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Distribution of the invasive hard clam, *Mercenaria mercenaria*, in the intertidal zone of Sanbanze in the inner part of Tokyo Bay

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Abstract： We investigated the distribution of the invasive hard clam, *Mercenaria mercenaria,* and other shellfish in the intertidal area of the tidal flats of Sanbanze in Tokyo Bay. We found that the distribution of *M. mercenaria* was negatively correlated with silt-clay content. Almost all the shellfish collected belonged to 1 of the following 3 species: *Ruditapes philippinarum* (41.6%), *M. mercenaria* (27.7%), or *Phacosoma japonicum* (24.1%). Within the study site, few sampling plots were completely occupied. Therefore, *M. mercenaria* did not strongly outcompete other species in this study site.

Keywords：*distribution, invasive species, Mercenaria mercenaria, Tokyo Bay*

1. Introduction

Invasive marine species often have a serious influence on native ecosystems and local fisheries. The invasive marine hard clam, *Mercenaria mercenaria*, was introduced into Tokyo Bay in Japan in the 1990s (KUROZUMI and OKAMOTO, 2002; NISHIMURA, 2005; HIWATARI and KOHATA, 2005; HIWATARI *et al.,* 2006; SUGIHARA *et al.,* 2012). This species is native to the east coast of North America, where it forms a very important fishery resource. In Tokyo Bay, *M. mercenaria* is distributed in highly eutrophic coastal areas and has not yet been observed in other areas in Japan (HIWATARI *et al.,* 2006). SUGIHARA *et al.* (2012) reported that the *M. mercenaria* populations in Tokyo Bay might be native to the Florida Peninsula. Further, it has been shown that *M. mercenaria* has excellent filter-feeding ability (TENORE *et al.,* 1973) and tolerance for environmental changes, such as hypoxic and low salinity conditions (HIWATARI and KOHATA, 2005). Such advantages have probably enabled this species to survive after its introduction into Tokyo Bay. It has been assumed that this clam may compete with native species, including commercially important species such as *Ruditapes philippinarum* (HIWATARI and KOHATA, 2005). Therefore, it is necessary to clarify the impact of this invasive clam on native ecosystems.

Only limited information is available about the environmental factors that affect the distribution of *M. mercenaria* in Japan. Obtaining

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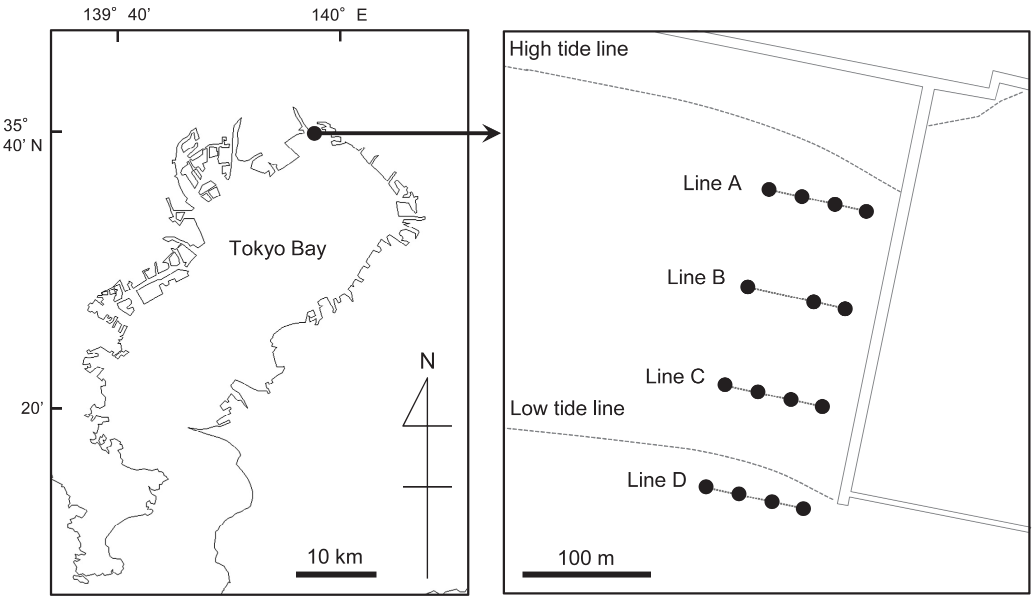

Fig. 1. The study site in Tokyo Bay and the location of the sampling plots. Sampling lines (A-D) were 80 m apart, and the sampling plots were created at 20-m intervals along the lines. The straight solid lines represent a dike.

more information about the distribution of both native and invasive clams should be the first step in evaluating the impact of the invasive clam on native ecosystems. This study was conducted to provide fundamental information for further ecological studies. We examined the tidal flats of Sanbanze in the inner part of Tokyo Bay in late summer, during which a large number of *M. mercenaria* can be found (NISHIMURA, 2005).

2. Materials and methods

Sanbanze is located in the inner part of Tokyo Bay and is one of the largest tidal flats in the bay. Most of the area has a muddy-sand bottom (ACHIARI and SAKAI, 2007). The survey was conducted at low tide, during which the 2.69-km^2 tidal flats were maximally exposed. (CHIBA PREFECTURE, 2012). It is difficult to access the tidal flats except via Funabashi Sanbanze Kaihin Park (35°40′N, 139°58′E). Therefore, the study site was prepared within the park boundary.

A spatial survey was conducted at low tide, between 10:00 a.m. and 1:00 p.m. on September 5, 2009, when the tidal flats were the most exposed (the lowest tide was at 11:18 a.m.). Four parallel sampling lines were set at 80-m intervals. The first line was set slightly below the high tide line, and the last line was set below the low tide line (Fig. 1). Sampling plots were created at 20-m intervals along each 80-m line by installing a 150-cm bar for determining the water depth at high tide. To analyze mollusk assemblages, sediment was collected using 30 × 30-cm quadrats at each sampling plot. Sediment collected 0 − 5 cm from the surface was sieved using a 2-mm mesh, and the animals obtained were fixed in 10% formalin immediately after sampling. To analyze environmental conditions, sediment samples were collected near each mollusk-sampling site by using 10 × 30-cm quadrats. Sediment temperature was measured at 3-cm depth by using a stick thermometer. A 0.8-cm-diameter corer was used to collect a 2-ml sediment core from 0 − 3 cm below the surface. This sediment was used to analyze water content. A 2.7-cm-diameter corer was used to collect a 100-ml sediment core from 0 − 3 cm below the surface. This sediment was used

Table 1. Density of animal species collected along sampling lines in Sanbanze (mean±standard deviation [SD]). Each sampling line contained 3－4 sampling plots.

Class	Family	Species	Density (individuals/m^2)				Number of plots where present
			Line A	Line B	Line C	Line D	
Bivalvia	Mactridae	*Mactra veneriformis*		3.7±6.4		8.3±10.6	3
	Tellinidae	*Macoma incongrua*			2.8±5.6		1
	Solenidae	*Solen strictus*				8.3±10.6	2
	Veneridae	***Mercenaria mercenaria***	**2.8±5.6**	**44.4±50.9**	**41.7±16.7**	**27.8±11.1**	**11**
		Phacosoma japonicum		3.7±6.4	11.1±15.7	77.8±76.4	7
		Ruditapes philippinarum		85.2±78.8	55.6±27.2	38.9±14.3	10
		Cyclina sinensis			2.8±5.6		1
Polychaeta		Polychaeta sp.	47.2±38.9	44.4±50.9	2.8±5.6	13.9±16.7	8

to determine sediment characteristics.

Animals were sorted into species or taxa. They were then counted and their density was calculated. To analyze sediment water content, samples were weighed before and after drying at 105℃ for 5 h, and the water content was calculated from the difference in mass. To analyze the sediment characteristics, samples were treated with 30% hydrogen peroxide to remove organic compounds and then dried at 60℃ for 96 h. The composition of sediment grain sizes was determined using a series of sieves with 2-, 1-, 0.5-, 0.25-, 0.125-, and 0.063-mm meshes, and the median grain size and silt-clay content were measured.

For homogeneity of variance and normality, all numerical data were log-transformed and percent data were arcsine transformed, as per the study by ZAR (1984). Differences between the means of environmental factors along each sampling line were compared using one-way analysis of variance (ANOVA) and Bonferroni's method. Pearson's product-moment correlation coefficient was calculated between major bivalve species. The correlation coefficient was tested using a t test. Stepwise multiple regression analysis was performed to examine correlations between the density of *M. mercenaria* and environmental conditions. Differences were considered significant if the associated p-value was less than 0.05.

3. Results and discussion

Seven bivalve species were found in the study site (Table 1). A vast majority of these bivalves were 1 of 3 species of filter feeders: Of the total number of bivalves found, 41.6% were *R. philippinarum*, 27.7% were *M. mercenaria*, and 24.1% were *Phacosoma japonicum*. *M. mercenaria* was found in a greater number of sampling plots (11 sampling plots) than any of the other species. Thus, the invasive clam, *M. mercenaria*, was one of the dominant species in the study site. The distribution of *M. mercenaria* was similar to that of *R. philippinarum*, and few sampling plots were entirely occupied by *M. mercenaria*. NISHIMURA (2005) reported that, although the number of individuals varied between seasons and places, 80% of all samples acquired in 2002 from Chiba Port in Tokyo Bay were *M. mercenaria*. We observed no such extreme dominance, and there was no negative correlation between the density of *M. mercenaria* and that of the other 2 major species, namely, *R. philippinarum* (R= 0.59; $P < 0.05$) and *P. japonicum* (R=0.17; $P >$ 0.05). These results imply that the presence of *M. mercenaria* did not exclude other species from the study site. Therefore, there was no evidence that *M. mercenaria* strongly outcompetes other species in the study site.

Sampling lines A－C in this study were placed in the intertidal zone, and line D was in

Table 2. Environmental conditions, species richness, and total density of collected animals along sampling lines in Sanbanze (mean ± standard deviation [SD]).

	Line A	Line B	Line C	Line D
Water depth at high tide (cm)	62.5±5.0[a]	95.0±10.0[b]	118.8±4.8[c]	NA*
Emersion (h)	7.1±0.3[a]	5.3±0.6[b]	4.2±0.3[c]	0.0±0.0[d]
Temperature (℃)	32.1±0.3[a]	30.3±1.2[b]	29.4±0.5[b]	26.1±0.8[c]
Water content (%)	23.0±2.0[a]	20.5±6.2[a]	21.1±2.6[a]	27.4±0.9[a]
Median grain size (μm)	78.3±6.5[a]	103.7±12.0[b]	88.4±6.0[ab]	82.4±4.2[a]
Silt-clay contents (%)	7.8±4.5[a]	0.3±0.2[b]	0.4±0.2[b]	0.7±0.3[b]
Species richness	1.0±0.0[a]	2.7±1.5[ab]	3.3±1.0[b]	4.5±0.6[b]
Density (individuals/m^2)	50.0±34.5[a]	136.1±107.5[b]	116.7±32.1[b]	175.0±80.8[b]

Different letters within rows indicate significant differences (n = 3−4; $P < 0.05$).
* The water depth at high tide on line D was deeper than 150 cm, which is the length of the bar used to measure water depth in this study.

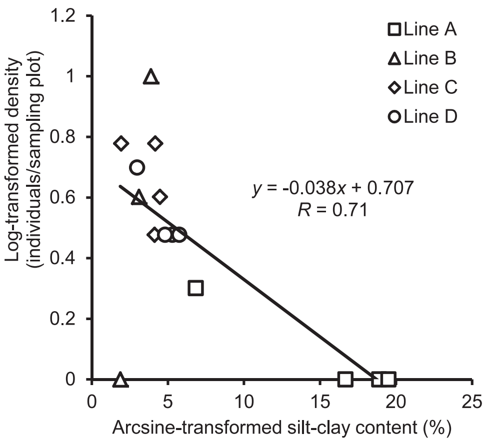

Fig. 2. Correlation between silt-clay content and the density of *M. mercenaria* in sampling plots (30 × 30 cm^2) located in Sanbanze mudflat in Tokyo Bay.

the subtidal zone. Although the water depth at high tide and emersion differed between lines B and C, the environmental conditions were similar along these 2 lines (Table 2). The following formula was obtained using stepwise multiple regression analysis:

$$y = -0.038x + 0.707\ (r = 0.71),$$

where y is the density of *M. mercenaria* and x is silt-clay content. The distribution of *M. mercenaria* in this study was negatively correlated with silt-clay content (Fig. 2). No other correlations were observed between environmental conditions and the abundance of *M. mercenaria*. The negative correlation between sediment silt-clay content and density of *M. mercenaria* was conspicuous along sampling line A (Table 1, 2). It has been reported that the presence of 44 mg/L of silt in the water decreases the growth rate of *M. mercenaria* (Bricelj *et al.*, 1984). Such changes in growth rate may depend on the efficiency of selection between nutritious substances and suspended sediments in the water (Bricelj and Malouf, 1984). It may be that the high silt-clay content observed in all the sediments was caused by a large quantity of suspended sediments in the water at high tide. The absence of *M. mercenaria* at line A may therefore be due to high silt-clay content of the water in that area. However, the silt-clay content of the sediments was approximately 8% or less along all sampling lines, including line A, which had the fewest individuals of this species. Contrary to the results of our study, Nishi *et al.* (2008) reported that *M. mercenaria* was found in several areas of Yokohama Port, which opens into in Tokyo Bay, where the silt-clay content was more than 15.7% and the substratum had low oxidation-reduction potential. Although our study results do not indicate the reason for this difference in silt-clay content between Sanbanze and Yokohama Port, it is assumed to be one of the reasons that the study site used by NISHI *et al.* (2008) and that used in our study were geomorphologically different. The study site used by NISHI *et al.* (2008) was located in a canal, whereas in our study, the site was an intertidal zone in a tidal flat. Sediments are well-raised from surf zone to swash zone (SHUTO, 1988), and the intertidal zone in this study was within these zones. Therefore, the study site used by NISHI *et al.* (2008) is assumed to be less affected by waves from the sea than the site used for our study. The negative correlation we observed between silt-clay content and the density of *M. mercenaria* may result from geomorphological features that cause a relatively large amount of suspended sediment in areas with high silt-clay content. In this study, we did not directly test this hypothesis. Therefore, a more detailed survey and additional experiments are necessary, even though it appeared that silt and clay were among the major factors that affected the distribution of *M. mercenaria* in the study site.

In this study, we investigated the distribution of *M. mercenaria* in a part of Sanbanze. *M. mercenaria* is abundant in Sanbanze, as it is in other places in Tokyo Bay (NISHIMURA, 2005; NISHI *et al.,* 2008). Our study data should help to clarify the distribution of *M. mercenaria* in Tokyo Bay. Furthermore, Sanbanze faces land subsidence; the area of the tidal flats has decreased by over 50% over the past 3 years, primarily because of the Great East Japan Earthquake (CHIBA PREFECTURE, 2012). Therefore, this study, which describes the biological distribution of bivalves before the Great East Japan Earthquake, can also be used as a baseline survey for tracing ecological and geographical changes after the earthquake. Further research is required to obtain more details about the distribution of the invasive species, *M. mercenaria.*

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