Appendix 1

Publication 1: TBT-pollution in the Gulf of Thailand: a re-inspection of imposex incidence after 10 years

Marine Pollution Bulletin 58 (2009) 526–532

Contents lists available at ScienceDirect

Marine Pollution Bulletin

journal homepage: www.elsevier.com/locate/marpolbul

TBT-pollution in the Gulf of Thailand: A re-inspection of imposex incidence after 10 years

Cornelis Swennen *, Uraiwan Sampantarak, Nukul Ruttanadakul

Faculty of Science and Technology, Prince of Songkla University, Pattani 94000, Thailand

ARTICLE INFO

Keywords:
Tributyltin
Imposex
Neogastropods
Gulf of Thailand
Southeast Asia

ABSTRACT

Imposex in neogastropods was used to determine the relative TBT distribution in the Gulf of Thailand in 2006. To identify the imposex prevalence, 8757 specimens, belonging to 22 species from five families of neogastropods, were collected at 56 sites in 13 sample areas. These areas were located between the Bight of Bangkok in the north and the Malaysian border in the south. A contamination model was developed to compensate for differences in TBT sensitivity among species and to make comparisons among areas. At every area imposex was found in neogastropods. The highest incidence was in the east side of the Bight of Bangkok off Si Racha and Pattaya and in the southern part around Pattani. The same areas showed the highest frequency of imposex in 1996. While the frequency of imposex appeared to have slightly decreased in these areas, increases were found elsewhere. In Pattani Province this could be related to dumping of highly polluted harbour sediments in the shallow coastal area. The overall frequency of imposex in the Gulf of Thailand significantly increased from 1996 to 2006.

1. Introduction

In the late 1960s, tributyltin (TBT) came in use as biocide in anti-fouling paints on ships and installations at sea. TBT provides a long lasting protection against fouling organisms such as barnacles, mussels, hydrozoans and algae. Adverse bio-impacts to organisms in the environment were not expected. However, gradually severe negative effects to organisms in non-target areas in the marine environment were reported over the next 40 years.

Some years after the introduction of TBT use, Blader (1970) discovered that females of the snail *Nucella lapillus* (Linnaeus, 1758) developed a small penis that was followed by a decline in the affected populations (Bryan et al., 1986, 1987). Subsequently, more snail species were discovered where females showed a small penis and pallial vas deferens. Additional research showed that these morphological changes were caused by TBT (Smith, 1981; Gibbs and Bryan, 1986; Oehlmann et al., 1993; Mensink et al., 1996). The phenomenon was first named masculinization or pseudo-hermaphroditism. However, the imposition of a mini-penis and mini pallial vas deferens in gastropods soon became commonly known as imposex. In the late 1970s, abnormalities in the shells of cultured oysters in Arcachon Bay cost the French oyster culture industry billions of US$, shown to be caused by TBT (Alzieu et al., 1981, 1986). Similar cases were found at other sites and countries (Phelps and Pace, 1997).

All these cases were close to harbours and large marinas. In particular yachts, moored for long periods in harbours, were suspected to increase TBT level in the water sufficiently to result in negative effects. This forced governments to ban the use of TBT containing anti-fouling paint on all vessels smaller than 25 m. France instituted this ban in 1982, followed by several other countries (Ten Hallers-Tjabbes, 1997). However, few localities have shown any positive effect of this ban (Dowson et al., 1993; Evans et al., 1996).

Identification of TBT (and other organotin compounds) in water is an expensive process when the distribution of TBT has to be established over large areas in the marine environment. The occurrence of imposex has commonly been accepted as a good bio-indicator of TBT pollution (Gibbs et al., 1987; Oehlmann et al., 1996). Monitoring of gastropods indicated that the abnormality is found on all continents close to harbours (Ellis and Pattisima, 1990; Horiguchi et al., 1991; Stewart et al., 1992). Most studies focused on gastropods that were collected by hand in or just below the intertidal zone. However, TBT also impacts gastropods in the open sea. The occurrence of imposex in *Buccinum undatum* Linnaeus, 1758 and the decline of this species in the North Sea could be related to shipping routes (Ten Hallers-Tjabbes et al., 1994). Similarly, imposex in 22 gastropod species appears to be related to shipping in south-east Asian waters (Strait of Malacca, Gulf of Thailand) up into the deepest possible sampling sites (Swennen et al., 1997). TBT has become a global pollution problem in marine habitats. This moved the Marine Environmental Protection Committee (MEPC) in 1996 to start discussions about a reduction of the use of TBT on large ships within five years with a total ban thereafter (Ten

* Corresponding author.
E-mail address: swennen@wxs.nl (C. Swennen).

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doi:10.1016/j.marpolbul.2008.11.028

Hallers-Tjabbes, 1997). The ban has been postponed, but in the meantime several vessels already no longer use anti-fouling paints containing TBT.

The present study is undertaken to determine if positive effects already occur in gastropods, specifically a decline in the frequency of imposex in the Gulf of Thailand. The main objectives were to: (1) establish the present levels of imposex in the area, (2) clarify possible differences in sensitivity among species, and (3) compare the results with data collected 10 years ago.

2. Materials and methods

The study was limited to key areas in the Gulf of Thailand (Fig. 1). Travel time and costs were reduced by dropping the Strait of Malacca from the sampling design, but this allowed collection of larger samples per site in the Gulf of Thailand. Neogastropods were collected from 56 sites in 13 areas in the Gulf of Thailand between June and November 2006. Specimens belonging to the neogastropod families Columbellidae and Volutidae were excluded, because absence of the imposex response has been reported in the former family (Gibbs et al., 1997), and females of the local genera *Cymbiola* and *Melo* of the Volutidae always show a small penis and vas deferens, (pseudo-imposex; Swennen and Horpet, 2008).

Samples were collected in both sublittoral and littoral environments. Specimens were hand-collected in the intertidal zone during low tide, but most were obtained from the by-catch of small, commercial fishing boats. The geographical positions of the sampling sites were determined by a hand-held GPS apparatus, while for those fishing trips in which we did not participate, the position was determined with help of the sea map on instruction of the skipper. The taxonomic nomenclature followed Swennen et al. (2001). A few species from the northern part not treated in Swennen et al. (2001) were identified by the curator of the Department of Malacology, University of Amsterdam (The Netherlands). The lengths (mm) of the gastropod shells were measured, and a part of the shell was removed to identify the sex and to check for imposex in females. Females with a short, curved penis and at least a part of a vas deferens were noted as showing imposex. An imposex index such as used by Mensink et al. (2002) was not used for avoiding errors, because the sizes of the specimens and the sizes and shapes of the male penis among the several species studied varied too much for correctly using the index on all species under local field conditions. Thus females with stage 1 and 2 were considered as showing no imposex. However, these stages were rarely found in specimens checked in the laboratory.

3. Statistical methods

Surveying a large area has the disadvantage that several habitats are included, each with a different assemblage of neogastropod species. No species was found at all sites. In the 1996 study, this imbalance was solved by calculating the imposex frequency over all females of species known to be sensitive for showing imposex. Since indications were found that species may differ in sensitivity

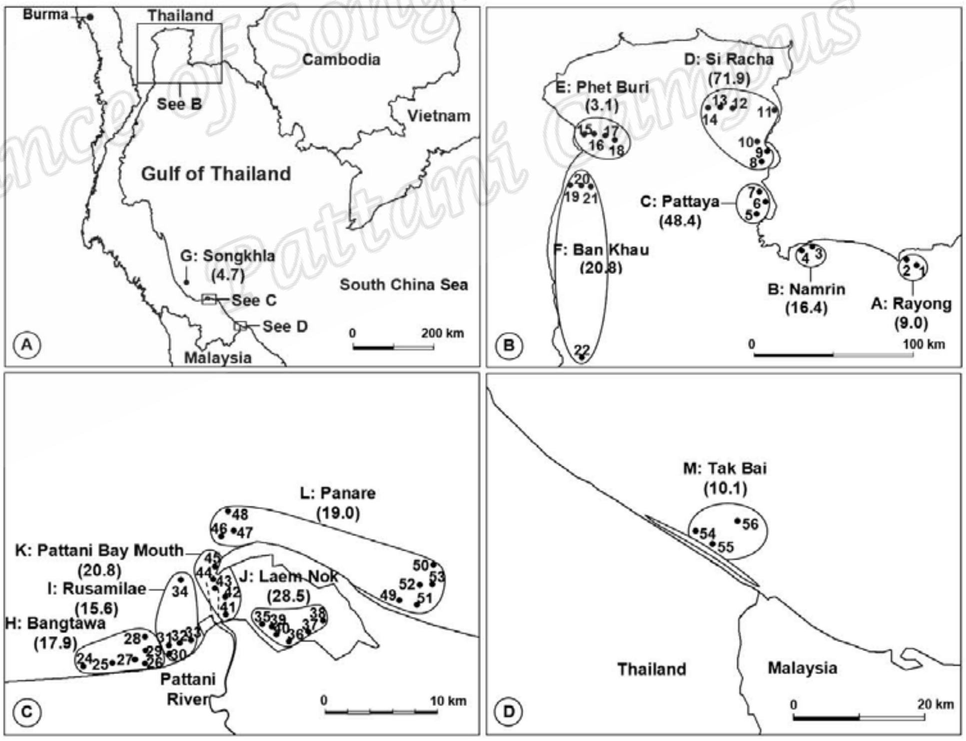

Fig. 1. The Gulf of Thailand with the study areas and sampling sites: A. overview; B. northern part of the Gulf of Thailand with study areas A–F; C. Pattani Bay and surroundings with study areas H–L; D. Narathiwat province with study area M. Dots show the sampling sites. Their number corresponds with the number in the detailed list appended to the online version. Value between brackets gives the adjusted imposex incidence in the area.

(Swennen et al., 1997), a contamination model is used that takes species into account for comparing imposex occurrence between areas.

The R statistical system (R Development Core Team, 2007; Venables and Ripley, 2002) was used for statistical model fitting. If p is the frequency of outcomes with a specific characteristic ("adverse" outcome) in a sample of size n, for large n, formula (1) is an asymptotically valid formula for the standard error. Formula (2) gives the 95% confidence interval (95% CI) for the frequency.

$$SE = \sqrt{\frac{p(1-p)}{n}} \qquad (1)$$

$$p - 1.96 \times SE, p + 1.96 \times SE \qquad (2)$$

For comparing two frequencies, we computed a p-value using Pearson's chi-squared test. We employed logistic regression (Hosmer and Lemeshow, 2000; Kleinbaum and Klein, 2002) to model the effects of multiple determinants on the occurrence of imposex. If there are two categorical determinants, and p_{ij} denotes the probability of the adverse outcome in categories i and j of these determinants, respectively, the simplest such model takes the additive form of formula (3). The method also gives a measure of the residual deviation between the model and the data and its corresponding number of degrees of freedom (df), defined as the number of cells into which the data are grouped minus the number of fitted parameters (Venables and Ripley, 2002 Chapter 7).

$$\ln\left(\frac{p_{ij}}{1-p_{ij}}\right) = a_i + b_j \qquad (3)$$

Formula (4) is used to express the frequency of the imposex outcome as a function of the two determinants. To eliminate redundant parameters, we took one of the categories of the second determinant as the referent and the corresponding parameter was set to 0. We fitted the model by using the maximum likelihood method with the observed data to estimate the parameters, providing standard errors for the fitted parameters. For each determinant, the method gives a p-value based on a chi-squared statistic for testing the null hypothesis that the outcome frequency is the same for each of its component categories. The method also gives separate p-values for comparing each parameter in the model with the mean (for the determinant of interest) or referent (for the covariate determinant).

$$p_{ij} = \frac{1}{1 + \exp(-a_i - b_j)} \qquad (4)$$

Logistic regression provides a straightforward method for adjusting a frequency that varies with a determinant of interest for a covariate determinant. To calculate the adjusted frequency for category i of the determinant of interest, the term b_j in formula (3) is replaced by a constant b, giving formula (5). The value of the constant b is chosen to ensure that the sum of the adjusted number of adverse outcomes is equal to the observed number, as expressed in formula (6), where n_i is the sample size in category i of the determinant of interest. This method extends straightforwardly to additional covariates.

$$p_i^* = \frac{1}{1 + \exp(-a_i - b)} \qquad (5)$$

$$\sum p_i^* n_i = \sum p_i n_i \qquad (6)$$

4. Results

A total of 8757 snail specimens were collected in the Gulf of Thailand in 2006. The specimens comprised 22 species belonging to five families of neogastropods. The full names of the species

Table 1
Names of families and species of the neogastropods studied, with a summary of the size range of males and females, number of females, percentage of females, and percentage of females showing imposex in 2006.

Species and families	Sample size	Shell height (mm)		No. of females	Female (%)	Imposex (%)
		Males	Females			
Family Muricidae						
1. *Murex altispira* Ponder and Vokes, 1988	359	40–97	41–125	286	79.7	0.7
2. *Murex occa* Sowerby II, 1824	888	24–90	26–98	417	47.0	40.8
3. *Murex trapa* Röding, 1798	1156	34–118	31–125	698	60.4	22.9
4. *Chicoreus banksii* (Sowerby II, 1841)	58	44–81	37–84	39	67.2	12.8
5. *Lataxiena blosvillei* (Deshayes, 1832)	721	21–44	22–45	366	50.8	88.8
6. *Semiricinula muricoides* (De Blainville, 1832)	753	13–22.5	13–28	521	69.2	50.9
7. *Thais bitubercularis* (Lamarck, 1822)	427	18–35	19–37	214	50.1	14.0
8. *Thais clavigera* (Kuester, 1860)	75	20–34	21–34	36	48.0	25.0
9. *Thais lacera* (Von Born, 1778)	587	15–57	15–70	306	52.1	21.9
10. *Thais rufotincta* Tan and Sigurdsson, 1996	22	15–26	18–22	10	45.5	10.0
11. *Morula musiva* (Kiener, 1835)	324	13–29	14–30	161	49.7	0.6
12. *Rapana rapiformis* (Von Born, 1778)	85	35–88	33–90	32	37.6	6.3
Family Buccinidae						
13. *Babylonia areolata* (Link, 1807)	314	38–64	20–70	158	50.3	1.3
14. *Nassaria pusilla* (Röding, 1798)	67	17.6–23	11–24	53	79.1	1.9
15. *Phos senticosus* (Linnaeus, 1758)	2	26	33	1	50.0	0.0
Family Nassariidae						
16. *Nassarius jacksonianus* (Quoy and Gaimard, 1833)	233	15–21	13–22	166	71.2	10.8
17. *Nassarius livescens* (Philippi, 1849)	62	16–29	16–25	29	46.8	6.9
18. *Nassarius siquijorensis* (A. Adams, 1852)	260	16–28	13–23	202	77.7	28.7
19. *Nassarius stolatus* (Gmelin, 1791)	533	11–22	13–23	306	57.4	4.9
Family Melongenidae						
20. *Pugilina cochlidium* (Linnaeus, 1758)	923	35–93	23–98	413	44.7	9.4
21. *Hemifusus ternatanus* (Gmelin, 1791)	368	38–105	27–131	225	61.1	15.6
Family Turridae						
22. *Turricula javana* (Linnaeus, 1767)	540	18–42	19–64	405	75.0	15.6
Total	8757			5044	57.6	25.2

Table 2
Results of the logistic model for imposex outcome among females in 2006.

Determinants		Coefficient	Standard error	*p*-Value
Area	A: Rayong	−2.119	0.389	<0.001
	B: Namrin	−1.429	0.369	<0.001
	C: Pattaya	0.138	0.231	0.551
	D: Si Racha	1.138	0.186	<0.001
	E: Phet Buri	−3.253	0.289	<0.001
	F: Ban Khau	−1.139	0.164	<0.001
	G: Songkla	−2.815	1.111	0.011
	H: Bang Tawa	−1.322	0.214	<0.001
	I: Rusamilae	−1.491	0.154	<0.001
	J: Laem Nok	−0.721	0.190	<0.001
	K: Pattani Bay Mouth	−1.140	0.230	<0.001
	L: Panare	−1.253	0.462	0.007
	M: Tak Bai	−1.987	1.251	0.112
Species	01: *Murex trapa* (referent)	0		
	02: *Murex altispira*	−2.961	0.834	<0.001
	03: *Murex occa*	0.697	0.186	<0.001
	04: *Lataxiena blosvillei*	1.674	0.271	<0.001
	05: *Semiricinula muricoides*	−0.215	0.211	0.308
	06: *Thais bitubercularis*	−0.494	0.289	0.087
	07: *Thais lacera*	0.097	0.203	0.633
	08: *Morula musiva*	−3.646	1.069	0.001
	09: *Babylonia areolata*	−2.398	1.243	0.054
	10: *Nassarius jacksonianus*	−0.700	0.291	0.016
	11: *Nassarius siquijorensis*	0.373	0.253	0.141
	12: *Nassarius stolatus*	−1.350	0.308	<0.001
	13: *Pugilina cochlidium*	−1.602	0.231	<0.001
	14: *Hemifusus ternatanus*	−1.504	0.247	<0.001
	15: *Turricula javana*	−0.323	0.208	0.120
	16: Other	−0.775	0.327	0.018

Table 3
Prevalence (%) of imposex by area before and after adjusting for species in the sampling of 2006.

Area	Total number of females	Number with imposex	Imposex (%)	Adjusted imposex (%)	
			(unadjusted)	Prevalence	95% CI
A: Rayong	190	9	4.7	9.0	4.9–13.0
B: Namrin	249	13	5.2	16.4	11.8–21.0
C: Pattaya	474	297	62.7	48.4	43.9–52.9
D: Si Racha	537	395	73.6	71.9	68.1–75.7
E: Phet Buri	417	13	3.1	3.1	1.4–4.7
F: Ban Khau	286	53	18.5	20.8	16.1–25.5
G: Songkla	171	1	0.6	4.7	1.5–7.8
H: Bang Tawa	735	128	17.4	17.9	15.1–20.7
I: Rusamilae	916	143	15.6	15.6	13.2–17.9
J: Laem Nok	507	157	31.0	28.5	24.5–32.4
K: Pattani Bay Mouth	266	50	18.8	20.8	15.9–25.6
L: Panare	140	9	6.4	19.0	12.5–25.4
M: Tak Bai	156	2	1.3	10.1	5.4–14.8

Table 4
Prevalence (%) of imposex per species before and after adjusting for areas in the sampling of 2006.

Species groups	Total females	No. of imposex	Imposex (%) unadjusted	Adjusted imposex (%)	
				Prevalence	95% CI
1. *Murex trapa*	698	160	22.9	30.1	26.7–33.5
2. *Murex altispira*	286	2	0.7	2.2	0.5–3.9
3. *Murex occa*	417	170	40.8	46.4	41.6–51.2
4. *Lataxiena blosvillei*	366	325	88.8	69.7	65.0–74.4
5. *Semiricinula muricoides*	521	265	50.9	25.8	22.0–29.6
6. *Thais bitubercularis*	214	30	14.0	20.8	15.4–26.3
7. *Thais lacera*	306	67	21.9	32.2	27.0–37.4
8. *Morula musiva*	161	1	0.6	1.1	0.0–2.7
9. *Babylonia areolata*	158	2	1.3	3.8	0.8–6.7
10. *Nassarius jacksonianus*	166	18	10.8	17.6	11.8–23.4
11. *Nassarius siquijorensis*	202	58	28.7	38.5	31.8–45.2
12. *Nassarius stolatus*	306	15	4.9	10.1	6.7–13.4
13. *Pugilina cochlidium*	413	39	9.4	8.0	5.4–10.6
14. *Hemifusus ternatanus*	225	35	15.6	8.7	5.1–12.4
15. *Turricula javana*	405	63	15.6	23.8	19.6–27.9
16 Other*	200	20	10.0	16.6	11.4–21.7

* *Chicoreus banksii*, *Thais clavigera*, *Thais rufotincta*, *Rapana rapiformis*, *Nassaria pusilla*, *Phos senticosus* and *Nassarius livescens* combined.

and families are presented in Table 1, which also summarises the size range of males and females, number of females, sex ratio, and percentage of females showing imposex. A list of the 56 sampling sites with location, depth, species, number and details of specimens per site is added to the online version.

The logistic regression model (formula (3)) for the imposex frequency used the 13 areas as the determinant of interest and the 16 species groups as the covariate. Species with fewer than 100 individuals were combined into a single group referred to as "other". The logistic regression model gave a residual deviance of 171.4 with 55 df based on the grouped data (the 84 combinations of area and species where samples were obtained). The chi-squared statistics for testing a common imposex frequency between areas and species groups were found to be 438.9 (12 df) and 453.0 (15 df) respectively, with corresponding highly statistically significant *p*-values (Table 2). From the results (Table 2), the adjusted imposex frequency was calculated using formula (5) with the corresponding 95% confidence intervals (formula (2)). Imposex frequency before and after adjusting for species is given in Table 3. When all species were combined, areas C and D had a high (>50%) imposex frequency, areas F, H, I, J, and K had medium (15–49%) frequency, whereas A, B, E, G, L, and M had low (<15%) frequency. After adjusting for the different sensitivities of the species, C and D still

Table 5
Number of female gastropods showing imposex and total number studied (imposex/ females) per area in the Gulf of Thailand in 1996 (Cited from Swennen et al., 1997).

Species	Area*							
	C	D	E	F	I	J	K	L	M
Murex trapa	–	57/59	–	–	–	–	–	–	–
Thais lacera	–	–	–	–	–	–	1/2	–	–
Babylonia areolata	13/15	–	0/27	0/28	1/42	1/1	–	–	0/71
Nassarius stolatus	–	–	–	–	–	–	1/1	–	–
Hemifusus ternatanus	6/7	3/5	–	–	–	1/3	1/5	–	1/34
Thais hippocastaneum	–	–	–	–	–	–	–	0/139	–
Murex sp.	28/136	19/19	–	0/28	–	–	–	–	0/11
Other	1/1	5/10	–	–	–	7/13	5/22	–	0/29
Total	48/159	84/93	0/27	0/56	1/42	9/17	8/30	0/139	1/145
Crude prevalence (%)	30.2	90.3	0.0	0.0	2.4	52.9	24.1	0.0	0.7
Adjusted prevalence (%)	33.5	84.8	–	–	0.3	57.8	26.7	–	0.1

* C: Pattaya; D: Si Racha; E: Phet Buri; F: Ban Khau; I: Rusamilae; J: Laem Nok; K: Pattani Bay Mouth; L: Panare; M: Tak Bai.

showed the highest values, but B and L moved up to the medium severity level of imposex frequency (Table 3). The location of the areas and the sampling sites are shown in Fig. 1 where the mean frequency is also given per area.

Sensitivity to imposex varied among species, *Lataxiena blosvillei* was found to be most prone to imposex (mean 69.7 ± 4.7%; 95% CI), whereas *Morula musiva* is least sensitive (mean 1.1 ± 1.6%; 95% CI) (Table 4). The 1996 data collected by Swennen et al. (1997) were grouped to the same areas as the present study to compare the results of this study with those of 1996 using the logistic regression model (Table 5). The results show an increase in five areas C (Pattaya), E (Pet Buri), F (Ban Khau), I (Rusamilae), and L (Panare) and a decrease in three areas D (Si Racha), J (Laem Nok), and K (Pattani Bay Mouth). The overall frequency significantly increased from 21.3% to 25.2% (Pearson chi-squared = 4.95 with 1 df, $p = 0.026$).

5. Discussion

Our investigation is currently the most extensive survey of imposex in the Gulf of Thailand. It shows that the TBT concentrations in the Gulf of Thailand are so high that imposex in female neogastropods was recorded in all 13 areas sampled. The overall imposex frequency has increased compared to 1996 (Swennen et al., 1997). Both studies identified the same locations with the highest occurrence of imposex, namely the eastern part of the Bight of Bangkok (C, D) followed by Pattani Province (H–K). Increases were also reported around Phuket in the Andaman Sea (Bech, 1999). Likewise, in the North Sea, the imposex frequency increased between surveys in 1991 and 1999 (Ten Hallers-Tjabbes et al., 2003).

The 1996 and 2006 studies differ somewhat in areas sampled, species composition sampled, and number of individuals collected, which makes a detailed comparison for each area difficult. Some changes can still be noted. The western part of the Bight of Bangkok (areas E, F), where no imposex was found in 1996, showed 3.1% imposex off Phet Buri (area E) and 20.8% imposex off Ban Khau (area F) in 2006. During both surveys not a single large ship was seen in these areas. The observed increase may be caused by a gradual dispersion of TBT from the eastern side. In Pattani Province in the southern part of Thailand, the species-adjusted imposex occurrence decreased slightly in Pattani Bay Mouth (area K), which is along the shipping route, and Laem Nok (area J) in Pattani Bay, but increased in Rusamilae (area I) and Bang Tawa (area H). All the areas in Pattani Province (H-K) are only accessible to small ships, mainly wooden fishing boats. Large commercial ships pass a long distance away from the coast. Contrary to the situation in 1996, no TBT containing paints were found on and around the local dry docks. A switch to vinyl copper paints may explain the decrease of imposex in J and K. The increase in H and I may have been caused by extensive suction dredging in the mouth of the Pattani River and Pattani Bay to construct a larger harbour and deepening the shipping route for large fishing boats to the Gulf of Thailand. The sediment has been deposited on the intertidal area west of the excavated channel against the mainland. That area was surrounded by a low stone dam that kept most of the sand fraction but allowed the heavily polluted fine material in suspension to disperse. Tidal movements spread the mud cloud that settled over the intertidal and subtidal area to the west. We speculate that the TBT contaminated mud from the shipping lane increased the imposex incidence in Rusamilae (area I) from only 0.3% during the 1996 study to 15.6% in 2006. A similar situation was found in the adjacent area Bang Tawa (area H) where no imposex was found in 1996 but 17.9% in 2006, and no imposex was found further westward in 1995 (C. Swennen pers. obs.). Considering the high incidence in the western sampling sites, TBT contamination could be present in areas that were off-limits in 2006 due to safety risk by local insurgents. These findings show that regulation for the safe disposal of contaminated sediments and of anti-fouling paints after cleaning ships in dockyards is urgently needed in Thailand. For the most southern part of the Gulf of Thailand (Tak Bai, area M), the difference between both surveys is negligible (one individual showed imposex in 145 females in 1996 and two individuals in 156 females in 2006). The observed frequency for imposex from Tak Bai was 1.3% (2 of 156 females sampled). According to the contamination model a much higher occurrence can be expected (Fig. 1D). The Tak Bai samples contained 156 females, but all but three were from species with very low sensitivity to imposex. If the samples from Tak Bai had the typical representation of the species sampled from the Gulf in this study, the risk of imposex would be higher than 1.3%, because the sample would then contain species with a higher sensitivity to imposex. Therefore, an estimated 10.1% risk at Tak Bai is quite plausible (see also Tables 3 and 4, and the basic data added to the online version).

The suspicion that the local species may differ in sensitivity to TBT (Swennen et al., 1997) has been strengthened by the present study involving larger samples per site. Although the local TBT contamination is the dominant factor, *M. musiva*, *Pugilina cochlidium*, and *Babylonia areolata* seems less sensitive for developing imposex while *Thais lacera*, *Murex occa*, and *L. blosvillei* seem most sensitive among the species used in this study (Table 4). The lower sensitivity in *Pugilina* and *Babylonia* was also found in the first survey. Bech (2002) found also that *M. musiva* is a less sensitive species. It is not clear what factor induces the difference in sensitivity to TBT. Imposex appears wide-spread in Southeast Asia and has been reported in intertidal gastropods from various Southeast Asian areas, such as the Andaman coast of Thailand (Bech, 1999; Bech et al., 2002), Singapore (Tan, 1999), Hong Kong (Blackmore, 2000; Leung et al., 2006), China (Shi et al., 2005), and the Asia-Pacific Mussel Watch showed sites with high TBT values in all countries between China and India (Sudaryanto et al., 2002).

TBT not only kills larvae of fouling organisms that will settle on ships and other installations at sea, but also affects a broad range of non-target organisms in the wider surroundings. TBT residues have been found in algae (Maguire et al., 1984), crabs (Lee, 1985), fish (Lee, 1985; Kannan et al., 1996; Kannan and Falandysz, 1997), sea birds (Guruge et al., 1997; Kannan and Falandysz, 1997), seals (Kim et al., 1996; Berge et al., 2004), dolphins (Iwata et al., 1995; Kannan and Falandysz, 1997; Berge et al., 2004), and in humans (Zaucke and Krug, 1997; Whalen et al., 2002; Grün and Blumberg, 2006) and is far from harmless in humans (Aluoch et al., 2006). In sublethal concentrations, it induces the growth of male characters such as a penis and a pallial vas deferens (imposex) in most neogastropods. As result of this the females of some affected species die and populations become extinct in shipping routes and around harbours. In about 200 gastropod species imposex has already been found (Shi et al., 2005). In bivalves it can induce shell abnormalities and economic damage is reported from oyster cultures (Alzieu et al., 1981, 1986; Phelps and Pace, 1997). Other effects usually remain hidden, but laboratory studies showed effects of sublethal concentrations on the growth and development of bivalve larvae (Stenalt et al., 1998; Coelho et al., 2001), meiobenthos (Gustafsson et al., 2000). In fish, a low TBT level induces masculinization (Shimasaki et al., 2003), disturbs visual and olfactory functions (Wang and Huang, 1999) and causes chromosomal aberrations (Cipriano et al., 2004). It means that the anti-fouling paints seriously disturb the food chains in the marine environment.

Regrettably, the International Maritime Organization (IMO) did not recommend a total ban of TBT on ships and structures in sea until September 2008, while discussions about this began in 1996. Preferably, the Thai government should have taken precautionary measures to reduce pollution of the Gulf of Thailand by banning the use of TBT containing anti-fouling paint on ships and structures at sea such as landing piers, gas and oil installations, in aqua-culture in their part of the Gulf. Furthermore, the Thai government could have requested appropriate compensation for using mooring facilities along the Thai coast from foreign vessels with these paints. The extensive TBT pollution in the Gulf of Thailand has been known since the 1990s (Swennen et al., 1996, 1997); even Thai newspapers paid attention to it. Thousand of local fishermen depend on marine products from these waters, and local people and tourists eat these sea foods. TBT contaminated food can impose a risk to both the local population and impact the tourist industry.

Acknowledgements

We like to thank the several anonymous fishermen that allowed us to collect the gastropods from their catches, Ni-hasem Waesalae, Somsak Buatip, Masakree Sani and Uma Suetae for their help in collecting and processing the gastropods. We thank Professor Don McNeil for suggesting the use of logistic regression to adjust imposex frequency for different species sensitivity, Dr. A.K.A. Holthuijzen and the anonymous referees for detailed comment that helped us to improve the final text.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.marpolbul.2008.11.028.

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