APPLIED ORGANOMETALLIC CHEMISTRY, VOL. 7, 149–157 (1993)

Matrix effects in the determination of butyltin compounds in environmental samples by GC AA after hydride generation

Ph Quevauviller,* F Martin, C Belin and O F X Donard
Université de Bordeaux I, Laboratoire de Photophysique et Photochimie Moléculaire, 351 Cours de la Libération, 33405 Talence, France

Experiments for the determination of mono-, di and tri-butyltin (MBT, DBT and TBT) by hydride generation/gas chromatography/atomic absorption spectrometry in various matrices (sediment, suspended matter, mussel, algae and water) have revealed that poor butyltin recoveries are obtained in sediments displaying high sulphur and hydrocarbon contents; very poor recoveries were also observed for TBT in sediments with high chlorophyll pigment contents as well as in algal samples. It was however not clear whether the hydride generation was inhibited by these infering compounds, as was previously assumed in the case of hydrocarbons, or whether interferences affected the atomization rate. Further studies were performed to solve this problem in order to validate this method in the case of analyses of, for example, oil-contaminated sediment and algae. This paper presents the results obtained. It is concluded here that the poor recoveries were due to an inhibition of hydride generation rather than to interference at the atomization stage.

Keywords: Butyltins, environmental matrices, hydride generation, matrix effects

INTRODUCTION

Concentrations of butyltin compounds in the marine environment are now routinely monitored in different matrices (water, sediment, biological tissues) in many laboratories. The well-known toxic impact of tributyltin (TBT) released from antifouling paints has led many countries to implement regulations and to carry out monitoring campaigns to control the levels of contamination.[1] The determination of TBT and its degradation products di- and mono-butyltin (DBT and MBT) is performed with a variety of analytical techniques involving different types of extraction (solvent or acid), derivatization (hydride generation, ethylation), separation (gas or liquid chromatography) and detection (atomic absorption spectrometry, inductively coupled plasma atomic emission spectrometry, flame photometric detection, mass spectrometry, etc.). These techniques have been found to be in good agreement for the analysis of organotin solutions and a TBT-spiked sediment.[2]

Some techniques, however, display sources of discrepancies which have still not been removed; this is the case for the linked technique of gas chromatography/atomic absorption spectrometry after hydride generation (HG/GC AA) which has been applied to a number of environmental studies since 1986[3] and which has been recently improved.[4] Difficulties have been experienced in the past with this technique; it has been shown that diesel fuel in seawater may inhibit the measurement of volatile tin hydrides by HG/GC AA in seawater solutions containing 1 $\mu g\,dm^{-3}$ of TBT.[5] In most cases, standard addition procedures allow us to take into account the different matrix effects and to measure the butyltin content without major difficulties; however, on some occasions the poor recovery observed due to the presence of interfering compounds may induce sources of error in the determination.

This paper presents experiments carried out with different types of matrices (suspended matter, sediments, mussel tissue, algae and water containing diesel fuel) in order to investigate the recovery of TBT, DBT and MBT and relate it to the presence of some possible interfering compounds (trace metals, organic carbon and sulphur); its aim is to demonstrate that this technique has to be applied with extreme care in

* Present address: Commission of the European Communities, Community Bureau of Reference (BCR), 200 rue de la Loi, 1049 Brussels, Belgium.

0268–2605/93/020149–09 $09.50

Received 17 August 1992
Accepted 19 October 1992

certain cases but may be used with success for a wide variety of analysis.

MATERIALS AND METHODS

Design of experiments

The aim of this study was to assess the recovery of butyltin compounds after hydride generation, GC separation and AA detection. Our purpose was to investigate the possible losses of butyltins occurring during the determination step and to relate this to interfering compounds existing in the analysed matrices; this is separate from the assessment of extraction recovery which represents another field of investigation.

The study was performed in two steps: in preliminary experiments, trace metal and sulphur contents were determined on dry sediments as well as the content of particulate organic carbon (POC) and butyltins. In a second stage, the matrix in which important inhibitory effects were observed for TBT (sample S6) was extensively studied. In both cases, the recovery of butyltins upon spiking was assessed.

Sample collection

A total of 13 samples was chosen for the experiments including suspended-matter and two sediment samples collected in the Rhine Estuary, The Netherlands, (SM1 and SM2, S1 and S2, respectively, in Tables 1–5); one sediment sample (S3) and one suspended matter (SM3) collected in the Scheldt Estuary, The Netherlands, in 1988; two sediment samples (S4 and S5) collected in the Sado Estuary, Portugal, in 1988 and an additional sample (S6) collected in 1989; two algae samples (A1 and A2) and one water sample (W) from the Arcachon Basin and one mussel sample (M) from the Bay of Lazaret, France. More details on the sample locations are given in the literature.[8] Intertidal samples (from the Sado Estuary) were collected from the shore at low tide whereas estuarine sediments were sampled with a Reyneck box core. The 2 cm top layer of the collected sediments were scraped with a Teflon spatula.

Suspended-matter samples were collected by centrifugation of *ca* 500 dm^3 of water with a Teflon-lined apparatus. The algae samples were collected by hand on intertidal flats of the Archachon Bay. All the samples were frozen prior to analysis and thereafter freeze-dried and ground. The water sample was collected at 20 cm below the surface, acidified with hydrochloric acid (0.1 mol dm^{-3}) and stored at 4 °C.

Analytical methods

Sample extraction

Only acidification is required for the analysis of filtered water (e.g. 0.5–1 cm^3 acetic acid in a 50 cm^3 sample). Sediment and biological samples are extracted using analytical-grade acetic acid (0.2–0.5 g in 20 cm^3) following two steps:[6] (a) agitation by stirring overnight and (b) ultrasonic extraction for 30 min (both with acetic acid). The solutions obtained are centrifuged at 4000 rpm for 5 min and the supernatant is collected in cleaned glass flasks. The extracts (2 cm^3) are directly injected in a 50 cm^3 water volume for analysis.

Determination

The butyltin determinations were performed by GC/AA after hydride generation. This technique has already been described in detail elsewhere:[4,6] butyltins react with a sodium borohydride solution (in the first stage of this study, 10 cm^3 of 40 g dm^{-3} $NaBH_4$ solution in a 50 cm^3 acidified sample volume; in a second stage, 30 cm^{-3} of 50 g dm^{-3} $NaBH_4$ solution) to yield the corresponding hydrides. Alkyltin hydrides are carried by a helium flow and cryogenically trapped in a U-column filled with chromatographic material (Chromosorb GNAW 60/80 mesh, coated with 3% SP 2100). After a 3 min purge time, the column is heated to 200 °C and the butyltin species are sequentially released. The compounds are atomized in a quartz furnace heated at 1000 °C and detected by AA at 224.6 nm (Perkin Elmer 5000) using an EDL source. Oxygen (O_2) and hydrogen (H_2) are introduced in the quartz cell (with respective flows of 20 and 200 cm^3 min^{-1}) to improve the rate of atomization.

The study was performed in two steps: in preliminary experiments, trace metal and sulphur contents were determined on dry sediments using X-ray fluorescence with a Phillips PW 1400/1510 apparatus. The content of particulate organic carbon (POC) was determined with the Strickland and Parsons method modified by Etcheber.[7] In a second stage, the matrix in which the most important inhibitory effects were observed (sample S6) was extensively studied by adding increas-

Table 1 Detection limits, repeatibility (r) and long-term reproducibility (R) of butyltins in water and sediment.

	Detection limit	r (%)	R (%)[a]
Water (ng dm^{-3} as Sn)			
TBT	1.2	13	18
DBT	0.5	6	10
MBT	0.6	7	12
Sediment (ng g^{-1} as Sn)			
TBT	1.8	14	20
DBT	1.0	8	12
MBT	1.2	7	12

[a] The reproducibility was assessed in solutions containing *ca* 200 ng dm^{-3} (as Sn) pf each of the three butyltin species.

ing volumes of leachate to a solution containing 50 ng of TBT for the assessment of the recovery.

Analytical conditions

The repeatibility of the technique for the different butyltin compounds (five replicate analyses of standard solutions in water and four replicate analyses of sediment and biological samples) and the detection limits are listed in Table 1. The accuracy of the technique was assessed for TBT in sediment samples[2,6] and was found to be acceptable. Previous experiments have shown that the extracts have to be analysed within two days to avoid losses of analytes.[6] The variations due to the analytical method were controlled by performing an analysis of standard solution in water every four sets of sample analyses. The same standard solution stored frozen at −20 °C and then defrosted at ambient temperature was used for all the calibrations and was verified at the end of the experiment with a fresh standard solution.

Table 2 Trace metal (copper, nickel, lead and zinc), sulphur and particulate organic carbon (POC) contents[a] in the samples studied

Sample	Pb	Cu	Zn	Ni	S	POC
S1	46.9	33.6	81.7	17.4	13.8	41.3
S2	109.2	94.7	292.8	48.1	4.3	20.4
SM1	110.6	94.5	293.1	46.4	4.5	33.8
SM2	114.7	84.8	269.0	33.7	5.3	55.8
S3	42.1	44.6	88.5	37.5	5.3	36.6
SM3	68.4	55.2	188.7	20.7	5.4	53.0
S4	44.0	34.0	75.9	15.1	6.1	38.6
S5	45.8	34.2	77.0	18.5	17.5	75.7
S6	—	23.8	53.5	4.9	—[b]	—

[a] Contents are given in μg g^{-1} (dry mass).
[b] —, Not analysed.

Table 3 Butyltin contents (in ng g^{-1} as Sn) in the samples studied

Sample	Amount[a]	MBT	DBT	TBT
S1	0.5 g	15 ± 0.5	16 ± 3.4	76 ± 6.3
S2	0.5 g	42 ± 1.6	28 ± 2.9	nd[b]
SM1	0.2 g	59 ± 2.6	44 ± 7.3	nd
SM2	0.2 g	253 ± 3	158 ± 30	152 ± 5.2
S3	0.5 g	51 ± 3.4	80 ± 11	140 ± 13
SM3	0.5 g	52 ± 8.3	20 ± 3.3	235 ± 25
S4	0.5 g	8 ± 0.2	36 ± 4.2	298 ± 7
S5	0.5 g	17 ± 0.7	34 ± 4.0	2803 ± 36
A1	0.5 g	nd	nd	nd
A2	0.5 g	nd	nd	nd
M	0.5 g	15 ± 1.6	120 ± 15	526 ± 11
W	50 cm^3	13 ± 1	97 ± 4.5	47 ± 3.8

[a] Amount of sample taken for analysis.
[b] nd, not detected.

The sample signals (peak area) were bracketed by the two standard levels considered (*ca* 2 and 5 ng in 50 cm^3).

Assessment of butyltin recovery

The butyltin recovery was assessed by comparing the results obtained with analyses of known amounts of standard solutions in 50 cm^3 water [respectively 1.8 and 4.5 ng of MBT (as Sn), 1.6 and 4 ng of DBT (as Sn) and 1.8 and 4.6 ng of TBT (as Sn)] with those of different samples spiked for 30 min with similar amounts of standard solutions. The experiments thus represented three separate steps: (a) the analysis of standard solutions, (b) the analysis of the samples and (c) the analysis of the samples spiked with standard solutions. All the analyses were performed in duplicate. The butyltin recoveries were calculated with the peak surface areas obtained as follows:

$$S_1 - S_2 = A_S \quad \%\mathrm{Rec} = 100 - A_S/S_s$$

where S_1 is the peak area of the spiked sample, S_2 is the peak area of the sample, %Rec the recovery and S_s is the peak area of the corresponding amount of standard.

Further experiments on TBT were performed with the sample where the most important inhibitory effects were observed (sediment sample S6, Sado Estuary, Portugal): increasing amounts of extracts were added to 50 cm^3 water spiked with 50 ng TBT and the percentage of inhibition was

Table 4 Recoveries of TBT obtained in the different samples.
The Table gives the peak areas obtained for the samples, the peak areas obtained for the samples spiked with 1.8 and 4.6 ng of TBT respectively and the recoveries obtained.

Location	Sample	Sample +1.8 ng TBT	Recovery (%)	Sample +4.6 ng TBT	Recovery (%)
S1	80	150	42	291	47
S2	nd[a]	180	109	419	93
SM1	nd	212	128	500	106
SM2	142	305	99	588	99
S3	331	486	94	811	106
SM3	525	669	87	979	100
S4	485	605	72	778	65
S5	450	460	6	485	8
A1	nd	45	27	115	25
A2	nd	62	38	154	34
M	737	840	62	990	56
W	240	410	103	750	112

[a]nd, not detected.
Note 1.8 ng of TBT had a peak area of 165, and 4.6 ng of TBT had a peak area of 452. In the other tables samples have bigger peak areas because they concern MBT or DBT, not TBT.

calculated as follows:

$$\% \text{ inhibition} = 100 \times (1 - A/B)$$

where A is the peak area obtained from the determination of 50 ng of TBT in the presence of sample S6, and B is the peak area obtained from the determination of 50 ng of TBT in water.

In addition, the extracts of the sample S6 were back-extracted in cyclohexane and analysed by spectrofluorimetry to detect the presence of organic compounds such as methyldibenzothiophene, methylfluoranthene and chlorophyl pigments.

Finally polyaromatic hydrocarbon (PAH) determinations were performed on the sample S6 in triplicate; the extraction was a Soxhlet extraction procedure followed by a Florisil microcolumn purification and the determination was by

Table 5 Recoveries of DBT obtained in the different samples.
The Table gives the peak areas obtained for the samples, the peak areas obtained for the samples spiked with 2.0 and 4.0 ng of DBT respectively and the recoveries obtained.

Location	Sample	Sample +2.0 ng DBT	Recovery (%)	Sample +4.0 ng DBT	Recovery (%)
S1	297	1537	59	4120	71
S2	870	3120	108	6385	103
SM1	512	2410	91	5710	97
SM2	1575	3250	80	6520	92
S3	2206	4151	93	7300	95
SM3	532	2340	87	5580	94
S4	723	2162	69	4364	68
S5	549	1710	56	3494	55
A1	nd[a]	750	36	1750	33
A2	nd	820	39	1850	34
M	3548	5650	101	8910	100
W	5700	7950	107	10 660	93

[a] nd, not detected.
Note 2.0 g of DBT had a peak area of 2085; 4.0 g of DBT had a peak area of 5355.

Table 6 Recoveries of MBT obtained in the different samples
The Table gives the peak areas obtained for the samples, the peak areas obtained for the samples spiked with 1.8 and 4.5 ng of MBT respectively and the recoveries obtained.

Location	Sample	Sample +1.8 ng MBT	Recovery (%)	Sample +4.5 ng MBT	Recovery (%)
S1	304	1750	69	3820	70
S2	1047	2793	84	5750	94
SM1	590	2319	83	5310	94
SM2	1514	2578	51	4240	54
S3	1248	2896	79	5960	94
SM3	1029	2267	59	5320	85
S4	101	1035	45	2315	44
S5	223	1230	48	2540	46
A1	nd[a]	910	44	2163	43
A2	nd	642	31	1543	31
M	391	2345	94	5014	92
W	654	2520	89	5389	94

[a] nd, not detected.
Note 1.8 ng of MBT had a peak area of 2088; 4.5 ng of MBT had a peak area of 5025.

reversed-phase liquid chromatography followed by fluorescence detection. The internal standard used was Pyrene d-10.

Notes on Figures and Tables

Table 1 r and R are defined in the table captions and the text: r = repeatibility assessed by five replicate analyses of standard solutions in water and four replicate analyses of sediment and biota samples; R = long-term reproducibility assessed in solutions containing *ca* 200 ng dm^{-3} (as Sn) of each of the three butyltin species (analysed at regular intervals over a period of three months).

Table 4 1.8 ng = 165 means e.g. that 1.8 ng of TBT in water had a peak area of 165 (the other tables have much bigger peak areas because they concern MBT and DBT respectively).

Table 7 Correlations obtained between the butyltin recoveries (Rec. MBT, Rec. DBT and Rec. TBT) and the other elements determined

	Pb	Cu	Zn	Ni	S	POC
Rec. MBT	0.3	0.5	0.5	0.8	−0.5	−0.7
Rec. DBT	0.7	0.8	0.8	0.8	−0.9	−0.7
Rec. TBT	0.6	0.7	0.7	0.7	−0.9	−0.7

Figures A_S is the surface area (as defined in the text).

Only samples which showed a poor recovery are identified (labelled) in the figures; labelling is not found to be necessary for the samples showing a good recovery as it would have given excessive detail.

The Tables indicate the peak areas of the non-spiked samples (second column) and the peak areas obtained after spiking, e.g. for TBT with +1.8 ng (third column) and +4.6 ng (fifth column), and the recoveries of the different tin species calculated as explained in the text (recoveries expressed in % in the fourth and sixth columns). The Figures plot the *peak areas* obtained upon spiking of the different samples (third and fifth columns in the tables) and not the recoveries; recovery data are not compared with the peak areas.

RESULTS AND DISCUSSION

Preliminary experiments

The results for some trace-metal (lead, copper, zinc and nickel), sulphur and particulate organic carbon (POC) determinations in sediment and suspended-matter samples are listed in Table 2. The samples appear to be weakly to moderately contaminated in trace metals and the range of

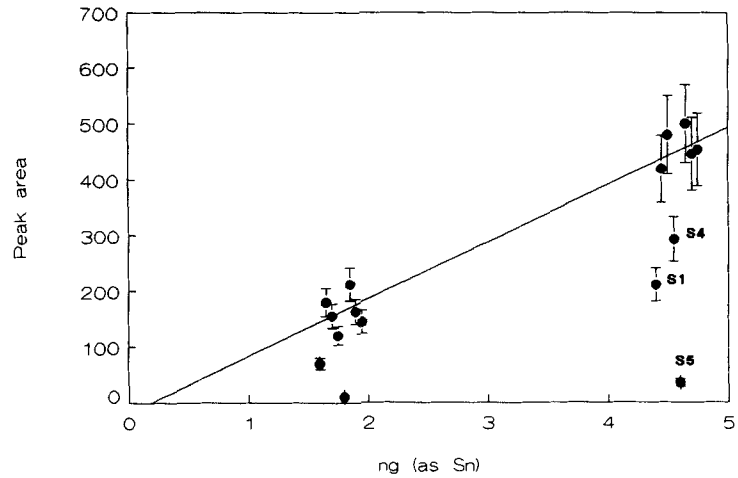

Figure 1 Recoveries of TBT in the samples S1, S2, SM1, SM2, S3, SM3, S4, S5 and M. The diagram plots the peak areas obtained upon spiking (surfaces area, A_S, defined in the text).

concentrations of POC is between 20.4 and 41.3 mg kg^{-1} except for three samples (SM2, SM3 and S5) which contain up to 50 mg kg^{-1} of POC. The sulphur contents range between 4.3 and 6.1 mg kg^{-1} except for the samples S1 and S5. The butyltin concentrations observed were representative of moderately to highly contaminated environments (Table 3).

The recoveries obtained for the three butyltin species in the different matrices studied are given in Tables 4–6 and plotted in Figs 1 to 6. A good correlation was observed between the recoveries obtained and the trace metal contents (Table 7); however, an inverse correlation was noted between the recoveries and the sulphur and POC contents. The Figures show that three sediment samples (S1, S4 and S5) displayed a poor recovery for the butyltin species, particularly for TBT. The recoveries appear to be better for MBT and DBT (Figs 2 and 3). In the case of algal samples, the three butyltin species display a very poor recovery whereas no losses are observed in water (Figs 4, 5 and 6).

The poor recoveries could be explained by three different factors: (i) an inhibition of the derivatization; (ii) an incomplete desorption of the species; and/or (iii) interferences in the atomization step. Sulphur may have an inhibitory effect on hydride generation, and/or this element interferes in the atomization step. This was supported by the inverse correlation observed for the butyltin recoveries and the sulphur and POC

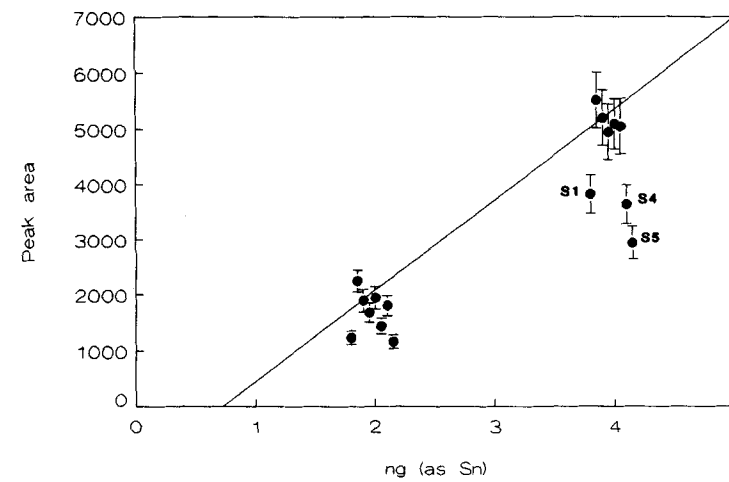

Figure 2 Recoveries of DBT in the samples S1, S2, SM1, SM2, S3, SM3, S4, S5 and M. The diagram plots the peak areas obtained upon spiking (surface areas, A_S, defined in the text).

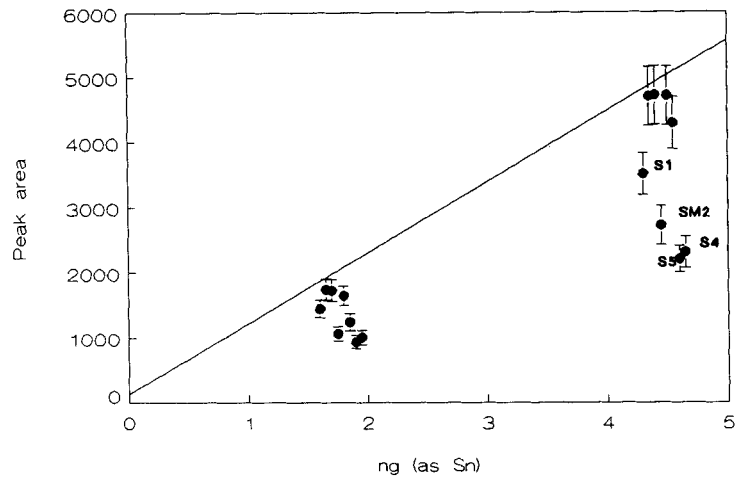

Figure 3 Recoveries of MBT in the samples S1, S2, SM1, SM2, S3, SM3, S4, S5 and M. The diagram plots the peak areas obtained upon spiking (surface areas, A_S, defined in the text).

contents (Table 7). The interferences could also originate from the presence of hydrocarbons which were presumably present in the three (harbour) samples (oily aspect). Indeed, inhibition of hydride generation in the presence of diesel fuel was suspected in the case of seawater analysis.[5] This was, however, not verified in the seawater sample from Arcachon (sample W) although it contained diesel oil. It is assumed that the actual effects of high organic matter content on inhibition of hydride generation as the $NaBH_4$ is to reduce the organic matter, and therefore the derivatization of tin species will not occur with a rate of 100%. This was confirmed by *Astruc et al.*,[9] who demonstrated that a better recovery could be obained by increasing the volume of $NaBH_4$ and therefore the yield of hydrides. The authors also recommended an increase in the concentration of the $NaBH_4$ solution.

Investigations on a complex sediment matrix

The second set of experiments was performed with the sample displaying the greatest analytical difficulties, i.e. the sample S6. The analyses were performed using more concentrated $NaBH_4$ solution (30 cm^3 of 5% solution). The results summarized in Fig. 7 clearly show that the percentage inhibition of the TBT signal increases with increasing extract addition, which could again be attributed either to an inhibition of hydride

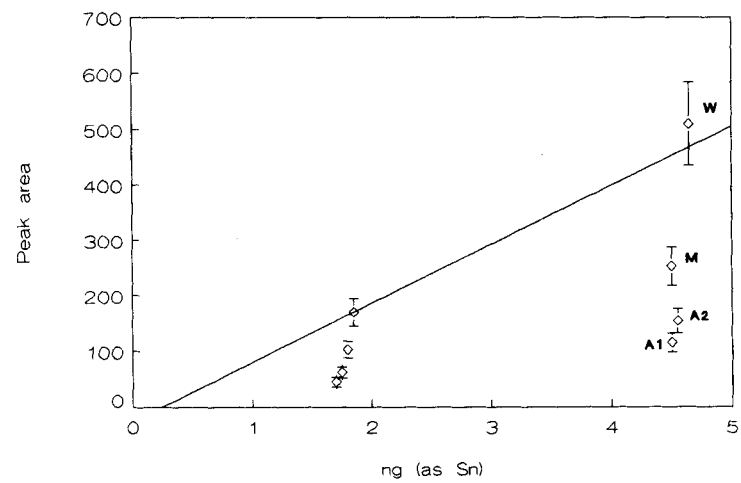

Figure 4 Recoveries of DBT in the samples A1 and A2 (algae), W (water) and M (mussel). The diagram plots the peak areas obtained upon spiking (surface areas, A_S, defined in the text).

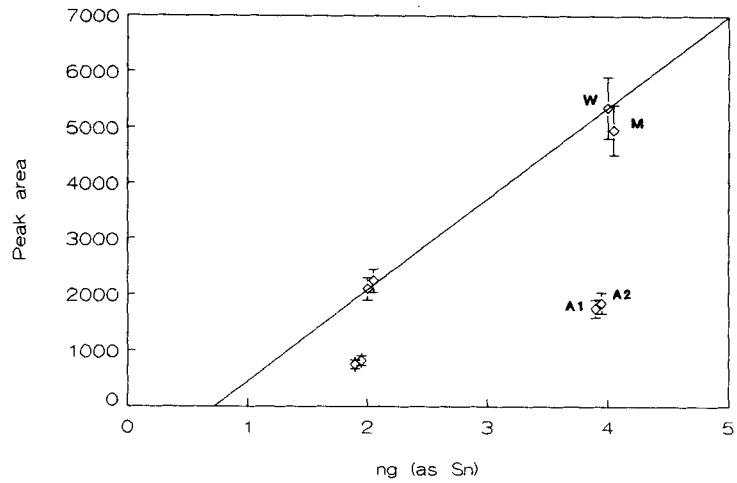

Figure 5 Recoveries of TBT in the samples A1 and A2 (algae), W (water) and M (mussel). The diagram plots the peak areas obtained upon spiking (surface areas, A_S, defined in the text).

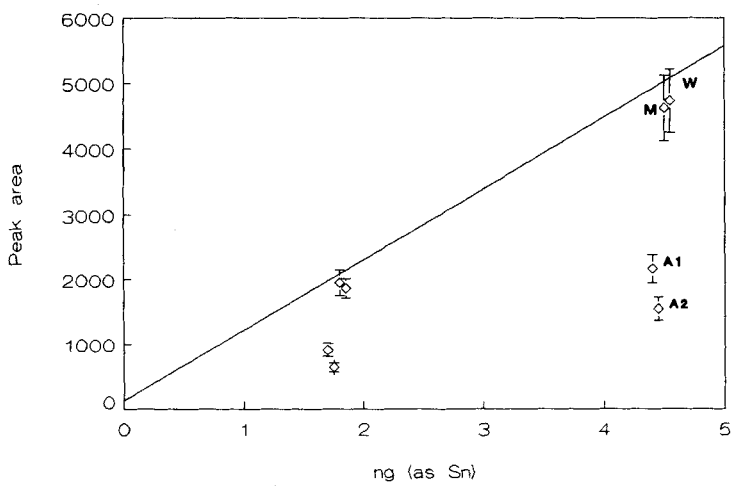

Figure 6 Recoveries of MBT in the samples A1 and A2 (algae), (water) and M (mussel). The diagram plots the peak areas obtained upon spiking (surface areas, A_S, defined in the text).

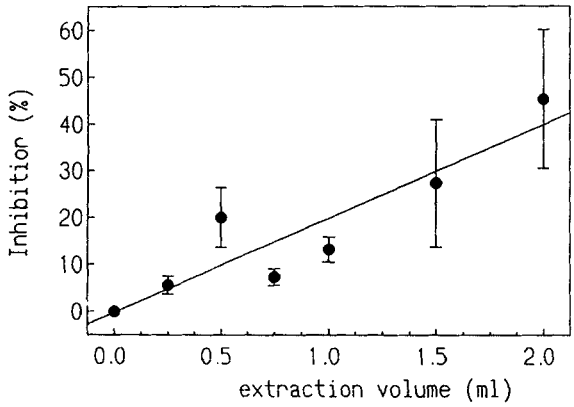

Figure 7 Inhibition of TBT signal in sample S6 in relation to the sample volume injected.

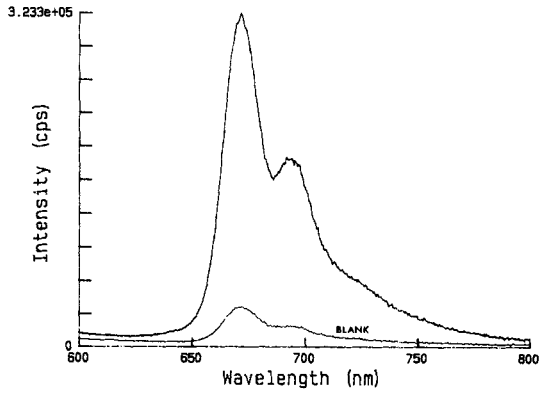

Figure 8 Emission spectra showing the presence of chlorophyl pigments in sample S6.

Table 8 Polyaromatic hydrocarbon (PAH) determinations in sample S6

PAH	Mean [ng g^{-1} (dry mass)]	Standard deviation
Pyrene	139	6
Benzo[*a*]anthracene	110	7
Benzo(*b*)naphtho[2,1-*d*]thiophene	266	14
Benzo[*e*]pyrene	149	9
Benzo[*b*]fluoranthene	52	5
Benzo[*h*]fluoranthene	43	3
Benzo[*a*]pyrene	73	1
Indeno[1,2,3-cd]pyrene	61	2

generation or to increasing interferences at the atomization step.

Spectrofluorimetric qualitative analysis has shown that organic compounds belonging to the groups of methyldibenzothiophene and methylfluoranthene were present at low concentrations whereas chlorophyl pigments could be observed at high concentration levels (Fig. 8). Chlorophyl pigments could have an inhibitory effect as poor recoveries were observed both in the case of the sample S6 which had high pigment content and in the case of algal analysis (samples A1 and A2). High polyaromatic hydrocarbon contents were also observed in this sample (Table 8). Considering the likelihood of interferences of sulphur and POC as were found in a similar sample (S5) and the ones suspected from both pigments and hydrocarbons, it is difficult to draw a firm conclusion regarding the compounds responsible from the inhibiting effects. It is likely that these effects are due to combined influences, which would explain why such low recoveries were obtained.

CONCLUSIONS

The recovery experiments performed in different types of matrices for the determination of butyltins by HG/GC AA clearly indicated that major analytical problems are observed in oil-sediment samples containing high amounts of sulphur and chlorophyl pigments. The authors believe that the inhibition is due to poor hydride generation rather than to atomization as it is assumed that sodium borohydride will first reduce the organic matter and the derivatisation of tin species may not occur at 100% yield in the case of organic-rich samples. Investigations are currently being conducted to confirm this hypothesis.

For the other matrices, the recovery obtained was generally very good. This analytical method may therefore be considered as an excellent technique for the determination of butyltins in a wide variety of matrices; special care is however to be taken when dealing with oily matrices in order to ensure that a good hydride yield is obtained and that no major interferences occur in the atomization step.

REFERENCES

1. Alzieu, C *Rapports Scient. Techn. IFREMER*, 1989, 17: 93
2. Quevauviller, Ph, Griepink, B, Maier, E A, Meinema, H and Muntau, H *Euroanalysis VII*, Vienna, abstract
3. Donard, O F X In: *Environmental Analysis using Chromatography Interfaced with Atomic Spectroscopy*, Harrison, R and Rapsomanikis, S (eds), Ellis Horwood, Chichester, 1988, chapter 7, p 189
4. Donard, O F X 5. *Colloquium Atomspektrometrische Spurenanalytik*, 1990, Welz, B (ed), Perkin-Elmer GmbH, 395
5. Valkirs, A O, Seligman, P F, Stang, P M, Homer, V, Lieberman, S H, Vafa, G and Dooley, C A *Mar. Pollut. Bull.*, 1986, 17: 319
6. Quevauviller, Ph and Donard, O F X *Fres. J. Anal. Chem.*, 1991, 339: 6
7. Etcheber, H *J. Rech. Oceanogr.*, 1981, 6: 37
8. Quevauviller, Ph and Donard, O F X *Appl. Organo.net. Chem.*, 1990, 4: 353
9. Astruc, A. Lavigne, R. Desauziers, V, Pinel, R and Astruc, M *Appl. Organomet. Chem.*, 1989, 3: 267