

## Butyltin Compounds Distribution in the Coastal Waters of Bohai Bay, People's Republic of China

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TBT is one of the most toxic chemicals to the aquatic organisms. It was reported that TBT at 2ng/l would inhibit calcification in *Crassostrea gigas* (Chagot et al., 1990) and induce imposex in the dogwhelk *Nucella lapillus* (Waldock et al., 1987). The decline of dogwhelk population on various coasts of France and the UK has been attributed to the occurrence of TBT in these waters (Bryan et al., 1986; Gibbs et al., 1986). Although TBT has been legislatively banned in antifouling paints from the late 1990s in most European and North American countries (Chau et al., 1997) due to its toxicity to aquatic life at low concentration, there is still no specific legislation controlling the use of TBT in China. Fragmentary data about the occurrence of organotin compounds in China showed that some aquatic environment had been seriously polluted by butyltin compounds (Jiang et al., 2001). Bohai Bay is a large, semi-enclosed shallow water basin located along the western region of the Bohai Sea in the northeastern part of China. While it has been suffered from growing pollution in its coastal areas for the last few decades, which led to the disappearance of many species, no knowledge about the occurrence of butyltin compounds in Bohai Bay is available.

Most of analytical methods for determination of organotin compounds are based on the use of hyphenated techniques that combine gas chromatography with element-selective detector. Ionic organotin compounds in environment generally have high boiling points and need to be derivatized into volatile forms prior to separation by GC. The derivatization methods included hydride generation with NaBH<sub>4</sub> (Jiang et al., 2001; Donard et al., 1986) and alkylation by NaBEt<sub>4</sub> (Eiden, et al., 1998; Millan, et al., 2000) or Grignard reagents (Fent et al., 1995). Generally, these methods all need extraction and cleanup with large amount of harmful organic solvent. Recently, headspace solid-phase micro-extraction (SPME), a one-step procedure, has been successfully combined with GC-FPD to determine butyltin compounds in environmental waters (Jiang et al., 2001) and sediments (Millan et al., 2000). In this paper, SPME-GC-MS combined hydride

generation with  $\text{NaBH}_4$  was developed and applied to determine the concentrations of TBT and DBT in coastal Bohai Bay, and the spatial distribution of their concentration was described by Kriging method, which allows immediate appreciation of the change in the contaminant with space and enables identification of areas that may contain hazardous concentrations (Markus et al., 2001).

## MATERIALS AND METHODS

Dibutyltin dichloride (DBT, 97% purity), tributyltin chloride (TBT, 95% purity) and their isotope internal standard DBT- $\text{d}_{18}$  and TBT- $\text{d}_{27}$  (100ppm in hexane, concentration is as chloride form), were all purchased from Hayashi Pure Chemicals (Tokyo, Japan). Sodium tetrahydroborate ( $\text{NaBH}_4 \geq 98\%$ ) was purchased from Shanghai chemical Co. (China). These reagents were used without further purification. Water was obtained by a compact ultrapure water system (EASY pure UV, USA). HPLC grade methanol was obtained from Fisher Scientific (New Jersey, USA). Individual stock solutions of DBT and TBT (1000mg/l as Sn) were prepared in methanol. A mixed butyltin working solution containing 200mg/l (as Sn) of each compound was also prepared using methanol as solvent. Dilution in methanol of the latter solution was used as required. All the solutions were stored at 4°C in the dark. Fresh  $\text{NaBH}_4$  solution of 3% (w/v) was prepared with ultrapure water just before use. Acetate buffer was made from acetic acid and sodium acetate solution and stored in a glass bottle at 4°C. The SPME holder and the fiber coated with 100 $\mu\text{m}$  thickness polydimethylsiloxane (PDMS) were obtained from Supelco (Bellefonte, PA, USA). A HP 6890 gas chromatograph equipped with a split/splitless injection port and coupled to a HP 5973 mass selective detector (Agilent Technologies Co., USA) using electron impact in the selected ion monitoring (SIM) mode was used. Separations were performed in a capillary column (HP-5MS, 60m x 0.32mm i.d.x0.25 $\mu\text{m}$  coating thickness) throughout the experiment. The carrier gas was helium of high purity (99.9999%). The whole system was controlled by G1701DA Enhanced ChemStations. Kriging method was used to map the spatial distribution of butyltin compound by Surface Mapping System Surfer Version 7.02.

An appropriate amount of the DBT-TBT mixed standard solution and its isotope internal standard were added into 21 ml ultrapure water (pH=3.77, adjusted by acetate buffers) in 40-ml amber glass vials sealed with PTFE-lined silicon septa (Supelco). After adding 1ml 3%  $\text{NaBH}_4$  solution, the vial was immediately closed and placed on the magnetic stirrer. The vial contents were stirred magnetically at the maximum rate allowed by the stirrer (CORNING PC-420, USA) with Teflon coated magnetic stirrer bar. When the constant stirring rate was obtained, the

SPME fiber was immediately exposed to the headspace over the vigorously stirred solution at room temperature. After 20 min, the fiber was withdrawn into the needle of the holder and SPME was placed in the GC injector. The injector and detector temperature were 250°C and 280°C, respectively. Desorption time was 3 min, and no carryover was observed after this desorption time. The temperature programs used for GC column were: 60°C for 1 min, 20°C/min to 100°C, then 8°C/min to 250°C, hold 0.5min. The concentrations of DBT and TBT in samples were determined by selecting ions as Table 1.

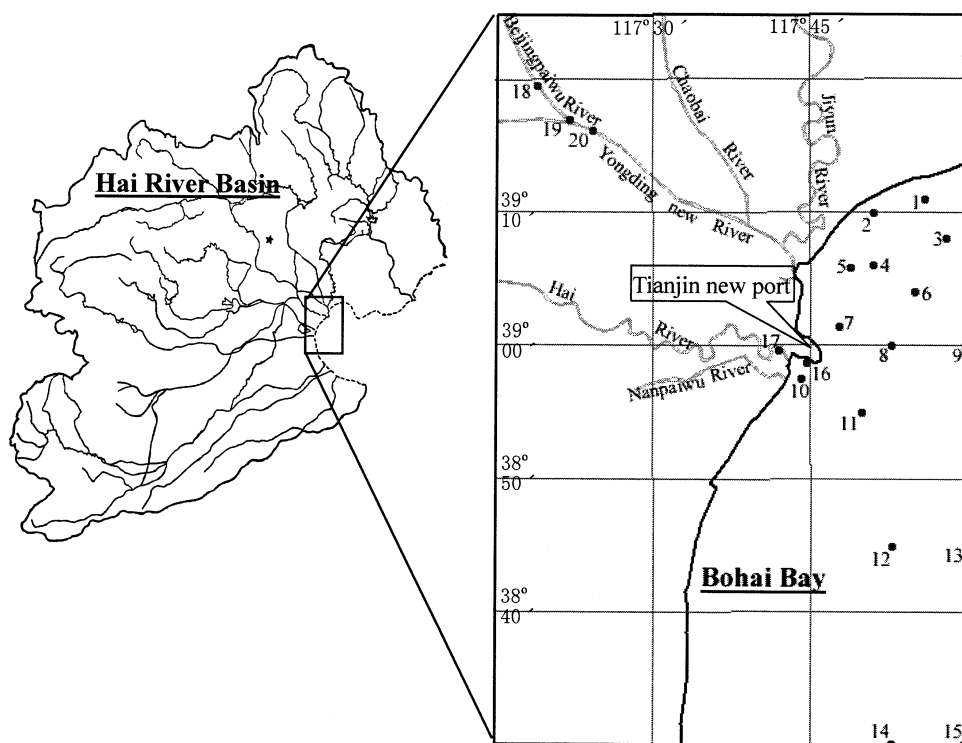
**Table 1.** Ions monitored in SIM mode

Time window (min)	Analyte	Molecular weight	Ion monitored
7.00 to 15.00	DBT-d <sub>18</sub>	321.83	118, 120, 121, 123, 186, 188
	DBT	303.83	118, 120, 121, 123, 177, 179
15.00 to 17.70	TBT-d <sub>27</sub>	352.51	120, 185, 186, 187, 188, 189, 251, 253
17.70 to 20.00	TBT	325.51	121, 175, 176, 177, 178, 179, 233, 235

Water samples were manually collected in summer of 2002 using amber glass bottles from just beneath the surface. To avoid microlayer contamination the bottle cap was carefully removed under water surface. The unacidified and unfiltered samples were stored at 4°C. Most of seawater samples were collected at various stations along the coastal Bohai Bay. Stations were chosen to provide good area coverage, including potential sources and fishing area. The station 16, that is to say, Tianjin New Port, is a potential pollution source. Tianjin New Port is located on the side of Bohai Bay. It is a maximal port in the north of China and the largest container-distributing center in China. Station 5 is located near a small fishing dock. Four samples were taken from Hai River, Beijingpaiwu River and Yongding New River which are all adjacent to Bohai bay. Geographical coordinates were determined with a mobile GPS at each sampling station. A sampling station map is shown in Fig.1.

## RESULTS AND DISCUSSION

Fig.2 shows the mass spectra of Bu<sub>2</sub>SnH<sub>2</sub> and Bu<sub>3</sub>SnH that were characterized by clusters of isotope ion at each fragment. A baseline separation was easily achieved for DBT and TBT under the GC conditions used in this study as shown in Fig.3 (a). An internal standard quantification strategy was employed to minimize the response variation. The isotope pattern created by ten tin isotopes contributions is particularly useful for recognition of any organotin compounds occurring in a

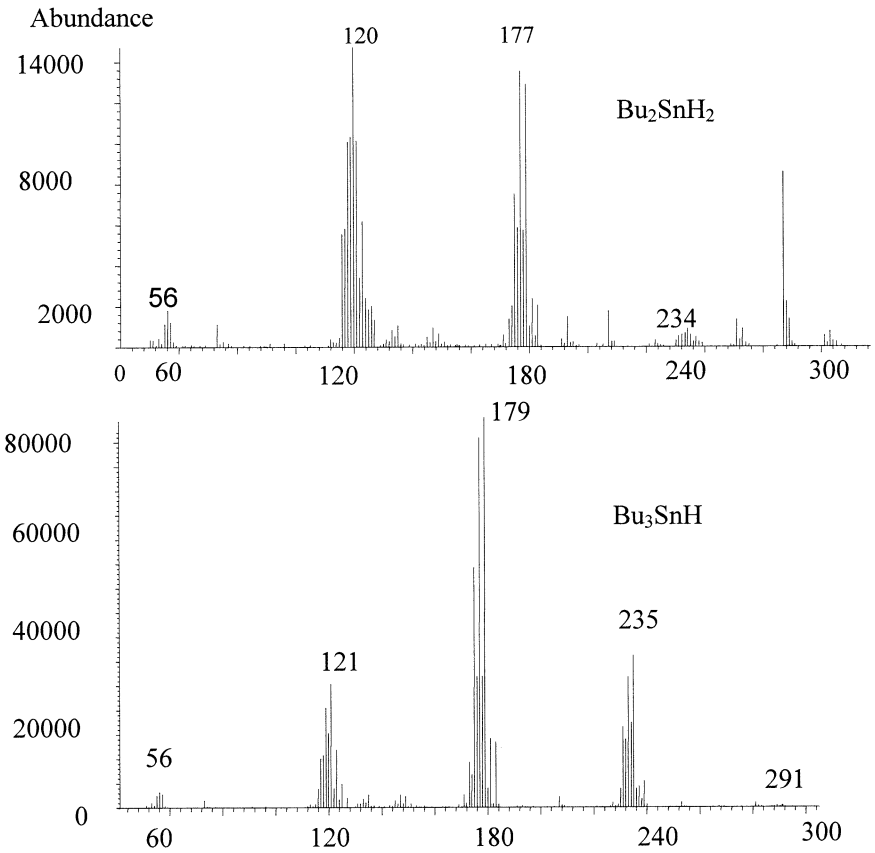


**Figure 1.** Map of sampling stations. Numbers indicate station No

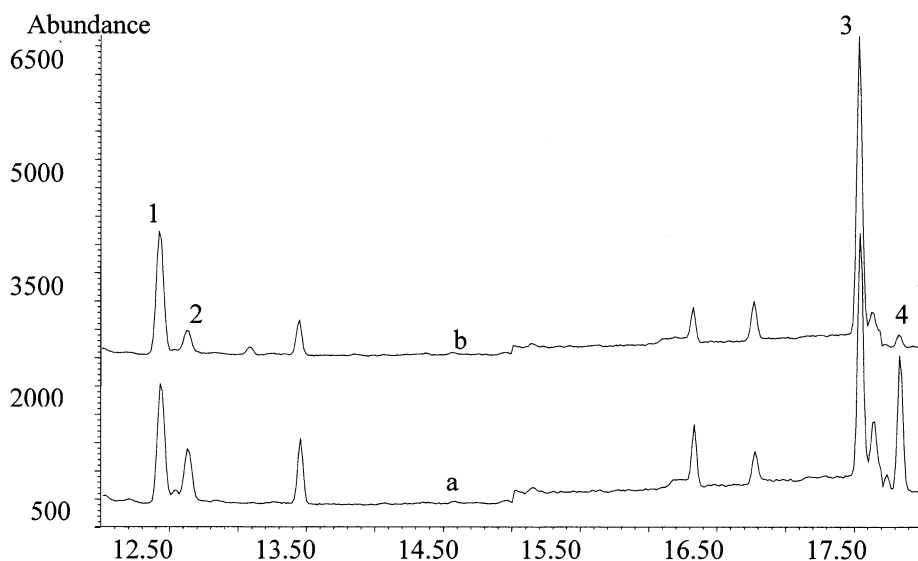
sample. Peaks in the chromatograms were assigned to individual organotin compounds on the basis of retention time. Peak areas were used to quantitative calculation for DBT and TBT. The detection limits of DBT and TBT in SIM mode by this method were 0.47, 1.25 ng Sn/l, respectively. The average recoveries of DBT and TBT based on standard spiking method were between 90 and 112% (n=5) for each compound.

Fig.3 (b) shows the GC-MS chromatogram of a seawater sample in selective ionization mode (SIM), which clearly indicated the presence of DBT and TBT. The concentrations of TBT and DBT compounds in unfiltered water samples from Bohai Bay were shown in Table 2. TBT and DBT compounds were detected in most of water samples: TBT in 13 out of 16 samples, DBT in all 16 samples. TBT concentrations varied from below the detection limits at stations 12, 13, 14 and 15 to the highest level of 14.23 ng Sn/l at station 16 (Tianjin New Port). The next high concentration of 9.07ng Sn/l was observed at station 10 followed by the station 11 (7.44 ng Sn/l). Besides, another relatively high concentration of TBT was observed at station 5 (7.02 ng Sn/l). Relatively low levels of TBT were found

at stations 3, 8 and 9, which were all below 3.20 ng Sn/l. The concentrations at stations 1, 2, 4, 6, and 7 were very similar (4.35-5.94 ng Sn/l), and these stations were all around station 5 or near coast. Collectively, levels of TBT in water samples declined with increasing distance from coast. On the other hand, relatively high levels of DBT were found at all stations, and ranged from the lowest level of 7.07 ng Sn/l at station 9 to the highest level of 64.84 ng Sn/l at station 10. The concentrations of TBT and DBT in Bohai Bay were similar with those in Tokyo Bay in January 1994 (Hashimoto et al., 1998). Moreover, TBT/DBT ratios at all stations were from 0.04 to 0.33, which were also similar with that in Tokyo Bay in January 1994 that was 0.16 (Hashimoto et al., 1998), but were lower than the ratio of 0.8 in Alexandria Harbors (Aly MA et al., 1995). In addition, four samples from three rivers adjacent to Bohai Bay were also detected to assess the possibility of TBT and DBT inputs via Hai River basin. Of four samples, TBT was detected only in the samples from Hai River, and the concentration was 3.36 ng Sn/l, which was considered that there were some boats



**Figure2.** Full scan EI spectra of  $\text{Bu}_2\text{SnH}_2$  and  $\text{Bu}_3\text{SnH}$ . Scan ranged from 50 to 300



**Figure 3.** GC-MS chromatogram of seawater sample from Tianjin new port. a, standard butyltin hydrides; b, Tianjin new port water sample. Peaks identified as: 1.  $\text{Bu}_2\text{SnH}_2\text{-d}_{18}$ ; 2.  $\text{Bu}_2\text{SnH}_2$ ; 3.  $\text{Bu}_3\text{SnH-d}_{27}$ ; 4.  $\text{Bu}_3\text{SnH}$

**Table 2.** Concentrations of DBT and TBT in unfiltered seawater from Bohai Bay and Hai River basin (ng Sn/l)

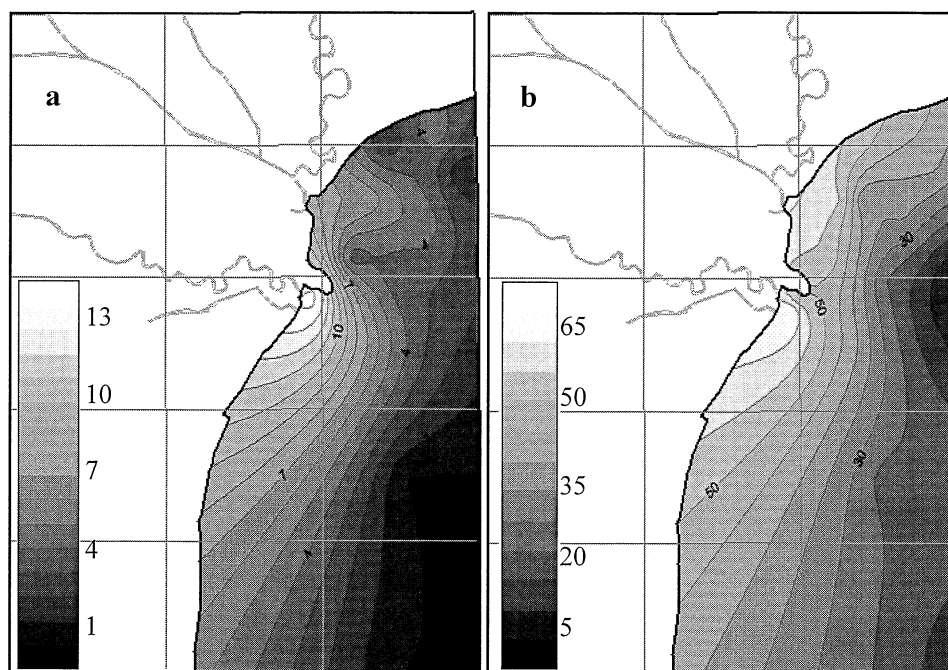
Sampling station	Position coordinate		Concentration (ng Sn/l)	
	North latitude	East longitude	DBT	TBT
1	39°10.99 ′	117°55.99 ′	45.27	4.81
2	39°10.00 ′	117°51.00 ′	51.59	4.35
3	39°8.00 ′	117°58.00 ′	39.98	1.43
4	39°6.00 ′	117°51.00 ′	35.15	5.94
5	39°4.99 ′	117°45.96 ′	55.53	7.02
6	39°3.99 ′	117°54.99 ′	34.60	4.85
7	39°0.99 ′	117°47.00 ′	53.23	5.29
8	38°59.93 ′	117°52.90 ′	25.67	3.14
9	38°59.99 ′	117°59.96 ′	7.07	2.36
10	38°58.00 ′	117°43.00 ′	64.84	9.07
11	38°55.00 ′	117°50.00 ′	54.79	7.44
12	38°44.91 ′	117°52.98 ′	23.08	ND*
13	38°45.11 ′	118°0.03 ′	25.71	ND
14	38°30.01 ′	117°52.96 ′	24.11	ND
15	38°30.08 ′	117°59.93 ′	16.54	ND
16	38°58.74 ′	117°44.66 ′	48.10	14.23
17	38°59.17 ′	117°42.77 ′	26.71	3.36
18	39°39.99 ′	116°53.36 ′	36.87	ND
19	39°16.92 ′	117°22.39 ′	33.48	ND
20	39°16.67 ′	117°22.82 ′	46.16	ND

\* ND indicated that the concentration was below the detection limit 1.25 ng Sn/l

activities in Hai River. DBT was detected in all four samples, which ranged from 26.71 ng Sn/l (station 17 in Hai River) to 46.16 ng Sn/l (station 20 in Yongding New River). The results suggested that the main source of TBT contamination in Bohai Bay was shipping activities, and the input from river basin is another source for DBT besides the TBT degradation in Bohai Bay.

Generally, TBT in the aquatic environment is a result of leaching from anti-fouling paints, although it also has been found as a result of other uses, such as the use as preservative for timber. DBT is usually a result of TBT degradation. However, because DBT is common stabilizer for PVC and catalysts for polyurethane foams, silicones, etc (Fent, 1996), there is a possibility of leaching from PVC and other materials. The existence of DBT in land stations, such as in Beijingpaiwu River and in Yongding New River, may be due to leaching from PVC or other materials.

Based on the above discussion, there is need to obtain more immediate appreciation of TBT and DBT change with space and enables identification of areas that may contain these two compounds. In this paper, TBT and DBT concentration levels from 16 monitoring stations were used to obtain the spatial patterns of TBT and DBT pollution. The optimum interpolation technique, Kriging, was used to describe the spatial distribution of TBT and DBT concentration. Fig.4 (a) and (b) depicted the spatial distribution of TBT and DBT compounds in seawater of Bohai Bay in summer of 2002. From the figure, it was intuitively clear that there were relatively high concentrations of TBT and DBT in coastal waters and decreased outside of the coast. Station 16 was a hot-spot of contamination and station 5 was a slight one, which were all attributed to some shipping activities. The concentrations in other stations decreased with the distance from the two hot-spots. There were also two hot-spots for DBT contamination at station 16 and station 5, respectively, but hot-spot at station 5 for DBT was more obvious than for TBT. Moreover, DBT distributed more widely than TBT, and its concentration declined more slowly with increasing distance from coast compared with TBT. Such a spatial distribution of DBT indirectly suggested there would be other sources for DBT besides the TBT degradation in Bohai Bay.



**Figure 4.** Spatial distribution of butyltin in coastal waters of Bohai Bay. (a) tributyltin (TBT), (b) dibutyltin (DBT). The numbers represent the concentrations of butyltin (ng Sn/l)

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