Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Chromatography A, 1048 (2004) 81–88
JOURNAL OF CHROMATOGRAPHY A
www.elsevier.com/locate/chroma

Liquid-phase microextraction of tributyltin and triphenyltin coupled with gas chromatography–tandem mass spectrometry Comparison between 4-fluorophenyl and ethyl derivatizations

Hirotaka Shioji[a], Shinji Tsunoi[a,*], Hiroya Harino[b], Minoru Tanaka[a]

[a] *Research Center for Environmental Preservation, Osaka University, Yamada-oka 2-4, Suita, Osaka 565-0871, Japan*
[b] *Osaka City Institute of Public Health and Environmental Sciences, Tennoji, Osaka 543-0026, Japan*

Received 31 March 2004; received in revised form 7 July 2004; accepted 9 July 2004

Available online 8 August 2004

Abstract

This paper describes the liquid-phase microextraction (LPME) of tributyltin (TBT) and triphenyltin coupled with gas chromatography–tandem mass spectrometry. The 4-fluorophenylation and ethylation reactions were used for the derivatization of the organotins. For the two derivatizations, the LPME parameters such as organic solvent, stirring rate, temperature, extraction time and the other additional conditions were examined. Using pure water, the calibration curves, method detection limits (MDLs) and reproducibilities (RSDs) of the two derivatizations were compared under the respective optimized procedures. The 4-fluorophenyl derivatization, which showed a lower MDL (0.36 ng/l) and better reproducibility (RSD = 11% at 10 ng/l) for TBT, was applied to the analysis of seawater. The TBT was detected in the range from 1.1 to 2.0 ng/l in the seawater samples collected in Osaka Bay.

Keywords: Ethylation; 4-Fluorophenylation; Gas chromatography–tandem mass spectometry; Liquid-phase microextraction; Seawater sample; Tributyltin; Triphenyltin

1. Introduction

Tributyltin (TBT) and triphenyltin (TPT) have been used as a biocide in antifouling paints over the past 30 years. As a result, their use brought about contamination of the seawater. At present, TBT and TPT are well known as endocrine disruptors and have an influence on marine organisms even at a low ng/l level [1].

In the analysis of organotins, despite the need for preliminary derivatization, gas chromatography (GC) is more widely used than high-performance liquid chromatography. Derivatized organotins are generally detected by the selective techniques such as mass spectrometry (MS) [2,3], tandem mass spectrometry (MS–MS) [4,5], flame photometry [6,7], inductively coupled plasma-mass spectrometry [8–10] and so on. Generally, alkylation with Grignard reagents and ethylation with sodium tetraethylborate are used for the derivatization. In 1998, propylation with sodium tetra(*n*-propyl)borate was proposed by Vandyck [11]. Recently, we reported that 4-fluorophenyl derivatization with sodium tetrakis(4-fluorophenyl)borate offered a high sensitivity for the GC–MS–MS determination of TBT [12].

With regard to the progress of extraction, liquid–liquid extraction (LLE), which requires large amount of toxic organic solvent and is a time-consuming method, has been replaced by solid-phase extraction (SPE). The SPE requires a lower amount of the organic solvent, but still requires an appreciable amount. In the early 1990s, Belardi and Pawliszyn developed a new solvent-free extraction technique, called solid-phase microextraction (SPME) [13]. This technique is much more rapid and simpler than the traditional methods because it integrates the extraction, concentration and injection into a single step. The application of the SPME to the analysis

* Corresponding author. Tel.: +81 6 6879 8977; fax: +81 6 6879 8978.
E-mail address: tsunoi@epc.osaka-u.ac.jp (S. Tsunoi).

0021-9673/$ – see front matter © 2004 Elsevier B.V. All rights reserved.
doi:10.1016/j.chroma.2004.07.016

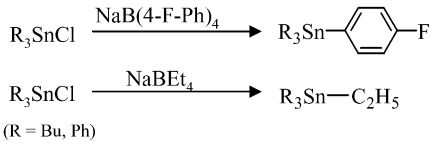

Fig. 1. Reactions of TBT and TPT with derivatizing reagents.

of organotins has already been reported [4,8–10,14]. However, the SPME has some drawbacks, e.g., its fiber is fragile and it needs some special instruments like an SPME holder. Recently, liquid-phase microextraction (LPME), which requires a small amount of organic solvent and is a simple and inexpensive method, has been developed [15,16]. The LPME is performed by directly immersing a small-microlitre drop of the organic solvent at the tip of a microsyringe in a stirring aqueous sample solution. The important feature of the LPME is that almost all of the organic solvent into which the analytes are extracted can be injected into the GC, while only part of the concentrated organic solvent is injected using LLE or SPE. In addition, apart from being inexpensive, only common laboratory equipment is required. Subsequently, some developed methods such as liquid–liquid–liquid microextraction [17,18], hollow fiber LPME [19] and headspace solvent microextraction [20] have been reported.

We investigated the LPME of TBT and TPT in an aqueous sample combined with GC–MS–MS. For the derivatizations, 4-fluorophenylation with sodium tetrakis(4-fluorophenyl)borate and ethylation with sodium tetraethylborate (Fig. 1) were used. These derivatizations that can be performed in aqueous media seem to be appropriate for the LPME. The two derivatizations were optimized and their analytical performances were compared with each other. Finally, an optimized procedure was applied to the analysis of seawater samples.

2. Experimental

2.1. Materials

Triphenyltin chloride (TPT-Cl) was purchased from Strem Chemicals (Bischeim, France), tributyltin chloride (TBT-Cl) from Wako (Osaka, Japan) and deuterated organotin compounds from Hayashi Pure Chemical Industry (Osaka, Japan). Individual stock standard solutions of TBT-Cl and TPT-Cl (1 mg/l as Sn) were prepared by diluting them in acetone. Working standard solutions were prepared by mixing the stock standard solutions, and further dilution was carried out with acetone. Surrogate standard solution, which calibrates the LPME process, was prepared by dissolving perdeuterated TBT-Cl and TPT-Cl in acetone. Hexyl TBT synthesized in our laboratory [5] was used as the internal standard and its standard solution was prepared by dissolving it in an extraction solvent. All the standard solutions were stored in the dark at 4 °C. Sodium tetrakis(4-fluorophenyl)borate dihydrate and sodium tetraethylborate were provided from Dojindo (Kumamoto, Japan) and Strem Chemicals, respectively. α,α,α-Trifluorotoluene was purchased from Aldrich (Milwaukee, WI, USA) and the other solvents and chemicals were from Wako. Buffer solutions with pH values that ranged from 3 to 8 were prepared by mixing acetic acid and sodium acetate. Water was processed through a Milli-Q VOC water purification system (Millipore, Bedford, MA, USA).

2.2. Instrumentation and conditions

All analyses were performed with a Finnigan MAT GCQ (San Jose, CA, USA) ion trap mass spectrometer equipped with a Finnigan GC. The column was a DB-5MS (30 m × 0.25 mm i.d., 0.25 μm d_f, J & W Scientific, Folsom, CA, USA). The carrier gas was high purity helium (99.9999%) with a constant linear velocity of 40 cm/s. The GC oven temperature was programmed as follows: 1 min at 50 °C, to 290 °C at 20 °C/min, 7 min at 290 °C (total analysis time, 20 min). The ion source, injection and transfer-line temperatures were set at 200, 270 and 290 °C, respectively. All injections were performed in the splitless-mode with the split vent closed for 1 min. The mass spectrometer was operated in the electron ionization mode. For MS–MS, the product ions were monitored by selected reaction monitoring. The optimized MS–MS conditions are shown in Table 1.

2.3. Optimized LPME procedures

A sketch of the LPME apparatus is shown in Fig. 2. The optimized LPME procedure for the 4-fluorophenyl derivatization is as follows: to a 4-ml water sample in a 5-ml glass sample vial, the standard solution of organotins (20 μl), surrogate solution (500 pg/μl, 5 μl), buffer solution (pH 3 (0.1 M), 100 μl) and tetrakis(4-fluorophenyl)borate (4 mg) were added. The vial was placed in the temperature-controlled water bath (14 °C) on a magnet stirrer, and the solution was stirred at 500 rpm. α,α,α-Trifluorotoluene (3 μl) containing 20 μg/l of hexyl TPT was loaded into a 10 μl mi-

Table 1
MS–MS conditions

Compound	Precursor ion (*m*/*z*)	Product ion (*m*/*z*)	CID voltage (V)
4-Fluorophenyl TBT	329	273	0.75
[$^2H_{27}$]4-Fluorophenyl TBT	347	283	0.75
4-Fluorophenyl TPT	369	197	1.2
[$^2H_{15}$]4-Fluorophenyl TPT	379	202	1.2
Ethyl TBT	291	235	0.7
[$^2H_{27}$]Ethyl TBT	318	254	0.7
Ethyl TPT	351	197	1.3
[$^2H_{15}$]Ethyl TPT	366	202	1.3
Hexyl-TPT	351	197	1.4

Other conditions: isolation time, 8 ms; excitation time of precursor ion, 24 ms for 4-fluorophenyl TPT and [$^2H_{15}$] TPT and 15 ms for others; CID: collision-induced dissociation.

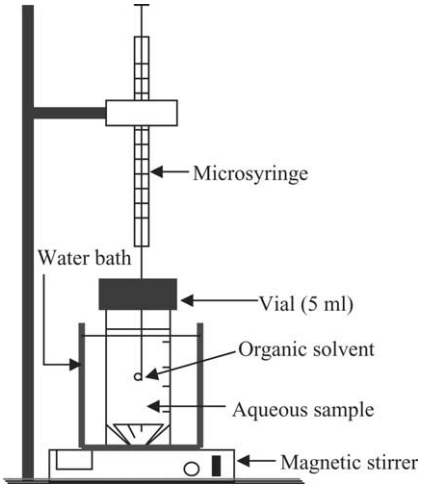

Fig. 2. LPME apparatus.

crosyringe with a 22° bevel (Hamilton), which was clamped above the vial. The syringe needle passed through the septum of the vial and a solvent drop was retained at the tip of the needle in the aqueous sample. After extraction for 60 min, the solvent drop was withdrawn back into the microsyringe, and 2 μl of the solvent was then injected into the GC.

The optimized LPME procedure for the ethyl derivatization was the same as the 4-fluorophenyl derivatization except for the concentration of the buffer (0.5 M) and amount of the derivatizing reagent (0.05% aqueous solution, 50 μl).

The average value of the peak area ratio of the derivatized organotin to hexyl TPT, which was obtained from triplicate analyses, was used for optimizing the LPME parameters. The calibration curve was evaluated by plotting the peak area ratio of the analyte to the corresponding surrogate standard versus the concentration of the analyte.

2.4. LPME experiment using pre-derivatized TBT and TPT

In order to confirm whether the buffer and derivatizing reagent affect the extraction, the LPME was performed using 4-fluorophenyl TBT and TPT synthesized in our laboratory [12]. When the effect of the buffer was examined, 4-fluorophenyl TBT and TPT were added instead of the standard and surrogate solutions and tetrakis(4-fluorophenyl)borate in the optimized procedure. In the case of the derivatizing reagent, 4-fluorophenyl TBT and TPT were added instead of the standard and surrogate solutions.

2.5. Enrichment factor and extraction efficiency

The enrichment factor (EF) was defined as the final analyte concentration C_{final} in the solvent drop divided by the initial sample concentration C_{initial}:

$$\text{EF} = \frac{C_{\text{final}}}{C_{\text{initial}}}$$

The extraction efficiency (EE) was defined as the total amount of the analyte in the solvent drop divided by the total amount in the initial sample:

$$\text{EE} = \left(\frac{C_{\text{final}} V_{\text{final}}}{C_{\text{initial}} V_{\text{initial}}}\right) 100 = \text{EF}\left(\frac{V_{\text{final}}}{V_{\text{initial}}}\right) 100$$

where V_{final} and V_{initial} are the volumes of the solvent drop and sample solution, respectively.

2.6. ^{119}Sn NMR study

TBT-Cl (10 mg, 0.03 mol), sodium tetrakis(4-fluorophenyl)borate dihydrate (100 mg, 0.4 mol) and toluene (2 ml) were added to pure water (5 ml) in a 10-ml glass sample vial. The mixture was then shaken at room temperature for 10 min. After phase separation, the organic phase was collected into an NMR tube via a short column of sodium sulfate. The solution was immediately identified by ^{119}Sn NMR (JEOL JNM-GSX-400). Chemical shifts were referenced to a tetramethyltin signal.

3. Results and discussion

3.1. Optimization of LPME using 4-fluorophenyl derivatization

The LPME parameters such as organic solvent, temperature, stirring rate, buffer, amount of sodium tetrakis(4-fluorophenyl)borate (TFB), solvent drop size, injection volume and extraction time were examined in order to achieve a sensitive analysis. The initial LPME conditions using TFB were as follows: temperature, 14 °C; stirring rate, 300 rpm; amount of TFB, 10 mg; solvent drop size, 2 μl; injection volume, 1 μl; extraction time, 30 min. The optimized parameters were used for the subsequent optimization.

3.1.1. Selection of organic solvent

A variety of solvents including toluene, octane, α,α,α-trifluorotoluene, xylene, ethylbenzene, cyclohexane, chloroform, hexane and benzene were tested in order to select the most suitable solvent. When hexane and benzene were used, bubble formation was observed in the solvent drop, and the growth of the bubble made the solvent drop rise up. This may

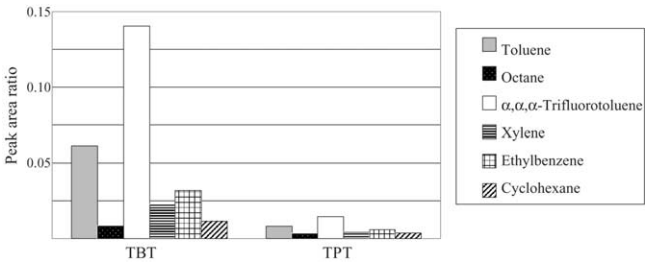

Fig. 3. Effect of solvent on 4-fluorophenyl derivatization.

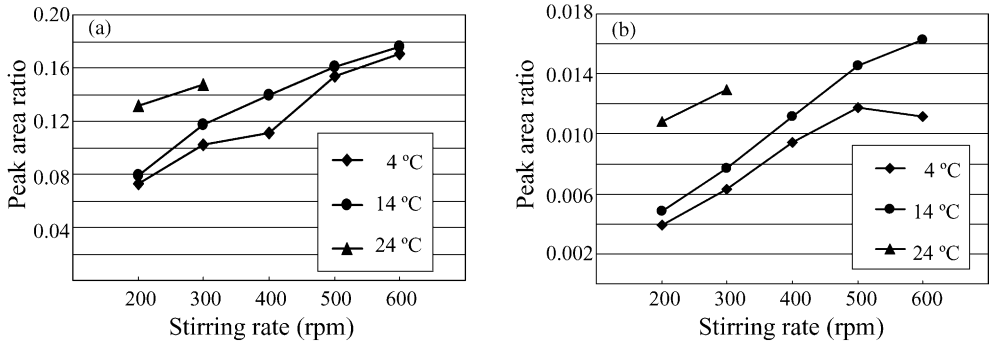

Fig. 4. Effects of temperature and stirring rate on 4-fluorophenyl derivatization of TBT (a) and TPT (b).

be due to their low boiling points and specific gravities. Since chloroform has a relatively high water solubility, the drop decreased to almost half size after the extraction. The results except for the chloroform, hexane and benzene are shown in Fig. 3. The aromatic solvents had higher sensitivities than the aliphatic solvents, and the highest peak area ratios were obtained with α,α,α-trifluorotoluene. α,α,α-Trifluorotoluene [21] having a relatively high boiling point (102 °C) and specific gravity (d = 1.199) was selected as the organic solvent. The reason for the high sensitivity with the aromatic solvents is described further (Section 3.1.3).

3.1.2. Effects of temperature and stirring rate

The effects of temperature (4–24 °C) and stirring rate (200–600 rpm) were evaluated (Fig. 4). The LPME performed at 24 °C caused the solvent drop to be unstable due to undesirable bubble formation in the solvent drop. At 24 °C, 300 rpm was the limitation for the stirring rate, while at 4 and 14 °C, it was possible to raise the stirring rate to 600 rpm. The highest peak area ratios were obtained at 14 °C and 600 rpm. The LPME sometime failed at 14 °C and 600 rpm, therefore 500 rpm was adopted. It was reported that a higher temperature allowed an increase in the extraction efficiency [22], however, a lower temperature was selected in order to stabilize the solvent drop in this study.

3.1.3. Effect of buffer

The pH of the buffer significantly affects the derivatization process using sodium tetraethylborate, so the effects of the pH and concentration of the buffer were evaluated by adding 100 μl of the acetate buffer into the aqueous sample (Fig. 5). Under the conditions of low pH and concentration (pH 3, 0.1 M), the highest peak area ratios were obtained. In order to examine whether these parameters affect the extraction step, a following experiment was carried out: to a 4-ml water sample, 4-fluorophenyl TBT and TPT synthesized and the buffer solution were added, and then the LPME was performed. By changing the pH and concentration of the buffer, there was no influence on the extraction efficiency, so it was considered that the pH and concentration of the buffer might affect the derivatization step. However, one curious result was obtained from the above experiments. At the same concentration (500 ng/l as Sn), the peak area ratios obtained from the LPME using 4-fluorophenyl TBT and TPT were one-third lower than those using TBT-Cl and TPT-Cl (data not shown). If TBT and TPT were extracted into the organic phase after the 4-fluorophenylation in aqueous media, higher or similar peak area ratios should be obtained by using 4-fluorophenyl TBT and TPT. We assumed that there might be an alternative derivatization path to enhance the extraction efficiency. To gain some insights into the 4-fluorophenylation, the reaction was monitored by ^{119}Sn NMR measurement. After shaking

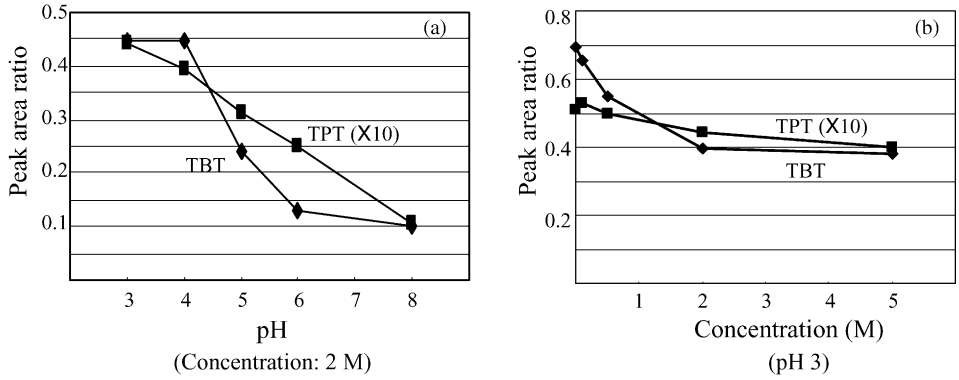

Fig. 5. Effects of pH (a) and concentration (b) of buffer on 4-fluorophenyl derivatization.

the mixture of TBT-Cl and TFB under the biphasic conditions (toluene–water) for 10 min, the organic phase was immediately subjected to the ^{119}Sn NMR measurement (Fig. 6a). The signal ascribed to 4-fluorophenyl TBT appeared at −39 ppm. The peak of TBT-Cl (145 ppm) was not observed. As seen in Fig. 6a, an unknown peak appeared at 99 ppm. After heating the tube at 40 °C for 1 h, the peak disappeared. However, no new peaks appeared (Fig. 6b), indicating that the unknown species changed to 4-fluorophenyl TBT. A similar result was obtained with TPT. These results indicate that the unknown species may probably be the ion pair of TFB and organotin, which is formed in the water phase and then extracted into the organic phase. We consider that the ion pair may be extracted into the organic phase more easily than the derivative.

The effect of the buffer is not clear at this stage, however, we consider that the concentration and pH of the buffer influence the formation of the ion pair.

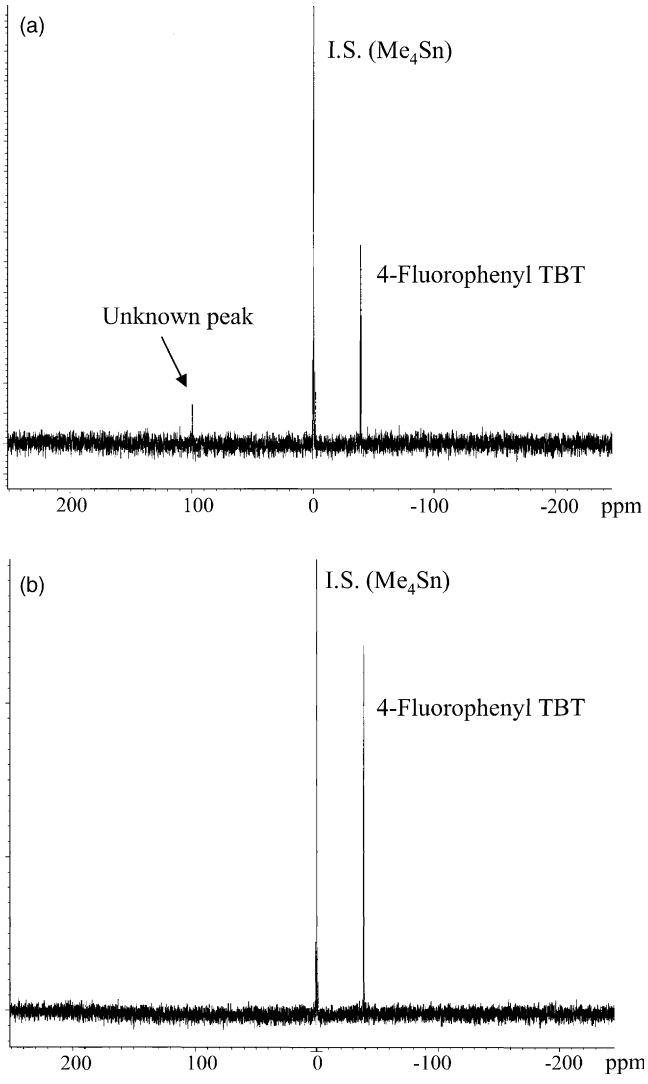

Fig. 6. ^{119}Sn NMR spectra of toluene extract after 4-fluorophenyl derivatization of TBT (a) and after the subsequent heating at 40 °C for 1 h (b).

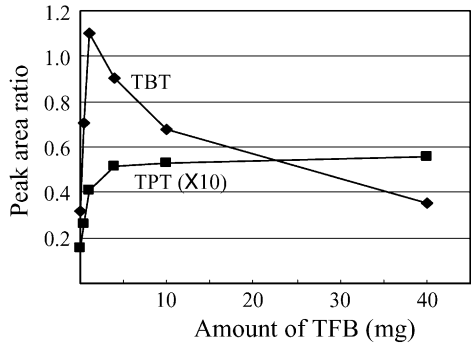

Fig. 7. Effect of TFB amount on 4-fluorophenyl derivatization.

The results obtained in the selection of solvent (Section 3.1.1) may be also explained by the ^{119}Sn NMR study. Aromatic solvents, especially α,α,α-trifluorotoluene, showed higher responses (Fig. 3). This may be attributed to the high affinity of the ion pair consisting of the organotin and TFB to α,α,α-trifluorotoluene.

3.1.4. Amount of derivatizing reagent

The range of the TFB amount investigated in this study was between 0.1 and 40 mg (Fig. 7). The peak area ratio of 4-fluorophenyl TPT increased with the increasing amount of TFB because the 4-fluorophenylation of TPT was relatively slow due to the steric effect of the phenyl groups. On the other hand, the peak area ratio of 4-fluorophenyl TBT decreased by adding more than 1 mg of TFB. When the effect of the TFB amount on the extraction step was examined using 4-fluorophenyl TBT and TPT synthesized, increasing the TFB amount resulted in a low extraction efficiency. This suggested that 4-fluorophenyl TBT may be stabilized by the excess TFB in the aqueous media, which makes it difficult to extract the derivative into the organic solvent. Taking both responses of 4-fluorophenyl TPT and TBT into consideration, 4 mg was selected for the spiking amount of the TFB.

3.1.5. Effect of solvent drop size

Using α,α,α-trifluorotoluene, the effect of the solvent drop size was investigated using the following four patterns: immersion/injection volume (μl/μl) = 2/1, 3/1, 3/2 and 4/2. When 3 μl of the organic solvent was immersed and 2 μl

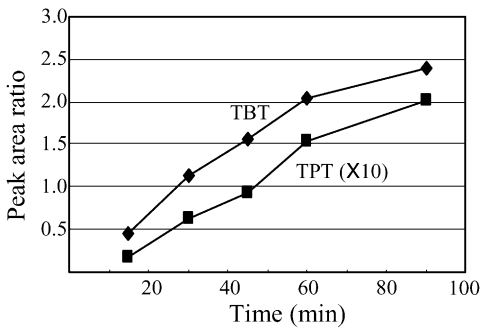

Fig. 8. Effect of extraction time on 4-fluorophenyl derivatization.

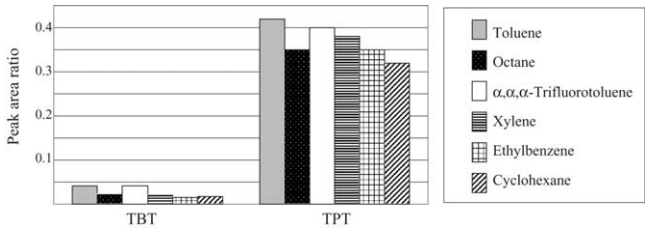

Fig. 9. Effect of solvent on ethyl derivatization.

was injected, the largest peak area was obtained. After the LPME for 30 min, 3 μl of α,α,α-trifluorotoluene decreased to 2.7 μl.

3.1.6. Effect of extraction time

The extraction time was studied up to 90 min (Fig. 8). The peak area ratio increased even at 90 min. The LPME is a process depending on equilibrium between the aqueous sample and the organic solvent unlike an exhaustive extraction such as LLE and SPE. For a quantitative analysis, it is not necessary to achieve equilibrium. In addition, the surrogate standard, which calibrates the analytical procedure, increases the precision.

The reproducibilities were compared between 30 and 60 min. As a result, the better reproducibilities (RSD = 10–11%) were obtained at 60 min, compared with those at 30 min (RSD = 18–21%). In addition, the change of the slope in Fig. 8 became gentle at 60 min, and therefore 60 min was selected for the extraction time.

3.2. Optimization of LPME using ethyl derivatization

The initial LPME conditions using sodium tetraethylborate were as follows: extraction time, 30 min; temperature, 14 °C; stirring rate, 500 rpm; buffer, pH 3 (1 M), 50 μl; amount of sodium tetraethylborate, 0.5% aqueous solution, 50 μl; solvent drop size, 3 μl; injection volume, 2 μl. For the stirring rate, temperature, solvent drop size and injection volume, the same values obtained from the optimization for the 4-fluorophenyl derivatization were used.

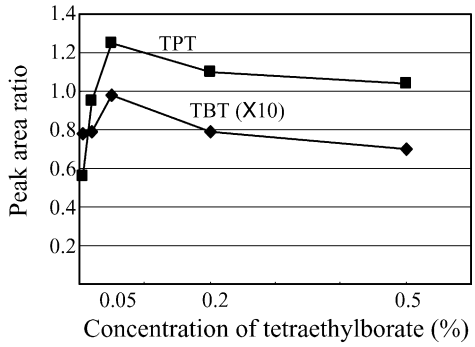

Fig. 11. Effect of tetraethylborate amount on ethyl derivatization.

3.2.1. Selection of organic solvent

Toluene, octane, α,α,α-trifluorotoluene, xylene, ethylbenzene and cyclohexane were tested. The results are shown in Fig. 9. Unlike the result from the 4-fluorophenylation, there are small differences among the six solvents. In this case, it is considered that ethylation may probably occur in the water phase since the ethylated organotins can be detected by headspace SPME [15]. α,α,α-Trifluorotoluene, which has the highest specific gravity among these solvents, was selected as the organic solvent.

3.2.2. Effect of buffer

The effects of the pH and concentration of the buffer were evaluated by adding 50 μl of acetate buffer (Fig. 10). Using more acidic buffer, higher peak area ratios were obtained. This tendency was the same as the 4-fluorophenylation and pH 3 was used for the subsequent experiment.

The concentration of the buffer had little effect on the peak area ratio, which was different from the 4-fluorophenylation result. The concentration of 0.5 M was chosen because of the good reproducibility at this concentration (data not shown).

3.2.3. Amount of derivatizing reagent

The amount of sodium tetraethylborate was investigated by adding 50 μl of an aqueous solution containing varying amounts of sodium tetraethylborate. As can be seen from Fig. 11, the peak area ratios of ethyl TBT and TPT decreased

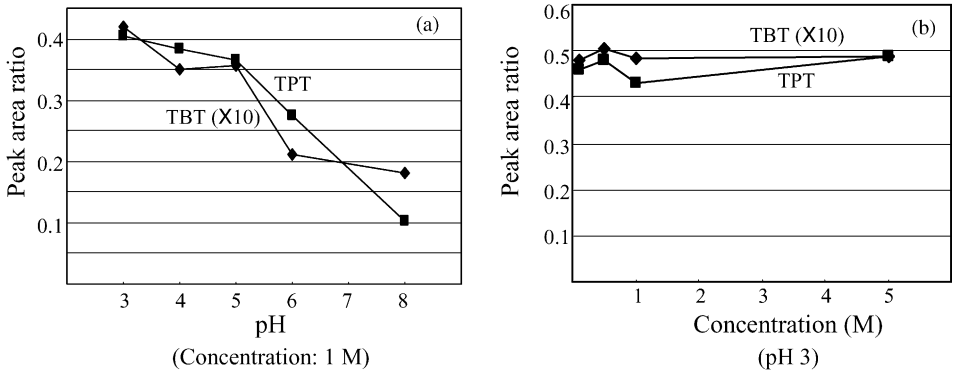

Fig. 10. Effects of pH (a) and concentration (b) of buffer on ethyl derivatization.

Table 2
Analytical performance

	Derivatization	Calibration range (ng/l)	Correlation coefficient (R)	RSD[a] (%)	Extraction efficiency[b] (%)	Enrichment factor[b] (fold)	MDL[c] (ng/l)
TBT	4-Fluorophenylation	1–5000	0.9996	11	3.5	140	0.36
TPT	4-Fluorophenylation	5–5000	0.9994	10	2.3	92	2.9
TBT	Ethylation	10–5000	0.9973	17	1.2	48	6.3
TPT	Ethylation	5–5000	0.9996	7.5	2.8	112	0.85

[a] Relative standard deviation at 10 ng/l for 4-fluorophenyl derivatives and 25 ng/l for ethyl derivatives (n = 5).
[b] Calculated at 500 ng/l.
[c] Calculated as standard deviation × t, where t = 1.860 from one-side t-distribution at 95% confidence level (n = 8, at blank for 4-fluorophenyl TBT, 10 ng/l for 4-fluorophenyl TPT, 25 ng/l for ethyl TBT and 2.5 ng/l for ethyl TPT).

Table 3
Comparison of the analytical performance for the analysis of TBT and TPT in water samples

Instrument	Extraction technique	Derivatizing reagent	MDL (ng/l) of TBT/TPT	Sample volume (ml)	Reference
GC–FPD	LLE	MeMgCl	0.5/2.0	1000	[6]
GC–MS	LLE	PeMgBr	1/1	200	[2]
GC–NICI–MS	LLE	–	0.10/0.13	200	[3]
GC–MS–MS	SPME	$NaBEt_4$	9/–	10	[4]
GC–ICP–MS	SPME	$NaBEt_4$	0.2/–	25	[10]
GC–MS–MS	LLE	$NaB(4\text{-F-Ph})_4$	0.35/2.2	50	[12]
GC–MS–MS	LPME	$NaB(4\text{-F-Ph})_4$	0.36/2.9	4	This work

FPD: flame photometric detection; NICI: negative ion chemical ionization; ICP: inductively coupled plasma.

Table 4
Recoveries of TBT and TPT from seawater and their concentrations

Compound	Absolute recovery[a] (%)	Relative recovery[b] (%)	Concentration (ng/l)			
			Kobe port	Nishinomiya port	Osaka south port	Osaka north port
TBT	63 (16)[c]	97 (9.7)[c]	1.1 (11)[c]	2.0 (15)[c]	1.7 (9.2)[c]	1.2 (7.1)[c]
TPT	73 (20)[c]	101 (10)[c]	nd	nd	nd	nd

[a] Seawater sample taken from Kobe port was spiked at 100 ng/l. Mean value (n = 5) calculated from peak area ratio to internal standard.
[b] Mean value (n = 5) calculated from peak area ratio to corresponding surrogate.
[c] Relative standard deviation (%) is in parentheses (n = 5).

by adding an excess amount of the tetraethylborate (over 0.05%). The same tendency has been reported in the purge trap GC-AAS analysis of organotins after the ethyl derivatization [23]. For the concentration of sodium tetraethylborate, 0.05% was chosen.

3.2.4. Effect of extraction time

The extraction time was evaluated up to 90 min. The extraction efficiencies increased even at 90 min. For a comparison with the 4-fluorophenyl derivatization, 60 min was selected.

3.3. Analytical performance

The linearity, reproducibility, enrichment factor, extraction efficiency and sensitivity results obtained with both the 4-fluorophenyl and ethyl derivatizations under the respectively optimized procedures are listed in Table 2. Good linear relationships (R = 0.9973–0.9996) were obtained in both cases. The relative standard deviations (RSDs) were obtained in the range from 7.5 to 17% for five replicates. The extraction efficiencies were only 1.2–3.5% for all compounds but more than 48-fold enrichments were achieved. The method detection limits (MDLs) of TBT and TPT were 0.36 and 2.9 ng/l with the 4-fluorophenylation and 6.3 and 0.85 ng/l with the ethylation, respectively. As for the 4-fluorophenylation, TBT showed a higher sensitivity than TPT, while for the ethylation, TBT had a lower sensitivity than TPT. This is ascribed to the bond-dissociation energy of Sn-aryl being stronger than that of Sn-alkyl [12].

Comparing the MDLs of TBT between the 4-fluorophenylation and ethylation, 4-fluorophenyl TBT had a lower MDL. In addition, the 4-fluorophenylation of TBT showed a better reproducibility than the ethylation of TBT due to much noise around the peak of ethyl TBT in the chromatogram. The worldwide use of TPT has been less than that of TBT, and TPT is hardly detected in seawater at present. The LPME coupled with the 4-fluorophenyl derivatization, which showed the higher sensitivity for TBT, was applied to the analysis of seawater.

In Table 3, the analytical performances reported by other methods were compared with the present work. Using only a low mililitre of water sample, the low MDL of TBT was obtained by the present work.

3.4. Application to seawater

To assess the feasibility of the optimized LPME procedure, the recovery test from seawater was carried out. Table 4 summarizes the recovery and reproducibility. The absolute recoveries calculated from the peak area ratio to the internal standard were in the range from 63 to 73%. When the addition of NaCl was examined using pure water, the extraction efficiencies also decreased to the similar level (data not shown). The relative recoveries calculated from the peak area ratio to the surrogate standard were obtained around 100% and the reproducibilities were found to be RSD 9.7–10%. The surrogate standards could successfully calibrate the LPME process.

Four seawater samples collected from the four ports in Osaka Bay were analyzed using the present method (Table 4). In all four samples, TBT was detected in the concentration range from 1.1 to 2.0 ng/l, and the RSD values were in the range from 7.1 to 15%. TPT was not detected in any of the samples. These concentrations are consistent with the previously reported values [12,24,25].

4. Conclusion

We have demonstrated the LPME of TBT and TPT in an aqueous sample combined with GC–MS–MS. Both 4-fluorophenylation and ethylation were used for the derivatization and the respective MDLs and RSDs from the optimized procedures were compared. 4-Fluorophenyl TBT showed one order of magnitude higher intensity than that of ethyl TBT. In addition, the LPME of 4-fluorophenyl TBT showed a similar MDL to the method using LLE or SPME. The GC–MS–MS analysis was suitable for the LPME because the cleanup step cannot be performed in the LPME. The LPME combined with 4-fluorophenylation has many advantages, e.g., simplicity, inexpensiveness and the use of a small amount of solvent. This technique was successfully applied to the seawater samples.

Acknowledgement

Thanks are due to the Instrumental Analysis Center, Faculty of Engineering, Osaka University, for assistance in obtaining NMR spectra on a JEOL JNM-GSX-400. One of the authors (H.S.) expresses his thanks for employment as a fellow in the 21st century COE program “Creation of Integrated Ecochemistry” of the Japan Society for Promotion of Science.

References

[1] P.E. Gibbs, G.W. Bryan, P.L. Pascoe, G.R. Burt, J. Mar. Biol. Ass. UK 67 (1987) 507.
[2] J.A. Stäb, W.P. Cofino, B. van Hattum, U.A.T. Brinkman, Fresenius J. Anal. Chem. 347 (1993) 247.
[3] K. Mizuishi, M. Takeuchi, T. Hobo, J. Chromatogr. A 800 (1998) 267.
[4] L. Dunemann, H. Hajimiragha, J. Begerow, Fresenius J. Anal. Chem. 363 (1999) 466.
[5] S. Tsunoi, T. Matoba, H. Shioji, L.T.H. Giang, H. Harino, M. Tanaka, J. Chromatogr. A 962 (2002) 197.
[6] I. Tolosa, J.M. Bayona, J. Albaigés, L.F. Alencastro, J. Tarradellas, Fresenius J. Anal. Chem. 339 (1991) 646.
[7] S. Díez, L. Ortiz, J.M. Bayona, Chromatographia 52 (2000) 657.
[8] L. Moens, T.D. Smaele, R. Dams, Anal. Chem. 69 (1997) 1604.
[9] H. Tao, R.B. Rajendran, C.R. Quetel, T. Nakazato, M. Tominaga, A. Miyazaki, Anal. Chem. 71 (1999) 4208.
[10] C. Bancon-Montigny, P. Maxwell, L. Yang, Z. Mester, R.E. Sturgeon, Anal. Chem. 74 (2002) 5606.
[11] T.D. Smaele, L. Moens, R. Dams, P. Sandra, J.V. der Eycken, J. Vandyck, J. Chromatogr. A 793 (1998) 99.
[12] S. Tsunoi, H. Shioji, M. Tanaka, Anal. Sci. 20 (2004) 101.
[13] R.P. Belardi, J. Pawliszyn, Water Pollut. Res. J. Can. 24 (1989) 179.
[14] E. Millán, J. Pawliszyn, J. Chromatogr. A 873 (2000) 63.
[15] M.A. Jeannot, F.F. Cantwell, Anal. Chem. 68 (1996) 2236.
[16] Y. He, H.K. Lee, Anal. Chem. 69 (1997) 4634.
[17] S. Pedersen-Bjergaard, K.E. Rasmussen, Anal. Chem. 71 (1999) 2650.
[18] L. Zhu, C.B. Tay, H.K. Lee, J. Chromatogr. A 963 (2002) 231.
[19] G. Shen, H.K. Lee, Anal. Chem. 74 (2002) 648.
[20] A.L. Theis, A.J. Waldack, S.M. Hansen, M.A. Jeannot, Anal. Chem. 73 (2001) 5651.
[21] A. Ogawa, D.P. Curran, J. Org. Chem. 62 (1997) 450.
[22] L. Zhao, H.K. Lee, J. Chromatogr. A 919 (2001) 381.
[23] Y. Cai, S. Rapsomanikis, M.O. Andreae, J. Anal. Atom. Spectrom. 8 (1993) 119.
[24] I. Takeuchi, S. Takahashi, S. Tanabe, N. Miyazaki, Mar. Environ. Res. 57 (2004) 397.
[25] H. Harino, M. Fukushima, S. Kawai, Environ. Pollut. 105 (1999) 1.