fluctuations. The density of P. inopinus varied little during the drought period and recovered quickly after the large flood in 2005. The successful reproduction of P. inopinus was suggested by the continuous occurrence of each life-cycle stages, even though nauplii were distributed more downstream under large freshwater discharges. A quick recovery of the P. inopinus population was observed after repetitive floods in summer 2006. In addition to the egg-carrying strategy, high temperature possibly accelerated the reproduction of the summer copepod P. inopinus (Suzuki et al., submitted). Pseudodiaptomus inopinus is known as a copepod that survived in a desalinated lake in Japan (Kawabata and Defaye, 1994) and settled in estuaries along the west coast of North America (Cordell et al., 1992, 2007). Although optimum ranges of environmental variables for P. inopinus remain unknown, P. inopinus is considered highly tolerant of a variety of environmental disturbances.

**CONCLUSIONS**

The present study contrasts the specific life history of S. sinensis with the flexible life history of P. inopinus by comparing the dynamics of stage-specific horizontal distribution along the macrotidal Chikugo River estuary. The reproduction of P. inopinus successfully tolerates large freshwater discharges by its egg-carrying strategy. The frequent occurrence of P. inopinus in Japan is probably attributable to its flexibility toward prevailing conditions in many estuaries. In contrast, the free-spawning strategy of S. sinensis is less likely to achieve successful reproduction under large freshwater discharges. Given the vulnerability of S. sinensis to droughts as well as floods, this species potentially prefers large-scale estuaries that could buffer discharge fluctuations. More importantly, S. sinensis consistently synchronizes its behavioral rhythms with the semi-diurnal and fortnightly tidal cycles (cf. Ueda et al., 2010). Feeding on plant detritus and tolerance for high turbidities could also characterize the ecology of S. sinensis (cf. Suzuki et al., submitted). As the free-spawning strategy is closely associated with
higher weight-specific fecundities and faster growth rates (Kiørboe and Sabatini, 1995), *S. sinensis* has the high potential to increase its population quickly under favorable conditions, i.e. well-developed ETM. Such ecological specializations should be the main reason why *S. sinensis* dominates only in macrotidal estuaries.

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