# Observations of eleven *Pseudo-nitzschia* species in Tokyo Bay, Japan

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Abstract: Appearance of *Pseudo-nitzschia* species was observed in samples collected biweekly in Tokyo Bay during April 2008—August 2009. Species identification was based on morphological characteristics observed under light microscope (LM) and transmission electron microscope (TEM). This is the first time that a periodic survey of this genus at species level was done in the area. The following 11 species were found: *P. americana, P. brasiliana, P. caciantha, P. caliantha, P. galaxiae, P. fraudulenta, P. multistriata, P. multiseries, P. pseudodelicatissima and P. pungens.* The presence of five species, *P. americana, P. brasiliana, P. caciantha, P. caliantha* and *P. galaxiae* is reported for the first time in Tokyo Bay.

Keywords: diatom, Pseudo-nitzschia species, Tokyo Bay

#### Introduction

Currently there are 32 species known in the genus *Pseudo-nitzschia* (AMATO and MONT-RESOR, 2008; CLEVE, 1897b; HASLE, 1965; HASLE, 1993; LUNDHOLM *et al.*, 2002a; LUNDHOLM and MOESTRUP, 2002; LUNDHOLM *et al.*, 2003; PRIISHOLM *et al.*, 2002; SKOV *et al.*, 1999; TAKANO, 1995). The type species of the genus, *P. seriata* was renamed from *Nitzschia seriata* CLEVE by PERAGALLO in (1897–1908). But it was only in 1993 that half, *i.e.*, 14 of the species were transferred from *Nitzschia* to *Pseudo-nitzschia* (HASLE, 1993).

Studies on this genus are numerous particularly in Europe and North America. In contrast studies of *Pseudo-nitzschia* in Southeast Asia remain to be scant. Notable are taxonomic works by LARSEN and NGUYEN (2004) that identified *Pseudo-nitzschia* species in Vietnam waters and PRIISHOLM et al. (2002) that described a new species, P. micropora found in Thailand. TAKANO and KUROKI (1977) found seven species of Pseudo-nitzschia, still under *Nitzschia* that time, in various coastal waters of Japan. These are P. delicatissima (N. actydrophila), P. fraudulenta, P. multiseries (N. pungens forma multiseries), P. pungens, P. pseudodelicatissima, P. subfraudulenta, and P. turgidula. Regular surveys of phytoplankton in Tokyo Bay have reported the presence of Pseudo-nitzschia, but their identification and enumeration have been done only at the genus level. As of this year 2010, there is no clear assessment of Pseudo-nitzschia species composition in Tokyo Bay. Thus the purpose of this study is to elucidate species composition of the genus Pseudo-nitzschia in Tokyo Bay. Descriptions and documentations put forth here are for morphometric characterizations of local populations and baseline data for future investigations of toxic or newly discovered taxa to further ecological studies of Pseudo-nitzschia in the area.

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Fig 1. Map of sampling stations. (http://gmt.soest. hawaii.edu/)

#### Material and Methods Field sampling

Samples collected by bucket from Odaiba station (35° 36'55"N, 139° 46'32"E) or by Van Dorn Water Sampler from Stations 3 (35° 33' 16"N, 139° 54'29"E), 8 (35° 25'24"N, 139° 51'42" E), 15 (35° 36'24"N, 139° 57'48"E) and Funabashi (35° 38'43"N, 139° 59'19"E) in Tokyo Bay (Fig. 1) during April 2008—August 2009 were assessed for presence of *Pseudo-nitzschia*. One liter of sample was preserved with neutralized formaldehyde at a final concentration of approximately 1 %. Another liter of sample remained unfixed for culture strains establishment.

#### Culture establishment

Pseudo-nitzschia cells were isolated by picking up single chains by capillary method (THRONDSEN, 1978). These were washed at least three times with sterilized sea water under a compound microscope and placed in 24-well plates filled with f/2 media (GUILLARD, 1975). When cells are numerous enough, these were then transferred to screw capped test tubes with the same media. The culture conditions were as follows: temperature 15°C (and/or 20 °C), light intensity 100  $\mu$  mol photons m<sup>-2</sup>s<sup>-1</sup> (daylight fluorescent lamps), salinity at 30 and 12L: 12D photoperiod.

# Species observation and species confirmation by TEM

Light microscope (LM) photomicrographs were taken of live cells from the field, in culture and preserved cleaned valves devoid of organic material. Organic material was removed from the cells as follows:

Aliquots of fixed Pseudo-nitzschia cells from field and live cells from cultures were transferred to 15 ml centrifuge tubes. The live cells were fixed with neutralized formaldehyde. Cells were washed with milliQ water at least three times prior to acid washing. Centrifugations were done at 35,000 rpm for 10 minutes. Organic material was dissolved following LUNDHOLM et al. (2002b). The condensed samples (1 ml) were treated with 0.2 ml of 30% sulfuric acid and 0.5-1 ml of saturated potassium permanganate (KMnO<sub>4</sub>) solution and were left for 24 hours. And then 1–2 ml saturated oxalic acid  $((COOH)_2)$  was added until the purple color turned out before the valves were washed several times with distilled water to wash out the acids. Drops of cleaned cells were mounted on formvar coated copper grids and allowed to dry. These were then observed under a JEM 2000EX JEOL Transmission Electron Microscope (TEM). General shape of a cell, its length and width, number of poroid rows, number of poroid in  $1 \,\mu$  m, number of interstriae in  $10 \,\mu$ m, number of fibulae in  $10 \,\mu$  m and inner poroid features were measured and noted. Pseudonitzschia species were identified the following several manuals and recent articles (HASLE and SYVERTSEN, 1997; SKOV et al., 1999; LUNDHOLM etal., 2002a and 2003; Amato and Montresor, 2008).

#### Results

#### **Culture strains**

Ten species out of the 11 species of *Pseudo-nitzschia* found in Tokyo Bay from sampling period starting April 2008—May 2009 were established into culture (Table 1). These were the following: *P. americana* (4), *P. brasiliana* (3), *P. caciantha* (1), *P. calliantha* (6), *P. delicatissima* (1), *P. fraudulenta* (6), *P. galaxiae* (1), *P. multistriata* (2), *P. multiseries* (2) and *P. pungens* (7). Numbers in parenthesis are the number of culture strains, all non-axenic, for



Figs. 2-11. LM micrographs of 10 species.

Fig. 2. *P. americana*, Fig. 3. *P. brasiliana*, Fig. 4. *P. galaxiae*, Fig. 5. *P. caciantha*, Fig. 6. *P. calliantha*, Fig. 7. *P. delicatissima*, Fig. 8. *P. fraudulenta*, Fig. 9. *P. multistriata*, Fig. 10. *P. multiseries*, Fig. 11. *P. pungens*. Figs. 2, 3, 9: LM micrographs of live samples; Figs. 4–8, 10: LM micrographs of cleaned valves of cells in culture. Fig. 11. DIC micrograph of cell in culture. Scale bar =  $10 \mu$  m.

each species. These were useful for morphometric measurements of species that rarely occurred e.g.: *P. delicatissima* and *P. galaxiae* and for tidy TEM photomicrographs. Dates of isolation provide information on the days a particular species appeared in the bay during the sampling period.

#### **Species Descriptions**

Descriptions of each species in alphabetical order follow herein. A summary of morphological characteristics of these species in Tokyo Bay is shown in Table 2. Except for *P. delicatis*- sima and *P. galaxiae* morphometric measurements of which are from newly established cultures, all measurements are from field samples in Tokyo Bay.

#### Pseudo-nitzschia americana (HASLE) FRYXELL in HASLE

Figs. 2, 12 LUNDHOLM *et al.*, 2002a. pp. 483–484, Figs. 1–20. ORLOVA and SHEVCHENKO, 2002, pp. 336–339, Fig. 1. KACZMARSKA *et al.*, 2005, pp. 4–7, Figs. 2–13, Table 1.

Synonym: Nitzschia americana HASLE

Name	Strain No.	Origin	Date of Isolation	Collector/Isolator
P. americana	TBaM01	Tokyo Bay, JP	16 March 2009	L. Yap-Dejeto
P. americana	TBaA01	Tokyo Bay, JP	04 April 2009	L. Yap-Dejeto
P. americana	TBaA02	Tokyo Bay, JP	04 April 2009	L. Yap-Dejeto
P. americana	TBaA03	Tokyo Bay, JP	04 April 2009	L. Yap-Dejeto
P. brasiliana	TBbA01	Tokyo Bay, JP	09 April 2008	L. Yap-Dejeto
P. brasiliana	TBbS01	Tokyo Bay, JP	24 September 2008	L. Yap-Dejeto
P. brasiliana	TBbS02	Tokyo Bay, JP	24 September 2008	L. Yap-Dejeto
P. caciantha	TBcc03	Tokyo Bay, JP	18 November 2008	L. Yap-Dejeto
P. calliantha	TBcl1101	Tokyo Bay, JP	11 May 2009	L. Yap-Dejeto
P. calliantha	TBcl1102	Tokyo Bay, JP	11 May 2009	L. Yap-Dejeto
P. calliantha	TBcl2502	Tokyo Bay, JP	25 May 2009	L. Yap-Dejeto
P. calliantha	TBcl2503	Tokyo Bay, JP	25 May 2009	L. Yap-Dejeto
P. calliantha	TBcl2507	Tokyo Bay, JP	25 May 2009	L. Yap-Dejeto
P. calliantha	TBc12508	Tokyo Bay, JP	25 May 2009	L. Yap-Dejeto
P. delicatissima	TBd05	Tokyo Bay, JP	07 July 2008	L. Yap-Dejeto
P. fraudulenta	TBfJ02	Tokyo Bay, JP	07 July 2008	L. Yap-Dejeto
P. fraudulenta	TBf1101	Tokyo Bay, JP	11 May 2009	L. Yap-Dejeto
P. fraudulenta	TBf1102	Tokyo Bay, JP	11 May 2009	L. Yap-Dejeto
P. fraudulenta	TBf2504	Tokyo Bay, JP	25 May 2009	L. Yap-Dejeto
P. fraudulenta	TBf2505	Tokyo Bay, JP	25 May 2009	L. Yap-Dejeto
P. fraudulenta	TBf2506	Tokyo Bay, JP	25 May 2009	L. Yap-Dejeto
P. galaxiae	TBg01	Tokyo Bay, JP	25 May 2009	L. Yap-Dejeto
P. multiseries	Kums	Kushimoto, JP	5 March 2008	T.OMURA/L. Yap-Dejeto
P. multiseries	TBms01	Tokyo Bay, JP	15 June 2008	L. Yap-Dejeto
P. multistriata	TBma01	Tokyo Bay, JP	21 October 2008	L. Yap-Dejeto
P. multistriata	TBma02	Tokyo Bay, JP	21 October 2008	L. Yap-Dejeto
P. pungens	TBpuA00	Tokyo Bay, JP	15 April 2008	L. Yap-Dejeto
P. pungens	TBpuA01	Tokyo Bay, JP	15 April 2008	L. Yap-Dejeto
P. pungens	TBpuA03	Tokyo Bay, JP	15 April 2008	L. Yap-Dejeto
P. pungens	TBpuJ01	Tokyo Bay, JP	07 July 2008	L. Yap-Dejeto
P. pungens	TBpuJ02	Tokyo Bay, JP	07 July 2008	L. Yap-Dejeto
P. pungens	TBpuJ03	Tokyo Bay, JP	07 July 2008	L. Yap-Dejeto
P. pungens	TBpu03	Tokyo Bay, JP	16 March 2009	L. Yap-Dejeto

Table 1. Strains of *Pseudo-nitzschia* and their corresponding origin and dates of isolation.

HASLE, 1964, pp. 41–44, Pl. 1, Fig. 4, Pl. 14, Figs. 13–19, Pl. 15, Figs. 7–10.

HASLE, 1974, p. 427. HASLE and SYVERTSEN, 1997, p. 324, Pl. 73.

#### Morphological Description:

Cells are linear to lanceolate with broadly rounded ends with no central interspace in valve view and almost linear with cut ends in girdle view. This cell's apical axis is between 15  $-21 \,\mu$  m and transapical axis about 2.3–2.9  $\mu$  m. Two rows of poroids, sometimes three are present in striae (circled with arrow, Fig. 12b). Poroids in 1  $\mu$  m are about 8–10. Interstriae in 10  $\mu$  m are about 38–43, and fibulae in 10  $\mu$  m are from 19–23.

#### Occurrence in Tokyo Bay:

This species was observed in March, April, June, July, August and September at temperatures of 14.5–27.5°C and salinity at 31–34. *P. americana* is reported to for the first time here. LUNDHOLM et al. (2002a) reported *P. americana* from Japan (34°50′, 139°31′) to be epiphytic to *Chaetoceros*. We also found *P. americana* to be epiphytic to some *Chaetoceros* species e.g.: *C. affinis, C. lorenzianous, C. socialis* among others from Tokyo Bay (Fig. 23).

#### **Remarks:**

This species was first described as *Nitzschia americana* by HASLE in 1964. It was recently redefined by LUNDHOLM *et al.* (2002a) to separate two species within *P. americana* complex, *P. brasiliana* and *P. linea*.

	Cleane	central	length	width	rows of	poroids	Interstriae	Fibulae	poroid
	Snape	interspace	$(\mu m)$	$(\mu m)$	poroids	in $1 \mu$ m	in $10\mu\mathrm{m}$	in $10\mu\mathrm{m}$	sectors
P. americana	1:	abaant	15 - 21	2.3 - 2.9	2-3	8-10	38-43	19-23	
(n=18)	imear	absent	16-24	2.5 - 4.5	2–3	8–10	26–31	18-24	
P. brasiliana	1:	- 1 4	23-52	2.4-3	2-3	7-8.5	24-26	19-24	
(n=14)	linear	absent	<i>12–65</i>	1.8–1.6	2–3	7–10	20–26	20-26	
P. caciantha	1		32–33	2.1-2.2	1	3–5	32-35	16-19	2-6
(n=6)	lanceolate	present	53-75	2.7–3.5	1	3.5 - 5	28-31	15-22	2–6*
P. calliantha	1:		69-87	1.3-2.3	1	3–5	32-39	15-20	4-13
(n=50)	imear	present	44–86	1.4 - 4.7	1	4–6	34–39	17-22	several
P. delicatissima	1:		50-65	1.3-1.5	2	11-12	36-38	23-25	
(n=20)	linear	present	40–78	1.1 - 2	2	10-12	36–41	19–25	
P. fraudulenta	spindle-		55-89	3.4-10	2-3	4.5-7	21-27	19-25	
(n=26)	shaped	present	50-119	4.5–10	2–3	5 - 7	18-24	12-24	
P. galaxiae	lomonoloto		24-25	1.5 - 1.6	—	—	55-57	15-16	
(n=20)	lanceolate	present	25–41	1.2 - 1.7			56-64	16-26	
P. multistriata	1:	- 1 4	47-53	2.3-3.2	2	9-12	40-42	24-29	
(n=44)	linear	absent	55 - 65	3.1–3.6	2	11**	37 - 42	23–26	
P. multiseries	linear-	abaant	76-89	2.9 - 5.6	3-6	5-6	11-16	12-16	
(n=22)	lanceolate	absent	68–140	3.4-6	3-4	4-6	10 - 19	10 - 19	
P. pungens	linear-	abaant	70-92	2.9-5	2	3-5	11-17	12-17	
(n=32)	lanceolate	absent	74–174	2.4 - 5.3	1–2	3–4	9–16	6–16	
P. pseudodeli-	1:		—	1.8	1	4-4.8	30	16	2
catissima (n=1)	mear	present	50-140	0.9–1.6	1	4–6	30–46	14–16	2

Table 2. Morphometric characteristics of the eleven species observed during April 2008- August 2009 in Tokyo Bay. Numbers in italics found below an entry are from original descriptions of the species. (\*AMATO et al., 2007; \*\* based from photomicrograph of original description)

Type locality of *P. americana* is in Atlantida, Uruguay (LUNDHOLM *et al.*, 2002a). *P. americana* is a common epiphyte on *Chaetoceros*, *Odontella* (LUNDHOLM *et al.*, 2002a) and sometimes *Bacteriastrum* (FRYXELL *et al.*, 1990; HASLE and SYVERTSEN 1997). It has been found in tropical to temperate waters in the world (LUNDHOLM *et al.*, 2002a).

#### Pseudo-nitzschia brasiliana Lundholm, HASLE and FRYXELL

Figs. 3, 13

LUNDHOLM *et al.*, 2002a, pp. 484–487, Figs. 21–45.

LARSEN and NGUYEN, 2004, pp. 30–31, Pl. III, Figs. 1, 3–4. VILLAC *et al.*, 2005, pp. 139–145, Figs.2–3. QUIJANO-SCHEGGIA *et al.*, 2009a, pp. 100–107, Fig. 2.

#### Morphological Description:

Cells are broadly linear, cigarette-shaped with broadly rounded tips in valve view and linear with truncate ends in girdle view. Apical axis is from  $23-52 \ \mu$  m and transapical axis at  $2.4-3 \ \mu$  m. No central interspace is found. Two rows of poroids, sometimes three near the border between valve face and mantle can be clearly seen under TEM (Fig. 13b). Poriods in  $1 \ \mu$  m are about 7-8.5 small and closely spaced. Interstriae in 10  $\mu$  m are about 24-26 and the fibulae in 10  $\mu$  m are in the range of 19-24. Overlap of chains is within 1/8-1/11 of the cell's length.

#### Occurrence in Tokyo Bay:

This species was observed during the months of April, July, and September at temperatures of  $14.5-26.4^{\circ}$ C and salinity at 33-34. *P. brasiliana* is reported for the first time here.

#### **Remarks:**

The approximately equal number of fibulae and striae, the coarser silicification, the single row of poroids in the girdle bands, the slightly longer length, the formation of stepped colonies and the planktonic mode of living are



Figs. 12-21. TEM micrographs of species observed in Tokyo Bay brought in culture.
A set of micrographs per species consists of: the whole valve (a), valve tip (b), and a section of the valve showing pores (c). Fig. 12. P. americana (arrow point to circled striae with 3 rows of poroids), Fig. 13. P. brasiliana Fig. 14. P. caciantha, Fig. 15. P. calliantha, Fig. 16. P. delicatissima, Fig. 17. P. fraudulenta, Fig. 18. P. galaxiae, Fig. 19. P. multistriata, Fig. 20. P. multiseries, Fig. 21. P. pungens. Scale bars: (a) = 10 μ m; valve section showing pores and tip (b and c) = 1 μ m. Arrowheads point to central interspaces, respectively.

features distinguishing *P. brasiliana* from *P. americana* under LM. *P. brasiliana* differs from *P. americana* under EM by the morphometric measurements found in Table 2.

Type locality of this species is in Sepetiba Bay, Brazil (LUNDHOLM *et al.*, 2002a). *P. brasiliana* is distributed mainly in warmer waters. Brazil, Vietnam, Gulf of Panama, Gulf of Mexico, Gulf of California, Vietnam, Indonesia (Jakarta Harbour), Thailand and Jinhae Bay, and S. Korea (LUNDHOLM *et al.*, 2002a).

#### Pseudo-nitzschia caciantha LUNDHOLM, HASLE and FRYXELL

Figs. 5, 14

LUNDHOLM *et al.*, 2003, pp. 806–809, Figs. 5 A-F, Table 2.

Synonym: *P. pseudodelicatissima* (HASLE) HASLE 1993 pro parte

#### Morphological Description:

Cells at valve view are lanceolate tapering from the middle towards the tips and appear almost asymmetrical. A central interspace is present (Fig. 14a, arrowhead). Its apical axis is around  $32\text{--}33 \ \mu$  m and around  $2.1\text{--}2.2 \ \mu$  m transapical axis. One row of poroid is found in striae. Poroids are large and are 3–5 in every  $1 \,\mu$  m partly closed by a membrane with 2–6 sectors of poroid hymen (Fig. 14c). Interstriae in 10  $\mu$  m are about 32–35, while fibulae in 10  $\mu$ m are about 16–19.

#### Occurrence in Tokyo Bay:

This species was observed in Tokyo Bay during the months of July and November at temperatures of  $16.9-24.1^{\circ}$ C and salinity at 31-33. *P. caciantha* is reported for the first time here.

#### **Remarks:**

This species was described by LUNDHOLM et al. (2003) from *P. pseudodelicatissima* complex. *P. caciantha* is distinguished from the "true" *P. pseudodelicatissima* by morphometric measurement observed under EM (Table 2).

The morphometric measurements of *P. caci*antha from this study matched up more with a newly described species, P. mannii (AMATO and MONTRESOR, 2008) than with P. caciantha in terms of length: measurements for this species was at  $32-38 \,\mu$  m, P. manni was at  $33-130 \,\mu$  m while P. caciantha was at  $53-75 \,\mu$  m; width: this study at 2.1–2.2 μm, P. manni at 1.7–2.6 μm, P. *caciantha* at 2.7–3.5  $\mu$  m; interstriae in 10  $\mu$  m: this study at 32–38. P manni at 30–40. P. caciantha at 28–31 and fibulae in  $10 \,\mu$  m: this study at 16-19, P. manni at 17-19, P. caciantha at 15-22. But according to AMATO and MONTRESOR (2008), P. manni is primarily differentiated from *P. caciantha* in three aspects: (1) the valve shape which is linear in P. manni while lanceolate in P. calliantha, (2) cell width, and (3) the pattern of sectors in *P. caciantha* where the poroids do not bear a central sector while P. manni has 3.6% poroids bearing central sectors. Our strain is lanceolate and all poroids do not have a central sector. Thus this was identified as P. caciantha.

Type locality of this species is Off Tuxpam, Mexico (LUNDHOLM *et al.*, 2003). This was found in Mexico, Andaman Sea, near Phuket Island Thailand (LUNDHOLM *et al.*, 2003).

#### **Pseudo-nitzschia calliantha** LUNDHOLM, MOESTRUP and HASLE Figs. 6, 15

LUNDHOLM *et al.*, 2003, pp. 801–804, Figs. 2, A-G, Table 2.

BARGU et al., 2002, pp. 480–482, Figs. 1–2. LARSEN and NGUYEN, 2004, pp. 32–33, Figs. 5–8. CAROPPO et al., 2005, pp. 763–774, Figs. 3a and c. FEHLING et al., 2006, pp. 32–33, Figs. 5–8. BESIKTEPE et al., 2008, p. 440, Figs. 2–3.

Synonym: *P. pseudodelicatissima* (HASLE) HASLE 1993 pro parte

HASLE et al., 1996, pp. 149–150, Figs. 26–29, 76– 81, Tables 1, 2, 8.

SKOV et al., 1999, pp. 13–15, Figs. 10A-G. PhLIPS et al., 2004, pp. 41–42, Fig. 2.

#### Morphological Description:

Cells at valve view are straight and linear and abruptly taper to rounded tips. A central interspace is present (Fig. 15a, arrowhead). A single row of poroids and about 3–5 large poroids in 1  $\mu$ m are found in striae. Each poroid is divided by hymen to form "flower (calliantha) like appearance, with one "central disc" and about four to 13 "petals" around it (Fig. 15c). Interstriae in 10  $\mu$  m are about 32– 39; fibulae in 10  $\mu$ m are from 15–20. Its apical axis is runs from 69–87  $\mu$  m and transapical axis at 1.3–2.3  $\mu$ m.

#### Occurrence in Tokyo Bay:

This species was observed during the months of May, June, July, August, September and November at temperatures of 17.8-27.5 °C and salinity at 31-33. *P. calliantha* is reported for the first time here.

#### **Remarks:**

*P. calliantha* is distinguished from the "true" *P. pseudodelicatissima* by the following features observed under EM: Poroids in  $1 \,\mu$  m for *P. calliantha* is 4–6 while for *P. pseudodelicatissima*, it was 5–6. Interstriae in  $10 \,\mu$  m for *P. calliantha* are 34–39 while *P. pseudodelicatissima* is at 36–43. Fibulae in  $10 \,\mu$  m for *P. calliantha* are at 17–22 while *P. pseudodelicatissima* is at 20–25. Inner poroid sectors of *P. calliantha* are 7–10 with central poroid sector while *P. pseudodelicatsima* has only two sectors.

Type locality of this species is in Ejby, Isefjorden, Sealand, Denmark (LUNDHOLM *et*  *al.*, 2003). This is cosmopolitan (SKOV *et al.* 1999, LUNDHOLM *et al.*, 2003).

#### *Pseudo-nitzschia delicatissima* (CLEVE) HEIDEN

Figs. 7, 16

HEIDEN and KOLBE, 1928.

HASLE et al., 1996, pp. 147–149, Figs. 23–25, 70– 75, Tables 1, 2, 7. SKOV et al., 1999, pp. 8–13, Figs. 9A-J. STEHR, et al., 2002, p. 59, Figs. 3G and H. CUSACK et al., 2004, pp. 58–71, Pl. V.

LARSEN and NGUYEN, 2004, pp. 35–36, Pl. IV, Figs 2, 4, 8. CAROPPO *et al.*, 2005, pp. 765, Fig. 3C. KACZMARSKA *et al.*, 2005, p. 8, Figs. 14–23. FEHLING *et al.*, 2006, pp. 95–97, Fig. B. LUNDHOLM *et al.*, 2006, pp. 467–470, Figs. 1A-G, Table 4. SCHNETZER *et al.*, 2007, pp. 377–378, Figs. 4c and d.

Synonym: *Nitzschia delicatissima* CLEVE CLEVE 1897 b. p. 24.

Nitzschia actydrophila HASLE

HASLE, 1965, pp. 35–37, Pl. 2, Fig. 10, Pl. 15,

Figs. 19–23, Pl. 16, Figs. 3–7.

Pseudo-nitzschia arenyesis QUIJANO-SCHEGGIA, GARCÉS, LUNDHOLM

QUIJANO-SCHEGGIA et al., 2009b, pp. 506–508, Figs. 30–34.

#### Morphological Description:

Cells in valve view are narrow and linearlanceolate with straight cut tips while linear with straight cut ends in girdle view. A central interspace is present (Fig. 16a, arrowhead). There are two rows of poroids (Fig. 16b) and about 11–12 small poroids in every 1  $\mu$  m. Interstriae in 10  $\mu$  m are about 36–38; fibulae in 10  $\mu$  m are about 23–25. Apical axis around 50– 65  $\mu$  m and transapical axis at 1.3–1.5  $\mu$  m. Overlap of chains is within 1/7–1/10 of the cell's length.

#### Occurrence in Tokyo Bay:

This species was observed once on the months July, 2008 at temperature of 24.1  $^\circ C$  and salinity at 31.

#### **Remarks:**

*P. delicatissima* can be easily distinguished from *P. pseudodelicatissima*, *P. caciantha* and *P. calliantha* by having two rows of poroids instead of one as the other three species have. It is more linear than *P. caciantha* and has wider transapical axis compared to *P. pseudodelicatissima* and *P. calliantha*. Its ends seem cut-off when viewed under LM (Fig. 7).

The name Pseudo-nitzschia arenyesis QUIJANO-SCHEGGIA, GARCÉS, LUNDHOLM is considered here as a synonym of *P. delicatissima*. Diagnosis for this name compared with P. delicatissima stipulates that there is no difference morphologically between P. delicatissima and P. arenysensis, but in helix I of ITS2 rDNA (QUIJANO-SCHEGGIA et al., 2009b). Considering that ITS2 is not transcribed and its high intraspecific variability, make this a weak criterion for species determination. Other regions of DNA should be investigated and compared. Further studies should be made to find distinguishing morphological characteristics that separates P. delicatissima and P. arenvesis.

Type locality for *P. delicatissima* is not designated. This is widely distributed in arctic (CAROPPO *et al.*, 2005; SKOV *et al.*, 1999), temperate (TAKANO and KUROKI, 1977; FRAGA *et al.*, 1998; SKOV *et al.*, 1999; CUSACK *et al.*, 2004; KACZMARSKA *et al.*, 2005; FEHLING *et al.*, 2006, SCHNETZER *et al.*, 2007), and subtropical waters (HASLE, 1965; SKOV *et al.*, 1999; LARSEN and NGUYEN, 2004).

#### **Pseudo-nitzschia fraudulenta** (CLEVE) HASLE Figs. 8, 17

HASLE *et al.*, 1996, pp. 144–146, Figs. 17–19, 57–61, Tables 1, 2, 6.

STEHR, et al., 2002, p. 59, Figs. 3G and H. CUSACK et al., 2004, pp. 58–72, Pl. IV, Figs. a-f. KACZMARSKA et al., 2005, pp. 8–11, Figs. 14–23. FEHLING et al., 2006, pp. 97–98, Fig. C. CONGRESTRI et al., 2008, p. 201, Figs. 2a-f.

Synonym: *Nitzschia fraudulenta* P.T. CLEVE CLEVE 1897a, p. 300, Fig. 11.

HASLE 1965, pp. 15–18, Pl.1, Figs. 2–3, Pl. 4, Figs. 8–10, Pl.6,

Figs. 5–10, Pl.8, Figs. 1 3. TAKANO and KUROKI, 1977, p. 43, Figs. 3, 14–18.

Pseudo-nitzschia seriata var. fraudulenta (P.T. CLEVE) H. PERAGALLO

H. PERAGALLO in H&M PERAGALLO, 1900.

#### Morphological Description:

Cells in valve view are symmetric and spindleshaped with straight cut tips. A central interspace is present (Fig. 17a, arrowhead). Two or a few times three rows of poroids and about 4.5–7 poroids with star-shaped membranes in every  $1\,\mu$  m are in striae (Fig. 17b). Interstriae in 10  $\mu$  m are about 21–27; 19–25 fibulae in 10  $\mu$  m. Apical axis is around 55–89  $\mu$  m and 3.4–10  $\mu$  m transapical axis.

#### Occurrence in Tokyo Bay:

This species was observed in May, June and July at temperatures of 18.8–24.1°C and salinity at 31.

#### **Remarks:**

*P. fraudulenta*, like *P. delicatissima* has striae which consisted of two rows of poroids and valves with central insterspace. But it is very different from *P. delicatissima* because it is wider more lanceolate or spindle-shaped; more heavily silicified than the latter, and has larger pores with star-patterned inner poroids within each pore.

Type locality of this species is in Plymouth Harbour, England (HASLE *et al.*, 1996). Reports of its occurrence include Atlantic Moroccan waters, N. Atlantic; Skagerrak (HASLE, 1965), and temperate waters (TAKANO and KUROKI, 1977; RINES *et al.*, 2002; CUSACK *et al.*, 2004; KACZMARSKA *et al.*, 2005; FEHLING *et al.*, 2006; ALMANDOZ *et al.*, 2007).

# *Pseudo-nitzschia galaxiae* LUNDHOLM and MOESTRUP

#### Figs. 4, 18

LUNDHOLM and MOESTRUP, 2002, pp. 596–601, Figs. 1–22.

CERINO *et al.*, 2005, pp. 347–39, Figs. 1A-E, 2A-H.

#### Morphological Description:

Cells in valve view are lanceolate with rostrate apices. A central interspace is present (Fig. 18a, arrowhead). Rows of poroids are tiny, about  $3-4\,\mu$  m wide. Poroids are very tiny (Fig. 18c) and spread all over the valve. These are difficult to count. Interstriae in 10  $\mu$  m is about 55–57 and fibulae in 10  $\mu$  m is about 15–16

and barely seen in LM. This tiny cell has an apical axis of around  $24-25 \,\mu$  m and  $1.5-1.6 \,\mu$  m transapical axis. Chains of about 2–4 cells were observed in culture.

#### Occurrence in Tokyo Bay:

This species was observed once in May, 2009 at temperature of 18.8°C and salinity at 31.

#### **Remarks:**

Type locality for this species is in Off Tuxpam, Mexico (LUNDHOLM and MOESTRUP 2002). This is also found in Caribbean coast of Mexico off Tuxpam; Sydney, N.S.W. Australia; Bay of Naples, Italy (LUNDHOLM and MOESTRUP 2002; CERINO *et al.*, 2005).

#### **Pseudo-nitzschia multistriata** (TAKANO) TAKANO

Figs. 9, 19

TAKANO, 1995, pp. 73–74. RHODES *et al.*, 1998, pp. 463–465, Figs. 1A-F. LARSEN and NGUYEN, 2004, pp. 41–43, Pl. VII, Figs. 1–4. Synonym: *Nitzschia multistriata* TAKANO, 1993, pp. 39–41, Figs. A-E, Table 1.

#### Morphological Description:

Cells at valve view are narrow, linear, tapering near the ends for a longer or shorter distance towards rounded apices. There is no central interspace. Two rows of poroids and about 9–12 poroids are found in 1  $\mu$  m (Fig. 19c). Interstriae in 10  $\mu$  m are about 40–42, and fibulae in 10  $\mu$  m range from 24–29. Apical axis is around 47–53  $\mu$  m and transapical axis at 2.3– 3.2  $\mu$  m. There are also two types of *P*. *multistriata* encountered here as originally described: one with uniform striations and another with uneven striations (Figs. 24 a and b).

#### Occurrence in Tokyo Bay:

This species was observed in June, July, August, September and October at temperatures of 19.9–27.5°C and salinity at 31–34.

#### **Remarks:**

*P. multistriata* may be distinguished by its curved ends or characteristic sigmoid shape when observed in girdle view under LM (Fig. 19). Valves of this species have striae which consisted of two rows of poroids similar to *P. fraudulenta* and *P. delicatissima*. Unlike these two species, *P. multistriata* has no central interspace.

Type locality for this species is in Fukuoka Bay, Japan (TAKANO, 1993). Reports of its distribution include China (QI *et al.* 1996), Gulf of Naples, Italy (SARNO and DAHLMANN, 2000), Japan, Korea, Malaysia, Vietnam (LARSEN and NGUYEN, 2004) and in Sea of Japan (ORLOVA *et al.*, 2008).

#### **Pseudo-nitzschia multiseries** (HASLE) HASLE Figs. 10, 20

SKOV et al., 1999, pp. 15–17, Figs. 11A, B, G, I. STEHR, et al., 2002, p. 58, Figs. 2 B and C.

KACZMARSKA et al., 2005, pp. 11–12, Figs. 30–34. Synonym: Nitzschia pungens f. multiseries G. R. HASLE 1965

HASLE, 1965, pp. 14–15, Pl. 2, Figs. 1–2, Pl.5, Figs. 10–12, Pl. 6, Fig.4, Pl. 7, Figs. 9–11.

TAKANO and KUROKI, 1977, p. 43, Figs. 2, 10, 12 -13 vix 9, 11.

Pseudo-nitzschia pungens f. multiseries G.R. HASLE 1993

HASLE, 1993, p.39. VILLAC *et al.*, 1993, pp. 215–217, Figs. 1–2.

#### Morphological Description:

Cells are symmetric and linear-lanceolate in both the valve and girdle views. There is no central interspace. Apical axes are at 76–89  $\mu$  m and transapical axis at 2.9–5.6  $\mu$  m. Three (Fig. 20c) and up to six (not in photo) rows of poroids are found in a striae with about 5–6 poroids in every 1  $\mu$  m (Fig. 20c). Interstriae in 10  $\mu\,{\rm m}$  are about 11–16, fibulae about 12–16 in 10  $\mu\,{\rm m}.$ 

#### Occurrence in Tokyo Bay:

This species was observed in April, May, June, July, August and September at temperatures of 14.5–27.5°C and salinity at 31–34.

#### **Remarks:**

P. multiseries was formerly called as P. pungens f. multiseries (HASLE, 1965). This is now considered as a separate species from P. pungens based on morphological, physiological and genetic data (HASLE, 1995). Morphological characteristics that separate P. multiseries from P. pungens are the different number of rows of poroids (HASLE, 1965). P. multiseries has three and as many as six rows of poroids especially near the border between valve face and mantle. P. pungens has three to four poroids in  $1 \,\mu$  m while P. multiseries has four to six poroids in  $1 \,\mu$  m.

Type locality of this species is in Drøbak, Oslofjord, Norway. Reports of its distribution are mainly in temperate waters (HASLE, 1965; SKOV *et al.*, 1999; TAKANO and KUROKI, 1977; KOTAKI *et al.*, 1996; ORLOVA *et al.*, 2008).

# *Pseudo-nitzschia pungens* (GRUNOW ex CLEVE) HASLE

Figs. 11, 21

HASLE et al., 1996, pp. 138–140, Figs. 3–6, 30–37, Table 1–3.

SKOV et al., 1999, pp. 17–19, Figs. 11C-F, H.
STEHR, et al., 2002, pp. 57–58, Figs. 2A and B.
CUSACK et al., 2004, pp. 58–63, Pl. 1, Figs. a-j,
Table 2. LARSEN and NGUYEN, 2004, pp. 43–45,
Pl. VIII, Figs. 1–3.

CHEPURNOV *et al.*, 2005, pp. 382–383, Figs. 3, 4– 6, Table 2. KACZMARSKA *et al.*, 2005, p. 16, Figs. 48–52.

Synonym: *Nitzschia pungens* Grunow ex P.T. CLEVE 1897b

HASLE, 1965, pp. 12–14, Pl. 1, Figs. 4–5, Pl. 5, Figs. 7–9, Pl. 6, Fig. 3, Pl. 7, Figs. 1–8.

#### Morphological Description:

Cells are symmetric and linear to lanceolate in both the valve and girdle views. Apical axis is around  $70-92 \,\mu$ m and transapical axis at 2.9-



Fig. 22. TEM micrograph of a valve section of P. *pseudodelicatissima* showing pores and inner pore feature divided into two. Scale bar =  $1 \,\mu$  m.

 $5 \,\mu$  m. No central interspace can be observed. Two rows of poroids are present (Fig. 21c) and about 3–5 poroids in every  $1 \,\mu$  m are counted (fig. 21b) under the TEM. Interstriae in  $10 \,\mu$ m are about 11–17; fibulae at 12–17. Overlap of chains is within 1/3–1/4 of the cell's length.

#### Occurrence in Tokyo Bay:

This species was observed in March, April, May, June, July, August and September at temperatures of 12.3-27.5°C and salinity at 31-34.

#### **Remarks:**

*P. pungens* has larger pores and is the most heavily silicified based on sharpness of TEM micrographs among all Pseudo-nitzschia



Fig. 23. LM micrograph of *P. americana* (shown by arrows) epiphytic to *Chaetoceros affinis*. Scale bar =  $20 \,\mu$  m.

	D A
18	1 1
38 '	
2 (D	
(B)	
18	0

Fig. 24. TEM micrograph of *P. multistriata* (a) regular striations of interstriae, (b) irregular striations of interstriae. Scale bar =  $1 \,\mu$  m.

species in this study. The interstriae of this species are widely spaced as another species, *P. multiseries.* This, plus the resemblance with size and shape made these two species appear similar. The number of rows of poroids distinguishes *P. pungens* from *P. multiseries* (Figs. 20c, 21c). *P. pungens* has two rows of poroids that consisted of larger poroids while *P. multiseries* has three or more.

Type locality of this species is in Yeddo Bay (Tokyo Bay), Japan (HASLE, 1965). HASLE and FRYXELL in 1995 reported this to be a cosmopolitan species. This was further supported by CASTELEYN *et al.* (2008) who reported global distribution of one strain of *P. pungens*.

**Pseudo - nitzschia pseudodelicatissima** (HASLE) HASLE Fig. 22 LUNDHOLM et al., 2003, p. 801. Figs. 1A-G, Table 2. non HASLE et al., 1996, pp. 149–150, Figs. 26–29, 76–81, Tables 1, 2, 8. non SKOV et al., 1999, pp. 13–15, Figs. 10A-G. non CUSACK et al., 2004, pp. 58–63, Pl. VI, Figs. a-f. non PHLIPS et al., 2004, pp. 41–42, Fig. 2. non FEHLING et al., 2006, pp. 95–97, Fig. C. KACZMARSKA et al., 2005, pp. 12–16, Figs. non 35–38 vix 39 non 10– 47.

Synonym: Nitzschia pseudodelicatissima G.R. HASLE 1976

TAKANO and KUROKI, 1977, p. 44, vix Figs. 21–25.

Nitzschia delicatula HASLE 1965

HASLE, 1965, pp. 37–40, Pl. 4, Figs. 4 –5, Pl. 16, Figs. 8–18, Pl. 17 Figs. 1–16 non 14–16.

#### Morphological Description:

There was only one broken value of this cell observed during the sampling period. It is linear in shap. A central interspace is present for this species (not found in micrograph). Transapical axis was at  $4.8 \,\mu$  m. Apical axis was not measured since an intact value was not observed. A single row of poroids was found in striae and poroids in  $1 \,\mu$  m is about 4–4.8, divided by a membrane into 2 sectors of inner poroids. Interstriae in  $10 \,\mu$  m were calculated to about 30 and fibulae in  $10 \,\mu$  m at 16.

#### Occurrence in Tokyo Bay:

This species was observed on July, 2009 at temperature of 24.5°C and salinity at 31.

#### **Remarks:**

P. pseudodelicatissima is recently emended by LUNDHOLM et al. (2003). This species was split in three; P. pseudodelicatissima, P. caciantha and P. calliantha based on inner poroid features among others. This publication distinguished these three species mainly by sectors dividing each poroid wherein 7–10 in P. calliantha, 2 in P. pseudodelicatissima and 4–5 or 2–6 (AMATO et al., 2007) in P. caciantha with no central perforated area. It was only P. calliantha which had a central perforated area surrounded by similar sized sectors inside its poroid. P. pseudodelicatissima at 50–140  $\mu$  m in length, 1.5–3.4  $\mu$  m in width, 30–46 interstriae and 14–26 fibulae per 10  $\mu$  m (HASLE, 1965) was emended by LUNDHOLM *et al.* (2003) to 54-87  $\mu$  m length, 0.9-1.6  $\mu$  m width, 36-42 and 20-25 in 10  $\mu$  m respectively.

Identification of *P. pseudodelicatissima* complex before and even after LUNDHOLM *et al.*, 2003 need to be restudied to ascertain proper identification of species. For example, Figs. 1– 16 in HASLE (1965) identifications in HASLE *et al.* (1996), SKOV *et al.* (1999) and PHLIPS *et al.* (2004) based on inner poroid features are clearly *P. calliantha.* Figures in, FEHLING *et al.* (2006) CUSACK *et al.* (2004) and KACZMARSKA *et al.* (2005) have four sectors and thus could be *P. caciantha* or *P. mannii* (AMATO and MONTRESOR, 2008). Inner poroid features of figures in TAKANO and KUROKI (1977) are not visible.

Type locality for this genus is in Denmark Strait. Other reports of it occurrence are near Iceland off Costa Nova, Portugal and Napoli, Italy (LUNDHOLM *et al.*, 2003).

#### Discussion

During the survey conducted in April 2008 -August 2009 in Tokyo Bay, 11 species of Pseudo-nitzschia which were critically identified using TEM are yielded. This report of 11 species found in one bay is higher compared to reports in other bays e.g. eight, in Irish waters al., 2004) and Argentina (CUSACK et(ALMANDOZ et al., 2007) and seven in Bay of Fundy, Canada (KAKZMARSKA et al., 2005). Further studies of Pseudo-nitzschia in this area, e.g., ecological in nature should prove productive. Species previously reported to produce domoic Acid are the following: P. calliantha (LUNDHOLM et al., 2003; BESIKTEPE et al., 2008), P. delicatissima (SMITH et al., 1991), P. fraudulenta (RHODES et al., 1998), P. galaxiae (CERINO et al., 2005), P. multistriata (SARNO and DAHLMAN, 2000; AMATO et al., 2010), P. multiseries (BATES et al., 1989; KOTAKI et al., 1996), P. pungens (RHODES et al., 1996; TRAINER et al., 1998). Most of toxic P. pseudodelicatissima have been found to be P. calliantha but there are still some unresolved toxic reports that attribute to this species (e.g. KACZMARSKA et al., 2005).

Five species are reported for the first time here. These are the following: *P. americana*, *P.* 

brasiliana, P. caciantha, P. calliantha and P. galaxiae. Morphometric characterizations of local populations and baseline data are provided herein for future investigations of toxic or newly discovered taxa. Methods are currently devised to aid species enumeration by LM to further ecological studies of *Pseudonitzschia* in the area.

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#### References

- ALMANDOZ, G.O., M.E. FERRARIO, G.A. FERREYRA, I.R. SCHLOSS, J.L. ESTEVES and F.E. PAPARAZZO (2007): The genus *Pseudo-nitzschia* (Bacillariophyceae) in continental shelf waters of Argentina (Southwestern Atlantic Ocean, 38–55° μ S). Harmful Algae, **6**, 93–103.
- AMATO, A. and M. MONTRESOR (2008): Morphology, phylogeny, and sexual cycle of *Pseudo-nitzschia* mannii sp. nov. (Bacillariophyceae): a pseudocryptic species within the *P. pseudodelicatissima* complex. Phycologia, 47, 487–497.
- AMATO, A., A. LüDEKING and W.H.C.F. KOOISTRA (2010): Intracellular domoic acid production in *Pseudo-nitzschia multistriata* isolated from the Gulf of Naples (Tyrrhenian Sea, Italy). Toxicon, 55, 157–161.
- AMATO, A., W.H.C.F. KOOISTRA, J.H. LEVIALDI GHIRON, D.G. MANN, T. PRöschold and M. MONTRESOR (2007): Reproductive isolation among sympatric cryptic species in marine diatoms. Protist, 158, 193–207.
- BATES, S. S., C. J. BIRD, A. S. W. DE FREITAS, R. FOXALL, M. GILGAN, L.A. HANIC, G.R. JOHNSON, A.W. MCCULLOCH, P. ODENSE, R. POCKLINGTON, M.A. QUILLIAM, P.G. SIM, J.C. SMITH, D.V. SUBBA RAO, E.C.D. TODD, J.A. WALTER and J.L.C. WRIGHT (1989): Pennate diatom Nitzschia pungens as the primary source of domoic acid, a toxin in shellfish from eastern Prince Edward Island, Canada. Can. J. Fish. Aquat. Sci., 46, 1203– 1215.
- BARGU, S., T. KORAY, and N. LUNDHOLM (2002): First

report of *Pseudo-nitzschia calliantha* LUNDHOLM, MOESTRUP & HASLE 2003, a new potentially toxic species from Turkish coasts. E.U. J. Fish. Aquat. Sci. **19**, 479–483.

- BESIKTEPE, S., L. RYABUSHKO, D. EDIGER, D. YILMAZ, A. ZENGINER, V. RYABUSHKO, and R. LEE (2008): Domoic acid production by *Pseudo-nitzschia* calliantha LUNDHOLM, MOESTRUP and HASLE (Bacillariophyta) isolated from the Black Sea. Harmful Algae, 7, 438–442.
- CAROPPO, C., R. CONGESTRI, L. BRACCHINI, and P. ALBERTANO (2005): On the presence of *Pseudo-nitzschia calliantha* LUNDHOLM, MOESTRUP and HASLE and *Pseudo-nitzschia delicatissima* (CLEVE) HEIDEN in the Southern Adriatic Sea (Mediterranean Sea, Italy). J. Plank. Res., 27, 763–774.
- CASTELEYN, G., V.A. CHEPURNOV, F. LELIAERT, D.G. MANN, S.S. BATES, N. LUNDHOLM, L. RHODES, K. SABBE, and W. VYVERMAN (2008): Pseudonitzschia pungens (Bacillariophyceae): a cosmopolitan diatom species? Harmful Algae, 7, 241– 257.
- CERINO F., L. ORSINI, D. SARNO, C. DELL'AVERSANO, L. TARTAGLIONE and A. ZINGONE (2005): The alternation of different morphotypes in the seasonal cycle of the toxic diatom *Pseudo-nitzschia* galaxiae. Harmful Algae, 4, 33–48.
- CHEPURNOV, V.A., D.G. MANN, K. SABBE, K. VANNERUM, G. CASTELEYN, E. VERLEYEN, L. PEPERZAK, and W. VYVERMAN (2005): Sexual reproduction, mating system, chloroplast dynamics and abrupt cell size reduction in *Pseudonitzschia pungens* from the North Sea (Bacillariophyta). Eur. J. Phycol., 40, 379–395.
- CLEVE, P.T. (1897a): Report on the phytoplankton collected on the expedition of H.M.S. "Research" 1896. Fifteenth Annual Report of the Fishery Board for Scotland, 3, 296–304.
- CLEVE, P.T. (1897b): A treatise on the phytoplankton of the Atlantic and its tributaries and on the periodical changes of the plankton in Skagerak. Upsala, pp. 1–27.
- CONGESTRI, R., S. POLIZZANO, and P. ALBERTANO (2008): Toxic Pseudo-nitzschia populations from the Middle Tyrrhenian Sea (Mediterranean Sea, Italy). In Algal toxins: nature, occurrence, effect and detection. NATO Science for Peace and Security. Series A: Chemistry and Biology. V. EVANGELISTA, L. BARSANTI, A.M. FRASSANITO, V. PASSARELLI and P. GUALTIERI (eds.), Springer, Netherlands, pp. 197–210.
- CUSACK, C., R. RAINE and J.W. PATCHING (2004): Occurrence of species from the genus *Pseudonitzschia* PERAGALLO in Irish waters. Proc. Royal Irish Acad., 104B, 55–74.
- FEHLING, J., K. DAVIDSON, C. BOLCH, and P. TETT

(2006): Seasonality of *Pseudo-nitzschia* spp. (Bacillariophyceae) in western Scottish waters. Mar. Ecol. Prog. Ser., **323**, 91–105.

- FRAGA, S., M.J. ALVEREZ, Á. MIGUEZ, M.L. FERNÁNDEZ, E. COSTAS, and V. LOPEZ-RODAS (1998): Pseudo-nitzschia species isolated from Galician waters: toxicity, DNA content and lectin binding assay. In Harmful algae. B. REGUERA, J. BLANCO, M.L. FERNÁNDEZ, and T. WYATT (eds.), Xunta de Galicia and the IOC of UNESCO, Paris, pp. 270–273.
- FRYXELL, G.A., M.E. REAP, and D.L. VALENCIC (1990): Nitzschia pungens GRUNOW f. multiseries HASLE: Observations of a known neurotoxic diatom. Beih. Nova Hedwigia, 100, 171–188.
- GUILLARD, R.R.L. (1975): Culture of phytoplankton for feeding marine invertebrates. *In* Culture of Marine Invertebrate Animals. SMITH W.L. and M.H. CHANLEY (eds.), Plenum Press, New York, USA, pp. 26–60.
- HASLE, G.R. (1964): Nitzschia and Fragilariopsis species studied in the light and electron microsocopes. I. Some marine species of the groups Nitzschiella and Lancaolatae. Skr. Norske Vidensk-Akad. I. Mat.-Nat. Kl. Ny Serie, 16, 1– 48.
- HASLE, G.R. (1965): Nitzschia and Fragilariopsis species studied in the light and electron microscopes. II. The group Pseudonitzschia. Skr. Norske Vidensk-Akad. I. Mat.-Nat. Kl. Ny Serie, 18, 1– 45.
- HASLE, G.R. (1974): Validation of the names of some marine planktonic species *Nitzschia* (Bacillariophyceae), Taxon, 23, 425–428.
- HASLE, G.R. (1993): Nomenclatural notes on marine planktonic diatoms. The family Bacillariaceae. Beih. Nova Hedwigia, **106**, 315–321.
- HASLE, G.R. (1994): Pseudo-nitzschia as a genus distinct from Nitzschia (Bacillariophyceae). J. Phycol., 30, 1036–1039.
- HASLE, G.R. (1995): Pseudo-nitzschia pungens and P. multiseries (Bacillariophyceae): nomenclatural history, morphology, and distribution. J. Phycol., 31, 428–435.
- HASLE, G.R. and G.A. FRYXELL (1995): Taxonomy of diatoms. *In* Manual on harmful marine microalgae. IOC Manuals and Guides No. 33. G.M. HALLEGRAEFF, D.M. ANDERSON, and A.D. CEMBELLA (eds.), UNESCO, pp. 339–364.
- HASLE, G.R., C.B. LANGE and E.E. SYVERTSEN (1996): A review of *Pseudo-nitzschia*, with special reference to the Skagerrak, North Atlantic, and adjacent waters. Helgolander Meeresun., **50**, 131–175.
- HASLE, G.R. and E.E. SYVERTSEN (1997): Marine diatoms. In Identifying marine phytoplankton. TOMAS, C.R. (ed.), Academic Press, San Diego, pp. 5–385.

- HEIDEN H. and R.W. KOLBE (1928): Die marinen diatomeen der Deutschen Sudpolar expedition 1901–3. Deutsche Südpolar-Expedition 8, 447–715.
- KACZMARSKA, I., M.M. LEGRESLEY, J.L. MARTIN and J. EHRMAN (2005): Diversity of the diatom genus *Pseudo-nitzschia* PERAGALLO in the Quoddy Region of the Bay of Fundy, Canada. Harmful Algae, 4, 1–19.
- KOTAKI, Y., K. KOIKE, T. OGATA, S. SATO, Y. FUKUYO, and M. KODAMA (1996): Domoic acid production by an isolate of *Pseudonitzschia multiseries*, a possible cause for the toxin detected in bivalves in Ofunato Bay, Japan. *In* Harmful and toxic algal blooms. T. YASUMOTO, Y. OSHIMA, and Y. FUKUYO (eds.) Intergov. Oceanogr. Comm., UNESCO, Paris, pp. 151–154.
- LARSEN J. and N.L. NGUYEN (2004): Potentially toxic microalgae of Vietnamese waters. Opera Botanica 140. Council for Nordi Publications in Botany, Copenhagen, Denmark, 216 pp.
- LUNDHOLM, N., G.R. HASLE, G.A. FRYXELL and P.E. HARGRAVES (2002a): Morphology, phylogeny and taxonomy of species within the *Pseudonitzschia americana* complex (Bacillariophyceae) with descriptions of two new species, *Pseudo-nitzschia brasiliana* and *Pseudonitzschia linea*. Phycologia, **41**, 480–497.
- LUNDHOLM, N., N. DAUGBJERG, and Ø. MOESTRUP (2002b): Phylogeny of the Bacillariaceae with emphasis on the genus *Pseudo-nitzschia* (Bacillariophyceae) based on partial LSU rDNA. Eur. J. Phycol. **37**, 115–134.
- LUNDHOLM N. and Ø. MOESTRUP (2002): The marine diatom *Pseudo-nitzschia galaxiae* sp. nov. (Bacillariophyceae): morphology and phylogenetic relationships. Phycologia, **41**, 594–605.
- LUNDHOLM, N., Ø. MOESTRUP, G.R. HASLE and K. HOEF-EMDEN (2003): A study of the Pseudonitzschia pseudodelicatissima/cuspidata complex (Bacillariophyceae): what is P. pseudodelicatissima? J. Phycol., 39, 797–813.
- LUNDHOLM, N., Ø. MOESTRUP, Y. KOTAKI, K. HOEF-EMDEN, C. SCHOLIN, and P. MILLER (2006): Interand intraspecific variation of the *Pseudonitzschia delicatissima* complex (Bacillariophyceae) illustrated by rRNA probes, morphological data and phylogenetic analyses. J. Phycol., 42, 464-481.
- ORLOVA, T.Y. and O.G. SHEVCHENKO (2002): The first finding of *Pseudo-nitzschia americana* (Bacillariophyta) in Russian seas. Russian J. Mar. Biol., 28, 336–339.
- ORLOVA, T.Y., I.V. STONIK, N.A. AIZDAICHER, S.S. BATES, C. LÉGER, and J. FEHLING (2008): Toxicity, morphology and distribution of *Pseudo*nitzschia calliantha, *P. multistriata* and *P.* multiseries (Bacillariophyta) from the

northwestern Sea of Japan. Bot. Mar., **51**, 297–306.

- PERAGALLO, H. and M. PERAGALLO (1897-1908): Diatomees marines de France et des disctricts maritimes voisins. Grez-sur Loing, pp.1-557.
- PHLIPS, E.J., S. BADYLAK, S. YOUN, and K. KELLEY (2004): The occurrence of potentially toxic dinoflagellates and diatoms in a subtropical lagoon, the Indian River Lagoon, Florida, USA. Harmful Algae, 3, 39–49.
- PRIISHOLM, K., Ø. MOESTRUP and N. LUNDHOLM (2002): Taxonomic notes on the marine diatom genus *Pseudo-nitzschia* in the Andaman sea near the island of Phuket, Thailand, with a description of *Pseudo-nitzschia micropora* sp. nov. Diatom Res., 17, 153–157.
- QI, Y., J. WANG, and L. ZHENG (1996): The taxonomy and bloom ecology of *Pseudo-nitzschia* on the coasts of China. *In* Proceedings IOC-WESTPAC Third International Scientific Symposium, Bali, Indonesia, 22–26 Nov., 1994. A. NONTJI, S. SOEMODIHARDJO, A.G. ILADUDE, D. SETIAPERMANA, D. P. PRASENO, M. K. MOOSA, and O. S. R ONGKOSONGO (eds.), pp. 88–95.
- QUIJANO-SCHEGGIA, S., E. GARCÉS, K. ANDREE, J.M. FORTUÑO, and J. CAMP (2009a): Homothallic auxosporulation in *Pseudo-nitzschia brasiliana* (Bacillariophyta). J. Phycol., 45, 100–107.
- QUIJANO-SCHEGGIA, S.I., E. GARCES, N. LUNDHOLM, Ø. MOESTRUP, K. ANDREE, and J. CAMP (2009b): Morphology, physiology, molecular phylogeny and sexual compatibility of the cryptic *Pseudonitzschia delicatissima* complex (Bacillariophyta), including the description of *P. arenysensis* sp. nov. Phycologia, 48, 492–509.
- RINES, J.E.B., P.L. DONAGHAY, M.M. DEKSHENIEKS, J.M. SULLIVAN, and M.S. TWARDOWSKI (2002): Thin layers and camouflage: hidden *Pseudonitzschia* spp. (Bacillariophyceae) populations in a fjord in the San Juan Islands, Washington, USA. Mar. Ecol. Prog. Ser., 225, 123–137.
- RHODES, L.L., C. SCHOLIN, I. GARTHWAITE, A. HAYWOOD and A. THOMAS (1998): Domoic acid producing *Pseudo-nitzschia* species educed by whole cell DNA probe-based and immunochemical assays. *In* Harmful algae. REGUERA, B., J. BLANCO, M.L. FERNANDEZ and T. WYATT (eds.), Xunta de Galicia and the IOC of UNESCO, Paris, pp. 274–277.
- RHODES L.L., D. WHITE, M. SYHRE and M. ATKINSON (1996): Pseudo-nitzschia species isolated from New Zealand coastal waters: domoic acid production in vitro and links with shellfish toxicity. In Harmful and Toxic Algal Blooms T. YASUMOTO, Y. OSHIMA and Y. FUKUYO (eds.), IOC of UNESCO, Paris, pp. 155–158.
- SARNO, D. and J. DAHLMANN (2000): Production of

domoic acid in another species of *Pseudo-nitzschia*: *P. multistriata* in the Gulf of Naples (Mediterranean Sea). Harmful Algae News, **21**, 5.

- SCHNETZER, A., P.E. MILLER, R.A. SCHAFFNER, B. STAUFFER, B. JONES, S.B. WEISBERG, P.M. DIGIACOMO, W. BERELSON, and D.A. CARON (2007): Blooms of *Pseudo-nitzschia* and domoic acid in the San Pedro Channel and Los Angeles harbor areas of the Southern California Bight, 2003–2004. Harmful Algae 6, 372–387.
- SKOV, J., N. LUNDHOLM, Ø. MOESTRUP and J. LARSEN (1999): Potentially toxic phytoplankton. 4. The diatom genus *Pseudo-nitzschia* (Diatomophyceae/Bacillariophyceae). *In* ICES identification leaflets for phytoplankton, Leaflet No. 185. LINDLEY, J.A. (ed.), International Council for the Exploration of the Sea, Copenhagen, pp.1–23.
- SMITH, J.C., K. PAULY, R. CORMIER, R. ANGUS, P. ODENSE, D. O'NEIL, M.A. QUILLIAM and J. WORMS (1991): Population dynamics and toxicity of various species of *Dinophysis* and *Nitzschia* from the southern Gulf of St. Lawrence. In Proceedings of the Second Canadian Workshop in Harmful Algae. D.C. GORDON Jr. (ed.), Can. Tech. Rep. Fish Aquatic Sci., p. 1799.
- STEHR, C. M., L. CONNELL, K. A. BAUGH, B.D. BILL, N. G. ADAMS and V.L. TRAINER (2002): Morphological, toxicological and genetic differences among *Pseudo-nitzschia* (Bacillariophyceae) species in inland embayments and outer coastal waters of Washington State, USA. J. Phycol., 38, 55–65.
- TAKANO, H. and K. KUROKI (1977): Some diatoms in the Section *Pseudo-nitzschia* found in coastal waters of Japan. Bull. Tokai Reg. Fish. Res. Lab., 91, 41–51.
- TAKANO, H. (1993): Marine diatom Nitzschia multistriata sp. nov. common at inlets of Southern Japan. Diatom, 8, 39–41.
- TAKANO, H. (1995): Pseudo-nitzschia multistriata (TAKANO) TAKANO, A new combination for the pennate diatom Nitzschia multistriata TAKANO. Diatom, 10, 73–74.
- THRONDSEN, J. (1978): Isolation of single cells. In Phytoplankton manual. SOURNIA, A. (ed.), UNESCO, Paris, p.132–135.
- TRAINER V.L., J.C. WEKELL, R.A. HORNER, C.L. HATFIELD and J.E. STEIN (1998): Domoic acid production by *Pseudo-nitzschia pungens. In* Harmful Algae. B. REGUERA, J. BLANCO, M.L. FERNÁNDEZ and T. WYATT (eds.), Xunta de Galicia and the IOC of UNESCO, Paris, pp. 337– 340.
- VILLAC, M.C., D.L. ROELKE, T.A. VILLAREAL, and G.A. FRYXELL (1993): Comparison of two domoic acid-producing diatoms - a review. Hydrobiologia, 269, 213-224.

VILLAC, M.C., S. MELO, M. MENEZES, and D. RIVERA (2005): Pseudo-nitzschia brasiliana (Bacillariophyceae), an opportunistic diatom on the coast of the state of Rio de Janeiro, Brazil. Atlântica, Río Grande, 27, 139–145.

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# 根室市三里浜沖海況の季節変化 I.水温構造

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# Seasonal Variations of the oceanic condition off Sanrihama Beach, Nemuro I. Temperature structure

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Abstract: The Nemuro City Fisheries Research Institute installed bottom temperature measuring sensors off Sanrihama Beach, Nemuro, in order to know seasonal variations of environmental circumstance of Hanasaki crabs. We set 8 stations along a straight line extended offshore, and the observations were made from December 28, 2005 through May 13, 2009. Depths of stations are 5, 10, 15, 20, 30, 40, 50, and 60m. STD observations were made at the time of replacement of measuring systems or at that of exchange of data loggers. Though some of measuring systems were often lost due to severe storms or due to fishing activities, we could clarify peculiar seasonal variations of temperature structure in the sea under consideration. In "summer season" or in "winter seasons", temperature is basically vertically uniform at each station, except in surface thermocline, but temperature decreases toward offshore in "summer season" and increases toward offshore in "winter season". These horizontal temperature gradients are the same as those in the flow regions of the East Hokkaido Warm Current and of the Coastal Oyashio, respectively. The phases of "summer season" and "winter season" appear to advance by about 3 months than that of the East Hokkaido Warm Current or of the Coastal Oyashio, respectively. Besides, melted water of sea ice which originated from the Okhotsk Sea cannot produce very cold water (say below 0°C) in March or in April. These findings would give valuable suggestion for studies of the East Hokkaido Coastal Current.

#### Keywords : Sanrihama, Nemuro City: Continuous measurement of bottom temperature: STD observation: Seasonal variation of temperature structure: The East Hokkaido Coastal Current

1. はじめに

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Marine Information Research Center, Japan Hydrographic Association (SakhNIRO)との共同研究プロジェクト「ハナ サキ・プロジェクト」の一環として、ハナサキガ ニの成体や幼生の動向と周辺環境の関係を調べる ため、根室市太平洋岸の落石岬西の三里浜の試験 操業地で水深 5m から 60m に至る 8 点の測点を 設けて、2005 年 12 月 28 日から 2009 年 5 月 13 日の間、周年にわたる底層水温の連続観測を行っ た。当海域をしばしば襲った嵐や、漁業活動によっ て計測装置が流失し、全測点での完全な同時観測 はほとんどできなかったが、3 年半に及ぶ測定結 果から、この海域の海況の特異な季節変化特性を 明らかにすることが出来た。この結果は今後のハ ナサキガニの生態研究の周辺環境資料として役立 つものと考えられる。また、道東沿岸流(沿岸親 潮と道東暖流)の水の起源や、その形成メカニズ ムの考察に対して重要な示唆を与えるものである。

#### 2. 観測地点と観測機器、観測経過

水温測定の位置と、ハナサキガニ試験操業地付 近の水深図を Fig. 1 に示す。観測点は、落石岬



Fig. 1. Positions of the stations of bottom temperature measurement off Sanrihama Beach, Nemuro. The cape extending southward is Cape Ochiishi. Stations from St. 1 through St. 8 are set at depths of 5m, 10m, 15m, 20m, 30m, 40m, 50m, and 60m, respectively. Bottom contours are shown at interval of 1m. The horizontal scale is given at lower right corner.



Fig. 2. Schematic view of the temperature measuring system. Temperature is measured about 50cm above the bottom.

西の三里浜から、ほぼ東経 145°29′E に沿って南 に延びる線上に配置されている。一番岸よりの水 深 5m のところに St. 1 が設けられ、St. 2 以下 St. 8 までの測点は、それぞれ水深 10m、15m、 20m、30m、40m、50m、60m のところに設けら れている。測点位置は最も岸よりの St. 1 が 43° 10.9'N、145°29.6'E、最も沖の St. 8 が 43°02'N、 145°02'E である。この海域では西向流が卓越す ることが知られており、St. 1 から St. 4 までは 落石岬による流れの影の部分にあり、St. 5 より 沖の測点とは若干海況特性が異なっている。

用いた観測装置を模式的に Fig. 2 に示す。水 温測定には温度センサーの付いた Onset Computer Cooperation Co. 製のデータロガー (UA-001-08 または TBI32-05-37) を用いた。温度セン サーは海底から約 50cm の高さにある。測定は 2005年12月28日から2009年5月13日まで実 施した。最初、一年間の連続記録を一気に取るこ とを計画したが、2006年10月7~9日に当地を 襲った嵐のため、岸よりの2点(St.1とSt.2) を除いて観測システムが流失し、測定記録も失わ れてしまった。2007 年からは、より太いロープ を使うなど若干の改良を加えるとともに、データ を頻繁に回収、再設置することにした。この作業 等の際に、Alex Electronic Co.製の STD (ATS 200-PK)を用いて水温・塩分の鉛直プロファイ ルを測定した。しかし、その後も悪天候や、漁船 の網に観測装置が引っかけられるなどの事故がし ばしば発生した。データの取得状況を Fig. 3 に 示すが、8測点全てで同時計測ができたのは、 2007年7月28日から12月1日の期間のみであっ た。しかし、全期間について最低2測点での同時 観測に成功している。データの取得状況から、 St. 1 と St. 2 のみの観測が行われた 2005 年 12 月28日から2006年12月7日までを期間Iとし、 St.1とSt.5のみの観測が行われた2006年12 月7日から2007年7月26日までを期間II、St.



Fig. 3. Periods of data acquisition for each observation station. Roman numbers shown at the bottom of the figure indicate the periods from I to V, which are set in the text used for convenience sake.

1からSt.8(一部St.6の記録を欠く)までの観 測が成功した2007年7月26日から2008年1月 30日までを期間IIIとする。さらにSt.1からSt. 5(一部St.5記録を欠く)の記録の得られた2008 年1月30日から2008年12月29日までを期間 IV、St.2とSt.3のみの記録の得られた2008年 12月29日から2009年5月13日までを期間V と呼ぶことにする。これらの期間はFig.3の下 部に示してある。2008年が閏年であるため、各 年を全て365日と整えて、図の軸等の表記には、 便宜上2006から2008年は各年の元日を0として、 2009年については2008年12月31日を0として、 それから数えた日番号で表示する。(参考のため 図の横軸下に月を示す。)

水温の記録間隔は1時間である。回収・再設置 作業には約2時間を要する。この作業等のため欠 測があった場合、3時間までの欠測は、線形補間 により内挿して連続記録にした。測定結果は、潮 汐周期等の変動を除くため、各日について0時か ら24時までの25個のデータを平均して、各日の 値として時系列を作成した。また、短周期変動を 除去するため、さらに15日間の移動平均を求め て使用するが、日平均値が1日得られていない場 合には、前後の日平均値の平均で代用して連続的 な時系列を作成し、移動平均を行った。

STD 観測を実施した日を Table 1 に示す。こ の表の各欄には、日付(日番号)、期間番号(ロー マ数字)が与えられている。水温・塩分は表面か ら 1m 間隔で海底近くまで観測されている。観測 は底層水温の各観測点の近傍で実施したが、測点 直上を避けて上記の各測点深度より、1~2m深め まで観測するように努めた。観測実施は 2007 年 5 月からであり、観測実施間隔は作業の都合から 一定ではないが、ほぼ周年にわたる観測を行うこ とができた。

#### 3. 各年の季節変化と基準季節変化

最も長期の連続観測が得られている水深 5m の St. 1 の記録を用いて、各年の季節変化を比較し たのが Fig. 4 である。St. 1 の記録が利用できな い 2008 年 12 月 29 日以降については、St. 2 (水 深 10m)の記録を代わりに用いた。日平均値の 記録には、かなりの振幅の短周期変動が現れ季節 変化の様相が分かりにくいので、この図では 15 日間の移動平均をかけたものを示している。各年 の変動にはなおも比較的短周期の変動が残ってい るものの、かなりスムースな季節変動が得られて いる。

季節変化は各年とも、正弦曲線からかなりはず れている。3月初め(日番号 30 付近)から7月 末(日番号210付近)までの昇温期の水温上昇が 直線的であるのに対して、9月中旬(日番号260 付近)からの下降期の曲線が、高温側に膨らむ形 をしている。北海道沿岸には多くの地点で、沿岸 水温の周年観測が行われており、観測結果は北海 道栽培漁業振興公社から、「北海道沿岸漁場観測 取りまとめ」として毎年公表されている。根室市 周辺の太平洋側でも、歯舞(根室市)や厚岸、釧 路に観測点があるが、これらの点での季節変化に は、このような正弦波からの著しい歪みは認めら れない。恐らく、三里浜が外洋に開けており、外 洋の条件に影響されやすく、海水の流動に伴う移 流の効果が大きいことが原因であろう。年による 変化は冬季水温の極小期と、夏季水温の極大期に 大きく現れている。

各測点間の水温変動を比較するとき、振幅の大 きな季節変動を除去しておくことが望ましい。各 年について St. 1 の移動平均曲線からの差を用い ることも考えたが、15 日の移動平均値に現れる 短周期の変動の現れ方も見るためには、どの年に

	2007	2008	2009
January		30 (29) III, IV	29 (29) V
February			26 (54) V
March			
April		3 (93), 30 (120) IV	
May	29 (147) II	28 (148) IV	
June		27 (178) IV	
July	28 (206) II, III	31 (211) IV	
August		28 (240) IV	
September		26 (269) IV	
October	1 (282) III	29 (302) IV	
November		26 (330) IV	
December	1 (334) III	29 (363) IV, V	

Table 1. Dates of STD observations. Date, day number (in bracket), and period number (in Roman number) are given in each column in this order.



Fig. 4. Seasonal variations of the sea bottom temperature (15 days running mean) at St. 1, for 2006 (thick full line), for 2007 (dotted line), for 2008 (dashed line) and for 2009 (thin full line). The observation at St. 1 is not available after December 29, 2008, so the temperature values at St, 2 are used. The temperature (°C) is taken in ordinate, and the day number is taken in abscissa The day number is counted from January 1 for 2006 through 2008, and from Dec. 31, 2008 for 2009.



Fig. 5. Seasonal variation of the bottom temperature, which was obtained by averaging the variations at St. 1 over the period from 2005 to 2009. This curve is used as a reference curve in the following analysis.

ついても同じ基準を取ることが望ましい。そこで、 Fig. 4 に示された各年の測定値を平均した季節変 動を基準にすることとした。平均した季節変動を Fig. 5 に示す。正弦波からの外れはこの図にも良 く現れている。平均した年数は少ないが、短周期 の変動も Fig. 4 に比べて小さくなっている。

#### 4. 各期間における底層水温変動特性

4-1. 期間 I および期間 II における水温変動 期間 I (2005 年 12 月 28 日から 2006 年 12 月 7
日) では St. 1 (5m) と St. 2 (10m) との 2 点、
期間 II (2006 年 12 月 7 日から 2007 年 7 月 26 日)
では St. 1 (5m) と St. 5 (30m) との 2 点しか観



Fig. 6. Temporal variations of bottom temperature at St, 1 and St, 2 in the period I (from December 28, 2005 to December 7, 2006). Temperature values are shown as deviations from the reference curve shown in Fig. 5. Full lines indicate the temperature at St, 1 and dotted lines that at St. 2. Thin lines indicate the daily mean, and thick lines the 15 days running mean of the daily values. Temperature in °C is taken in ordinate, and the day number counted from January 1, 2006 is taken in abscissa.



Fig. 7. Same as in Fig. 6 except for St. 1 (full lines) and St. 5 (dotted lines) in the period II (from December. 7 to July. 26, 2007). The day number is counted from January 1, 2007.

測されていない。それぞれの日平均観測値(細線) および15日間移動平均値(太線)について、基 準季節変化(Fig. 5)からの偏差をFig. 6とFig. 7に示す。St. 1に対する曲線を実線で、St. 2ま



Fig. 8. Temporal variations of bottom temperature at St, 1 through St. 8 in the period III (from July 28, 2007 to January 30, 2008). Temperature values are shown as deviations from the reference curve shown in Fig. 5. Temperature in °C is taken in ordinate, and the day number counted from January 1, 2007 is taken in abscissa (it is reset to 0 on January 1, 2008). Thin curves indicate 4 inshore stations (dotted and dashed line is used for St. 1, dashed line for St. 2, dotted lines for St. 3 and full lines for St. 4). Thick curves indicates 4 offshore stations (dotted and dashed line for St. 5, dashed line for St. 6, dotted line for St. 7, and full lines for St. 8), respectively.

たは St. 5 に対する曲線を点線で示す。これらの 図では、それぞれ、2006 年 1 月 1 日、2007 年 1 月 1 日を 0 とした日番号を横軸にとっている。水 温差の値の範囲や、観測期間の違いから、両者の 縦軸と横軸双方のスケールが違っていることに留 意されたい。

Fig. 6 と Fig. 7 の図の日平均値の曲線には、 短周期の変動が見られる。しかし、St. 1 と St. 2、 あるいは St. 1 と St. 5 を比べた時、短周期変動 間にはほとんど相関は認められない。15 日間移 動平均した曲線にも若干のやや長い短周期変動が 残っている。この変動については 2 地点間である 程度の相関が認められるが、対応関係は明確とは 言えない。季節変動に注目しているここでの議論 では、15日移動平均値の変動を議論の対象とする。

期間 I (Fig. 6)を見ると、15 日移動平均が利 用できる 2006 年 1 月 5 日(日番号 4)から 2 月 14 日(日番号 44)までの間は St. 1 の水温が St. 2 よりも低い。一方、2 月 26 日(日番号 56)か ら 10 月 2 日(日番号 274)までの間は St. 1 の方 が高い。その後、11 月 13 日(日番号 316)以降 になると、再び St. 1 の水温の方が低くなる。こ の岸側の水温が低い傾向は、そのまま期間 II の 初期にも引き継がれる。期間 II(Fig. 7)では 2007 年 3 月 9 日(日番号 67)まで St. 1 の水温 は沖側の St. 5 より低い。3 月 17 日(日番号 75) を過ぎると、St. 1 の水温は St. 5 より高くなっ



Fig. 9. Vertical temperature profiles measured with STD in the periods II (top figures) and III (bottom figures). Shown temperature range and interval of vertical lines are variable: range is from 2.0 to 7.0°C and interval is 1.0°C in top left figure (May 26, 2007), from 7.0 to 15.0 °C and 2.0°C in top middle figure (Jul. 26, 2007), from 12.0 to 16.0°C and 1.0°C in bottom left figure (October 1, 2007), from 6.0 to 8.0°C and 0.5°C in middle bottom figure (June 31, 2008), and from −1.0 to 0.0°C and 0.2°C in bottom right figure (January 30, 2008), respectively. Depth range is from 0 to 61m for all figures. Thin curves indicate 4 inshore stations (dotted and dashed lines are used for St. 1, dashed lines for St. 2, dotted lines for St. 3 and full lines for St. 4). Thick curves indicate 4 offshore stations (dotted and dashed lines for St. 5, dashed lines for St. 6, dotted lines for St. 7, and full lines for St. 8), respectively.

ている。すなわち、両年とも初冬期から厳冬期に かけて(以下では簡単のため「冬季」と呼ぶ)深 い方、沖側の水温の方が高くなり、春先から秋に かけて(以下では「夏季」と呼ぶ)は深い方、沖 側の水温の方が低くなる。

温度成層が発達する亜熱帯域であるならば、水 温は深さとともに低下するのが普通である。高緯 度では、深さと共に水温が降下しても、塩分鉛直 分布の効果で安定化されることが起こりやすい。 しかし、現在検討している三里浜沖の状況は、次 に論じる STD 観測の結果が示すように、水温・ 塩分の鉛直構造が主役を演じてはいなかった。表 1に示すように期間 II の期間中に、5月28日 (日番号 147) と7月26日(日番号 176、期間 II の終わり)に STD 観測を実施している。この測 定結果をふくめ、期間 II と期間 IIIにおいて得ら れた水温の鉛直分布を Fig. 9に示す。期間 II に とられたものが上段の2つである。この両観測は、 St. 1の水温が St. 5の水温より高い時期(「夏季」) にとられている。この両図ともに 15m 以浅に明 瞭な水温躍層が現れている。したがって、水深の 浅い方が高温であることには、水温躍層が寄与し ていることが考えられる。しかし、15m より深 い躍層の下側では事情が異なる(St. 5の水深は 30m)。そこでは等温層が発達しており水温の鉛 直勾配は小さい。この等温層の各測点の水温を比 較すると、水温は沖に向かうほど低下する。すな わち、海底水温の沖方向への減少は、水温の鉛直 勾配に起因するのではなく、主として水温の岸一 沖方向の水平水温勾配に起因していることが示唆 されている。

ただし、水温鉛直分布の曲線は、互いに交差す る場合もあり、複数の測点の曲線がほとんど重な る場合ある。また、鉛直プロファイルは瞬間値で あるのに対して、底層水温値は15日間の移動平 均であるから、底層の水温記録と単純に比較する ことはできない。

#### 4-2. 期間 III における水温変動

期間 III (2007 年 7 月 28 日~2008 年 1 月 30 日) では、2007 年 12 月 1 日 (日番号 334) 以降では St. 6 の記録が得られていないが、ほぼ全 8 測点

での水温記録が得られている。各測点の底層水温 の変化を、基準季節変化(Fig. 5)からの偏差と して示したのが Fig. 8 である。最も沖の St. 8 の 海底水温(太い実線)に注目してみよう。その水 温偏差値は11月11日(日番号314)に0となり、 それ以前の期間では8点の中で最低の水温値を示 しており、それ以後ではほぼ最高の水温値を示し ている。一方、St.1の水温値は、11月11日の前 では最高値を示し、その後では最低値を示してい る。したがって、前節で述べた「夏季」で海底水 温は沖に向かって低下する傾向は期間 III におい ても明らかに認められる。ただ、St. 5の水温 (太い一点破線)がSt.4の水温(細い実線)より 高くなっていることに見られるように、水温は沖 向きに単調に低下しているわけではない。この 「夏季」の底層水温の変化特性は、次の4つの測



Fig. 10. Temporal variations of bottom temperature at St. 1 through St. 5 in the period IV (from January 30 to December 29, 2008). Temperature values are shown as deviations from the reference curve shown in Fig. 5. Thin dotted and dashed line is used for St. 1, thin dashed line for St. 2, thin dotted lines for St. 3 and thin full lines for St. 4, and thick dotted and dashed line for St. 5, Temperature in °C is taken in ordinate, and the day number counted from January 1, 2008 is taken in abscissa.



Fig. 11. Same as in Fig. 9, except for the period IV (April 3 to August 28, 2008). Temperature range is from −0.4 to 0.8°C and interval of vertical lines is 0.2°C in top left figure (April 3, 2008), from 0.0 to 6.0 °C and 1.0°C in top middle figure (April 30, 2008), from 3.0 to 6.0°C and 1.0°C in top right figure (May 28, 2008), from 5.0 to 9.0°C and 1.0°C in left bottom figure (June 7, 2008), from 7.0 to 13.0°C and 1.0°C in bottom middle figure (July31, 2008), and from 12.0 to 15.0 °C and 1.0 in bottom right figure (August 28, 2008), respectively.

点グループ、(1) St. 1~St. 2、(2) St. 3~St. 5、 (3) St.6、(4) St.7~St.8、に分けられる。Fig. 1の地形図から分かるように、St.4より岸側は 卓越する西向流に対して落石岬の影となる部分に あり、St. 5 が境界で、St. 6~St. 8 はほとんど落 石岬の影響を受けない地点と考えられる。複数の 測点を含むグループ(1)、(2)、(4)のそれぞれ のグループ内での水温差は非常に小さい。グルー プ(2) とグループ(4) との間の水温差は大きい が、その間のSt.6(グループ(3))の水温は、 短周期の振動に伴って(2)と(4)の水温の間を 往復する形になっている。(2)と(4)の間に水 温フロントが存在し、短周期の振動に伴って、こ のフロントがSt.6の位置を繰り返して通過した ものと考えられる。このようなグループ分けを行 い、グループ内の小さな水温差を無視すると、水 温は沖に向かってほぼ単調に低下している。11 月11日(日番号314)以降については、St.3の 水温が12月後半から著しい高温を示すのを除け ば、St. 7とSt. 8が高い水温を、中間のSt. 2~St. 5が中位の水温を、最も岸よりのSt. 1が 低い水温を示している。この場合も単調に水温が 沖に向かって上昇しているわけではないが、グルー プ分けをして見ると、沖方向に水温が上昇すると いう「冬季」の特性を見てとれる。各測点におけ る水温値偏差は1月に入ると低下を始め、期間 IVの2月(Fig. 10)になると全て負になり、そ のまま次の「夏季」の状態につながってゆく。こ の年の「冬季」の終期は2月頃と厳冬期に起こっ ている。

STD 観測は期間 III の「夏季」においては、 2007 年 7 月 26 日と、2007 年 10 月 1 日の 2 回行 われている。7 月 26 日については期間 II と III の境目で期間 II の章ですでに述べた。10 月 1 日 の結果は Fig. 9 下段左に示してあるが、39m 以 深で St. 7 と St. 8 のプロファイルが、20m 以浅



Fig. 12. Same as in Fig. 9, except for the period from September 26, 2008 to February 26, 2009 (period IV and period V). Temperature range is from 11.0 to 16.0°C and interval of vertical lines is 1.0°C in top left figure (September 26, 2008), from 9.0 to 13.0°C and 1.0°C in top middle figure (October 29, 2008), from 6.0 to 9.0 °C and 1.0°C in top right figure (November 26, 2008), from 3.0 to 5.0°C and 1.0°C in left bottom figure (December 29, 2008), from 0.0 to 2.0°C and 1.0°C in bottom middle figure (January 29, 2009), and from 1–1.0 to 0.0°C and 1.0°C in bottom right figure (February 26, 2009), respectively.

で St. 4 と St. 5 のプロファイルが、それぞれ重 なっているが、沖方向に水温が減少する傾向は明 瞭に認められる。

「冬季」の時期にも 2007 年 12 月 1 日(日番号 334) および 2008 年 1 月 30 日(日番号 29)の 2 回の STD 観測が行われている。観測された水温 鉛直プロファイルを Fig. 9 下段中と下段右にそ れぞれ示す。「冬季」の水温プロファイルは、各 測点において等温層が「夏季」よりも発達してい る。この等温層部分あるいは St. 6、St. 7、St. 8 では等温層より下の部分で、水温が沖に向かって 上昇する傾向が明瞭に見られ、「冬季」の底層水 温が沖に向かって上昇することの原因が、水温の 水平勾配に起因することを示している。

期間 IV (2008 年 1 月 30 日~12 月 29 日) では、

初めの4月3日までのSt.5を除き、ほぼSt. 1~St.5までの記録が得られている。各観測点で の、海底水温の基準季節変化(Fig.5)からの差 をプロットしたのが、Fig.10である。St.5にお いて15日移動平均値が利用できるのは5月8日 (日番号128)以降である。

期間 IV における St.3 における底層水温の変 化は他の測点の変化に比べて特異である。4 月中 頃(日番号 105)までは、St.1の水温とほぼ同 じ値を示しているが、その後水温はどんどん降下 していき、以降は St.5の水温に近い低い値で推 移していく。その原因は分からないが、全体の特 性を見るとき、4 月中旬以降に異常に低い水温値 を示す St.3の水温は考慮しないでおく。

期間 III の終わり頃から各測点の水温が低下しているが、期間 IV に入って、2月1日(日番号32)には、観測されている4測点全てで水温偏差

は負になっている。4月中頃までの部分で水温は 沖に向かうほど降下する「夏季」の状態になって いたと考えられる。ただ、4月のSTD観測によ ると、30m水深に非常にシャープな水温躍層が見 られており、少なくとも4月において、岸よりの 4測点のデータから「夏季」の状態にあったと結 論することは困難である。この現象については後 に6章で論ずる。

St. 5の値が利用できる4月25日(日番号115) から9月末(日番号270)の期間では、異常な St. 3を除き、St. 1からSt. 5まで水温は沖向き に単調に低下する典型的な「夏型」が現れている。 それ以後も、St. 4とSt. 5の水温を比較してみ ると10月末(日番号300前後)までSt. 5の水 温の方が低く「夏季」の特性が認められる。それ 以後ではSt. 4とSt. 5の水温カーブは、ほとん ど重なってしまうが、他の岸側の3点の水温より は明らかに高く、「冬季」の特性が見られる。こ の年の「夏季」から「冬季」への移行は、10月 下旬頃に起こったと考えられる。

期間 IV とそれに続く期間 V において、2008 年4月3日から2009年2月26日までの約1年間 の間にほぼ1ヶ月の間隔で12回のSTD 観測を 実施することができた(Table 1)。その結果を Fig. 11 と Fig. 12 に示す。Fig. 11 の上段左の 4 月3日の水温プロファイルには明確なステップ構 造が現れており、上段中央の4月30日のプロファ イルにもその影響が残っている。このステップ構 造については6章で改めて論じる。2008年5月 28日 (Fig. 11上段右)から、10月 29日 (Fig. 12 上段中)までが、「夏季」の状態の時にとられ たものであり、2008年11月26日(Fig. 12上段 右)から2009年2月26日(Fig. 12下段右)ま でが「冬季」にとられたものである。STD 観測 は、全測点で実施されているので、海底水温が得 られなかった St. 6 から沖の情報も得られている。 「夏季」と「冬季」の全ての水温プロファイルに、 等温層の発達が見られる。この部分に注目すると、 「夏季」では測点が沖に向かうほど水温が低下し、 「冬季」では上昇する傾向がみとめられる。

5月28日のSt.3、St.2のプロファイルを見 ると底層に沿って低温水が侵入してきている。こ のため、底層近くでSt.3、St.2のプロファイル はSt.4のそれと交わっている。このような現象 が、水温の沖向きの低下が単調に起こらない原因 の1つであろう。

#### 4-4. 期間 V における水温変動

期間 V (2008 年 12 月 29 日~2009 年 5 月 13 日) では St. 2 と St. 3 の記録しか得られていない。 両測点での海底水温の基準季節変化(Fig. 5)からの偏差値の変動を Fig. 13 に示す。この図では、2008年2月5日以降の期間 IV の変化も含めてある。時期 V の少なくとも 2009年2月頃までは、他の年であれば、「冬季」に当たる期間であるが、全期間にわたって、沖合の St. 3の方が低温になっている。この傾向は、2008年の期間 IV の「夏季」の状態がそのまま続いているように見える。

しかし、STD 観測の結果を見ると、先に述べ たように 2009 年 11 月 26 日 (Fig. 12 上段右) には「冬季」の水温構造になっていた。2008年 11月26日ではSt.2の水温プロファイルは、St. 3とSt.4のプロファイルの間にあり、Fig.13に 示された沖側の St. 3の水温が低かったことと整 合性をもってはいる。しかし、2008年12月29 日 (Fig. 12 下段左) と 2009 年 2 月 26 日 (Fig. 12段右)では、プロファイルの等温層の水温は 沖に向かって上昇しており、沖側の方の水温が高 い「冬季」の特性を示している。2009年1月29 日の観測結果(Fig. 12下段中)は特異であり、 St. 6、St. 7、St. 8 の 3 つのプロファイルは約 30mの深さで互いに交差している。この3測点 で交差点より上では水温は沖向きに上昇しており、 下では沖向きに低下している。これは上層では 「冬季」の型であるが、下層では「夏季」の型を 示していることになる。また、St. 1~St. 5まで のグループは、その中で見る限り沖向きに水温が 上昇しているが、より沖側の3点の水温よりも著 しく高い。いわば、「夏季」の特性と、「冬季」の 特性が入り混じった形になっている。このように STD 観測で得られた水温構造は非常に変動して いる。これに対して、底層水温2測点の関係は長 期にわたって維持されている。<br />
期間 V において は底層水温の変動特性と、STD 観測の結果とを 結び付けるのは困難である。しかし、期間 V に おける STD 観測の結果は、2009 年1月 29 日の 複雑な構造を除外すると、11月以降、他の年と 同様に、「冬季」の状態にあったことを示してい る。

#### 5. 得られた結果の総括と沖合海況との関連

5-1. 水温の沖向きの水平勾配

三里浜沖の水深 60m 以下の極沿岸域の海況の 季節変化を調べてきたが、その特徴の1つに、表 層の季節水温躍層の部分を除くと、季節を問わず 等温層が発達していることがあげられる。この等 温層の水温は沖に向かって降下していく時期と、 上昇していく時期があり、ここでは前者を「夏季」、 後者を「冬季」と定義した。

三里浜を含む道東沿岸域の沖側には、道東沿岸



Fig. 13. Temporal variations of bottom temperature at St, 2 and St. 3 in period V (from December 29, 2008 to May 13, 2009). Variations at these points in period IV is also shown after February 5. Temperature values are shown as deviations from the reference curve shown in Fig. 5. Full lines indicate the temperature at St, 2, and dotted lines that at St. 3. Temperature in °C is taken in ordinate, and the day number counted from December 31, 2008 is taken in abscissa.

流(沿岸親潮・道東暖流)が流れており、冬季に は低温・低塩分の水(沿岸親潮)が、夏季には高 温・高塩分の水(道東暖流)が、岸に沿う形で南 西方向に流れている。その出現状況の季節変化に ついては永田ら(2009a)が、その水塊の季節変 化については永田ら(2009b)が詳しく検討して いる。永田ら(2009a)は、水塊の季節変化を調 べるのに、50m 深の水温・塩分を調べることを 推奨している。この論文で扱っているのは極沿岸 部ではあるが、最も沖の観測点 St. 8の水深は 60m に達しており、沖合の流れとの関連をある 程度は考察できるであろう。道東暖流の内部では、 水温は沖向きに降下しており、沿岸親潮の内部で は水温は沖向きに上昇している。「夏季」の沖向 きの水温勾配は道東暖流のそれに連続する形になっ ており、「冬季」の水温勾配は沿岸親潮のそれに 連続する形になっている。これまでの議論で、春 後半から夏の期間を「夏季」、秋後半から冬の期 間を「冬季」と呼んできたのは、夏を中心に現れ る道東暖流と、冬を中心に現れる沿岸親潮を意識 したためでもある。

5-2. 「冬季」・「夏季」の出現時期と沿岸親潮・ 道東暖流の出現時期

永田ら(2009a, 2009b)は、釧路水産実験場の 2か月おきの観測資料を用いて、2月~6月が沿 岸親潮の時期であり、8~12月が道東暖流の時期 であることを示している。また、沿岸親潮に関し ては、1月には発生していると一般に考えられて いる。

4章の議論をもとに、「冬季」・「夏季」それ ぞれの各年でのおよその生起・消滅の時期をまと めたのが Table 2 である。4 章で論じてきたよう に、「夏季」から「冬季」へ、あるいは「冬季」 から「夏季」への遷移の仕方は、年によって多様 である。しかし、上述の各期間についての議論の 中で考察したように、「冬季」、「夏季」の始まり と終わりのおよその時期を決めることができる。 Table 2 では「冬季」、「夏季」の始期と終期を月 の上旬、中旬、下旬あるいは何月頃という形で示 してある。(この表で2007年秋から2008年に至 る欄で、「夏季」の始まりを1月下旬としている が、4月に、「冬季」とも「夏季」ともつかない 構造が見られたので、その後を夏季の始まりと見 れば、5月中旬が始まりとすることもできる。こ こでは1月末にすでに「夏季」的な構造が現れた

	"winter season"		"summer season"				
	srart	end	srart	end			
$2005 \sim 6$		middle of February	late February	early November			
$2006 \sim 7$	middle of November	early March	middle of March	middle of November			
2007~8	late October	early February	end of January	around November			

early February

Table 2. Starting time and ending time of "winter season" and of "summer season".

#### ことを重視している。)

 $2008 \sim 9$ 

Table 2 は「冬季」は 11 月頃に始まり 2 月頃 に終わり、「夏季」は 2~3 月頃に始まり、11 月 頃に終わることを示している。この「冬季」・ 「夏季」の出現時期と、上に述べた沿岸親潮・道 東暖流の出現時期との間にはかなりのずれがあり、 沿岸親潮の始まりを 1 月とすると「冬季」の始ま りは 2 か月程度ほど先行し、「道東暖流」の始ま りは 2 か月程度ほど先行し、「道東暖流」の始ま りた 6 月とすると「夏季」の始まりは 4 か月ほど 先行していることになる。ただし、2 か月、4 か 月といっても、その決定の精度は高いとはとても 言えない。ここでは、「冬季」・「夏季」の出現 時期は親潮・道東暖流の出現時期より早く、位相 のずれは約 3 か月であると一応結論しておく。

#### 5-3. 測点間に見られる位相差

岸一沖方向の位相の遅れは、三里浜沖の測点間 にも見られる。例えば、Fig. 7 において、2007 年2月から3月に「冬季」から「夏季」に移る時 期、St. 1 と St. 5 の水温偏差値は互いに平行す る形で減少するが、偏差値が0 になるのは岸側の St.1 の方が早い。Fig. 8 の 10 から 11 月、Fig. 10 の 9 から 10 月の「夏季」の終末期に、各測点の 水温偏差の曲線は平行する形で上昇するが、これ らの曲線が0を切る時間は岸よりの測点の方がは 早く、沖の測点はど遅くなっており、岸に近づく ほど位相が先行している。

季節変動を含めた 2007 年の生の水温変化を St. 5 ~St. 8 までの沖合 4 測点について、Fig. 14 に太線で示す。上から下にSt. 5、St. 6、St. 7、 St. 8 の順に示すが、各図に比較のためSt. 1 の 水温変化を細線で示してある。St. 5 では年間を 通して観測が行われており、5 月の初め頃から 8 月の初めごろまで、水温はSt. 1 に比べて水温が 低く、「夏季」の状態が生じていたことが分かる。 St. 6 以下では「夏季」の始まり部分の観測値は 得られていないが、沖の測点ほど水温値が低下し ていく様子が見られる。「夏季」の終わりの時期 に注目すると、測点の深さが増大するほど遅れる 傾向が認められる。最も沖のSt. 8 では 11 月に なっても、僅かであるが St. 1 よりも低い水温値 が認められる。 このような、測点間で生じる岸近くほど位相が 先行するという現象は、「冬季」・「夏季」の出 現時期が親潮・道東暖流の出現時期より約3か月 先行することを直接説明するものではないが、関 連する現象として興味あるところである。

#### 5-4. 沿岸親潮水の起源について

従来から沿岸親潮は、その水の低温・低塩分性 からその起源をオホーツク海の海氷の融解水に求 める見解がある(例えば、大谷、1971)。その根 底には、沿岸親潮を涵養するに十分な量の一定の 水塊特性を持つ低温・低塩分水が、海氷の融解で 作り出されることを前提としており、そのような 水がオホーツク海から流出してくると考えている。 しかし、均質な水が流出してきたとすると、三里 浜の極沿岸域まで水温の水平勾配を持つような構 造を作り出すことは考えにくい。

永田(2009)は道東海域を30分メッシュの領 域にわけ、過去のデータからそれぞれの領域にお ける水温の季節変化を調べている。42°N線沿 いに東方へ並ぶ5つの領域 AI、AII、AIII、 AIV、AVの季節変化を比べると、2~3月の沿 岸親潮期の最低水温は東に進むほど高くなる傾向 を示す。また、8・10月の道東暖流期の水温は東 に進むほど低くなる傾向を示す。この水温の東西 勾配の特性が、東西に圧縮された形で道東沿岸域 にもたらされるとすると、道東沿岸流の構造を説 明できる。今後さらにアリューシャン列島南岸沿 いの海況を調べる必要があると考えられる。池田 実(私信)は DNA の解析から、知床半島の東西 に住むハナサキガニは異なったグループに属して いることを示し、この2つのグループは数万年前 に分離されたことを示している。三里浜で見出さ れた、極沿岸域においても沖向きの水温勾配が存 在するいう特性も、根室水道等から流氷の溶融水 が直接この海域に流入してくると言う考えを否定 していると考えられる。最近、根室水道が浅く、 幅も狭いため、沿岸親潮水が、オホーツク海の水 の直接的な流入によって形成されることに対して、 否定的な見解が示されるようになってきている。 特に Oguma et al. (2008) は、化学物質の分析 から、同様の結論を得ている。次に述べる 2008



Fig. 14. Variations of bottom temperature measured at St. 5 through St. 8 (from top to bottom) in 2008. Thin curve drown in each figure shows temperature variation at St. 1.

年4月3日に観測された事象も、海氷の溶融水が 沿岸親潮を作り出せないことを示唆している。

#### 6. オホーツク海起源の海氷の溶解水

2008年の3月、オホーツク海の流氷が大量に 根室海峡を通って太平洋に流出してきて、釧路周 辺の海岸におびただしい流氷が着岸した。このよ うな流出は数年に1度くらいしか起こらない現象 である。三里浜沖の観測域にも、3月3日に多量 の流氷が回遊してきて、沖側の4つの観測点 (St.5からSt.8)の底層水温測定システムが流 失した。2008年4月3日に復旧作業を行い、そ の際にSTD観測を行っている。その時の水温プ ロファイルはFig.11の上段左に示すが、30m水 深付近に非常にシャープな水温躍層が認められる。 この水温躍層はシャープな塩分躍層をともなって おり、最も躍層が発達している St. 7 では、31m 深では水温 0.08℃、塩分 32.25 であるのに対して、 32m 深では、水温-0.11℃、塩分 32.41 と急変し ている。この躍層から表層 10m の季節躍層季節 躍層底部の間には水温 0.1~0.2℃、塩分が 32.2 前後の等温・塩分層が見られる。この層の水は、 その低い塩分値から考え、太平洋に流出した海氷 の溶融水と推定される。躍層の下側では、St. 6 とSt.8の水温プロファイルがほぼ重なりあって おり、St. 7の水温値がこれより低温側にある。 したがって、「冬季」的な状態であったのか「夏 季|的状態であったたのかの判定できない。しか し、流氷の溶融水の影響を受けていないと考えら れる下層の水温が、流氷の溶融水よりも低温であ ることは注目すべきであり、少なくとも3~4月 においては、溶融水はそれほど低温の水を作りえ

ないこと示している。1例だけでは結論を得られ ないが、沿岸親潮の水が流氷の溶融水ではありえ ないことを示唆していると考えられる。

4月30日に観測された水温プロファイル (Fig. 11上段右)では30m以浅に温度躍層が認 められる。これは、溶融水による躍層の名残であ る可能性が高い。そうだとしても、4月3日に見 られた流氷溶融水層が4月30日にはほとんど消 滅していることになる。したがって、流氷溶融水 から作られた層の寿命は高々1月と短いものであっ たと推定される。量的にも流氷溶融水が沿岸親潮 を形成するとは考え難い。

#### 7. おわりに

この論文では、8地点に設置した海底水温計測 装置による水温の連続測定の結果と、STD 観測 の結果を報告した。ハナサキガニが生息する三里 浜沖の環境水温の周年的な変化特性を明確にする ことを目的に行われたものである。ここでは水温 構造の季節変化に焦点をあて、塩分構造・密度構 造の検討は次の論文で扱う。3年間の成果は、初 期の目的を十分果たすものであると考える。この 結果は、ハナサキガニの生態・行動の研究に十分 な指針を与えうるものと確信している。さらに、 ここで見出された三里浜沖に現れる特異な季節変 化特性、「夏季」および「冬季」の水温鉛直プロ ファイルには表層を除き等温層が発達しているこ と、この等温層の水温は、「夏季」では沖に向かっ て水温が低下し、「冬季」では沖に向かって水温 が上昇すること、そうしてこれらの水温の水平勾 配は、沖合を流れる道東沿岸流(沿岸親潮と道東 暖流)の特性にそのままつながっていると考えら れること等の知見は、重要な成果と考える。また、 「夏季」・「冬季」の発生時期は、沿岸親潮・道 東暖流の発生時期に約3か月先行する。極沿岸水 域内でも、「夏季」・「冬季」の現象は、岸近く が先行し、沖方向に遅れるという位相差が認めら れる。また、流氷の溶融水はそれほど低温(0℃ 以下)にならないことが示唆された。これらの知 見は、道東沿岸流の起源水を求める研究に貴重な 示唆を与えるものと考えられる。

#### 引用文献

- 大谷清隆(1971):噴火湾の海況変動の研究Ⅱ 噴火湾に流入、滞留する水の特性。北大水 産彙報、22,58-66.
- 永田豊(2009):根室周辺海域の海況の季節変化. うみ (La mer), **46、**135–140.
- 永田豊・小熊幸子・長瀬桂一・相川公洋・田中伊
   織・中多章文・夏目雅史(2009a):道東沿
   岸流(沿岸親潮・道東暖流)の季節変化。うみ(La mer), 47, 29–42.
- 永田豊・小熊幸子・長瀬桂一・相川公洋・田中伊 織・中多章文・夏目雅史(2009b):道東沿 岸流の水塊の季節変化。うみ(La mer), 47, 67-73.
- Oguma, S., T. Ono, A. Kusaka, H. Kasai, Y. Kawasaki, and T. Azunaya (2008) : Isotopic Tracers for Water masses in the Coastal Region of Eeastern Hokkaydo. J. of Oceanography, 64, 525–539.

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# Marking of tiger shrimp *Penaeus monodon* (Fabricius) juveniles: Comparison among inexpensive tagging options

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Abstract: Three low-cost marking methods (staining with food color, uropod trimming, and Tbar tagging) were tested on 2-mo old juvenile tiger shrimps Penaeus monodon with  $8.42 \pm 0.1$ mm mean carapace length (CL) for 8 weeks. Together with an unmarked control group, marker retention and effects on shrimp' s survival and growth were monitored. There was no significant difference in specific CL growth rates among treatments (p>0.05). Survival was not significantly different among staining (60%), uropod trimming (51%), and control (58%) throughout the experiment, but T-bar tagging showed significantly lower survival from week 6 until 8 (33%) (p<0.05). Marker retention was significantly highest for T-bar tags (100%) at the end of 8 weeks, followed by uropod trimming at 65%, while staining was already 0% from week 3. Food color stains are poor shrimp markers because of weak retention. T-bar tags are not effective markers despite having excellent tag retention because of low shrimp survival after 6 weeks. Uropod trimming is a more practical option given that shrimps showed comparable survival with the control group and marker distinction through unique uropod regrowth was relatively high. Further work on modifications on these methods is needed to increase efficiency while maintaining lower cost. This study is viewed to have practical applications for community-based shrimp stock enhancement monitoring.

Keywords: molting, shrimp tagging, stock enhancement, tag retention

#### 1. Introduction

World capture fisheries is seen to have become static in the recent years and signs of decline have been reported in some countries (FAO-FIES, 2008; FAO, 2009). In the Philippines, artisanal fish capture production quality and quantity have declined in the past years from various fishing grounds (MINES *et al.*, 1986; LIM *et al.*, 1995; KATON *et al.*, 1998; EVASCO, 2000; FERNANDEZ *et al.*, 2000; PALMA *et al.*, 2002; NEDA, 2005). Specifically for example, the Batan Estuary in the northern Panay Island in central Philippines was known for abundant wild shrimps, including the highlypriced tiger shrimp *Penaeus monodon*, but decreasing catch in the recent decades has become very evident (ALTAMIRANO, 2007; ALTAMIRANO and KUROKURA, 2008). The steady loss and overexploitation of local fishery resources directly affects the livelihood of the already poor fishers.

Shrimp restocking can be an effective tool in enhancing wild stocks (DAVENPORT *et al.*, 1999; WANG *et al.*, 2006) and, at the same time, in alleviating poverty among subsistence fishers through a direct increase in shrimp catches (ALTAMIRANO and KUROKURA, 2008). While the practice of fish stock enhancement has been done since the 1960s, crustacean stock enhancement programs especially for shrimps, have been limited (BELL *et al.*, 2005). For example, shrimp restocking projects were implemented for *Penaeus japonicus* in Japan (HAMASAKI

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Cracical	Montron on Tor	Setup	Size	Survival	Reten-	Defenence
species	Marker or Tag	(Duration)	(mm CL)	(%)	tion (%)	Reference
Neocaridina denticulate (S)	Laboratory	Trypan blue/red	15-30 BL <sup>2</sup>			Niwa <i>et al.</i> , 1998
Penaeus monodon (S)	Tank (5-6 mo)	Streamer	15 18	31-35 37-70	100 100	Benzie et al., 1995
monouon (b)	Pond (5-6 mo)	Streamer	15 15 18	30-44 44-59	100	
P esculentus (S)	Tank (~2 mo)	Streamer	12-19	~80		Hill and Wassenberg 1985
P nleheius (S)	Pond $(2 \text{ mo})$	Streamer	17-27	35-48		Montgomery and Gray 1991
P. monodon (S)	Tank (2 mo)	Streamer	16-18 18-21	90 47		Primavera and Caballero, 1992
P. esculenlus (S)	Tank (2-3 mo)	Streamer	12-17 15-20	75-90 90-93		Wassenberg and Kerr, 1990
P. merguiensis(S)	Tank (2-3 mo)	Streamer	11-17	85-100		
Munida rugosa (L)	Tank (2 mo)	Streamer	<27 to >32	29	100	Claverie and Smith, 2007
0		T-bar tags		52	100	
		V.I. Elastomer		95	100	
Macrobrachium	Tank (2.5 mo)	V.I. Elastomer	$0.01 \ \mathrm{BW^3}$	100	79	Brown <i>et al.</i> , 2003
rosenbergii (S)		V.I. Alpha	$< 5.5 \ \mathrm{BW^3}$	~99	60	
Jesus	Tank (37 mo)	T-anchor, dart	90-104	43-51	78-82	Montgomery and Brett, 1996
verreauxi (S)		Uropod trim		47	77	
Homarus	Trays (3 molts)	Micro wire	5-8	82	97	Linnane and Mercer, 1998
gammarus (L)			12-16	97	97	
		V.I. Elastomer	5-8	68	100	
			12-16	97	100	
		Rostrum cut	5-8	80	0	
		<b>D</b> 11	12-16	99	0	
		Branding	12-16	57	0	
		0	16-19	90	0	
D :(0)	<b>T</b> 1 (9 )	Streamer	16-19	97	100	C 1: / 1 100C
P. vannamei (S)	Tanks (3 mo)	V.I. Elastomer	$< 3.92 \text{ B W}^{-1}$	99-100 100	91-93	Godin <i>et al.</i> , 1996
Litoponaque	Tople (~2 mo)	VI Alpho	21-41 D W 2.7 B W <sup>3</sup>	100 81 09	95 02 08	$\Delta roo at al 2003$
vannamei (S)	Talik ( 2 1110)	v.i. Alpha	2.7  DW $21.5 \text{ BW}^3$	68 74	92-90 80 83	Aice et ut., 2005
bunnumer (b)	Pond (~1 mo)		21.0  DW $2.7 \text{ RW}^3$	90-93	93_97	
	1 0110 ( 4 1110)		$21.1 \text{ BW}^{3}$	80-88	83-85	
Callinectes	Lah (1.5 mo)	V.I. Elastomer	6-25	~58-60	~81-84	Davis <i>et al.</i> $2004$
sapidus (C)		Micro wire	6-25	~47-60	~92-93	
Litopenaeus setiferus (S)	Tank (1 mo)	Micro wire	30-90 BL <sup>2</sup>	81-100	95-100	Kneib and Huggler, 2001
Litopenaeus vannamei (S)	Lab (4 mo)	Uropod cut	13-16 28-29	~98 ~63	0 ~88	Leano and Liao, 2006
		Uropod trim	13-16	~99	~91	
		-	28-29	~53	100	
Penaeus	Lab (~2.5 mo)	Uropod cut	$21-73 \text{ BL}^2$	92	96	Miyajima <i>et al.</i> , 1999
<i>japonicus</i> (S)	Sea (12 mo)	Uropod cut	$35-75 \text{ BL}^2$		$2.4^{4}$	

Table 1. Some studies on different marking and tagging methods for crustaceans.

1 (S) =shrimps, (L) =lobster, (C) =crab; 2 BL=Body Length (mm); 3BW=Body Weight (g); 4 recaptured

and KITADA, 2006), *Penaeus chinensis* in China (WANG *et al.*, 2006), *Penaeus monodon* in Taiwan (Su and LIAO, 1999) and Sri Lanka (DAVENPORT *et al.*, 1999), and *Penaeus esculentus* in Australia (LONERAGAN *et al.*, 2006). Although various methods and technology have been applied, the problems in monitoring still remain to be a challenge, even for a financially-capable community.

To evaluate the success or failure of shrimp

restocking initiatives, an efficient method of marking shrimps is important in sorting recaptured stocks from wild catch. The main challenge for shrimp tagging is the difficulty imposed by molting. A number of marking options for shrimps have been tested and improved since the late 1950s, to include various modes of staining, internal or external tags, and cutting of body parts (NEAL, 1969; FARMER, 1981). Table 1 summarizes some of the studies on marking and tagging of crustaceans, especially shrimps. Most of these studies use sub-adults and larger juveniles, requiring longer pre-release culture that increases cost. On the other hand, other mass shrimp restocking programs use post-larvae shrimps that cannot be marked because of their very small size. Therefore, shrimps that are to be released should be big enough to be tagged but should not require long culture periods. In this study, juvenile shrimps of about 10 mm CL or about 60 days old were marked.

The main concern in choosing a marking method or tag is its efficiency in terms of retention and minimal effects on the commodity's growth, movement and survival. It has been recommended that visual implant elastomers (VIE) and micro wire tags are the more viable options for crustaceans (GODIN *et al.*, 1996; LINNANE and MERCER, 1998; DAVIS *et al.*, 2004); however, the main limitation for these tags is the cost required for its use. Except those involving ablation of body parts, majority of shrimp marking and tagging studies use specialized branded materials that are expensive.

In addition, stock-enhancement programs are also viewed to have better efficiency when implemented and managed by its direct beneficiaries — the local communities (GARAWAY *et al.*, 2006). Although community-based stock enhancement may have positive prospects, its implementation is mostly restricted financially. Therefore, for local stock enhancement programs, the availability and cost of tags and methods are also very important considerations.

In this study, we attempted to use and compare low-cost and readily available options for tagging and marking small juvenile shrimps P. *monodon*. Instead of specialized stains, we opted to use ordinary natural food color (McCormick and Company, Inc., Maryland) that has also been described in some reports and applied to stain other taxa like nematodes (THIES *et al.*, 2002). Ordinary plastic T-bar tags (Bano'k, Japan), commonly used in tagging clothing merchandise, were also tested as a possible substitute for streamer tags and specialized T-bar tags (Hallprint Pty Ltd., Australia). Uropod trimming, discussed by various authors (MIYAJIMA *et al.*, 1999; TOYOTA *et al.*, 2003; LEANO and LIAO, 2006), was also tested because it involves no additional costs for materials.

#### 2. Materials and Methods

This experiment was a part of a series of studies on the stock enhancement of tiger shrimp *Penaeus monodon* in the Batan Estuary, Aklan, Philippines led by the Laboratory of Global Fisheries Science, the University of Tokyo, Japan. This study was conducted at the Brackishwater Aquaculture Station of the College of Fisheries and Marine Science (CFMS), Aklan State University, New Washington, Aklan, Philippines.

#### 2.1. Set-up and monitoring

Hatchery-bred tiger shrimp P. monodon porstlarvae (PL15) were acquired (approx. 1000 pcs) from a local hatchery (wild broodstock were sourced locally). These were reared until 2 mo old in hapa nets inside an open pond with half-opened gates to allow regular water change from a nearby creek. In a separate experiment, 2-month old juveniles were observed to be the optimal age of shrimps for release in the immediate local water conditions of the Batan Estuary. At this age, the study showed that P. monodon juveniles have a carapace length of about 8-10 mm. From the reared stocks, shrimp juveniles measuring 8.42  $\pm$  0.1 (s.e.) mm mean carapace length (CL) and weighing  $3.4 \pm 0.02$  (s.e.) mg mean body weight (BW) were used in the experiment. A total of 12 aquaria (140 L) were prepared with washed fine sand as substrate (2 cm deep) then filled with filtered water (120 L) from the same rearing pond and creek. Water from each aquarium was drained by 10% daily using a siphon tube. Water was refilled through individual distribution pipes connected to a central holding tank where the filtered source water was collected daily, aided by a submersible electric pump. Constant aeration was supplied using airstones powered by an electric blower. There were three replicate aquaria for each marking treatment, namely: staining, uropod trimming, and T-bar tagging. Another three aquaria were designated as unmarked control replicates. These 12 aquaria were placed adjacent to a wide window in the laboratory to allow natural diel cycles, arranged in one row and alternating among treatments.

Prior to the actual experiment, tagging trials were done as practice runs for each tagging method to achieve minimal time in marking and to reduce stress of shrimps while out of the water. In the final run, twenty shrimp juveniles were tagged/marked accordingly (as described below) and placed in each aquarium for a day to acclimate. The same number was also stocked untagged in each control aquaria. Few dead shrimps, as a possible result of tagging shock or acclimation after about 24 h was noted as immediate mortality and removed. Those that survived were further randomly reduced to 15 individuals per aquarium as the final number used in this study for the next 8 weeks. Visual observations were carried out daily. Monitoring for growth was conducted weekly in terms of carapace length measured with a digital caliper, and wet body weight using a digital top loading scale (0.01 g sensitivity). Shrimp's survival and mortality were assessed from weekly total count. Examination of marker retention and/or visibility was also done every week. The experiment lasted for 8 weeks, from 20 April to 15 June, 2008.

Finely chopped fish meat was provided as feed daily (1800 H) *ad libitum*. Uneaten food and unwanted particulates in the aquaria were siphoned-out daily during every water change. Water quality data (salinity, temperature, DO, pH) were monitored daily (0900–1000 H) using a Horiba UX-21 multi-parameter probe. A data logger (ACT-HR, Alec Electronics, Japan) was also used to monitor a detailed (10 min interval) trend in salinity and temperature.

#### 2.2. Staining

Biological staining has been used to mark shrimps since the 1950s (MENZEL, 1955; NEAL, 1969) and has evolved into using some expensive patented materials. In this experiment, however, the cost of staining agent must be affordable and locally available. Hence, the common natural food color (McCORMICK) was tested. Using a small (6.35 mm) sterile tuberculin syringe and a 30-gauge needle, a small amount (about 0.01 ml) of red food color was injected ventrally through the articular membrane of the shrimp between the 1st and 2nd abdominal segments. Stained shrimps were placed in a holding pan with aeration for about 20 min. Stains spread beneath the exoskeleton and mostly accumulate in the gills. Only those that appeared stable and survived the initial shock of staining were replaced in each aquarium (20 shrimps aquarium<sup>-1</sup>) for a day of acclimation. Then, only the final number of 15 individuals was retained per aquarium for monitoring throughout the experiment.

#### 2.3. Uropod trimming

Trimming of shrimp's uropod was chosen rather than complete removal of uropod. Trimmed uropod usually creates unique regrowths after molting that can be used as a marking indicator (LEANO and LIAO, 2006). Trimming was done using a pair of small finetip surgical scissors. Half of the left outer uropod was cut, adapting the methods described by TOYOTA et al. (2003) and LEANO and LIAO (2006). Trimmed shrimps were placed in an aerated collection pan for a few minutes before replacing in their respective aquarium. After given about 24 h to stabilize and acclimate, the number of individuals in each aquarium was reduced to 15, setting day zero of the experiment. Observations and monitoring proceeded as described above.

#### 2.4. T-bar tagging

Physically-attached tags have been extensively used for stock enhancement monitoring purposes and a number of these have been utilized for tagging crustaceans, including shrimps (NEAL, 1969; HOWE and HOYT, 1982; HILL and WASSENBERG, 1985; TEBOUL, 1993; LINNANE and MERCER, 1998; ARCE *et al.*, 2003; BROWN *et al.*, 2003; CLAVERIE and SMITH, 2007). Most of these special tags however, require considerable capital to acquire, making it impractical for low-budget studies. In this experiment, ordinary T-bar tags, commonly used in clothing's price label, were used. The plastic Tbar tags were about 0.3 mm thick and 11 mm long, giving enough room for shrimp's growth until marketable size. Specifically, the Bano'k 303XL T-bar injector gun was chosen because of its thin and long injector needle. This injector gun drives the T-bar tags from a set of cartridges.

A number of tagging locations have been used for physical tags, such as a dorso-ventral insertion of T-bar tags between the cephalothrax and 1st abdominal segment for squat lobsters M. rugosa (CLAVERIE and SMITH, 2007), and horizontal tagging of streamer tags between abdominal segments on P. monodon (BENZIE et al., 1995) primarily to minimize difficulty of shedding exuviae during molting. However, the present study adapted the methods for streamer tags on P. monodon by WASSENBERG and KERR (1990) and PRIMAVERA and CABALLERO (1992) where the tag was inserted laterally through the left side of the middle of the 2nd abdominal segment, exiting and of the body. This tagging location provided the least resistance for movement of the shrimps. WASSENBERG and KERR (1990) also found that this location has the fastest wound healing effect. The T-bar tag was injected through the guide needle with a single squeeze of the injector gun, carefully avoiding further tissue damage, and was slowly removed after the tag was locked in place. Same as the other treatments, T-bar tagged shrimps were held in aerated pans before replacing in the aquaria. The final number of 15 individuals was maintained a day after tagging and monitored throughout the experiment.

#### 2.5. Data analysis

Water quality data (water pH, DO, temperature and salinity) were subjected to two-way analysis of variance (ANOVA) with marking treatment and measurement week as fixed factors. Initial and weekly growth parameters in terms of CL and BW, as well as total count of surviving shrimps, were recorded to determine growth and survival rates, respectively. Weekly specific growth rates (SGR) were computed for CL and BW using the formula adapted from Y<sub>E</sub> *et al.* (2009):

SGR (% d<sup>-1</sup>) = 100 × (ln  $x_t$  - ln xi) / d,

where SGR is Specific Growth Rate for CL (%CL d<sup>-1</sup>) or BW (%BW d<sup>-1</sup>), xi is initial measurement for CL or BW,  $x_t$ = measurement for CL or BW at a given time t, and d = number of days between measurements. A similar measure for SGR was also used by PRIMAVERA and CABALLERO (1992) to present CL and BW growth rates of P. monodon marked with streamers tags.

Survival was represented as a percentage of remaining shrimps from the initial 15 individuals during each weekly monitoring. Marker retention and visibility was examined and recorded as a percentage from among the surviving shrimps per monitoring week.

Data were initially tested for normality using the Shapiro-Wilk test, and for homoscedasticity using Levene's test. Percentage data were further transformed to arcsine prior analyses to improve homogeneity of variances (ZAR, 1999). Then, data analysis proceeded with ANOVA. When significant differences were detected, further analyses with Tukey's Honestly Significant Difference (HSD) multiple comparison tests were performed. Comparisons were made to determine differences among treatments and control on the survival and growth of shrimps, as well as on the tag retention. Significance was established at p < 0.05. Statistical analyses were performed with SPSS statistical software version 14 (SPSS Inc., Chicago, IL).

#### 3. Results

#### 3.1. Water condition

Water pH, dissolved oxygen (DO), salinity and temperature were constantly monitored and showed no significant differences among treatments on every monitoring week (twoway ANOVA, treatment\* week, p>0.05). Average measurements (mean  $\pm$  s.d.) of these are as follows: pH,  $7.77\pm0.89$ ; DO,  $3.83\pm0.66$  mg  $L^{-1}$ ; salinity, 18.80 ± 1.77; and temperature, 26.96 ± 2.59 °C. Since aeration was sustained and water exchange was maintained, DO was relatively stable throughout the experiment, as well as pH. Some fluctuations in temperature and salinity were recorded by the data logger especially during the sustained rains in May but no drastic changes occurred that may have affected the shrimps.

#### 3.2. Shrimp's growth

Specific growth rates in terms of % CL d<sup>-1</sup> showed no significant differences among treatments and control within the 8-week period (ANOVA, p>0.05) (Fig. 1).

The trend in CL changes between monitoring weeks suggests two molting stages within 8 weeks (Fig. 1). Results confirmed rapid molting and growth within the first 2 weeks after marking, reaching an overall average CL change of 0.75  $\pm$  0.03 %CL d<sup>-1</sup> after week 1 and  $1.29 \pm 0.14$  %CL d<sup>-1</sup> on week 2. Observations also indicated that about 75% of the experimental shrimps molted within these first two weeks. On week 3, zero growth rate was recorded which was significantly lower (ANOVA, Tukey's HSD, p < 0.05) than the previous week indicative of intermolt period (Fig. 1). After week 3, an increasing growth rate was again manifested until week 6 (0.69  $\pm$  0.11 %CL d<sup>-1</sup>), highlighting the second molt stage. Although less pronounced and statistically not significant (ANOVA, Tukey's HSD, p>0.05), another carapace rigidity stage followed on week 7 as denoted by another zero average specific growth rate (Fig. 1). A slight increase ini-



Fig. 1. Specific growth rates (SGR) in terms of carapace length (%CL d<sup>-1</sup>) of 2-mo old *Penaeus monodon* juveniles marked through staining (STN), T-bar tagging (TBT), and uropod trimming (URT) together with untagged control (CON) within 8 weeks. Molting periods are shown, terminating during the carapace rigidity stages (zero growth) at weeks 3, then week 7. Similar superscripts among weeks are not significantly different (ANOVA, p>0.05). Also, values among marking methods were not found to be significantly different.

Table 2. Specific growth rate (SGR) in	n terms of BW	(mean $\pm$ s.e.,	%BW d <sup>-1</sup> ) of 2-n	no old <i>Penaeus moi</i>	10don juve-
niles marked through staining (	(STN), T-bar	tagging (TBT	.), and uropod	trimming (URT)	, including
untagged control (CON) within 8	3 weeks.				

unuagged control				
Week	CON	STN	URT	TB T
1	$3.45\pm0.38$	$3.72\pm0.73$	$3.57\pm0.34$	$3.14\pm0.52$
2	$1.80\pm0.24$	$0.58 \pm 0.45^{*}$	$3.41\pm0.26$	$1.43\pm0.22$
3	$2.75\pm0.35$	$2.28\pm0.29$	$1.44\pm0.22$	$2.07\pm0.38$
4	$1.48\pm0.27$	$0.58\pm0.49$	$2.23\pm0.72$	$2.50\pm0.28$
5	$1.37\pm0.30$	$0.82\pm1.12$	$0.35\pm0.57$	$1.01\pm0.68$
6	$0.42\pm0.46$	$-0.51 \pm 0.73$	$1.17\pm0.92$	$1.50\pm0.18$
7	$0.73\pm0.46$	$-0.79 \pm 1.00$	$0.83\pm0.33$	$-0.49\pm0.45$
8	$0.75 \pm 0.33$	$1.02 \pm 0.47$	$-0.08 \pm 1.04$	$0.82\pm0.23$

\* significant difference (p<0.05) from among treatments on the same week.

tiated again on week 8 (0.12  $\pm$  0.08 %CL d^{-1}) that may have been the start of the 3rd molting cycle.

Specific growth rates in terms of %BW d<sup>-1</sup> showed a generally decreasing trend, from an overall mean of  $3.47 \pm 0.12$  %BW d<sup>-1</sup> during the first week to only  $0.63 \pm 0.24$  %BW d<sup>-1</sup> during the last week (Table 2). Generally, no significant differences in BW-SGR were found among treatments and control throughout the study period, except only for staining at week 2.

#### 3.3. Shrimp's survival

The small initial size (mean  $CL \pm s.e.$ , 8.42  $\pm$  0.1 mm; mean  $BW \pm s.e.$ , 3.4  $\pm$  0.02 mg) of shrimp juveniles was a challenge for tagging. As a result, high immediate mortality was observed especially for staining and T-bar tagging during the practice runs as a direct effect of mishandling. However, tagging efficiency greatly increased after familiarization of the tagging procedure. During the actual experiment, immediate mortality (<1 hr after marking) was very low even for T-bar tagging (1 to 3 individuals).

Survival steadily declined and was not significantly different (ANOVA, p>0.05) among treatments up to week 5 (control, 84%; staining, 73%; uropod trimming, 71%) (Fig. 2). Although similarly not significantly different from other treatments, T-bar tagging showed a lower survival of 67% after 5 weeks. The decrease in survival continued with similar trend until week 8 for control (58%), staining (60%), and uropod trimming (51%). However, T-bar tagging showed significantly lower (ANOVA, Tukey's HSD, p<0.05) survival rates of shrimps from week 6 (47%) until week 8 (33%).

#### 3.4. Marker retention

Marker retention from survived shrimps was significantly highest (ANOVA, Tukey's HSD, P<0.05) up to the last week for T-bar tags, where 100% were retained throughout the 8week experiment (Fig. 3). However, visibility of trimmed uropods steadily declined until 65% at the end of 8 weeks, while only 14% of stained shrimps were identified as having been marked after the first week and none showed signs of staining from week 3.

For uropod trimming, marker retention was determined through abnormal regrowth of the trimmed portion (Fig. 4). The decline in marker visibility for this method was caused by normal regeneration of uropods after molting on some shrimps. In weeks 2 and 3, after the first molt, 88% of the survived shrimps (14) out of 15 individuals) showed identifiable abnormal uropod regrowths; while from week 4 until week 7 (after the second molt), only 75% of the 10 surviving shrimps from each aquarium retained abnormal uropod regrowth. Lastly, at week 8, only 65% were recognizable as having had a trimmed uropod. For this method, it is important that the observer be properly trained to differentiate an untrimmed



Fig. 2. Survival (%) of *Penaeus monodon* juveniles marked through staining (STN), T-bar tagging (TBT), and uropod trimming (URT), including untagged control (CON) within 8 weeks. Asterisks (\*) denote significant difference from among other treatments in the same monitoring week (ANOVA, p<0.05).



Fig. 3. Tag retention/visibility (%) from survived *Penaeus monodon* juveniles marked through staining (STN), T-bar tagging (TBT), and uropod trimming (URT), including untagged control (CON) within 8 weeks of laboratory experiment. Similar superscripts denote no significant difference among treatments in the same monitoring week (ANOVA, p>0.05).



Fig. 4. (A) *P. monodon* juvenile marked by uropod trimming (trimmed portion represented by dotted line). (B) After molting, abnormal regrowth from the trimmed uropod can be observed (arrow).

normal uropod and that of the regenerated uropod and that careful scrutiny is necessary during monitoring.

On the other hand, staining showed very significantly lower (ANOVA, Tukey's HSD, P<0.01) marker retention rate than the other methods even on the first week where only 14% of survivors were able to retain the red food color stain, mostly in the gills. At week 2, only 3% were observed to have a tinge of red in their body. Then, from week 3 onwards, no identifiable stain marks were observable from the experimental subjects.

#### 4. Discussion

Although some studies already recommended the use of more expensive visual implant elastomers (VIE) and micro wire tags for crustaceans (GODIN *et al.*, 1996; LINNANE and MERCER, 1998; DAVIS *et al.*, 2004; LEBATA *et al.*, 2009), the current study on the other hand, evaluated the utility of more economical options for marking shrimps. Three marking categories were tested in this experiment: staining, tagging with physical tags, and ablation of body parts. The use of common food color as a staining agent and tagging with ordinary plastic T-bar tags have never been mentioned in published literature, especially for small shrimp juveniles.

Various stains such as trypan blue (Hartman-Leddon Company, Philadelphia) and similar others have been injected to accumulate in the gills of shrimps such as Neocaridina denticula (NIWA et al., 1998). Literature search of current studies on shrimp marking by staining returned no results. In a review by NEAL (1969), he mentioned that stains appeared to have less utility as these tend to fade after a few days although, in some cases, it can last for about a week. The results in the current study also confirmed this observation. Only 14% of shrimps retained their stain after one week and were completely faded starting the third week. However, stains have practical applications for laboratory studies of a few days. The current experiment also showed that even ordinary food color can have the same short-term effectiveness as that of more expensive stains although a direct comparative experiment is needed to really prove this. In addition, proper staining with natural food color seemed to have very minimal effect on shrimp survival and growth, where current results for staining were not significantly different from the control group (Figs. 1, 2).

Physical cutting or modifying body parts such as rostrum ablation in lobsters were tested by LINNANE and MERCER (1998) but complete regeneration was only as short as three molts. Trimming of uropods in shrimps, however, showed higher retention rates since molting often resulted in distinct abnormal growths from cut or trimmed uropod in Litopenaeus vannamei and Penaeus japonicus (MIYAJIMA et al., 1999; TOYOTA et al., 2003; LEANO and LIAO, 2006). Similar results were found in the present study for P. monodon juveniles. Abnormal regrowths were noted, often characterized by a branched protruding extension from the trimmed uropod (Fig. 4). The first molt and regeneration was observed within 1 week after trimming which was similar with that of L. vannamei (LEANO and LIAO, 2006). Aside from unique regrowth, MIYAJIMA et al. (1999) also mentioned noticeable differences on coloration of the regenerated uropod of bigger (around 60 mm BL) P. japonicus but this was not clearly observed in our samples, probably because of relatively smaller sizes. Survival of shrimps for this method was high, reaching 98% for juvenile L. vannamei after 4 mo (LEANO and LIAO, 2006). In the present study, survival for uropod trimming (51%) was not significantly different from the control group (58%) after the 8-week experiment. This suggests that the relatively low survival rate may be attributed to external factors such as water quality and natural mortality, rather than a direct effect of the marking method. One noticeable immediate effect of uropod trimming on shrimps was the initial difficulty in swimming. Shrimps were observed to swim sideways because of the unbalanced trimmed uropods. However, after a few hours, some were able to correct their balance and swim upright. It is therefore recommended to allow the uropod-trimmed shrimps to stabilize for a few hours or days before actual release in the open to minimize possible susceptibility from predation.

Through-the-body external markers were widely used, especially the streamer tags, noting that larger shrimps have lower short-term or immediate mortality rate than smaller juveniles (HILL and WASSENBERG. 1985: WASSENBERG and KERR, 1990; MONTGOMERY and GRAY, 1991; PRIMAVERA and CABALLERO, 1992; BENZIE et al., 1995). Anchor tags or T-bar tags are smaller; hence, short-term mortality was significantly lower than streamer tags as used in squat lobster Munida rugosa (Claverie and SMITH, 2007) and Jesus verreauxi (MONTGOMERY and BRETT, 1996). The main advantages of external tags are high retention rates and ease in tag detection. In the present study, observations for marker retention and/or visibility were easiest for T-bar tags because of the externally protruding plastic were easy to spot. Marker retention for this method was 100% from the survived shrimps in the present study. This high retention (100%) of the similar T-bar or T-anchor tags was also true for *M. rugosa* after  $2 \mod (CLAVERIE and$ SMITH, 2007), while 78-82% was retained for shrimp J. verreauxi after a long-term study of 37 mo (MONTGOMERY and BRETT, 1996).

One concern in having tags inserted through the body is the negative effect during molting. LINNANE and MERCER (1998) observed that cast exoskeleton frequently became entangled on the streamer tags during molting of Homarus gammarus and that tagged shrimps needed more flicks to discard old exoskeleton on P. esculentus (BENZIE et al., 1995). However, this effect seems to be only minimal. BENZIE et al. (1995) also showed that time to complete molting was not affected, while molting and general growth rate was also not significantly different between streamer-tagged and untagged shrimps (HILL and WASSENBERG, 1985; BROWN et al., 2003). The same observations were also noted in the present study for P. monodon juveniles, where exuviae sometimes get entwined with the T-bar tag; however, growth rates and intermolt periods were still not significantly different than the control group or other marking methods (Fig. 1, Table 2.). P. monodon has been known to molt at an average of every 5 days during post-larval stage and about every 14 days for sub-adults (KIBRIA, 1993). For this reason, longer studies and actual release experiments employing throughthe-body markers like T-bar tags may eventually have adverse effects on survival after these multiple moltings. As shown in the current study, the main drawback in using Tbar tags was in the survival of shrimps. A significantly lower survival (47%) was recorded for T-bar tagging after 6 weeks compared with other treatments (62–73%) (Fig. 2). This lower survival was estimated to roughly coincide with the second molting of the shrimps at about 6 weeks although no further confirmation was conducted. Other studies using a specialized T-bar or T-anchor tags produced the same results, such as for M. rugosa with 52% survival after 2 mo (CLAVERIE and SMITH, 2007), and J. verreauxi with 43–51% after 37 mo (MONTGOMERY and BRETT, 1996). These high mortalities, especially after a month or so can be the effects of delayed infection and some studies applying antibiotics did not seem to have any major advantage (NEAL, 1969). WASSENBERG and KERR (1990) also observed that wounds inflicted by the inserted tags and its constant movement hinders the quick healing of the injuries and thereby increasing the chances for irritation and infection. High mortalities were also recorded during tagging of smaller shrimps (>10 mm CL) in our experiment during the practice tagging runs. During the actual experiment; however, survival rates were comparable with the control group in the first 5 weeks. For P. monodon, the minimum size of juveniles for T-bar tagging was established to be about 9-10 mm CL in this study but smaller shrimps where also successfully tagged. WASSENBERG and KERR (1990) even estimated a higher critical size limit for streamer tags for P. esculentus (18 mm CL) and P. merguiensis (17 mm CL). This means that Tbar tags, streamer tags, and similar throughthe-body tags may only be practically used on larger juveniles or sub-adults unless significant modifications on the tag size or applicator device can be developed. However, T-bar tags have practical applications for restocking studies in smaller areas that expect full recapture within a month or so.

Although the present study was done in a laboratory, natural water conditions were attempted to be simulated. Hence, only creek water was used with no other treatments made aside from filtration. In effect, changes in natural water conditions were also manifested in the aquaria through daily water exchanges. Water condition is also an important factor that affects shrimps growth and survival. At 30°C, JACKSON and WANG (1998) showed that P. monodon can grow double in weight than that at 20°C within 180 d. Abrupt changes in salinity also affects mortality although younger prawns (< 40 days post-metamorphosis) can tolerate short acclimation periods of 6 h to 3 days and can survive in low salinity water better than older animals (CAWTHORNE et al., 1983). Some fluctuations in both temperature and salinity were observed during the study but these were still within tolerable ranges and were not observed to have drastic effects on the shrimps. Dissolved oxygen was also far from the lethal level of  $0.9 \text{ mg L}^{-1}$ (ALLAN and MAGUIRE, 1991), as this was maintained at about 4 mg  $L^{-1}$  with aeration. Other biological and chemical factors may also have affected the growth and natural mortality of the shrimps in this study. However, limited by resources, no measurements were done on these aspects. Nevertheless, all treatments were subjected to the same water condition, hence, comparisons among marking treatments and control can still be done, negating other external causes.

Generally, results in the present study showed comparable outcomes with those made with conventional vet more expensive tagging materials, as discussed above. Of course, direct comparisons among different studies cannot be accurately made because of differences in laboratory setup, shrimp's initial age and size, stocking density and water condition. It is interesting to see that shrimp marking activities do not necessarily need proprietary tags and sophisticated equipment to implement. In terms of cost, the most expensive equipment used in our experiment was perhaps the Bano'k 303XL injector gun, purchased for about US\$25. Of course, a similar local brand would have even cost less. Other materials like food color and syringe for staining, T-bar cartridges, and small scissors for uropod trimming can be easily and cheaply acquired.

#### 5. Conclusion

Staining is not an effective marker for shrimps because marker retention only lasts for a few days or at most a week or two, limiting its use for very short-term studies. Externally visible tags like T-bar tags have very high retention rates but there is a significant decline on shrimps' survival after about 6 weeks. This maybe used for studies requiring high marker retention but also only limited for about a month. For initial restocking trials in the current site, the Batan Estuary, this method can be applied because tag distinction is fairly easy even for untrained local fishers. Also with the current status of fishing pressure in this small area (ALTAMIRANO, 2007), recapture of released stocks can be expected to be in the order of a few weeks only. On the other hand, the more practical method may as well be the trimming of uropods as this provides both good retention and survival of shrimps even for longer tagging experiments. However, more study is recommended on finer modifications of

these methods for applications on specific restocking sites to primarily increase efficiency but with equal consideration on cost of materials and methods for community-based stock enhancement programs.

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#### References

- ALLAN, G.L. and G.B. MAGUIRE (1991): Lethal levels of low dissolved oxygen and effects of short-term oxygen stress on subsequent growth of juvenile *Penaeus monodon*. Aquaculture, 94, 27–37.
- ALTAMIRANO, J.P. (2007): Declining mangroves and fisheries in the Batan Estuary, Panay Island, central Philippines. Bulletin of Marine Science, 80, 916-917.
- ALTAMIRANO, J.P. and H. KUROKURA (2008): Shrimp restocking as incentive for estuarine rehabilitation. Proceedings of the World Fisheries Congress 2008, Yokohama, Japan, 130.
- ARCE, S.M., B.J. ARGUE, D.A. THOMPSON and S.M. Moss (2003): Evaluation of a fluorescent, alphanumeric tagging system for penaeid shrimp and its application in selective breeding programs. Aquaculture, 228, 267–278.
- BELL, J.D., P.C. ROTHLISBERG, J.L. MUNRO, N.R. LONERAGAN, W.J. NASH, R.D. WARD and N.L. ANDREW (2005): Restocking and stock enhancement of marine invertebrate fisheries. Advances in Marine Biology, 49, 1?370.
- BENZIE, J.A.H., S.D. FRUSHER, M. KENWAY and L.

TROTT (1995): Utility of streamer tags to assess survival and growth of juvenile tiger prawns (*Penaeus monodon*) in aquaculture environments. Aquaculture, **136**, 57–69.

- BROWN, J.H., S. MCCAULEY, B. ROSS, A. TAYLOR and F. HUNTINGFORD (2003): A test of two methods for marking larvae and postlarvae of the giant freshwater prawn, *Macrobrachium rosenbergii*. Aquaculture Research, 34, 49–54.
- CAWTHORNE, D.F., T. BEARD, J. DAVENPORT and J.F. WICKINS (1983): Responses of juvenile *Penaeus* monodon Fabricius to natural and artificial sea waters of low salinity. Aquaculture, **32**, 165–174.
- CLAVERIE, T. and I.P. SMITH (2007): A comparison of the effect of three common tagging methods on the survival of the galatheid *Munida rugosa* (Fabricius, 1775). Fisheries Research, 86, 285– 288.
- DAVENPORT, J., S.U.K. EKARATNE, S.A. WALGAMA, D. LEE and J.M. HILLS (1999): Successful stock enhancement of a lagoon prawn fishery at Rekawa, Sri Lanka using cultured post-larvae of penaeid shrimp. Aquaculture, 180, 65–78.
- DAVIS, J.L.D., A.C. YOUNG-WILLIAMS, A.H. HINES and O. ZMORA (2004): Comparing two types of internal tags in juvenile blue crabs. Fisheries Research, 67, 265–274.
- EVASCO, F. (2000): Overfishing depletes fish population in the Manila Bay. Canopy International, 26, 5–9.
- FAO (2009): The State of World Fisheries and Aquaculture 2008. Food and Agriculture Organization of the United Nations, Rome.
- FAO-FIES (2008): FAO Yearbook Fishery and Aquaculture Statistics 2006. Food and Agriculture Organization of the United Nations, Rome.
- FARMER, A.S.D. (1981): A review of crustacean marking methods with particular reference to penaeid shrimp. Kuwait Bulletin of Marine Science, 2, 167–183.
- FERNANDEZ, P.R.J., Y. MATSUYA and R.F. SUBADE (2000): Coastal area governance system in the Philippines. Journal of Environment and Development, 9, 341-369.
- GARAWAY, C.J., R.I. ARTHUR, B. CHAMSINGH, P. HOMEKINGKEO, K. LORENZEN, B. SAENGVILAIKHAM and K. SIDAVONG (2006): A social science perspective on stock enhancement outcomes: Lessons learned from inland fisheries in southern Lao PDR. Fisheries Research, 80, 37–45.
- GODIN, D.M., W.H. CARR, G. HAGINO, F. SEGURA, J.N. SWEENEY and L. BLANKENSHIP (1996): Evaluation of a fluorescent elastomer internal tag in juvenile and adult shrimp *Penaeus vannamei*. Aquaculture, **139**, 243–248.
- HAMASAKI, K. and S. KITADA (2006): A review of kuruma prawn *Penaeus japonicus* stock

enhancement in Japan. Fisheries Research,  $80,\,80{-}90.$ 

- HILL, B.J. and T.J. WASSENBERG (1985): A laboratory study of the effect of streamer tags on mortality, growth, molting and duration of nocturnal emergence of the tiger prawn *Penaeus esculentus* (Haswell). Fisheries Research, 3, 223–235.
- HOWE, N.R. and P.R. HOYT (1982): Mortality of juvenile BROWN shrimp *Penaeus aztecus* associated with streamer tags. Transactions of the American Fisheries Society, **111**, 317–325.
- JACKSON, C.J. and Y.G. WANG (1998): Modelling growth rate of *Penaeus monodon* Fabricius in intensively managed ponds: effects of temperature, pond age and stocking density. Aquaculture Research, 29, 27-36.
- KATON, B.M., R.S. Pomeroy, M. RING and L.R. GARCES (1998): Mangrove rehabilitation and coastal resource management: A case study of Cogtong Bay, Philippines. NAGA, The ICLARM Quarterly, 21, 46–62.
- KIBRIA, G. (1993): Studies on molting, molting frequency and growth of shrimp (*Penaeus monodon*) fed on natural and compound diets. Asian Fisheries Science, 6, 203-211.
- KNEIB, R.T., and M.C. HUGGLER (2001): Tag placement, mark retention, survival and growth of juvenile white shrimp (*Litopenaeus setiferus* Perez Farfante, 1969) injected with coded wire tags. Journal of Experimental Marine Biology and Ecology, 266, 109-120.
- LEANO, E.M. and I.C. LIAO (2006): Tagging of *Litopenaeus vannamei* (Boone) by uropod cutting: a comparison of two methods. Aquaculture Research, 37, 885–890.
- LEBATA, M.J.H.L., L. LE VAY, M.E. WALTON, J.B. BINAS, E.T. QUINITIO, E.M. RODRIGUEZ and J.H. PRIMAVERA (2009): Evaluation of hatchery-based enhancement of the mud crab, *Scylla* spp., fisheries in mangroves: comparison of species and release strategies. Marine and Freshwater Research, **60**, 58–69.
- LIM, C.P., Y. MATSUDA and Y. SHIGEMI (1995): Problems and constraints in Philippine municipal fisheries - the case of San-Miguel Bay, Camarines-Sur. Environmental Management, 19, 837–852.
- LINNANE, A. and J.P. MERCER (1998): A comparison of methods for tagging juvenile lobsters (*Homarus gammarus* L) reared for stock enhancement. Aquaculture, 163, 195-202.
- LONERAGAN, N.R., Y.M. YE, R.A. KENYON and M.D.E. HAYWOOD (2006): New directions for research in prawn (shrimp) stock enhancement and the use of models in providing directions for research. Fisheries Research, 80, 91–100.
- MENZEL, R.W. (1955): Marking of shrimp. Science,

121, 446-446.

- MINES, A.N., I.R. SMITH and D. PAULY (1986): An overview of the fisheries of San Miguel Bay, Philippines. *In* The First Asian Fisheries Forum. MACLEAN, J.L., DIZON, L.B., HOSILLOS, L.V.S (eds.), Asian Fisheries Society, Manila, Philippines, pp. 385–388.
- MIYAJIMA, T., Y. HAMANAKA and K. TOYOTA (1999): A marking method for kuruma prawn *Penaeus japonicus*. Fisheries Science, **65**, 31–35.
- MONTGOMERY, S.S. and P.A. BRETT (1996): Tagging eastern rock lobsters *Jasus verreauxi*: Effectiveness of several types of tag. Fisheries Research, 27, 141–152.
- MONTGOMERY, S.S. and C.A. GRAY (1991): Effects of sizes of streamer tags on mortality and growth of juvenile eastern king prawns *Penaeus plebejus*. Marine Ecology-Progress Series, **76**, 33– 40.
- NATIONAL ECONOMIC AND DEVELOPMENT AUTHORITY (2005): Success Stories in Coastal Resource Management. devPulse, 10, 1–2.
- NEAL, R.A. (1969): Methods of marking shrimp. FAO Fisheries Reports, 57, 1149–1165.
- NIWA, N., M. UENO and Y. INOUE (1998): Accumulation of trypan blue and trypan red in nephrocytes of the fresh-water shrimp *Neocaridina denticulata* (Decapoda: Atyidae). Journal of Crustacean Biology, 18, 666–672.
- PALMA, A.L., A.S. DIAMANTE and P.M. Pol (2002): An assessment of fishery resources of Laguna de Bay. Aquatic Ecosystem Health and Management, 5, 139–126.
- PRIMAVERA, J.H. and R.M. CABALLERO (1992): Effect of streamer tags on survival and growth of juvenile tiger prawns, *Penaeus monodon*, under laboratory conditions. Australian Journal of Marine

and Freshwater Research, 43, 737-743.

- SU, M.S. and I.C. LIAO (1999): Research and development of prawn stock enhancement in Taiwan. *In* Stock Enhancement and Sea Ranching. HOWELL, B.R., E. MOSKNESS, and T. SVASAND (eds.), Blackwell Science, Oxford, pp. 379–392.
- TEBOUL, D. (1993): Internal monofilament tags used to identify juvenile *Penaeus monodon* - tag retention and effects on growth and survival. Aquaculture, 113, 167–170.
- THIES, J.A., S.B. MERRILL and E.L. CORLEY (2002): Red food coloring stain: New, safer procedures for staining nematodes in roots and egg masses on root surfaces. Journal of Nematology, 34, 179–181.
- TOYOTA, K., T. YAMAUCHI and T. MIYAJIMA (2003): A marking method of cutting uropods using malformed regeneration for kuruma prawn *Marsupenaeus japonicus*. Fisheries Science, 69, 161–169.
- WANG, Q.Y., Z.M. ZHUANG, J.Y. DENG and Y.M. YE (2006): Stock enhancement and translocation of the shrimp *Penaeus chinensis* in China. Fisheries Research, 80, 67–79.
- WASSENBERG, T.J. and J. KERR (1990): Woundhealing in prawns tagged with streamer tags, and the effect of tag placement and prawn size on survival rate. Fisheries Research, 9, 209–218.
- YE, L., S. JIANG, X. ZHU, Q. YANG, W. WEN and K. WU (2009): Effects of salinity on growth and energy budget of juvenile *Penaeus monodon*. Aquaculture, **290**, 140-144.
- ZAR, J.H. (1999): Biostatistical Analysis, 4th ed. Prentice Hall, New Jersey.

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### 資 料

# 第48巻第1号掲載欧文論文の和文要旨

#### Leni G. Yap-DEJETO<sup>182</sup>, 大村卓朗<sup>1</sup>, 長濱幸生<sup>1</sup>, 福代康夫<sup>1</sup>:東京湾で採集された 11 種の Pseudo-nitzschia

2008 年 4 月から 2009 年 8 月の期間に、東京湾で得られたサンプルについて、光学顕微鏡および透過型電子顕微鏡 を用いて形体観察をおこなった。その結果、東京湾からこれまでに報告の無い 5 種(Pseudo-nitzschia americana, P. brasiliana, P. caciantha, P. calliantha, P. galaxiae)を含む 11 種(残りの 6 種: P. delicatissima, P. fraudulenta, P. multistriata, P. multiseries, P. pseudodelicatissima, P. pungens)が観察された。

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#### ジョン・P・アルタミラノ、黒倉 寿: ウシエビ *Penaeus monodon* (Fabricius) 稚エビの標識法―安価な標識法 の比較

コストのかからないエビ類の標識法として、染色、尾肢の一部切除、T-barタイプのタグについて検討した。実験 には2ヶ月令のウシエビ、Penaeus monodon(頭胸甲長 8.42±0.10)を用い、無標識の対照群を含めて、各種の標 識法で標識した個体を8週間飼育した。頭胸甲長の成長には、対照群を含めて、各処理間で有意な違いは見られなかっ た。染色処理群(60%)、尾肢一部切除群(51%)、対象群(58%)の間には、生存率の違いも見られなかったが、Tbarタグ群の生残率は有意に劣っていた(33%)。標識の保持率はT-barタグ群で極めて高く(100%)、これに尾肢一 部切除群(65%)でつづいていた。しかし、染色群の標識保持率は実験開始後 3 週間で 0%となった。以上のことか ら、染色法は標識保持時間が短く長期にわたる標識としては使えないこと。T-barタグは標識保持率は高いものの生 残に強い影響を与え、標識方法として不適切なことが明らかになった。尾肢の一部切除は再生した尾肢が特有の形態 となるため、比較的識別性が高く、生残率も対照群と大きく異ならないことから、現実的に利用可能な標識法である 物と考えられた。今後、費用がからない形でこの方法を改善し、生残率・保持時間をさらに高めていく事が必要であ る。本研究は、途上国において地域レベルで資源添加を行いその結果を追跡していくことを可能にするために行われ た研究である。

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#### 学会記事

- 2010-2011年度評議員(24名)選出 荒川久幸 有元貴文 石丸 隆 磯田 豊 今脇資郎 神田譲太 北出裕二郎 小池勲夫 小松輝久 河野 博 斎藤誠一 関根義彦 千手智晴 田中祐志 寺崎 誠 中田英昭 長島秀樹 森永 勤 門谷 茂 柳 哲雄 八木宏樹 山口征矢 山崎秀勝 吉田次郎 内田 裕\* 小池康之\* 鈴木秀和\* 溝端浩平\* (\*は会長推薦)
- 2010-2011年度会長選出 会長 今脇資郎会員
- 3. 2010年度役員·学会賞委員
  - 会 長:今脇資郎
  - 副会長:森永 勤 小松輝久
  - 幹 事:(庶務)河野 博 荒川久幸,(会計)神田穣太
     山崎秀勝,(編集)田中祐志 北出裕二郎,
     (研充)石丸 隆 鈴木秀和,(渉外)小池康之,
     (広報)溝端浩平 内田 裕
     監 事:長島秀樹 小池 隆
     編集委員長:吉田次郎
  - 学会賞委員:小松輝久 荒川久幸 河野 博 和泉 充 門谷 茂 神田穣太 石丸 隆 田中祐志 市川 香
- 2010年5月17日(月)日仏会館会議室において2010 年度第1回幹事会が開かれた。
- 審議事項
- 1)2010・2011年度の体制について 24名の評議員,会長の選出,幹事の依頼,賞推薦委員 の選出について,資料に基づいて論議した。研究担当 幹事として鈴木秀和会員(東京海洋大学)を加えるこ とで了承された。なお,新体制については,評議員会 で承認を得る必要がある。
- 2) Kobe2010について
  - ・「Techno-Ocean 2010」(TO2010)の,日仏海洋学 会関係の世話人は,引き続き,河野幹事が務める。 Kobe2010の中の「第14回日仏海洋学シンポジウム第 1部」の世話人は小松副会長が務め,創立50周年記念 式典・講演の世話人は森永副会長が務めることとした。 「Kobe2010実行委員会」の副委員長は,八木前副会長 に代って小松副会長が務めることとした。

TO2010の基調講演者をHenocque氏とすることが了

承され、小池幹事が連絡をとることとした。「第14回 日仏海洋学シンポジウム第1部」の基調講演者を Bailly氏と柳哲雄氏とすることが了承され、Bailly氏 には小池幹事が、柳氏には小松副会長が連絡をとるこ ととした。創立50周年記念講演者をSempere氏とする ことが了承され、小松副会長が連絡をとることとした。 ・「第14回日仏海洋学シンポジウム第1部」の内容を 検討するために、日仏合同組織・科学委員会を設置す る。日本側の主査は小松副会長が務める。詳細はフラ ンス側の意向を聞きながら企画することで了承された。 投稿論文の審査は当委員会が担当し、必要に応じて 「Kobe2010実行委員会」の総務担当委員に審査を依頼 する。

・Kobe2010の,TO2010でのセッション・タイトルお よび会場の看板について論議した結果,フランス語表 記にすることとし,表記案を小松副会長が考えること とした。

・記念式典での祝辞をお願いする方について論議した。 その結果, Ceccaldi氏には今脇会長が, 日仏会館には 小池幹事が, フランス大使には小松副会長が連絡する こととした。なお, 感謝状の贈呈に関しては森永副会 長が担当することとした。また, 祝辞の言語について, 母国語がフランス語の方には, 日本語訳を作成・配布 しフランス語で祝辞を, それ以外の方には英語で祝辞 をいただくこととした。

・名誉会員や元会長の方々の招待については, 荒川幹 事が担当することとした。

・助成・寄附については森永副会長が、日仏学者交流 事業については小松副会長が、引き続き折衝を行うこ ととした。

- 3)「第14回日仏海洋学シンポジウム第2部」について 「第14回日仏海洋学シンポジウム第2部」を2010年10 月19日(火)午後3時~5時30分の予定で日仏会館に おいて開催することとした。世話人は小松副会長が務 める。
- 4)2010年度の評議員会・総会などについて 今年度の評議員会・総会・賞贈呈・受賞記念講演を, 上記シンポジウムの直前に,2010年10月19日(火)午 後1時~2時30分の予定で日仏会館において開催する こととした。世話人は荒川幹事が務める。
- 5) 2009年度の決算および2010年度の予算について 2009年度の決算案および監査報告がなされ,了承され た。また,2010年度の予算案が示され,若干の修正を

行った上で了承された。なお,会費の自動引落しの可 能性を調べることとした。

- 6)「うみ」創立50周年記念号の編集について 記念号に、学会の50年間の活動を記録した「50年の歩み(仮称)」を掲載する方向で作業を進めることとした。海洋学・水産学分野での日仏間の交流、今回で14回となる日仏海洋学シンポジウムの歴史、日仏交換学者の記録、歴代賞受賞者一覧その他を資料とし、小池 幹事・森永副会長・小松副会長が中心となって執筆する。
- 7)第13回日仏海洋学シンポジウムの報告について 2008年10月にマルセイユとパリで開催された第13回日 仏海洋学シンポジウムの報告書を,7月末を目処に完 成させることとなった。
- 8) 学会ホームページなどの充実について

学会の活動としてホームページの充実は極めて重要で あることを確認した。「うみ」掲載論文のpdfファイ ルを(発刊から一定期間後に)ホームページに載せる ことに関連して、コピーライトの問題点などを内田幹 事が検討することとした。「うみ」掲載論文を国立情 報学研究所のCiNii (NII論文情報ナビゲータ)に集録 してもらえるかどうかを、吉田編集委員長が調査する こととした。また、会員への電子メールによる連絡を 促進するために、会員のメールアドレスの情報を集め ることにした。

- 9)学会のフランス語名について 現在の学会のフランス語名が、フランスの仏日海洋学 会とまったく同じであることの問題点が指摘され、同 学会と区別するために、最後に「、Tokyo」を付ける 案が検討された。この件は、引き続き検討することに した。
- 10) 追悼記事について

2009年に逝去された,字野寛元会長の追悼記事を小池 幹事の担当で,また森田良美名誉会員のそれを神田幹 事の担当で,それぞれ「うみ」に掲載することとした。

#### 5. 新入会員

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溝端浩平	東京海洋大学 〒108-8478 東京都港区港南4-5-8

退会(逝去者含む)

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8. 寄贈図書および資料

農工研ニュース(農村工学研究所); No.63, 64, 65

- FRAN NEWS (水産総合研究センター); Vol.20, 21
- 広島観光コンベンション; Vol.76, 77
- Ship & Ocean Newsletter (海洋政策研究財団); No.218-226
- なつしま (JAMSTEC); 第63号通巻283-287
- 水産技術;第2巻1号
- J-STAGE NEWS (独立行政法人科学技術振興機構); No.21, 22
- ニュースレター(東京大学海洋研究所); No.19
- 「海一自然と文化」(東海大学海洋学部); Vol.7, No.2 Techno-ocean News (テクノオーシャンネットワー ク); No.35, 36
- Bulletin of the national museum ; 34 (4), 36
- 日本海ブロック資源研究会報告(日本海区水産研究所); No.44
- 勇魚(日本捕鯨協会); No.36
- 中国海洋大学学報(中国海洋大学); Vol.39, No.169-174.
- Progress in Fishery Sciences (中国水産学会); Vol. 30, No.5-6.
- Meereswissenchaftliche Berichte Marine Science Repoorts
- (Leibniz-Institut Fur Ostseeforschung Warnemun de) ; No.77, 78
- Annual Report (KoreaOceanResearch&Institute); 2009

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