Appendix 5

Standard Operating Procedure

for the USGS Reston, Virginia Environmental Organic Geochemistry Laboratory

Instrumental Analysis for the Long-Chain Alkylbenzenes

Note: A version of this Standard Operating Procedure was accepted by the National Oceanic and Atmospheric Administration (NOAA) in August 1993 as part of the Analytical Chemistry Quality Assurance Plan for the Southern California Natural Resource Damage Assessment.

Contents

Instrumental Analysis for the Long-Chain Alkylbenzenes

1. **Application:** This document describes procedures to be used for determination of long-chain alkylbenzenes (LCABs) in marine sediment samples using gas chromatography/mass spectrometry (GC/MS). The procedures presented here are for 26 individual linear alkylbenzenes (LABs) + 5 surrogates (RS_n, where n = 1-5) and 12 targeted peaks of the tetrapropylene-based alkylbenzenes (TABs). A description of the standards used in this procedure is given below (section 4.). It is assumed that the samples were previously extracted according to procedures described in Appendix 4 and fractionated according to procedures given in Appendix 2.

a. ***Tested concentration range.*** This procedure is only one part of a complete analytical method. Expected concentrations of the total linear alkylbenzenes (*i.e.* summation of all 26 compounds; ΣLAB_{26}) in the marine sediments to be tested are 0.16 to 1.97 µg/dry g based on measurements made by Eganhouse *et al.* (2000) on sediments collected near LACSD station 3C in 1992 (*i.e.* core 124B1-DDT). Total LAB concentrations in sediments near 6C are predicted to range from 1.2 to 15.0 µg/dry g. TAB concentrations in sediment (*i.e.* the sum of the twelve TAB analyte peaks, ΣTAB_{12}) from the vicinity of station 3C could range from 0. 24 to 1.94 µg/dry g. ΣTAB_{12} concentrations in sediment from the vicinity of station 6C are expected to range from 2.4 to 16.34 µg/dry g.

b. ***Sensitivity.*** Following is a tabulation of peak areas (using the respective quantitation ions; *cf.* Table 5 below) obtained near the detection limit based upon method detection limit (MDL) experiments conducted for the long-chain alkylbenzenes in 1993 (Table 1). All injections were one microliter in volume, and the data represent average results obtained from three replicate injections. These data were obtained with an ion trap detector not used in the **Palos Verdes Remediation Project**. Therefore, they serve only as a general guide for the sensitivity that might be obtained using the Agilent 5973 mass selective detector, which was used in the **Palos Verdes Remediation Project**. Obtaining similar data based on an MDL experiment with the Agilent system was not possible in the **Palos Verdes Remediation Project** due to time limitations.

Table 1. Tabulation of peak areas of long-chain alkylbenzenes near the method detection limit[a].

Compound	Peak Area
5-phenyl-C_{10}	1426
4-phenyl-C_{10}	848
3-phenyl-C_{10}	890
2-phenyl-C_{10}	2008

Table 1. Tabulation of peak ares of long-chain alkylbenzenes near the method detection limit[a] cont'd.

Compound	Peak Area
6-phenyl-C_{10}	474
5-phenyl-C_{11}	2998
4-phenyl-C_{11}	321
3-phenyl-C_{11}	1134
2-phenyl-C_{11}	366
6-phenyl-C_{12}	4677
5-phenyl-C_{12}	4016
4-phenyl-C_{12}	3796
3-phenyl-C_{12}	2979
2-phenyl-C_{12}	3629
7&6-phenyl-C_{13}	5217
5-phenyl-C_{13}	3978
4-phenyl-C_{13}	2965
3-phenyl-C_{13}	2935
2-phenyl-C_{13}	5659
7-phenyl-C_{14}	1940
6-phenyl-C_{14}	2199
5-phenyl-C_{14}	2071
4-phenyl-C_{14}	1664
3-phenyl-C_{14}	1406
2-phenyl-C_{14}	2696

[a]Data obtained with Finnigan MAT model 800A Ion Trap Detector. Note: the nomenclature used with secondary phenylalkanes is **n**-phenyl-$C_{\mathbf{m}}$, where **n** = position of substitution of benzene ring on the alkyl chain and **m** = number of carbons in the alkyl chain.

c. ***Detection limits.*** Limits of detection have been determined for the 26 linear alkylbenzenes in marine sediments according to procedures described in 40 CFR Part 136 (USEPA, 1992). The results are based on analysis of seven replicate processed samples of precombusted sand (1.0 g/sample) and are given in the following table (Table 2).

Table 2. Method detection limits for linear alkylbenzenes.

Compound	MDL (ng/dry g)
5-phenyl-C_{10}	1.76
4-phenyl-C_{10}	1.73

Table 2. Method detection limits for linear alkylbenzenes, cont'd.

3-phenyl-C_{10}	0.48
2-phenyl-C_{10}	2.56
6-phenyl-C_{11}	2.86
5-phenyl-C_{11}	6.79
4-phenyl-C_{11}	4.07
3-phenyl-C_{11}	5.09
2-phenyl-C_{11}	8.94
6-phenyl-C_{12}	10.7
5-phenyl-C_{12}	9.53
4-phenyl-C_{12}	6.44
3-phenyl-C_{12}	7.67
2-phenyl-C_{12}	4.06
7&6-phenyl-C_{13}	12.6
5-phenyl-C_{13}	8.83
4-phenyl-C_{13}	6.13
3-phenyl-C_{13}	6.11
2-phenyl-C_{13}	7.47
7-phenyl-C_{14}	4.96
6-phenyl-C_{14}	6.01
5-phenyl-C_{14}	5.55
4-phenyl-C_{14}	3.24
3-phenyl-C_{14}	1.15
2-phenyl-C_{14}	4.09

Note: the nomenclature used with secondary phenylalkanes is **n**-phenyl-$C_{\mathbf{m}}$, where **n** = position of substitution of benzene ring on the alkyl chain and **m** = number of carbons in the alkyl chain.

d. ***Interferences.*** The long-chain alkylbenzenes are identified and measured using GC/MS. Eganhouse *et al.* (1983b) have shown that analysis of complex hydrocarbon fractions for the LABs by GC/MS is effective at avoiding interferences. Quantitation of the LABs is performed using selected ions (*e.g.* m/z = 91, 92 and 105) such that the signal-to-noise ratio for these compounds, even in complex mixtures such as the F2 fraction, is high (*e.g.* m/z = 91: ~10:1, m/z = 92: ~80:1, m/z = 105: ~5:1). The principal interference problem comes from partial coelution of the two groups of long-chain alkylbenzenes within the F2 fraction: the linear alkylbenzenes (LABs) and the tetrapropylene-based alkylbenzenes (TABs).

Zeng *et al.* (1998) conducted an investigation of the potential effects of TAB interference with individual LABs when the ions 91 (LABs) and 119 (TABs)

were used for quantitation of the long-chain alkylbenzenes in a sediment core collected on the Palos Verdes Shelf. They found that even in this non-optimized case, the effects on ΣLAB_{26} concentrations were minimal and did not significantly affect sediment concentration profiles. There are only 26 LABs within the $C_{10\text{-}14}$ range, and all have simple structures and well established mass spectra, many of which are in the NIST (National Institute of Standards and Technology) library. By contrast, the TABs are a complex mixture of alkylbenzenes with a high degree of branching, notably at the α carbon. Consequently, ions such as m/z = 119 ($C_9H_{11}^+$ ion) are often diagnostic of the TABs. In short, the two compound groups can be differentiated only by mass spectrometry. A thorough discussion of this problem and its solution is provided in Eganhouse *et al.* (1983b) and Eganhouse (1986). As will be discussed below, the quantitation of the LABs relies upon the use of m/z = 91, 92, 105, and (occasionally) a more diagnostic, but less abundant, ion, whereas m/z = 105 and 119 are used for quantitation of the twelve TAB analyte peaks. There are five LAB peaks which could, in principle, suffer from interference from the TABs. These are 2-phenyl-C_{10}, 6-phenyl-C_{11}, 4-phenyl-C_{11}, 3-phenyl-C_{11} and 2-phenyl-C_{11} (Note: the nomenclature used with secondary phenylalkanes is **n**-phenyl-$C_{\mathbf{m}}$, where **n** = position of substitution of benzene ring on the alkyl chain and **m** = number of carbons in the alkyl chain). In all cases, quantitation ions have been selected for these LABs that are either not present or are in very low abundance in the TAB peaks with which they coelute.

e. ***Analysis Rate.*** Chromatographic runs take approximately 85 minutes including cool down time for the gas chromatograph. Since there are approximately 7 runs for 4 field samples/compound class (2 single point calibration runs, ½ equivalent blank, ½ matrix spike/matrix spike duplicate equivalent + 4 samples), it takes approximately 10 hours to complete a run of 4 field samples. This does not include final preparation of the samples (~30 minutes/sample) and other miscellaneous preparatory activities (instrument checkout, computer/data system checkout, tuning and calibration of the mass spectrometer). Reduction of the data is completed after the acquisition(s). Each chromatographic run is inspected for quality of the chromatogram, obvious interference problems, peak identification, baseline and proper integration. This would be expected to take approximately 4-6 hours. If necessary, reprocessing (or reanalysis) is carried out. Hence, two eight-hour days is a conservative estimate of the time required to complete the instrumental analyses for 4 samples.

2. **Chemistry:** No chemical reactions are involved in these procedures.

3. **Apparatus:** All analyses of long-chain alkylbenzenes are performed by GC/MS. Confirmation of the identifications made by the Agilent ChemStation software are based on retention times (relative to that of the internal standards) and mass spectral information produced with the GC/MS (*i.e*. qualifier ion/quantitation ion abundance ratios are compared with reference mass spectra from the calibration standard). Confirmation requires that a peak fall within the targeted retention time window (± 0.3 min) and satisfy the specified qualifier ion/quantitation ion abundance ratio criterion (± 20% of that reported for the calibration standard). Following is a description of the instrument.

GC/MS: The instrumentation includes an Agilent 6890 capillary gas chromatograph equipped with cool on-column and split/splitless injectors with electronic pressure control. The analytical column, a 30 m x 0.25 mm (id) DB-5 fused silica capillary with a 0.25 μm film thickness (J& W Scientific), is identical to that used for the GC/ECD analyses of the F2 fractions (*cf*., Appendix 7). The capillary column is directly interfaced to an Agilent 5973 mass selective detector by way of the standard heated transfer line. The mass spectrometer interface is held isothermal at 285 °C, and the manifold and ion source regions are held isothermal at 150 °C and 250 °C, respectively. Analyte molecules are ionized by electron impact at 70 eV. The instrument is operated in full scan mode, scanning from 50 to 500 amu at 1.68 scans/second. The GC/MS is controlled by Agilent ChemStation software, D01.00 Build 75, running under Windows XP Professional (SP1). Data are acquired by the ChemStation data system and stored on the hard disk of a Dell GX270 Optiplex desktop computer. Mass spectral confirmations and peak quantitations are performed post-acquisition using Agilent quantitation programs running under ChemStation. Acquired runs and subsequent quantitation files are backed up on a separate external hard disk. Prior to sample analyses, an Autotune is performed on the system using PFTBA (perfluorotributylamine).

4. Standards:

a. ***Calibration standards.*** The LABs (not including the surrogates and internal standards-see below) and the TABs are commercially produced technical mixtures that were obtained from Monsanto Company in the early 1980s. A 'master mixture' of LABs containing all 26 secondary phenylalkanes having alkyl chains with 10 to 14 carbons was made by combining several LAB mixtures having different chain lengths. Concentrations of the 26 individual secondary phenylalkanes in the LAB 'master mixture' were determined by replicate analysis (n = 5) of this solution following incorporation of an internal (quantitation) standard, 1-phenyldodecane, by gas chromatography/flame ionization detection (GC/FID; Eganhouse *et al.*, 1983a, 1983b). Hence, the LAB calibration solutions described below are secondary standards. Five 1-phenylalkanes (1-phenyldecane, 1-phenylundecane, 1-phenyldodecane, 1-phenyltridecane, and 1-phenyltetradecane) were added as surrogates and two 1-phenylalkanes (1-phenylnonane and 1-phenylpentadecane) were added to the LAB 'master mixture' as internal (quantitation) standards to create a single-point calibration standard (LAB-CS-05-1/5; *cf*., chromatograms below and Table 3). Thus, the LAB single-point calibration solution contains 26 + 5 + 2 =33 alkylbenzenes (*cf*., Figure 1 showing chromatogram below, Table 3). The multipoint calibration solutions (Table 4) do <u>not</u> contain these surrogates, but they do contain the two 1-phenylalkanes (1-phenylnonane and 1-phenylpentadecane) used as internal (quantitation) standards. Therefore, they contain 26 + 2 =28 alkylbenzenes.

There are two types of calibration standards being used in this procedure: 1) single-point calibration standards, one each for the linear alkylbenzenes (LABs; LAB-CS-05-1/5; *cf*., Table 3) and the tetrapropylene-based alkylbenzenes (TABs; TAB-CS-01-1/1; *cf*., Table 3), and 2) a set of multipoint calibration standards for the linear alkylbenzenes (five levels: LAB-MP-03-1/n; where n=1,2,4,16,64 corresponding to the levels; *cf*., Table 4). The use of two calibration standards for

the LABs (*i.e*. a single point calibration standard and a set of multipoint calibration standards) was necessitated by the fact that the multipoint calibration standard solutions originally prepared for the analyses conducted for the NOAA-sponsored study in 1992-944 did not contain the reovery surrogates. They only contained the 26 secondary C_{10-14}-benzenes + 2 internal quantitation standards as discussed above. Thus, a single-point calibration standard containing the 26 secondary C_{10-14}-benzenes + the 5 surrogates + 2 internal quantitation standards was created to separately quantify the surrogates. Use of a single-point calibration for quantitation of the surrogates is justified because the multipoint calibration curves (with forcing through the origin) for the long-chain alkylbenzenes are highly linear ($R^2 > 0.999$). On the same basis, multipoint calibration for the TABs was deemed unnecessary because the LABs and TABs are closely related compounds whose chromatographic/mass spectroscopic behaviors are essentially identical. Thus, in the original NOAA-sponsored studies (1992-94) and in the EPA-sponsored **Palos Verdes Remediation Project** (2006) the LABs are used to represent both types of long-chain alkylbenzenes (LABs and TABs) in the multipoint calibration exercise. Individual component concentrations for the single-point calibration standards range from 11 to 60 ng/μL (LAB-CS-05-1/5; *cf*., Table 3) and 9-65 ng/μL (TAB-CS-01-1/1; *cf*., Table 3), whereas the nominal component concentrations in the multipoint calibration standard solutions range from ~1-290 ng/μL (LABs only; *cf*., Table 4).

In the case of the TABs, a stock solution was made by weighing a portion of the TAB technical mixture obtained from Monsanto Company into a volumetric flask and bringing it to volume in hexane (concentration ≈ 0.47 μg/μL). The abundances of twelve individual TAB peaks in the TAB mixture were determined by adding a known amount of 1-phenyltetradecane to the solution and then performing repetitive GC/FID analyses (n = 7) using 1-phenyltetradecane as an internal quantitation standard. The single-point calibration standard solution used in this study (TAB-CS-01-1/1; *cf*. figure showing chromatogram to follow, Table 3) consists of the TABs + two internal (quantitation) standards (1-phenylnonane, 1-phenylpentadecane). Note that no surrogates are included in the TAB single-point calibration standard because recovery is assessed in field samples by quantitation using the single-point LAB calibration standard (LAB-CS-05-1/5; *cf*., Table 3).

Table 3. Compositions of single-point calibration standards for the long-chain alkylbenzenes (LAB-CS-05-1/5, TAB-CS-01-1/1).

	LAB-CS-05-1/5	
Component	Description	(ng/μl)
5-phenyl-C_{10}	analyte	21.19
4-phenyl-C_{10}	analyte	16.65
3-phenyl-C_{10}	analyte	15.88
2-phenyl-C_{10}	analyte	15.89

Table 3. Compositions of single-point calibration standards for the long-chain alkylbenzenes (LAB-CS-05-1/5, TAB-CS-01-1/1) cont'd.

6-phenyl-C_{11}	analyte	24.27
5-phenyl-C_{11}	analyte	40.55
4-phenyl-C_{11}	analyte	30.80
3-phenyl-C_{11}	analyte	28.50
2-phenyl-C_{11}	analyte	24.19
6-phenyl-C_{12}	analyte	45.95
5-phenyl-C_{12}	analyte	39.95
4-phenyl-C_{12}	analyte	27.21
3-phenyl-C_{12}	analyte	20.48
2-phenyl-C_{12}	analyte	11.47
7&6-phenyl-C_{13}	analyte	58.56
5-phenyl-C_{13}	analyte	36.89
4-phenyl-C_{13}	analyte	27.24
3-phenyl-C_{13}	analyte	26.23
2-phenyl-C_{13}	analyte	26.45
7-phenyl-C_{14}	analyte	22.15
6-phenyl-C_{14}	analyte	19.58
5-phenyl-C_{14}	analyte	19.26
4-phenyl-C_{14}	analyte	14.67
3-phenyl-C_{14}	analyte	14.12
2-phenyl-C_{14}	analyte	12.42
1-phenyl-C_{9}	IS1	23.41
1-phenyl-C_{10}	RS1	20.25
1-phenyl-C_{11}	RS2	27.11
1-phenyl-C_{12}	RS3	25.21
1-phenyl-C_{13}	RS4	30.22
1-phenyl-C_{14}	RS5	20.22
1-phenyl-C_{15}	IS2	26.95

TAB-CS-01-1/1		
Component	**Description**	**(ng/μl)**
TAB-1 (TAB1)	analyte	33.28
TAB-2 (TAB2)	analyte	19.03
TAB-3	analyte	11.21
TAB-4	analyte	65.01
TAB-5	analyte	32.79
TAB-6	analyte	20.07
TAB-7	analyte	21.41
TAB-8 (TAB3)	analyte	50.20
TAB-9	analyte	8.56
TAB-10	analyte	45.04
TAB-11	analyte	20.24
TAB-12	analyte	13.62

1-phenyl-C_9	IS1	25.13

Table 3. Compositions of single-point calibration standards for the long-chain alkylbenzenes (LAB-CS-05-1/5, TAB-CS-01-1/1) cont'd.

1-phenyl-C_{15}	IS2	66.37

Note: RS=surrogate
IS=internal quantitation standard

Table 4. Composition of multipoint calibration standard solutions for the linear alkylbenzenes (LAB-MP-03-1/n; where n=1,2,4,16,64).

Component	Description	LAB-MP-03-1/1 (ng/µl)	LAB-MP-03-1/2 (ng/µl)	LAB-MP-03-1/4 (ng/µl)	LAB-MP-03-1/16 (ng/µl)	LAB-MP-03-1/64 (ng/µl)
5-phenyl-C_{10}	analyte	105.95	52.98	26.49	6.62	1.66
4-phenyl-C_{10}	analyte	83.25	41.63	20.81	5.20	1.30
3-phenyl-C_{10}	analyte	79.40	39.70	19.85	4.96	1.24
2-phenyl-C_{10}	analyte	79.45	39.73	19.86	4.97	1.24
6-phenyl-C_{11}	analyte	121.35	60.68	30.34	7.58	1.90
5-phenyl-C_{11}	analyte	202.75	101.38	50.69	12.67	3.17
4-phenyl-C_{11}	analyte	154.00	77.00	38.50	9.63	2.41
3-phenyl-C_{11}	analyte	142.50	71.25	35.63	8.91	2.23
2-phenyl-C_{11}	analyte	120.95	60.48	30.24	7.56	1.89
6-phenyl-C_{12}	analyte	229.75	114.88	57.44	14.36	3.59
5-phenyl-C_{12}	analyte	199.75	99.88	49.94	12.48	3.12
4-phenyl-C_{12}	analyte	136.05	68.03	34.01	8.50	2.13
3-phenyl-C_{12}	analyte	102.40	51.20	25.60	6.40	1.60
2-phenyl-C_{12}	analyte	57.35	28.68	14.34	3.58	0.90
7&6-phenyl-C_{13}	analyte	292.80	146.40	73.20	18.30	4.58
5-phenyl-C_{13}	analyte	184.45	92.23	46.11	11.53	2.88
4-phenyl-C_{13}	analyte	136.20	68.10	34.05	8.51	2.13
3-phenyl-C_{13}	analyte	131.15	65.58	32.79	8.20	2.05
2-phenyl-C_{13}	analyte	132.25	66.13	33.06	8.27	2.07
7-phenyl-C_{14}	analyte	110.75	55.38	27.69	6.92	1.73
6-phenyl-C_{14}	analyte	97.90	48.95	24.48	6.12	1.53
5-phenyl-C_{14}	analyte	96.30	48.15	24.08	6.02	1.50
4-phenyl-C_{14}	analyte	73.35	36.68	18.34	4.58	1.15
3-phenyl-C_{14}	analyte	70.60	35.30	17.65	4.41	1.10
2-phenyl-C_{14}	analyte	62.10	31.05	15.53	3.88	0.97
1-phenyl-C_9	IS1	23.41	23.41	23.41	23.41	23.41
1-phenyl-C_{15}	IS2	26.95	26.95	26.95	26.95	26.95

b. ***Surrogates (recovery).*** As noted above, five surrogates (all 1-phenylalkanes) are used for recovery assessment and correction (if necessary). The reason for using this number of compounds is that the LABs and TABs are semi-volatile, and there is a significant amount of variation in vapor pressure and solubility over their chain length distribution (Sherblom *et al.*, 1992). It is commonly found that the LABs with shorter chain lengths can be preferentially lost during sample workup. Consequently, recovery correction is sometimes necessary and beneficial (Note: Recovery correction is performed if calculated recoveries are below 80% . This was not necessary in the **Palos Verdes Remediation Project**.). In this case however, correction of LAB concentrations of a given chain length (*e.g.* C_{10}) is performed with the surrogate bearing one less alkyl carbon (*i.e.* C_9). This is because a given secondary phenylalkane and the 1-phenylalkane having one less carbon on its alkyl chain have similar physical properties (Sherblom *et al.*, 1992). The scheme used for recovery correction is summarized in Table 5. Several solutions of surrogates were prepared in anticipation of widely varying concentrations (*cf.* Appendix 4). Quantitation of surrogate recovery in samples is carried out using the LAB single-point calibration standard (LAB-CS-05-1/5).

Table 5. Scheme used for recovery correction of long-chain alkylbenzene concentrations.

Surrogate	Analytes Corrected[a]
1-phenyldecane (RS1)	5-phenyl-C_{10}, 4-phenyl-C_{10}, 3-phenyl-C_{10}, 2-phenyl-C_{10}, 6-phenyl-C_{11}, 5-phenyl-C_{11}, 4-phenyl-C_{11}, 3-phenyl-C_{11}, 2-phenyl-C_{11}
1-phenylundecane (RS2)	6-phenyl-C_{12}, 5-phenyl-C_{12}, 4-phenyl-C_{12}, 3-phenyl-C_{12}, 2-phenyl-C_{12}
1-phenyldodecane (RS3)	7&6-phenyl-C_{13}, 5-phenyl-C_{13}, 4-phenyl-C_{13}, 3-phenyl-C_{13}, 2-phenyl-C_{13}
1-phenyltridecane (RS4)	7--phenyl-C_{14}, 6--phenyl-C_{14}, 5-phenyl-C_{14}, 4-phenyl-C_{14}, 3-phenyl-C_{14}, 2-phenyl-C_{14}
1-phenyltetradecane (RS5)	none
1-phenylundecane (RS2)	TAB peaks

[a]Nomenclature used with secondary phenylalkanes in this table is **n**-phenyl-$C_{\mathbf{m}}$, where **n**=position of substitution of benzene ring on the alkyl chain and **m**=number of carbons in the alkyl chain.

c. ***Internal (quantitation) sandards.*** One internal (quantitation) standard solution (LAB-IS-03-1/10) consisting of 1-phenylnonane (IS1) and 1-phenylpentadecane (IS2) in hexane was prepared for this study. This solution contains the two 1-phenylalkanes at concentrations of 46.8 and 53.9 ng/µL, respectively. These compounds were selected because they elute in gas chromatograms away from the LABs (*cf.*, Figure 1 with chromatograms below). Of the two internal standards,

1-phenylnonane is the only one that coelutes within the TAB range, but it is easily distinguished from the TABs by its distinctive mass spectrum (base peak at m/z = 92).

5. Procedure:

a. ***Preparation of F2 fractions for instrumental analysis.*** As discussed in Appendix 7 entitled, "**Instrumental Analysis for Chlorinated Hydrocarbons**", an aliquot consisting of 70% by volume (700 µL out of 1000 µL) of the F2 fraction is used for the LCAB split. This is reduced in volume to approximately 100 µL under a stream of dry nitrogen gas after which it is transferred to an autosampler microvial (National Scientific, #C4000-2W, #C4010-629L) followed by three successive rinses with 50 µL of dichloromethane. Care is taken to ensure quantitative transfer of the LCAB F2 split, and the rinses are alternated with reduction in volume in the microvial using a stream of dry nitrogen gas. Once the transfer is complete, the solvent is gently evaporated to dryness under a stream of dry nitrogen, and the LCAB split is immediately taken up in an appropriate volume of the internal standard solution (LAB-IS-03-1/10; *cf*., section 4.c.). An equal volume of hexane is added to bring internal standard compounds to the same concentration as in the multipoint and single point calibration solutions (see above). The sample is then ready for analysis or temporary (<24 hours) storage in the freezer.

b. ***Instrumental analysis.*** Following are general instructions for the analysis of the LCAB F2 splits by GC/MS.

GC/MS: Long-chain alkylbenzenes in sediments are analyzed by GC/MS using an Agilent 6890 high resolution gas chromatograph interfaced to an Agilent 5973 mass selective detector. Conditions of analysis are given in the following table (Table 6).

Table 6. Conditions used for GC/MS analysis of sediment F2 fractions for long-chain alkylbenzenes.

Parameter	Setting
Column:	Agilent p/n 122-5032
Phase	DB-5
Length	30 meters
ID	0.25 mm
Film thickness	0.25 µm
Gas Chromatograph:	Agilent 6890 Plus
Injector	Agilent 7683 autosampler + split/splitless injector, liner #18740-80220
Injector temperature	285 °C
Transfer line temperature	285 °C

Table 6. Conditions used for GC/MS analysis of sediment F2 fractions for long-chain alkylbenzenes cont'd.

Initial oven temperature	50 °C, 3 minute isothermal hold
Program 1	50 °C to 100 °C @ 25 °C/min
Program 2	100 °C to 285 °C @ 4 °C/min with 40 min isothermal hold
Carrier, linear velocity	helium (upc), 43 cm/sec
Mode	constant flow (1.4 mL/min)
Mass Spectrometer:	Agilent 5973
Electon multiplier voltage	1717 volts
Mass range, scan rate	50-500 amu, 1.68 Hz
Source temperature	250 °C
Quad temperature	150 °C
Data System:	ChemStation (D01.00 Build 75)
Sampling rate	20 Hz
Method Name	LCABSA-PVR1.m

Injection is splitless, and data acquisition is in full scan mode. In the case of the LABs, quantitation is performed by the internal standard method using ions that are either base peaks in the mass spectra or, in cases of potential TAB interference (*cf.* section 1.d.), ions that are abundant in the target LAB analytes but are either not found or are minor ions in the mass spectra of interfering TABs. In the case of the TABs, twelve major peaks whose abundance in the TAB mixture is known, are quantitated. Two of these peaks, hereafter referred to as TAB1 and TAB2 (aka TAB-1, TAB-2; *cf.* Figure 1), elute in a region of the chromatogram free of LABs and the 1-phenylalkane surrogates and internal standards. A third, TAB3 (aka TAB-8), partially coelutes with 1-phenyldecane (RS1). However, the mass spectra of TAB3 and 1-phenyldecane are sufficiently different to permit selection of ions that reduce the error to insignificant levels. Once the concentration of the twelve individual TAB peaks is known, the summed concentrations of the twelve analyte peaks (ΣTAB_{12}) can be computed. Table 7 lists the ions used for quantitation of the 26 LABs + five surrogates and the twelve TAB peaks along with the internal standard employed for each analyte. The analyte tabulation is by order of elution. Figure 1 shows high resolution gas chromatograms (GC/MS-total ion current) of the LAB and TAB calibration standard solutions with each peak labelled accordingly. Please note that owing to space limitations, the LABs are labeled in this figure **n-m**, where **n** = the position of substitution, **m** = chain length.

Figure 1. High Resolution Gas Chromatograms (GC/MSD, total ion current) of LAB and TAB Calibration Standards.

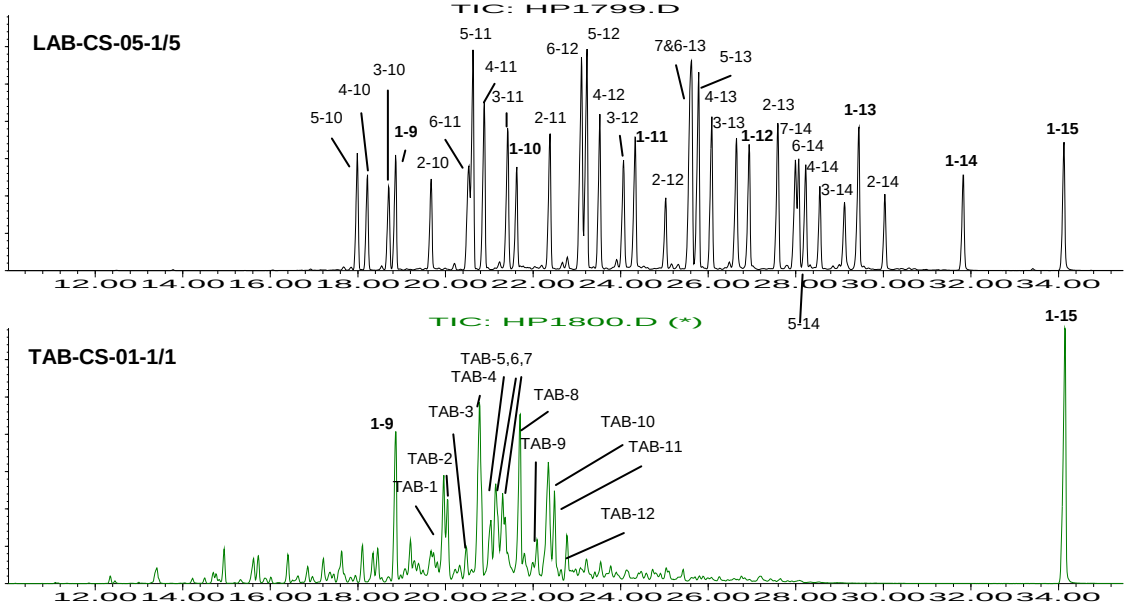

Table 7. Ions used for GC/MS quantitation of long-chain alkylbenzenes.

Analyte[a]	Quant ion used (m/z)[b]	Qualifier ions	IS compound used
5-phenyldecane	91	147, 161, 218	1-phenylnonane (IS1)
4-phenyldecane	91	133, 175, 218	"
3-phenyldecane	91	119, 189, 218	"
1-phenylnonane (IS1)	92	91, 204	---
2-phenyldecane	105	218, 91, 106	1-phenylnonane (IS1)
TAB-1 (TAB1)	105	161, 189, 246	"
TAB-2 (TAB2)	105	147, 203, 246	"
TAB-3	119	91, 105, 246	"
6-phenylundecane	161 (91)	105, 91, 232	"
5-phenylundecane	91	147, 175, 232	"
TAB-4	119	105, 133, 246	"
4-phenylundecane	189 (91)	133, 91, 232	"
TAB-5	119	105, 91, 246	"
TAB-6	119	105, 133, 246	"
TAB-7	105	133, 217, 246	"

Table 7 . Ions used for GC/MS quantitation of long-chain alkylbenzenes cont'd.

Compound	Quant ion used (m/z)[b]	Qualifier ions	IS compound used
3-phenylundecane	119 (91,203)	203, 91, 232	"
1-phenyldecane (RS1)	92	218, 91	"
TAB-8 (TAB3)	105	119, 133, 246	"
TAB-9	119	105, 133, 260	"
TAB-10	119	91, 133, 260	"
2-phenylundecane	105 (232)	232, 106, 91	"
TAB-11	119	91, 105, 246	"
TAB-12	119	91, 105, 246	"
6-phenyldodecane	91	161, 175, 246	"
5-phenyldodecane	91	147, 189, 246	"
4-phenyldodecane	91	133, 203, 246	"
3-phenyldodecane	91	119, 217, 246	"
1-phenylundecane (RS2)	92	232, 91	"
2-phenyldodecane	105	246, 106, 91	"
7&6-phenyltridecanes	91	105, 189 274	1-phenylpentadecane (IS2)
5-phenyltridecane	91	147, 203, 260	"
4-phenyltridecane	91	133, 217, 260	"
3-phenyltridecane	91	119, 231, 260	"
1-phenyldodecane (RS3)	92	246, 91	"
2-phenyltridecane	105	260, 106, 91	"
7-phenyltetradecane	91	175, 189, 274	"
6-phenyltetradecane	91	161, 203, 274	"
5-phenyltetradecane	91	147, 217, 274	"
1-phenyltridecane (RS4)	92	274, 91	"
1-phenyltetradecane (RS5)	92	288, 91	"
1-phenylpentadecane (IS2)	92	91, 133, 288	---
2-phenyltetradecane	105	274, 106, 91	"

[a] Compounds listed in order of elution.

[b] Ions in parentheses are alternates to be used in cases where significant TAB interferences are present.

6. Calculations: Computation of alkylbenzene concentrations is carried out by the ChemStation data system. Previous multipoint calibrations with the solutions described here using GC/MS have been linear over the entire concentration range (2-275 ng/µL). Consequently, we use a linear model with forcing through the origin. This applies to both multipoint and single-point calibrations for the LABs and the single-point

calibration for the TABs. Under these circumstances, the following equation is employed for computation of alkylbenzene concentrations in sediment samples.

The equation used for computing concentrations of the analytes using a linear model with forcing through the origin (not recovery corrected) is given below.

$$[C_i]_S = \left[\frac{A_i \times C_{IS}}{A_{IS}}\right]_S \times RRF_{CS}^{-1} \times \frac{V_{final} \times DF}{M_S} \qquad (1)$$

where: $[C_i]_S$ = concentration of analyte *i* in the sediment sample (ng/dry g),
A_i = area of analyte peak *i* in the final sample solution,
C_{IS} = concentration of the internal standard in the sample solution (ng/µL),
A_{IS} = area of the internal standard peak in the final sample solution,
RRF_{CS}^{-1} = relative response factor of the calibration standard run,
V_{final} = total volume of the sample solution at time of GC analysis (µL),
DF = dilution factorincorporating fraction of total extract fractionated, split taken of F2 fraction and subsequent dilutions, if any,
M_S = dry mass of sediments extracted (dry g).

The equation for estimating recovery is given below.

$$R_i = \left[\frac{C_{RSi} \times M_S}{M_{RSi}}\right] \times 100 \qquad (2)$$

where: R_i = recovery of surrogate *i* in per cent,
C_{RSi} = concentration of surrogate *i* in the sample (ng/dry g),
M_S = dry mass of sediments extracted (dry g),
M_{RSi} = mass of surrogate *i* added to the sample (ng).

The equation for correcting analyte concentrations for recovery is given below.

$$C_i' = [C_i]_S \Big/ 0.01R_i \qquad (3)$$

where: C_i' = recovery corrected concentration of analyte *i* (ng/dry g),
$[C_i]_S$ = concentration of analyte *i* in sample before recovery correction (ng/dry g),
R_i = recovery of surrogate appropriate to analyte *i*.

7. **QA/QC Considerations:** Information on the number of blanks, SRMs (standard reference materials), MS/MSDs (matrix spike/matrix spike duplicates), *etc.* that are processed along with a given number of samples or per batch can be found in Appendix 4 entitled, "**Extraction of Sediments for Analysis of Trace Organics**". The quality control criteria that must be met with regard to the analysis of sediment samples for chlorinated hydrocarbons are listed in the **Southern California Damage Assessment Analytical Chemistry Quality Assurance Plan** (Manen, 1994) and will not be further elaborated here. In general, these criteria are also observed for the long-chain alkylbenzenes. As stated in the QA Plan under section 7.0, "When the data from the analyses of any quality control sample exceeds the project specified control limits… or indicates that the analytical method is drifting out of control, it is the immediate responsibility of the analyst to identify and correct the anomaly before continuing with sample analysis. Either a narrative or a completed corrective action report form...describing the anomaly noted, the steps taken to identify and correct the anomaly and the treatment of the relevant sample batch, *i.e.* recalculation, reanalysis, reextraction must be submitted with - and accompany - the relevant data package through all steps in the data validation and archival process." In view of the many possible situations that could fall within this category, it is impossible to list all of the corrective actions that would or conceivably could be taken. No SRM exists for which LAB and TAB concentrations have been certified. However, in 2000 Hartmann *et al.* published a paper on the determination of LABs in SRM1941a. Their data are compared with results obtained in this study.

8. **Health, Safety, and Waste-Disposal Information:**

a. ***Personal protection.*** Safety glasses and protective gloves are recommended whenever reagents or samples are handled. For other precautions and safety procedures, consult the Material Safety Data Sheets (MSDS) for each chemical used. They are on file in the laboratory; *http://www.ilpi.com/msds/#Manufacturers* provides links to MSDSs of most chemical companies.

b. ***Electrical hazards.*** Electrical systems must conform to the National Electric Code, the National Fire Protection Association Code (NFPA 70-1971), and the American National Standards Institute (ANSI) Code (C1-1971). Consult the U.S. Geological Survey's Safety and Environmental Health Handbook (U.S. Geological Survey, 2002). Shock hazards exist inside the instruments. Only an authorized service representative or an individual with training in electronic repair should remove panels or circuit boards where voltages are greater than 20 V. The instruments require a third-wire protective grounding conductor. Three-to-two wire adapters are unsafe for these instruments.

c. ***Chemical hazards.*** Hexane, dichloromethane, and methanol are solvents used in cleaning glassware and the preparation of clean adsorbents and reagents. They also are evaluated for purity. Gloves should be worn when handling organic solvents and, whenever possible, manipulations should be conducted in a fume hood. Waste solvents accumulated during rotary evaporation or other cleaning operations should be stored in a capped glass bottle (satellite accumulation point) and arrangements made for its disposal through the USGS Materials Management Office.

d. ***Gas cylinder handling*****.** Compressed gas cylinders must be handled and stored according to the Safety and Environmental Health Handbook (U.S. Geological survey, 2002). Each cylinder must be 1) carefully inspected when received, 2) securely fastened at all times with an approved chain assembly or belt, 3) capped at all times when not in use, 4) capped when transported, 5) transported only by a properly designed vehicle (hand truck), and 6) stored separately with other full, empty, flammable, or oxidizing tanks of gas, as appropriate.

e. ***Sharps.*** Microsyringes with fixed or removable needles should be handled with care to avoid accidental skin punctures.

9. **<u>References</u>:** Following are citations from this SOP along with some additional sources of information (marked in bold) about the procedures that have been described here.

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