The Determination of Pesticidal and Non-Pesticidal Organotin Compounds in Water Matrices by *in situ* Ethylation and Gas Chromatography with Pulsed Flame Photometric Detection

Otis Evans, Peter Kauffman[1], Anne P. Vonderheide, Larry J. Wymer and Jeffrey N. Morgan

Chemical Exposure Research Branch
Microbiological and Chemical Exposure Assessment Research Division
National Exposure Research Laboratory

[1]*National Council on Aging*

U. S. Environmental Protection Agency
26 West Martin Luther King Drive
Cincinnati, Ohio 45268 USA

[*]Author to whom correspondence should be addressed:

Jeffrey N. Morgan

U.S. Environmental Protection Agency, MS-564

26 W. Martin Luther King Drive

Cincinnati, OH 45268

Morgan.jeffrey@epa.gov

513-569- 7738 (office)

513-569-7757 (FAX)

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Abstract

The concurrent determination of pesticidal and non-pesticidal organotin compounds in several water matrices, using a simultaneous *in situ* ethylation and liquid-liquid extraction followed by splitless injection mode capillary gas chromatography with pulsed flame photometric detection, is described. The speciation analysis of nine organotin compounds includes low molecular weight - low boiling (non-pesticidal) and high molecular weight - high boiling analytes (pesticidal) of significant environmental interest. The minimum time for sodium tetraethylborate alkylation, using mechanical agitation, is determined to be fifteen minutes in order to ensure the complete derivatization of the complete list of analytes. The utilization of a "hot needle" and a rapid injection rate is shown to be an efficacious means to eliminate "mass" or "needle" discrimination when determining the mixture of organotin compounds. Method detection limits are calculated to be in the low ng L^{-1} range. The final method is applied to various water samples; storm water from the Cincinnati area demonstrated low native levels of three of the organotin compounds.

Keywords: pesticidal organotins, non-pesticidal organotins, *in situ* ethylation, liquid-liquid extraction, GC-PFPD, comproportionation

Introduction

Organotin compounds may be sub-divided into two distinct classes of compounds - pesticidal and non-pesticidal. These two groups of compounds exhibit dissimilar chemical and physical properties. Non-pesticidal organotins are generally smaller alkyl entities that are both polar and non-labile. Pesticidal organotins, on the other hand, are typically larger alkyl and aryl compounds that are non-polar, hydrophobic and labile. Toxicity characteristics of the two classes also differ; the non-pesticidal organotinsare immuno- and neurotoxins whereas the pesticidal compounds have documented deleterious effects on marine animals.[1; 2; 3] In spite of these differences, most papers to date primarily investigate the pesticidal organotins[3; 4; 5], possibly with the inclusion of dibutyltin.[6] Conversely, many papers that study the non-pesticidal organotinsinclude the pesticidal tributyltin[7; 8]; nevertheless, very few consider the methylated non-pesticidal organotins.

The differences in properties contribute to the versatility of organotin compounds, and as a consequence, their utility and applicability in a diverse range of applications. For instance, pesticidal organotins have been used as insecticides, fungicides (e.g., fungicidal wood preservatives), herbicides, acaricides, disinfectants (e.g., textiles), anti-foulant coatings/biocides (e.g., applied to ship and boat hulls, docks, fishnets, and buoys to discourage the growth of marine organisms), and preservatives for many different types of materials. As a result, they have been widely used in certain industries, such as paper and pulp mills, cooling towers (electric power generation), breweries, textile mills, leather processing plants and other facilities.

In the plastics industry, health and safety requirements have mandated a removal of toxic heavy metals (e.g., lead [neurotoxin], cadmium [renal toxin]) and the non-pesticidal organotins have been substituted in their place. Polyvinyl chloride (PVC) has found major applications in today's society and the non-pesticidal organotins are employed as important heat stabilizer additives in PVC (e.g., pipes, tubing, food packaging) to protect it from heat, light, oxidation and mechanical stress. Other industrial uses of non-pesticidal organotins are as catalysts in producing polyurethane foams and silicone elastomers, esters used as plasticizers, lubricants, and heat transfer fluids, glass coatings, and anthelminthics for poultry.

In spite of the intended benefits, the non-specificity, extreme toxicity, persistence and bioaccumulation tendency of the pesticidal organotins, and the increasingly significantly diverse and pervasive applications of the non-pesticidal organotins, have raised concerns regarding occurrence. The toxicity assessment of organotins is dependent, in part, on whether they may enter the environment in concentrations that could potentially lead to human exposure with concomitant adverse health effects. Further, toxicity characteristics (both human and ecological) require an analytical methodology capable of providing quantitative analysis for both classes of organotins in aqueous samples at very low limits of detection in water (ng L^{-1}).[4; 5; 6; 7; 8]

Alkylation of organotin salts for subsequent gas chromatographic (GC) analysis can be accomplished by several reactions. The Grignard reaction[9; 10; 11] and alkylation with

a sodium alkylborate reagent[3; 4; 5; 6; 7; 8] have been often employed. One particular advantage of the latter is that the derivatizing reagent is stable in water, unlike Grignard reagents, and, hence the reaction can be conducted in aqueous matrices. In the instrumental realm, GC is the primary technique used to separate organotin analytes and several detection schemes have been coupled with this separation method in pursuit of sensitive and selective detection. Many are based on element-specific detection, such as a flame photometric detection[3; 5], pulsed flame photometric detection[4] and inductively coupled plasma-mass spectrometry[7; 8]; however, electron ionization mass spectrometry[6] has also been shown to be useful.

Our objective in this work was the development of a sensitive and selective method for the simultaneous analysis of both pesticidal and non-pesticidal organotin compounds. An *in situ* derivatization with tetraethylborate coupled with liquid-liquid extraction that had previously delivered low detection limits for the non-pesticidal organotinswas used as the basis for sample preparation.[3; 4; 5; 6; 7; 8] Key parameters were investigated for the best possible recovery of the analytes of interest. Analysis was accomplished with capillary gas chromatography and various injection techniques were investigated for optimal sensitivity. Pulsed flame photometric detection was employed as it has been shown to be more sensitive than the conventional flame photometric detector.[12] The newly developed method was applied in the analysis organotin compounds in drinking and surface waters.

Experimental

Reagents and chemicals

Methanol, acetone, hexane, sodium sulfate, acetic acid, sodium acetate, hydrochloric acid and nitric acid were purchased from Fisher Scientific (Fairlawn, NJ, USA).

Sodium tetraethylborate (STEB) (97%) was obtained from Aldrich Chemical Company (Milwaukee, WI, USA) in one gram bottles. A 1% solution was prepared by dissolving the entire contents of each bottle in cold deionized-distilled water and diluting to a final volume of 100 ml. The actual amount of chemical reagent was determined by differences in weight between full and empty bottles, generally 1± 0.05 g. Subsequently, the derivatization solution was quickly transferred, in 25 ml portions, to Teflon™ bottles, frozen and stored at -74 °C (Forma Scientific BioFreezer (Marietta, OH, USA)). The solutions were retrieved as needed, thawed under ambient conditions, and immediately used in the alkylation reactions. The buffer (pH 5.0, acetate) and the derivatizing agent (sodium tetraethylborate, STEB) were extracted, with hexane, to remove potentially interfering organic tin (Sn) species.[13]

All mono-, di-, and trisubstituted organotin compounds were obtained in the chloride form. Monomethyltin trichloride (MMT, 97%), dimethyltin dichloride (DMT, 97%), mono-n-butyltin trichloride (MBT, 95%, liq.), di-n-butyltin dichloride (DBT, 95+%), tri-n-butyltin chloride (TBT, 97+%, liq.), tri-n-propyltin chloride (NTPrT, 95%, liq.), diphenyltin dichloride (DPhT, 95%), and triphenyltin chloride (TPhT, 95+%) were obtained from Alfa Aesar (Ward Hill, MA, USA). Tricylohexyltin (TCyHT, 97%), tetrabutyltin (TEBT, 93%, liq.) and tetrapentyltin (TEPT, 97%, liq.) were purchased from

Aldrich Chemical Company. (Mono)phenyltin trichloride (MPhT, 98%, liq.) was obtained from Gelest, Inc., (Morrisville, PA, USA) and Aldrich Chemical Company (Milwaukee, WI, USA). The organotin concentrations reported in this paper are expressed as mass of Sn per volume unit. The organotin stock solutions were prepared in the following solvents: the methyltin, propyltin, and butyltin compounds were dissolved in methanol containing 2% hydrochloric acid (HCl). The phenyltin compounds and tricyclohexyltin were dissolved in acetone containing 2% HCl and the fully alkylated tetrabutyltin and tetrapentyltin compounds were dissolved in hexane. The entire list of analytes is given in Table 1.

Surrogate and Internal Standards.

The surrogate utilized in this work was tri-n-propyltin chloride (NTPrT). Tetrabutyltin (TEBT) and tetrapentyltin (TEPT) were used as internal standards. Given the range of sample types we analyzed, reported here and elsewhere, and the various measurement phenomena that may be encountered while using splitless injection, it is advisable to employ multiple internal standards.

Sample preparation

A 1 L water sample was placed in a 1 L separatory funnel. The separatory funnel contained a Teflon™ stopcock and the glass stopper was replaced with a polyethylene stopper. A volume of 10 mL of a buffer solution, 1.0 mL of sodium tetraethylborate (1% w/v) and 4 mL of hexane were added in succession. The volume of hexane, used for extraction, was intended to be kept to a minimum, but adequate for extracting the selected

aqueous sample volume. The surrogate, NTPrT, was added and the sample was shaken manually, vented and then agitated mechanically. Initial experiments were shaken for 20 minutes; however, 15 minutes was found to be optimal as discussed in the Results and Discussion section. A Glas-Col Model S60012 Bench Top Shaker (Terre Haute, IN, USA), equipped with variable speed motor (0 to 250 revolutions minute-1), a shaker head, a BT6000 mounting platform, and a Model 099A BT 1000S holder for 1 L separatory funnels, was used. The aqueous phase was discarded and the hexane layer was dried with anhydrous Na_2SO_4 contained in a 6 ml glass reaction tube with a Teflon™ frit. The height of the Na_2SO_4 bed in the 6 ml glass reaction tube (column) was ~ 18-20 mm. The hexane extracted was adjusted to a 5 mL final volume and the internal standards, TEBT and TEPT, were added.

Instrumentation

Gas chromatography was performed with a Varian CP-3800 gas chromatograph (Walnut Creek, CA, USA), equipped with a Varian CP-8400 AutoSampler and a Varian Pulsed Flame Photometric Detector (PFPD) System [with a BG-12 Sulfur filter and an R647 Photomultiplier Tube (6, 7)], a three mm i.d. quartz combustor and a Varian 1079 split/splitless temperature programmable injector. Chromatographic separation was performed on a DB-17 fused silica capillary column (30 m x 0.25 mm i.d.) coated with 50%-(phenyl)methylpolysiloxane (0.25 μm film thickness) (J&W Scientific, Folsom, CA, USA) with helium as the carrier gas at a flow rate of 2.0 mL min^{-1}. The injector and detector port temperatures were 220 °C and 350 °C, respectively. The column temperature was held constant at 40 °C for 3 min, subsequently the temperature was

increased at a rate of 8 °C min^{-1} to 280 °C and held at 280 °C for one min. Complete instrumental conditions are detailed in Table 2.

Results and Discussion

Reaction time studies (in situ derivatization)

The non-pesticidal and pesticidal organotins are ionic in nature and must be derivatized in order to make them amenable to GC analysis. It is important to know the minimum amount of time for complete conversion of the organotins to the fully alkylated compounds. Most researchers report reaction times ranging from 2-30 minutes[5], even up to 16 hours[14], depending, in part, on the sample matrix. Our determinations involved evaluating reaction times from 5 to 20 minutes, in five minute increments, using mechanical agitation. Figure 1 portrays the normalized responses for non-pesticidal and pesticidal organotins, respectively, from 5 to 20 minutes and actual normalized values are given in Table 3. A multiple comparison procedure, Benjamini-Hochberg, was used to interpret these results and the table in Figure 1 displays this data. Only changes from 5 or 10 minutes to 20 minutes are significant, and then only for the pesticidal compounds. Because there was no significant change between the times of 15 and 20 minutes, optimal reaction time was chosen to be 15 minutes.

Choice of surrogate and internal standards

Tri-n-propyltin chloride was chosen as the surrogate. It proved to be an efficacious agent for mimicking the response of organotins in a wide range of sample types, including high total organic content (TOC) water (> 10 mg/L) and stormwater (discussed later). We

chose to employ the use of multiple internal standards, TEBT and TEPT, for the determination of both the non-pesticidal and pesticidal organotins. The absolute analytical response for TEBT essentially paralleled that obtained for TEPT. However, experiments with injection technique demonstrated more similarity in behavior between TEPT and the pesticidal compounds and so it was employed with the pesticidal compounds while TEBT was used with the non-pesticidal analytes. Figure 2 demonstrates the elution order and retention times for the organotin compounds under the chosen chromatographic conditions. All analytes were completely resolved.

Injection technique

Splitless injection mode is an ideal technique for low level environmental analyses with applicability to a range of analytes; however, it is known to have inherent limitations associated with sample vaporization in hot injection ports. If one injection port temperature is used to vaporize all of the analytes in a sample that encompass a wide range of molecular weights and boiling points, the higher boiling and higher molecular weight tend to give reduced peak area and peak height responses. This results in non-linear signals and lower response slopes when compared to the lower boiling and lower molecular weight analytes. This phenomenon is known as "needle" or "mass" discrimination[15] and has often been attributed to losses inside the syringe needle that results in the unequal delivery of sample components of different volatilities. This is characterized by high boiling (less volatile) and high molecular weight components either exiting the syringe needle at lower concentrations and at a slower rate than the solvent and more volatile (low boiling and low molecular weight) components or the less volatile

components being left behind coating the inner surface of the needle. As a consequence of the incomplete transfer of needle content, chromatographers observe peak areas or peak heights for high boiling sample constituents which are too small, i.e., a loss of sensitivity, when compared to the lower boiling sample components.

This work investigated alternate injection techniques based on the boiling points of the ethylated organotins.[16] Because each of the compounds was included as the same concentration of Sn, an element-based detection should produce similar areas for all. However, this discrimination is apparent in Figure 3A, which displays the gas chromatogram for the organotins with a "cold" needle injection. The sample was drawn into the barrel, with an air gap, so that an empty needle was inserted into the injection port. The sample injection was performed immediately at a rate of 5 μL sec^{-1} with a subsequent 6.5 sec post injection delay, i.e., the syringe needle remained in the injection port for the specified time, prior to its removal. The results reveal that several of the analytes, specifically peaks 10, 12 and 13 (tetrapentyltin [TEPT], tricyclohexyltin [TCyHT] and triphenyltin [TPhT]), undergo significant "discrimination", apparently due to the "cold" needle injection, when compared to the more volatile organotins and the surrogate, tri-n-propyltin (NTPrT, peak 5). Peak 11 (diphenyltin [DPhT]), although yielding a higher response compared to the aforementioned pesticidal analytes, demonstrates a diminished response under these injection conditions relative to the other non-pesticidal organotins.

Figure 3B presents the gas chromatogram for the tetraalkylated organotins for a "hot" needle injection. The sample was drawn into the syringe barrel, followed by an air gap, and the empty needle was inserted into the injection port. In this instance, the needle was heated 5 seconds prior to injection, the sample subsequently injected at a rate of 5 μL sec^{-1} and the syringe needle immediately withdrawn from the injection port. The intent was to allow the needle to equilibrate in temperature with the injection port prior to sample injection. The response for DPhT (peak 11), in this case, is virtually comparable to the non-pesticidal, lower boiling and lower molecular weight organotins. Furthermore, there is an increase in the response of the internal standard TEPT (peak 10) and noticeably increased responses in the pesticidal organotins, TCyHT (peak 12) and TPhT (peak 13). However, the latter three continue to exhibit, albeit to a lesser degree, the characteristic behavioral response of compounds subject to needle "discrimination". Experiments conducted employing post-injection delays did not give significantly different chromatographic responses.

Figure 3C presents results obtained with a sample injection rate of 50 μL sec^{-1} and confirms that the combination of a "hot" needle and a rapid injection rate (rapid plunger depression) lessens the severity of high boiler "needle discrimination". The rapid plunger depression rate forces the liquid sample into the "hot" needle, with a concomitant and coinstantaneous increase in solvent vapor pressure. The higher pressure inside the syringe needle produces a rapid expulsion of the sample into the injection port. This apparently prevents the deposition and / or retention of the higher boiling organotins on the inner surface of the needle. As can be seen, the responses for TCyHT and TPhT,

peaks 12 and 13 respectively, are further enhanced and are nearly comparable to the responses for the non-pesticidal organotins. Additionally, the responses for the internal standards TEBT (peak 9) and TEPT (peak 10), in this instance, are nearly comparable (cf. to Figure 3A). Increasing injection rates up to 100 μL sec^{-1} did not result in further enhancement of the analytical signal response. Using a post-injection delay, irrespective of its duration, did not contribute to an increase in the analytical signals.

During the course of performing "cold" needle injections, it was observed that while one internal standard, TEPT, gave smaller responses that may be attributable to "needle discrimination", the other internal standard, TEBT, was relatively unaffected. TEBT has a calculated boiling point of 285 °C at 760 mmHg pressure[16], higher than the fully alkylated non-pesticidal organotins but significantly lower than DPhT, TCyHT and TPhT.[16] And, it has a molecular weight that lies between DPhT and TPhT. Under the "cold" needle experimental conditions, the responses of the internal standards are somewhat divergent and inconsistent, with the response for TEBT appearing to increase relative to TEPT. The explanation we proffer for this apparent behavior is that it further shows the "needle" discrimination effect on the higher boiling internal standard. However, under "hot" needle injection experimental conditions with rapid sample injection (cf. Figure 3C), the response of TEPT increased and was comparable to the signal for TEBT.

Comproportionation of monophenyltin trichloride

During the injection experiments, it was noted that diphenyltin (DPhT) dichloride, a high boiling and high molecular weight compound, did not exhibit the response of the other compounds undergoing "mass" discrimination. At the same time, monophenyltin (MPhT) trichloride appeared to give a somewhat smaller response than expected. Comproportionation, also known as symproportionation, is sometimes defined as the reverse of disproportionation. Disproportionation may be a redox reaction where a reactant acts both as an oxidant and a reducing agent, i.e., an element in an intermediate oxidation state is transferred to a higher and a lower oxidation state, depending on the reaction conditions. Lytle, et. al.[17], in a recent ecological study, reported on the reversible comproportionation reaction in which tributyltin (TBT) and monobutyltin (MBT) are produced from two molecules of dibutyltin. Nguyen Van found both MPhT and DPhT to be unstable when stored in methanol and to undergo a similar redistribution.[18]

In our work, we found MPhT standards contained a significant amount of DPhT while the DPhT solutions were found to be stable and free of MPhT and any other organotin compounds (Figure 4A). The manufacturer's literature stated the analytical standard was 98% MPhT at receipt. Figure 4B represents the analysis of MPhT calibration standards prepared via dilution ~ 6 months after receipt. Figure 4C shows the results of analysis of MPhT calibration standards, prepared from the aforementioned primary stock standard (Figure 4B), ~ 7 months later. In the latter figure, it is conclusively demonstrated that the major component, in standard solutions of MPhT at the time of measurement, is DPhT. The calculated concentrations of DPhT at the expected concentration levels of MPhT (in

the extracts: 0.2, 0.5, 1.0, 2.0, and 5.0 μg L^{-1} as Sn) are as follows: 0.17, 0.22, 0.33, 0.66, and 1.57 μg L^{-1} as Sn. We believe the results indicate that MPhT, in both the neat solution (reagent grade material) and primary standard solutions, irreversibly comproportionates to DPhT.

Figures 5B and 5C shows calibration curves for MPhT prepared from the reagent grade chemical obtained from another manufacturer. The material safety data sheet (MSDS) from the manufacturer stated the material is >95% MPhT and <5% other organotins. Figure 5A is the calibration curve for DPhT, and attests to the stability of the organotin solution, prepared from the primary stock standard after approximately 20.5 months. This somewhat longer time frame clearly shows that DPhT did not comproportionate to TPhT, nor disproportionate to MPhT. Figure 5B shows the immediate preparation (upon receipt), dilution and analysis of MPhT standard solutions from the second manufacturer. Figure 5C presents the analysis of freshly prepared standards, from the primary stock standard (Figure 5B), about 4 months later. The calculated concentrations of DPhT in the extracts are: 0.11, 0.16, 0.22, 0.34 and 0.80 μg L^{-1} as Sn. These results appear to corroborate that MPhT, in both the neat material and standards (Figures 5B and C), tends to irreversibly comproportionate to DPhT under homogeneous solution conditions. We wondered if MPhT could also simultaneously disproportionate to form Sn (IV). There was no definitive evidence to show that Sn (IV) was produced by the disproportionation of MPhT. The determinative evidence would have been a larger response for tetraethyltin (TEET) following the alkylation/derivatization reaction with STEB. We also analyzed standard solutions of triphenyltin (TPhT) trichloride to determine if the compound had a

tendency to disproportionate to DPhT. The analyses did not show the presence of DPhT in the concentration range of the calibration curve.

Method detection limits

The GC parameters relating to the injector volume, injection temperature, oven temperature, detector temperature, incl. photomultiplier voltage, gate delay, gate width, trigger level, oxygen pressure parameters, etc., where applicable, were optimized individually to maximize the signal-to-noise ratio. Figure 2 presents a typical chromatogram of the ethylated products of the organotin analytes obtained under final instrumental conditions.

Method detection limit (MDL) determinations were made using the U. S. EPA single concentration procedure[19]. This method produces a concentration result which is unlikely (p=0.05) to be obtained when the analyte is absent. Eight deionized water samples were fortified with 0.2 ng L^{-1} and the calculated values were used in the MDL determination. The concentration of the series of standards used in these determinations lie within the suspected range of the MDL (EPA single concentration procedure) and occur in the lower 20% range of the calibration curve. Table 4 lists the U.S. EPA MDL values.

Analysis of aqueous matrixes for organotins

High carbon content can be problematic when utilizing this detector[20] and Table 5 presents the results of analyses for a tapwater sample with low total organic carbon

(TOC) content (<2.5 mg L^{-1}) and tapwater with a high TOC content (>10 mg L^{-1}). The samples were fortified with the respective organotin compounds at 5 ng L^{-1}. The tapwater samples were analyzed without the prior addition of a dechlorinator. As reported by others[21], an emulsion was formed during the course of the extraction of the high TOC content sample and may, in part, account for the somewhat lower recoveries of the mono- and di-alkyltin analytes. There are a number of analytical measures that may be applied to address emulsion formation; however, none were employed other than vigorous shaking of the emulsion. Finally, a storm water sample collected from the Cincinnati metropolitan area was analyzed and demonstrated native concentrations for three of the non-pesticidal organotins of interest: monomethyltin, 1.85 ng L^{-1}, dimethyltin, 13.2 ng L^{-1} and monobutyltin, 1.70 μg L^{-1}. Fortification of this same matrix with 10 ng L^{-1} of the organotin compounds produced recoveries ranging from 79.5 to 124%. The chromatographic results for this analysis are displayed in Figure 6.

Conclusion

The minimum time for the STEB ethylation reaction is 15 minutes when using mechanical agitation. The specified chromatographic, detection and liquid-liquid extraction conditions allow the complete separation and quantitative analysis of nine organotin analytes. Furthermore, adequate sensitivity, for analytical measurements at the low parts-per-trillion level, is obtained for all analytes while minimizing baseline noise and analysis time. The utilization of a “hot needle” and a rapid injection rate has been shown to be an efficacious and simple means to eliminate “mass” or “needle” discrimination when determining a mixture containing low boiling and low molecular

weight (non-pesticidal) and high boiling and high molecular weight (pesticidal) organotin compounds. It is important to check neat or reagent grade MPhT, and primary standard and other solution dilutions, for possible comproportionation to DPhT, in order to determine if the material is suitable for the preparation of accurate calibration standards for quantitative environmental analyses. The U.S.EPA MDLs demonstrated values in the low ng L^{-1} range. Adequate recoveries were obtained for analysis of low TOC and high TOC content water without additional sample treatment measures.

DISCLAIMER

The United States Environmental Protection Agency through its Office of Research and Development funded and managed the research described here. It has been subjected to the Agency's administrative review and approved for publication. The mention of commercial products does not constitute endorsement or recommendation for use by the United States Environmental Protection Agency.

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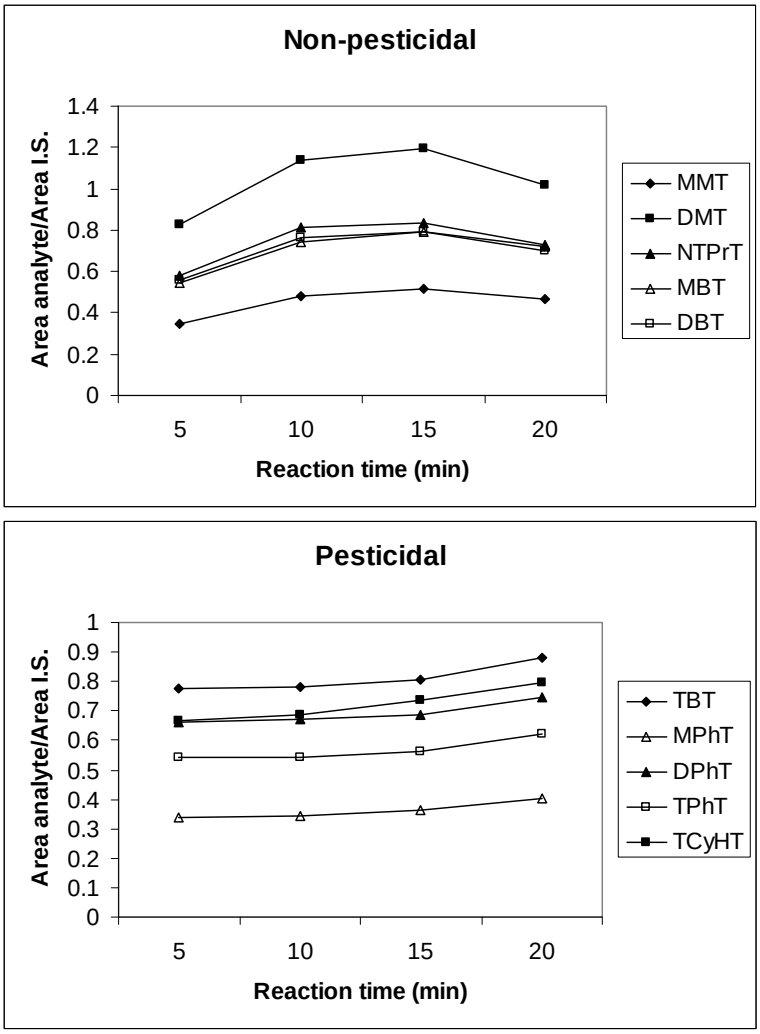
Figure 1 – Comparison of time of reaction (STEB) on the responses of the organotin analytes.



	Differences (Bold Indicates False Discovery Rate[a] < 0.05 via Benjamini-Hochberg Comparison)									
	MMT	**DMT**	**NTPrT**	**MBT**	**DBT**	**TBT**	**MPhT**	**DPhT**	**TPhT**	**TCyHT**
10 vs 5 m	0.13	0.32	0.23	0.20	0.20	0.01	0.01	0.01	0.00	0.02
15 vs 5 m	0.16	0.37	0.26	0.24	0.24	0.03	0.02	0.02	0.02	0.07
20 vs 5 m	0.11	0.20	0.15	0.17	0.14	**0.10**	**0.07**	**0.08**	**0.08**	**0.13**
15 vs 10 m	0.03	0.05	0.03	0.04	0.03	0.02	0.02	0.01	0.02	0.05
20 vs 10 m	-0.02	-0.12	-0.08	-0.03	-0.06	0.10	**0.06**	**0.07**	**0.08**	**0.11**
20 vs 15 m	-0.05	-0.18	-0.11	-0.07	-0.09	0.07	0.04	0.06	0.06	0.06

[a]False discovery rate is defined as expected proportion of false positive findings among all the rejected hypotheses (i.e., the proportion of significant findings that may be false).[22]

Figure 2 – GC-PFPD chromatogram of ethylated organotin analytes.

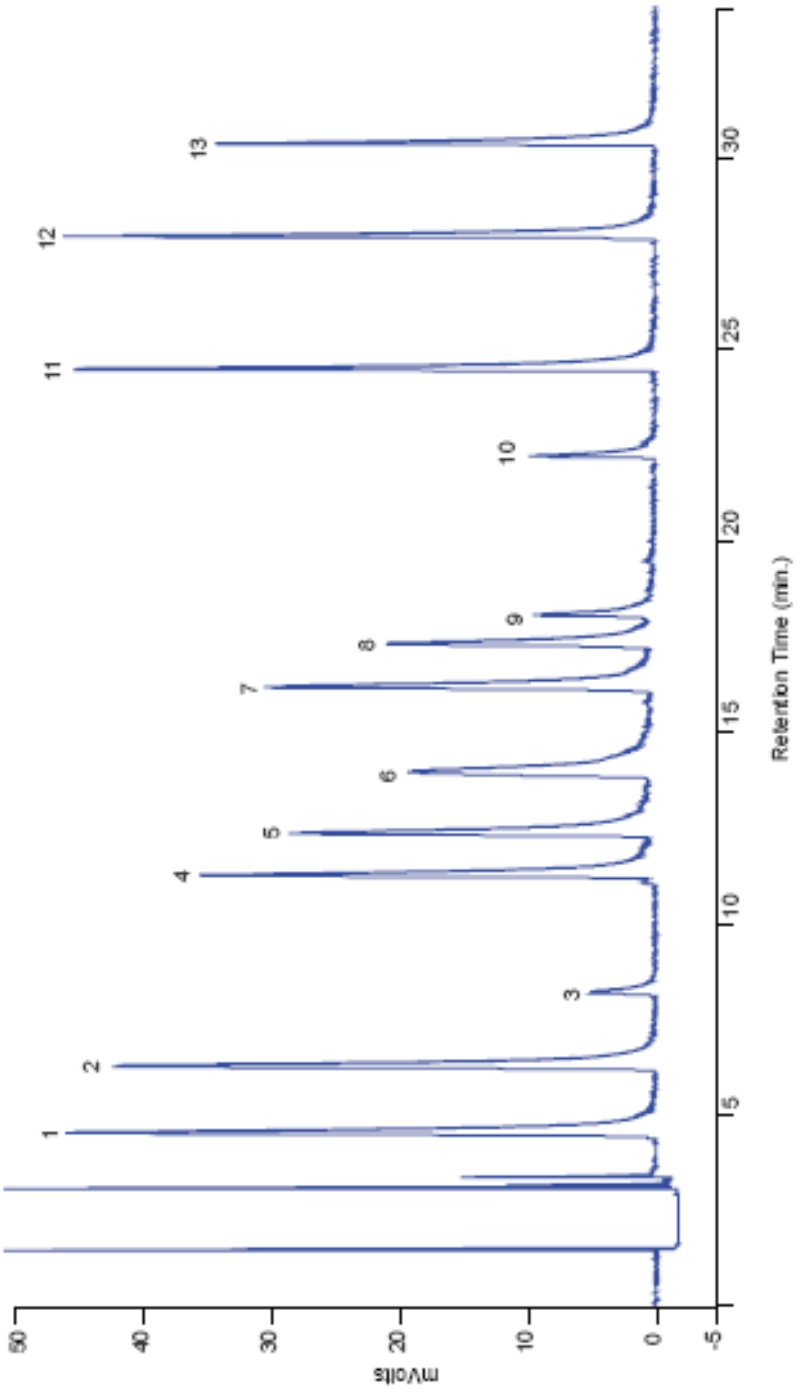


Figure 3 – Injection speed and injection delay ("cold needle" vs. "hot needle") **(A)** 5 µl sec^{-1}, 6.5 sec post injection delay - "cold needle", **(B)** 5 µl sec^{-1}, 5 sec pre-injection delay - "hot needle", and **(C)** 50 µl sec^{-1}, 5 sec pre-injection delay - "hot needle".

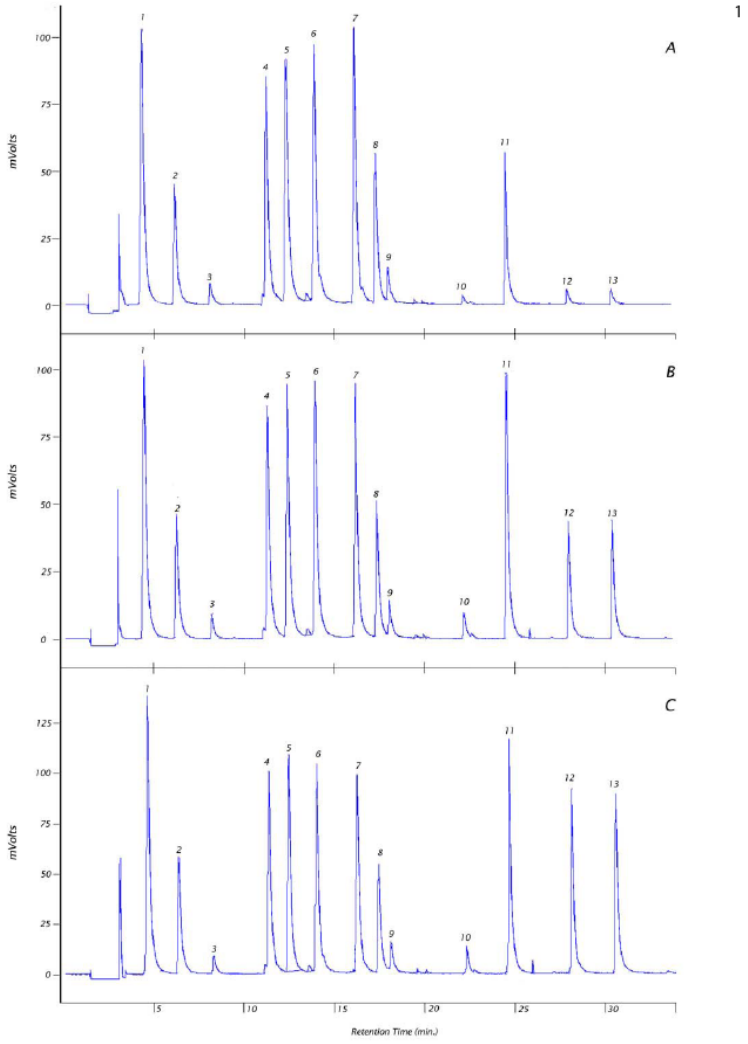


Figure 4 – A) Calibration curve for DPhT(primary stock standard prepared ~16.5 months earlier). **B)** Calibration curve for MPhT(~ 6 months after receipt) from manufacturer #1. **C)** Calibration curve for MPhT (~7 months after Figure 4B).

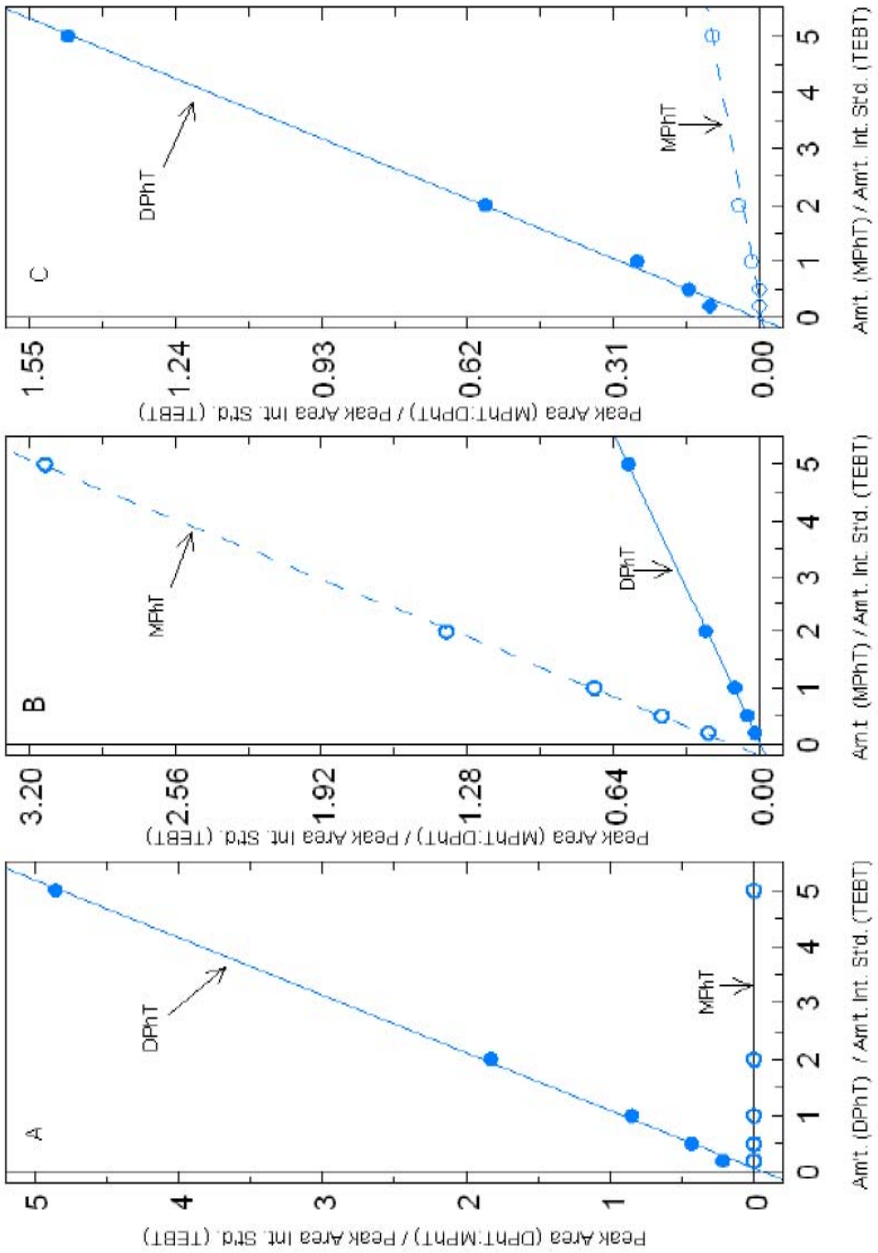


Figure 5 – A) Calibration curve for DPhT (primary stock standard prepared ~20.5 months earlier). **B)** Calibration curve for MPhT (immediate analysis upon receipt) from manufacturer #2. **C)** Calibration curve for MPhT (~4 months after Figure 5B).

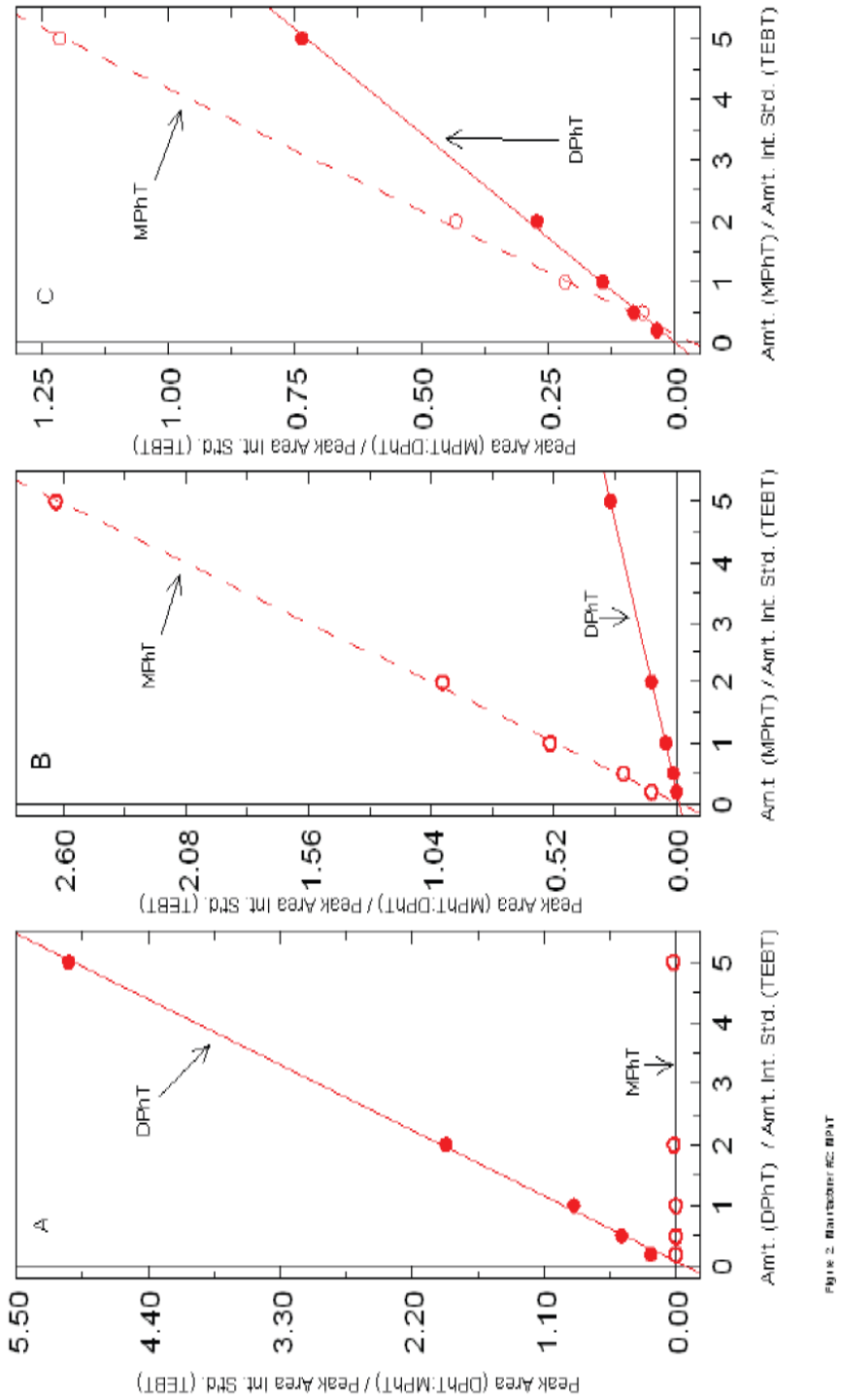


Figure 6 – Chromatogram for storm water. Dimethyltin, monomethyltin and monobutyltin were detected in the sample. The analytical data for the original sample and the fortified sample are presented in Table 5. Peak 14 is an unidentified sulfur compound.
A) Storm water sample. B) Fortified storm water sample.

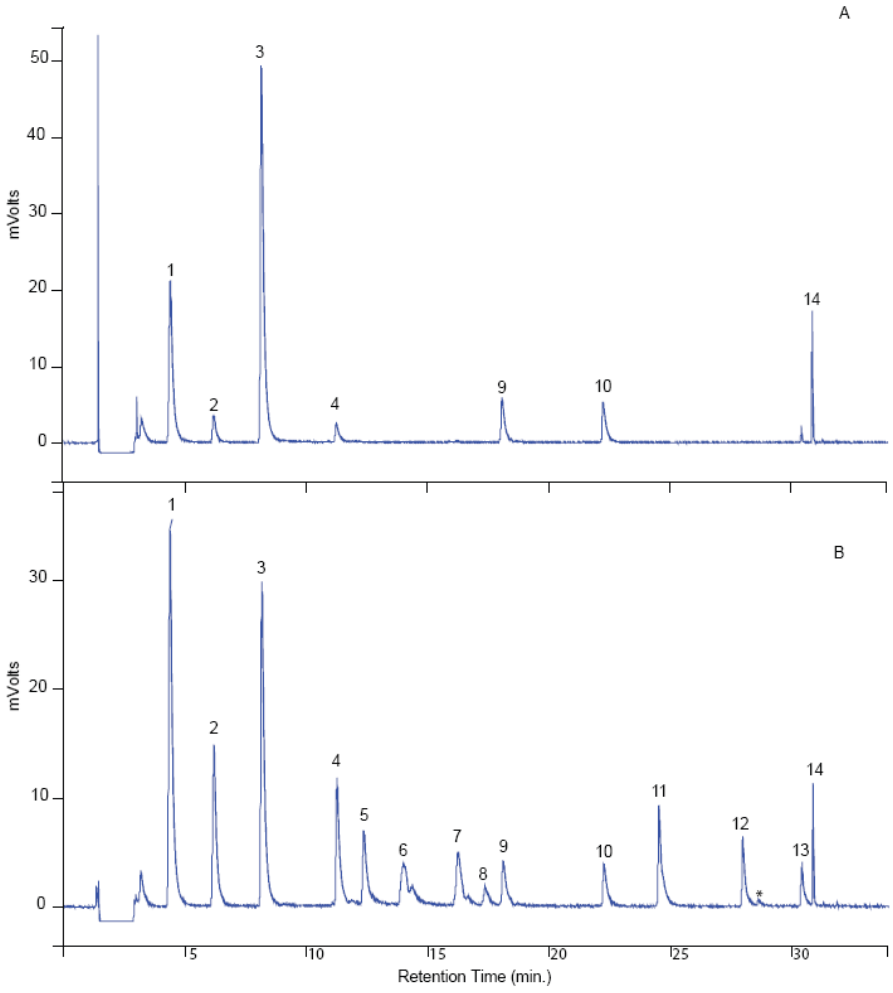


Table 1— List of Organotin Compounds

Peak #	Compound	Ethylated Compound	Formula Weight[1]	Retention time (min)	W ½ (sec)[2]
	Non-pesticidal				
2	Methyltin trichloride (MMT)	Triethyl methyltin	240.08 (220.9)	6.21 ± 0.12	10.91 ± 1.31
1	Dimethyltin dichloride (DMT)	Diethyl dimethyltin	219.67 (206.88)	4.32 ± 0.25	12.08 ± 2.48
4	Butyltin trichloride (MBT)	Triethyl butyltin	282.17 (262.99)	11.34 ± 0.01	9.51 ± 0.71
6	Dibutyltin dichloride (DBT)	Diethyl dibutyltin	303.83 (291.04)	14.07 ± 0.06	14.70 ± 2.50
	Pesticidal				
7	Tributyltin chloride (TBT)	Ethyl tributyltin	325.49 (319.1)	16.27 ± 0.02	11.57 ± 1.78
8	Phenyltin trichloride (MPhT)	Triethyl phenyltin	302.16 (282.98)	17.45 ± 0.01	9.53 ± 0.63
11	Diphenyltin dichloride (DPhT)	Diethyl diphenyltin	343.81 (331.02)	24.66 ± 0.01	8.66 ± 0.34
13	Triphenyltin chloride (TPhT)	Ethyltri phenyltin	385.46 (379.07)	30.55 ± 0.01	8.74 ± 1.03
12	Tricyclohexyltin chloride (TCyHT)	Ethyltricyclo hexyltin	403.61 (397.22)	28.10 ± 0.00	7.84 ± 0.26
	Other standards				
5	Tri-n-propyltin chloride[3] (NTPrT)	Ethyltri-n-propyltin	283.41 (277.02)	12.49 ± 0.04	14.27 ± 2.29
9	Tetrabutyltin[4] (TEBT)	Tetrabutyltin	347.15	18.16 ± 0.01	10.54 ± 1.00
10	Tetrapentyltin[4] (TEPT)	Tetrapentyltin	403.26	22.32 ± 0.01	10.37 ± 0.81
3	Tetraethyltin[5] (TEET)	Tetraethyltin	234.94	7.56 ± 0.06	--

[1]Formula weight of ethylated compound in parentheses.
[2]W ½,(sec), peak width at half height (n=10, n=9 for TPhT).
[3]Surrogate
[4]Internal Standards
[5]Impurity.

Table 2 — GC-PFPD Chromatographic Conditions

Injection mode	Splitless (*glass liner 3.4 mm i.d.*)
Injection volume	3 μL[1]
Injector temperature	220°C (*Isothermal)*
Column	DB-17: 30 m x 0.25 mm i.d. x 0.25 μm film thickness
Flow rate (Helium)	2.0 mL min^{-1}
Oven temperature	40°C for 3 min; 8°C min^{-1}; 280°C for 1 min
Detector temperature	350°C
Detector trigger level	200 mV
Detector high voltage	550 V
Gate Delay	4.5 msec
Gate Width	5.0 msec
Detector gases	Hydrogen and Air
Gas flows	Air 1: 17.0 mL min^{-1}
	Air 2: 10.0 mL min^{-1}
	Hydrogen: 13.0 mL min^{-1}
Total run time	34.0 min

[1]An injection volume of 3 μL allowed the optimum analytical signal with minimum baseline noise via adjustment of the gate delay and gate width.

Table 3 – Reaction Time Data (n=4)

	MMT	DMT	NTPrT	MBT	DBT	TBT	MPhT	DPhT	TPhT	TCyHT
5 min	0.3494	0.8244	0.5800	0.5479	0.5570	0.7773	0.3366	0.6624	0.5441	0.6678
std. dev.	0.1141	0.2755	0.1963	0.1866	0.1790	0.0421	0.0152	0.0377	0.0338	0.0362
RSD %	32.66	33.42	33.84	34.06	32.14	5.42	4.51	5.69	6.25	5.42
10 min	0.4822	1.1405	0.8101	0.7459	0.7616	0.7830	0.3422	0.6706	0.5403	0.6853
std. dev.	0.0318	0.0667	0.0518	0.0643	0.0551	0.0581	0.0288	0.0364	0.0330	0.0418
RSD %	6.59	5.85	6.39	8.62	7.23	7.42	8.42	5.43	6.11	6.10
15min	0.5142	1.1948	0.8359	0.7904	0.7934	0.8048	0.3614	0.6852	0.5641	0.7356
std. dev.	0.0227	0.0222	0.0117	0.004570	0.009672	0.0128	0.0100	8.7737e-3	8.9212e-3	0.0185
RSD %	4.41	1.86	1.40	0.58	1.22	1.59	2.77	1.28	1.58	2.51
20 min	0.4642	1.0196	0.7278	0.7183	0.6998	0.8794	0.4047	0.7440	0.6223	0.7975
std. dev.	0.1026	0.2515	0.1583	0.1509	0.1493	0.0309	0.0151	0.0145	0.0353	0.0251
RSD %	22.10	24.67	21.75	21.01	21.33	3.51	3.73	1.95	5.67	3.15

*Data represented graphically in Figure 1.

Table 4 – Method Detection Limits

Analyte	Mean observed (ng L^{-1})	Standard Deviation (ng L^{-1})	Relative Standard Deviation (%)	Mean Accuracy (% true Conc.)	MDL[1] (ng L^{-1})
MMT	0.23	0.010	4.3	115	0.03
DMT	0.20	0.004	2.2	100.	0.01
MBT	0.19	0.003	1.7	95.0	0.01
DBT	0.27	0.007	2.5	135	0.02
TBT	0.21	0.003	1.6	105	0.01
MPhT	0.12	0.005	4.5	60.0	0.02
DPhT	0.17	0.009	5.1	85.0	0.03
TPhT	0.30	0.012	4.0	150	0.04
TCyHT	0.23	0.003	1.2	115	0.01

[1]MDL = t * standard deviation, where t = Student's t-value, 2.998 for 7 degrees of freedom.

Table 5 – Recoveries of Organotins from Water Samples

		Organotin Analytes, μg L^{-1} in sample extract									
		DMT	MMT	MBT	NTPrT	DBT	TBT	MPhT	DPhT	TCyHT	TPhT
Cincinnati tap water, <2.5 mg L^{-1} total organic carbon content	Mean observed, μg L^{-1}	1.11	.097	1.07	1.11	1.06	1.12	1.03	1.03	.097	1.01
	Standard deviation	.024	001	.017	.009	.021	.012	.034	.059	.070	.001
	RSD (%)	2.17	.013	1.69	.084	1.98	1.04	3.34	5.75	7.23	.013
	% Recovery	111	96.7	101	112	106	112	103	103	97.1	101
Water with high TOC, >10. mg L^{-1} total organic carbon content	Mean observed, μg L^{-1}	1.09	.074	.075	.093	.073	.091	.071	.074	.081	.088
	Standard deviation	.039	.016	.017	.008	.037	.047	.018	.013	.021	.016
	RSD (%)	3.62	2.16	2.23	.082	5.14	5.13	2.59	1.80	2.59	1.83
	% Recovery	109	74.5	74.9	93.0	72.7	90.8	70.8	74.3	81.4	88.3

*Fortified organotins, 5 ng L^{-1} in 1 L sample; 1 μg L^{-1} in the hexane extract.

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