MRI

FINAL REPORT

Range Finding Study: Analysis of Interstitial Water and Sediment for Surfactants by Liquid Chromatography/Mass Spectrometry (LC/MS)

For
Mr. Alvaro J. DeCarvalho
The Soap and Detergent Association
1500 K Street, NW
Suite 300
Washington, D.C. 20005

MRI Project No. 310287

October 4, 2002

Midwest Research Institute

425 Volker Boulevard
Kansas City, Missouri
64110-2299
(816) 753-7600

solutions through science and technology

MIDWEST RESEARCH INSTITUTE
425 Volker Boulevard
Kansas City, Missouri 64110
Telephone (816) 753-7600
Telefax (816) 753-8420

October 4, 2002

Mr. Alvaro J. DeCarvalho
The Soap and Detergent Association
1500 K Street, NW
Suite 300
Washington, D.C. 20005

Subject: MRI Project No. 310287, "Range Finding Study: Analysis of Interstitial Water and Sediment for Surfactants by Liquid Chromatography/Mass Spectrometry (LC/MS)"

Dear Mr. DeCarvalho and Members of the Sediment Task Force:

Results from the range-finding study to determine residual surfactant concentrations in interstitial water and sediment samples from Little Miami River (Ohio) samples are presented herein. This report also includes results from additional experiments designed to measure spike recovery and stability of surfactants in preserved sediment over a 14-day storage period. Extractions for all residual test samples were initiated within 14 days of sample collection to be consistent with the stability experiments.

The experimental design for extraction of alkyl ethoxylates (AE) in the large-volume downstream interstitial water sample was modified to collect additional "breakthrough" solid-phase extraction cartridges for analysis. Figures 1 and 2 present an overview of the Range Finding Study and these additional experiments.

1. Range Finding Experiment for Surfactants in Interstitial Water and Sediment

The purpose of this task was to measure background levels of alkyl ethoxylate (AE), alkyl sulfate/alkyl ethoxysulfate (AS/AES), and linear alkyl benzene sulfonate (LAS) surfactants in interstitial water and sediment samples. Sediment samples, upstream and downstream from a test site on the East Fork of the Little Miami River (Ohio) site were collected. Interstitial water from the two sediment samples was separated using special equipment right after collection and submitted separately for analysis. The water and sediment samples were preserved with 3% formalin, and shipped to MRI under refrigerated storage conditions.

Surfactant concentrations in the samples were determined using methods based on previously validated or published studies, as described below. Due to the large number of homologues associated with surfactants, the scope of this study was limited to processing and reporting data for only representative subsets from each class of chemicals, specifically: AE surfactants C12, EO=0, 1, 2, 3, 6, 9, 12, 15; C13-15, C18, EO=0, 1, 2, 6, 9, 15; AS/AES surfactants C12-C15, EO=0, 2, 4, and 8; and LAS homologues C10, C11, C12, C13 and C14 (integrated as total area for each homologous series).

1.1 Analysis of Interstitial Water Samples for AE

Aqueous samples were analyzed using the procedure described in the MRI Report "Method Validation Study for the Analysis of Alkyl Ethoxylates in Water Effluents and Influents," Revised Report, dated June 29, 2000 (MRI Project No. 305224.1.001). This AE analytical method is summarized below:

AE Analysis of Aqueous Samples

- The aqueous sample (4 L) was siphoned through a pre-conditioned C2 cartridge to extract AE from the interstitial water.
- After the aqueous sample had passed through the cartridge, the C2 cartridge was dried by pulling air through the cartridge for a minimum of 8 hours.
- AE was eluted from each C2 cartridge as two fractions and the eluant was also passed through pre-conditioned SCX/SAX cartridges connected in series.
- The C2/SCX/SAX (in series) catridges were first eluted with 30-mL acetonitrile and collected as the first fraction.
- The same C2/SCX/SAX series was then eluted with 10-mL methanol/ethyl acetate/ water (78:20:2, v/v/v) and collected separately as the second fraction.
- The second fraction was taken to dryness under nitrogen and reconstituted in the first fraction (acetonitrile).
- The combined extract was spiked with ~ 15 micrograms internal standard ($C_{13}D_{27}$AE), then 0.2-0.3 g of 2-fluoro-1-methyl pyridinium-*p*-toluenesulfonate was added, followed by the addition of 100 microliters triethylamine. The mixture was stirred and allowed to derivatize for at least 2 hours at ambient temperature.
- The derivatized solution was evaporated to dryness under nitrogen, reconstituted in HPLC mobile phase, and analyzed by positive ion electrospray LC/MS using a Supelcosil TPR-100 column.

The experimental design for AE in water was modified for the downstream interstitial water sample to measure breakthrough (i.e., column overloading) as presented in Figures 3 and 4. The "breakthrough" C2 cartridges were then eluted with the organic solvents (Fractions 1 and 2), and extracted separately and analyzed with the other water extract samples.

Table 1 presents results for AE in interstitial water along with associated QC samples. The water samples were all 4 liters initial volume. The downstream and upstream samples required an extensive amount of time (~16 - 24 hours) to siphon through the C2 cartridge. The downstream sample required 4 sets of C2 (plus backup C2) cartridges to complete the extraction. The upstream sample required three C2 cartridges. The method blank and laboratory control sample (LCS) were extracted using two C2 cartridges each in about 8 hours time.

Data were processed by integration of peaks at the appropriate mass ion, calculating the relative response times and response factors versus the corresponding internal standard peak, and determining sample concentrations from the standard data by linear regression. For samples with low responses, concentrations were calculated from the average response factor from the standard data.

Sample results are reported in µg/L for water samples and µg/g dry weight for sediment samples. Spike recovery determinations were corrected for background or residual concentrations (as appropriate) to obtain the net increase in concnetration.

In cases where chromatographic interferences were present, an objective approach was used to assess the data. Chromatographic peaks that were within ~ 3% relative retention time of standards were included as "hits." Those peaks that were between ~ 4% to ~10% of the standard RRT were considered an interferent peak and the calculated value was reported as a "less than (<)" value. Peaks falling outside the ~10% window or were of poor shape were considered non-detect for the target analyte. Some sample results that were calculated to be less than zero by linear regression were recalculated using an average response factor generated from the standard data.

Calibration data for all analytes exhibited correlation coefficients 0.99 or better, but a significant interference with the C13 EO 2 chemical increased its reporting limit by an about 1 order of magnitude.

The internal standard responses for the upstream, downstream, and downstream breakthrough samples were consistently reduced at about 25% to 30% of the typical response exhibited by the QC samples, standards, and sediment samples. This anomaly may be associated with ion suppression from the field samples since all reagents and sequence of events were the same throughout sample preparation and analysis. If the ion-suppression affect is consistent throughout the chromatogram, calculation of sample results using the internal standard technique should minimize the impact on sample results.

1.2 Analysis of Interstitial Water Samples for LAS and AS/AES

The aqueous sample extraction method used for this study was based on combined information from two papers. The analytical procedure for measuring LAS in water is described in "Use of Isomer Distributions to Characterize the Environmental Fate of LAS," Morrall, et al., The Procter & Gamble Company. The AS/AES procedure is described in the "Determination of Alkyl Sulfates and Alkyl Ethoxysulfates in Wastewater Treatment Plant Influents and Effluents and in River Water Using Liquid Chromatography/Ion Spray Mass Spectrometry," Popenoe, et al., Analytical Chemistry, 1994, Vol. 66, pp. 1620-1629.

These two sample preparation techniques are similar in the use of the C2 SPE cartridge for extraction of surfactant from water and alcohol elution of surfactant from the C2 cartridge. Because of the similarities, MRI performed a simultaneous extraction of the water samples for both LAS and AS/AES using a modified combined procedure.

The extracted samples were analyzed for LAS and AS/AES using one set of LC/MS operating conditions. The LC/MS operating parameters used for this study are based on the MRI Draft Report, dated March 2, 2001 "The Development and Validation of an Analytical Method for the Determination of Alkyl Sulfates and Alkyl Ethoxylate Sulfates in Environmental Sediments Using Liquid Chromatography/Mass Spectrometry," Robaugh, et al. Analysis of LAS extracts using the same general instrumental operating conditions was demonstrated in previous method evaluation work (MRI Project 310220). The modified combined method for the determination of LAS and AS/AES in aqueous samples is presented below.

The referenced AS/AES paper included an optional filtering step (Fisher, fluted, 0.45 micrometer) to remove suspended solids prior to extraction on the C2 cartridge. Removal and analysis of suspended solids from an aqueous sample may need to be addressed separately due to strong sorption characteristics of surfactants. Water samples in this study were not filtered.

LAS & AS/AES Analysis of Aqueous Samples

- Sample volumes of 200-mL were extracted using this method.
- A C2 cartridge was pre-conditioned with 10-mL methanol, 10-mL methanol / 2-propanol (80:20, v/v), and 10-mL Milli-Q® filtered water.
- The aqueous sample was siphoned through the pre-conditioned C2 cartridge.
- AS/AES and LAS are eluted from the column using 10-mL methanol / 2-propanol (80:20, v/v), followed by 5-mL methanol.
- The combined eluates are evaporated to dryness under nitrogen at ambient temperature.
- The residue was reconstituted in 1-mL HPLC mobile phase (acetonitrile/water mixture with 0.3 mM ammonium acetate).
- The samples were spiked with internal standards (d_4-C_{12}-LAS and sodium dodecyl-d_{25} sulfate) and analyzed by negative ion electrospray LC/MS using a C8 Phenomenex Prodigy® column.

Table 2 presents residual concentrations of LAS and AS/AES found for the interstitial water samples along with the reagent blank and reagent spiked sample that were prepared with the field samples. Both the sample results and spiked recovery values were corrected for the small amount of AES found in the reagent blank. Separate spiked reagent quality control (QC) samples were prepared for the AES and LAS at 10 times (10X) the estimated residual concentrations from earlier studies.

The AS/AES concentrations are based on standards prepared from a formulated product (NEODOL® 25-3S, Knavish) with an activity value of 16.2%. The final concentrations were corrected for the activity value.

1.3 Analysis of Sediment Samples for AE

Sediment samples were prepared and analyzed for AE using the method described in the MRI Report "Method Validation / Preservation Study of Alkyl Ethoxylates in Sediment by LC/MS," Final Report, dated August 31, 2001.

Table 3 presents the analysis results for AE in residual sediment samples. The results are reported on a dry-weight basis. Precision values are reported based on triplicate analysis of the upstream and downstream samples. No AE compounds were detected in the method blank (reagent only), except for C13 alcohol which was detected at a relatively low 1.0 ng/g equivalent basis. The sample results were corrected for this minor background concentration.

Matrix spike recovery and precision results are presented in Table 4. These samples were prepared and analyzed with the non-spiked sediment samples. Individual downstream sediment samples were spiked with an AE standard at 10X the estimated background concentration. The recovery of the laboratory control reagent spiked sample is also shown. Recovery values have been corrected for residual background levels found in the non-spiked downstream sample.

1.4 Analysis of Sediment Samples for LAS & AS/AES

Sediment samples were prepared and analyzed for LAS & AS/AES using the method described in the MRI Report "Method Evaluation for the Analysis of LAS in Sediment by LC/MS," dated August 13, 2001.

Table 5 presents the results for LAS/AES in sediment. The upstream and downstream samples were each analyzed in triplicate. Precision values are listed for chemicals that were found by analysis. Portions of the downstream sediment were spiked at 10X the estimated residual concentration from earlier studies. Recovery and precision data for the spiked sediment samples are also included in Table 5.

2. Preservation Study for AE in Sediment

A preservation study was performed to measure the recovery of spiked surfactant in sediment that is preserved with 3% formalin and stored under refrigerated conditions for up to 14 days. This information is intended to support field sample collection activities and establish sample preservation and holding times. The preservation studies were all performed using the downstream sediment sample.

2.1 Study Design

The study design for this preservation test is based on testing one bulk sediment sample for each of the 3 types of surfactants at 0, 7 and 14 days. The sediment was preserved with formaldehyde at the time of collection and stored under refrigerated conditions from time of collection to extraction. The experimental design for this study is presented below:

Preservation Study of Surfactants (AE, AS/AES, LAS) in Sediment

- The sediment was preserved with formaldehyde (3%) at the time of collection and shipped to MRI by overnight courier under refrigeration (e.g., refrigerant packs).
- Upon receipt, the sediment was homogenized by manual mixing. The sediment was moist, but contained no overlay water.
- Three (3) bulk samples were spiked separately at 30 times the estimated residual surfactant concentrations based on earlier studies on samples from the Little Miami River.
- The spiked bulk samples were manually mixed with a spatula and allowed to equilibrate for 1 hour, then duplicate 20-g wet-weight samples were removed.
- These initial samples (designated "Day 0") were freeze-dried and extracted using the procedures referenced in Section 1.3 and 1.4.
- The Day 0 sample extracts were stored at ~ 4-6°C until extraction of the Day 7 and Day 14 sample sets.
- The bulk spiked sediment samples were returned to cold storage (~ 4-6°C) until the next stability time point.
- After 7 and 14 days of bulk sample preparation, the spiked sediments were again homogenized, duplicate samples of ~ 20-g were removed for extraction, and the remaining sediment returned to cold storage.
- All the extracted samples (Day 0, 7 and 14), along with associated reagent blanks and spikes, were processed through SPE clean-up, derivatized (AE only), and analyzed at the same time to minimize any analytical variability.
- The sample extracts were analyzed by LC/MS using the procedures referenced in Sections 1.3 and 1.4.

2.2 AE Stability Study Results

Table 6 presents results of the AE stability study. Stability results are based on the relative response versus Day 0 samples. AE recovery for the Day 0 samples is also presented in the table. The spiked sediment samples were useful in the examination of the non-spiked sediment to help identify target chemical peaks in cases where chromatographic interferences were present.

Table 7 presents the laboratory control spike recoveries for all stability time points. The QC samples were reagent only, spiked at the same 30X concentration as the stability sample, processed through the SPE cartridges, and stored under the same conditions and for the same length of time as the stability samples. Table 8 shows results from the reagent blanks throughout the stability study. The reagent blanks were extracted through SPE cartridges and stored under the same conditions as the test samples.

2.3 AES / LAS Stability Study Results

Table 9 presents results from a 2-week stability study in which separate bulk sediment samples (downstream) were spiked at 30X the background concentrations estimated from previous studies.

The sediment samples were preserved with 3% formalin and were stored at ~ 4 to 6°C throughout the storage time. Day 0 samples were extracted ~ 1 hour after spiking the target chemicals. The AES and LAS stability studies were performed using separate spiked bulk samples.

Chemical responses are calculated relative to the appropriate LAS or AES internal standard, normalized to actual sample weight, and performed in duplicate at each time point. No correction for background or residual concentrations was applied to these results.

Table 10 presents results spiked recovery results from the Day 0 stability study samples that were prepared in duplicate. The spiked sediment recovery results are compared to a laboratory control sample (solvent only) that was spiked at the same concentration and extracted with the stability test samples.

Table 11 presents LCS recoveries for the 14-day stability test on LAS/AES in sediment. The LCS samples are spiked solvent (no sediment) that were processed with each batch of samples at the Day 0, 7 and 14 time points. These QC samples, spiked at the same concentration as the stability samples, were stored with the stability samples and analyzed to demonstrate that there was no significant degradation from extraction to time of analysis. The table combines results from separate LCS samples (one fortified with a LAS standard, the other fortified with an AES standard) for each of the stability time points.

3. Discussion

Based on the results of this range finding study, both the upstream and downstream interstitial water samples from this site exhibited relatively low levels of surfactants. Total AE found in the upstream and downstream interstitial water samples were measured to be 2.7 and 2.1 µg/L, respectively. The downstream "breakthrough" sample was measured to be about 20% of the primary extraction at about 0.4 µg/L total AE. Total AS/AES found in the upstream sample was 4.8 µg/L and 1.0 µg/L in the downstream sample. Total LAS concentrations were 5.9 and 5.4 µg/L for the upstream and downstream samples, respectively.

The AE method is able to measure down to a range of 0.005 - 0.0001 µg/L per component. The lowest calibration standard concentrations ranged from 0.0001 ug/L (C16 EO=1) to 0.06 µg/L (C15 EO=0); the majority of homologs were ~ 0.005 µg/L. In general, the alcohols in each series were the highest concentrations in the standard curve because the formulated products used as reference standards were fortified with additional alcohol standards to run as a single calibration curve (rather than separate calibration curves for the formulated material and alcohols). Sensitivity was affirmed with the lowest concentrated standard exhibiting good response (generally better than 10 times baseline) with good peak shape.

There were significant chromatographic differences between actual environmental water samples and control samples (e.g., standards, QC spikes, etc.)--affecting some AE homologs more than others. Interstitial water sample extracts have additional background peaks and noise. The large volume (4 L) interstitial water samples exhibited ion suppression when analyzed, resulting in only about 25% to 35% of the expected internal standard responses (across all internal standard compounds). The ion suppression appears to be matrix-related because the same internal standard solution was used throughout the study. Also, this anomaly was not evident in other samples (e.g., standards, aqueous QC samples, or sediment sample extracts) and the samples were analyzed as essentially one continuous batch.

There were a few difficulties with the AE interstitial water samples that may impact the results. For example, siphoning large volume samples (4 L) through multiple C2 cartridges is a very slow step that took as much as 24 or more hours to complete. Some possible steps to address the difficulties with the water samples may be going to a larger C2 SPE cartridge to increase flow rate, pre-filtering the sample and extracting the particulate and filter separately (combined results), reducing the sample volume, or re-spiking extracts with derivatized AE standard for confirmation purposes.

Surfactant concentrations in sediment were slightly higher in the downstream sample compared to the upstream sample. Total AE in sediments were 138 ng/g in the upstream sample and 250 ng/g for the downstream sample. Total AS/AES in sediments were 3.6 ng/g for the upstream sample and 10.5 for the downstream sample. Total LAS was 35.2 for the upstream sample compared to 119 ng/g for the downstream sample.

With few exceptions, quality control results were good. Recovery of the higher-carbon, lower-ethoxylated AE chemicals were low (11% to 28%) for the water extractions and also low (6% to 38%) for the some of the higher ethoxylated AE in the sediment extractions. Background levels of laboratory method blanks were low indicating glassware decontamination procedures were effective.

Preservation study results show that surfactant spiked sediments were stable for up to 14 days when stored at cold temperatures (4-6°C) and stabilized with 3% formilin. These data suggest that field samples may be extracted within 14 days of collection if stored under the same conditions.

Respectfully submitted,

Dennis Hooton
Senior Chemist

Approved for:

Midwest Research Institute

Thomas M. Sack, Ph.D.
Director, Chemical Sciences Division

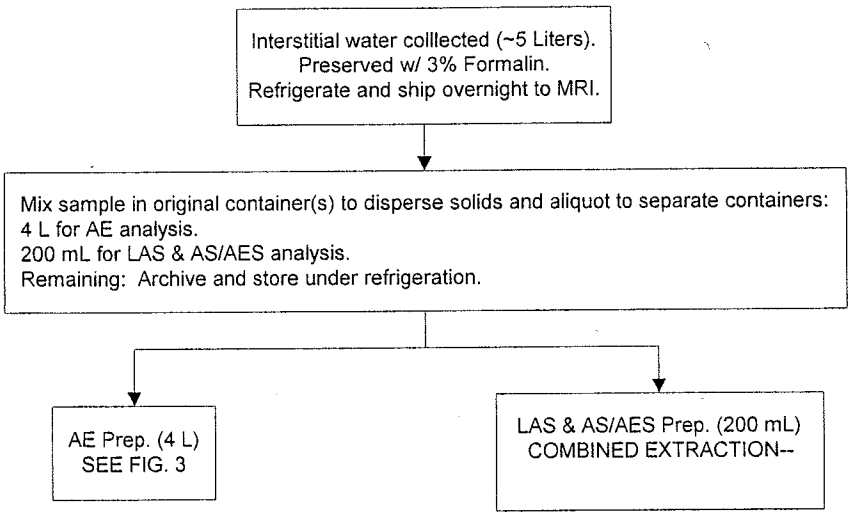


FIGURE 1. INTERSTITIAL WATER PROCESSING

FIGURE 2. SEDIMENT PROCESSING

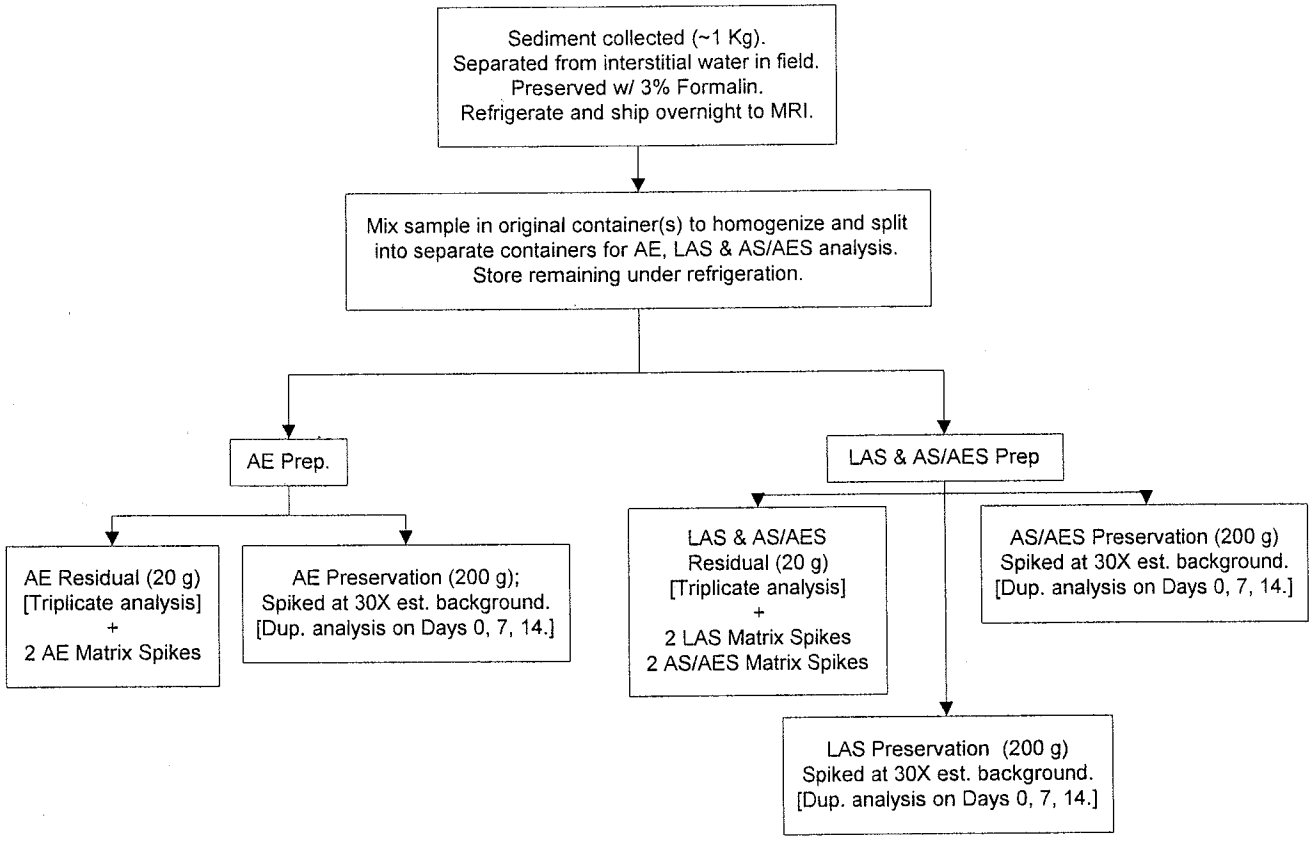
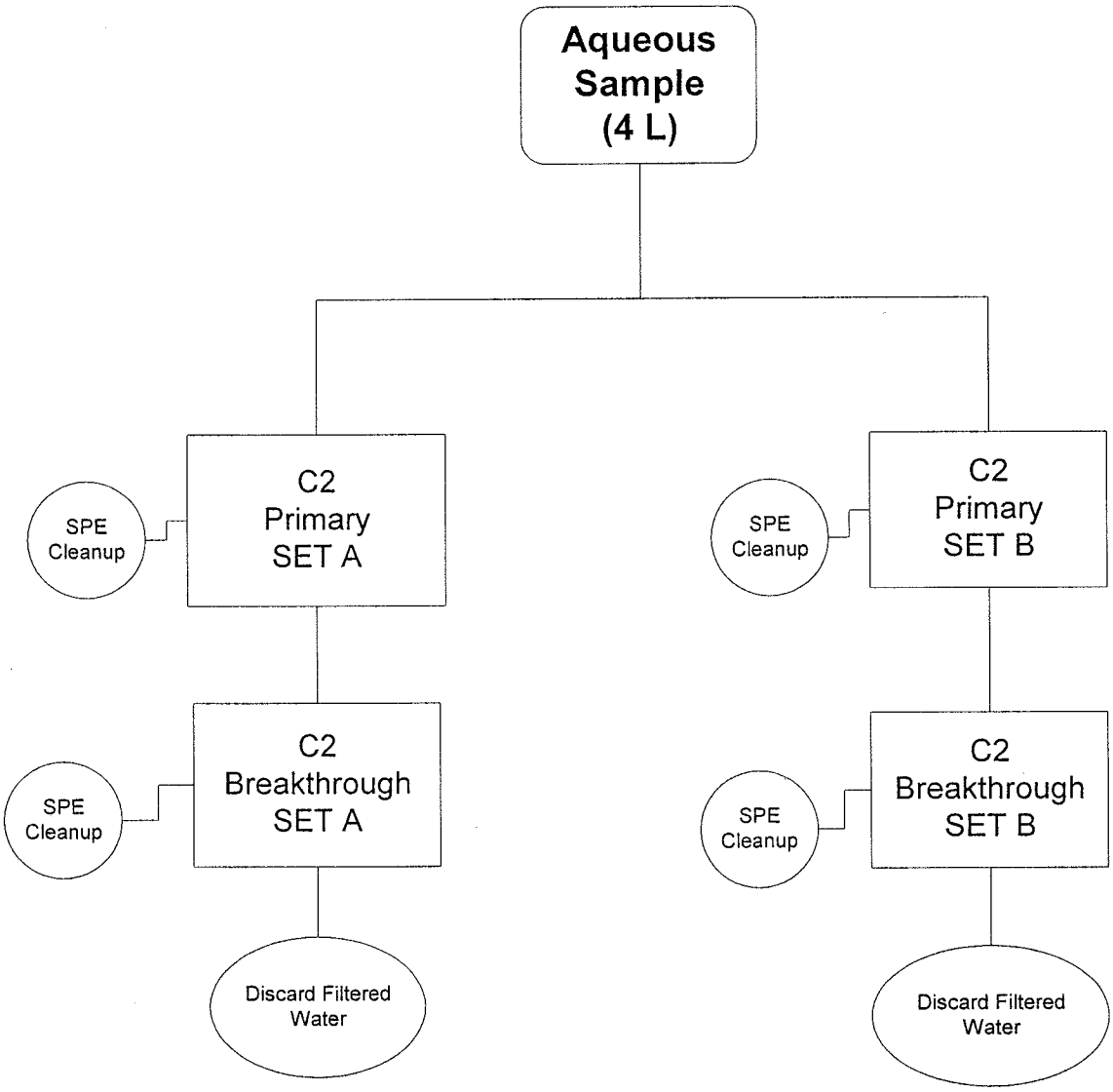


FIGURE 3. EXTRACTION / BREAKTHROUGH DETERMINATION OF AE IN WATER

Note: This "breakthrough" extraction design (i.e., addition of Set B cartridges) was performed on the downstream interstitial water sample.



Extraction of Interstitial Water using C2 Cartridge:

1. Mix the sample aliquot (in secondary container) to disperse any suspended solids.
2. Set up parallel C2 cartridge pairs (primary & breakthrough) as needed.
3. The second C2 cartridge is connected in series for *the downstream sample only for determination of AE breakthrough.*
4. Pour or siphon aqueous sample through pre-conditioned C2 SPE cartridge pairs.
5. Perform quantitative transfer with Milli-Q water of empty sample container and add to *the primary* C2 cartridge.
6. After extraction, dry the C2 cartridges (*single or as pair*) by pulling vacuum for minimum of 8 hrs (overnight).
7. Each C2 cartridge (primary or breakthrough) is eluted separately (see Figure 4).
8. Dry the secondary sample container with nitrogen stream to remove water.

FIGURE 4 SPE ELUTION & CLEAN-UP OF EXT. C2 CARTRIDGE FOR AE

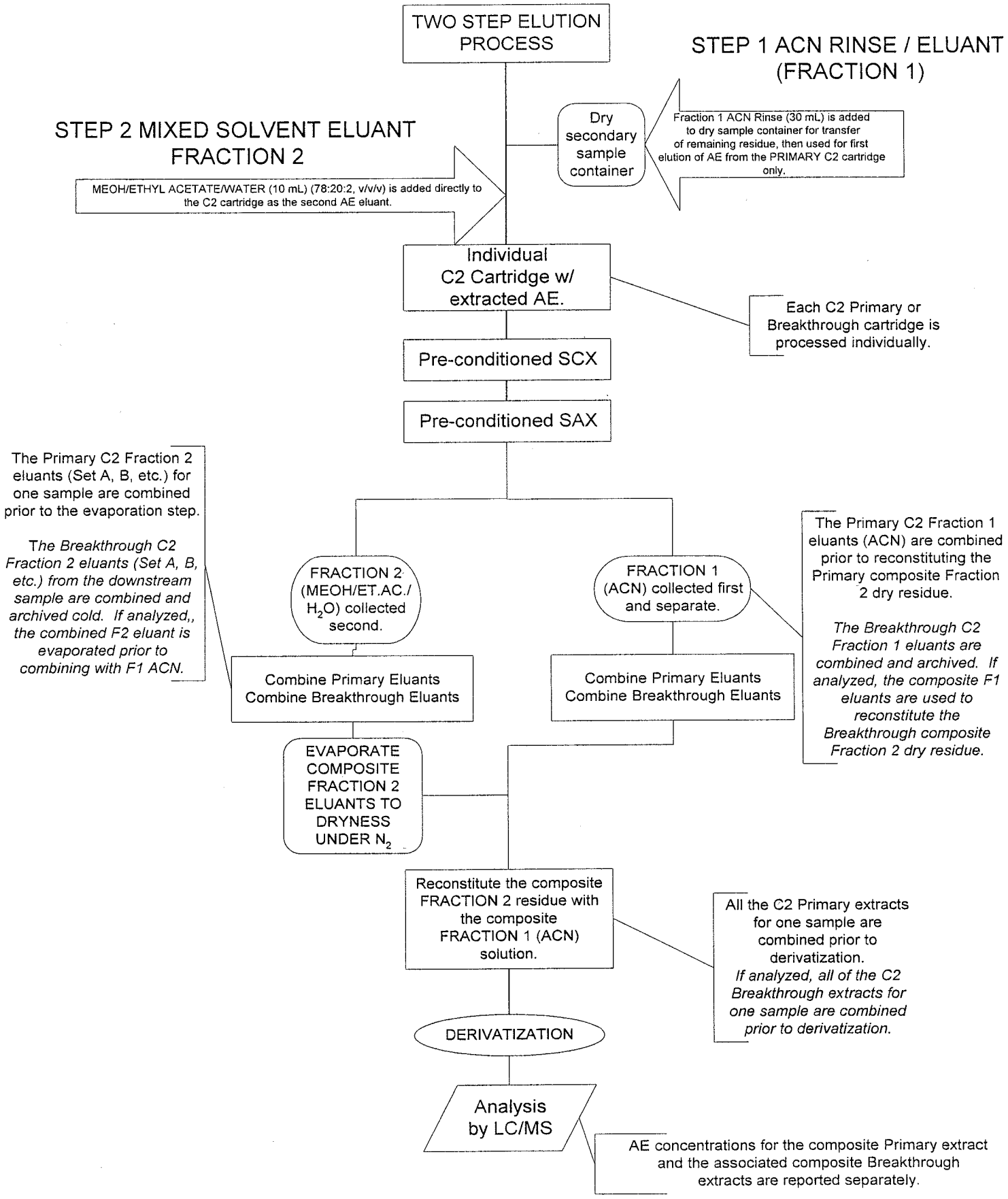


Table 1. AE Results for Interstitial Water Samples

Chain Length	EO Unit	Upstream Sample Conc'n (ng/L)	Downstream Sample Conc'n (ng/L)	Downstream SPE Breakthrough Sample Conc'n (ng/L)	Lab Control Spiked Conc'n (ng/L)	Lab Control Spike Recovery (%)	Reagent Blank Conc'n (ng/L)	Est. Detection Limit (ng/L)
12	0	875 E	530	13.2	81.8	88	17.5	1.4
12	1	3.6	90.3	1.5	9.6	90	1.0	1.0
12	2	INT<343	49.8	46.3	17.1	61	8.4	3.4
12	3	60	21.6	INT <116	25.2	68	6.0	5.0
12	6	102	103	3.6	50.9	74	-	10.2
12	9	11*	7.5 E*	4.0 *	72.4	83	-	14.5
12	12	13*	11 *	12.4	71.4	76	-	7.1
12	15	-	20.1	16.0	46.7	82	-	9.3
13	0	221	36.8	9.3	83.1	46	-	2.8
13	1	3.9	12.8	1.1 *	9.6	52	-	1.0
13	2	<58 E*	<67 E*	INT	INT	INT	-	50.0
13	6	6.7 *	<11 *	-	54.6	69	-	1.8
13	9	7.4	<3.5 *	-	74.3	183	-	2.1
13	15	<20 E*	-	-	47.9	59	-	9.6
14	0	172	113	4.9	283.6	24 *	-	1.1
14	1	<1.7	19.7	-	10.0	26	-	2.0
14	2	109 E	<341	-	15.3	36	-	3.1
14	6	-	-	-	38.2	35	-	1.5
14	9	-	-	-	59.9	128	-	2.4
14	15	-	-	-	38.6	44	-	7.7
15	0	324	65	27 *	580.7	16 *	-	1.9
15	1	19.4	15.7	-	8.9	28	0.6	1.8
15	2	5.6	251	<85.0	10.6	12	-	2.1
15	6	-	-	-	38.2	15	-	1.5
15	9	-	-	-	52.1	98	-	2.1
15	15	-	-	-	33.6	26	-	6.7
16	0	42	119	17.1	385.2	17 *	-	0.8
16	1	2.1	2.3	1.7	1.0	12 *	-	0.2
16	2	10.2	12.1	3.3	2.9	23	-	0.6
16	6	<5.2	1.5	-	16.9	26	-	1.1
16	9	<3.5 *	2.4	-	35.1	114	-	1.8
16	15	-	-	-	40.6	49	-	8.1
18	0	130	165	18.3	478.6	20 *	-	1.0
18	1	0.5	0.30	<0.1 *	2.4	11 *	-	0.2
18	2	60	24	6.7	7.1	120	-	1.4
18	6	4.7 *	1.7 *	-	39.6	33	-	1.1
18	9	9.9 *	10.6 *	-	82.7	115	-	1.8
18	15	<69	15.8 *	-	95.4	37	-	9.5
TOTALS		2693	2125	438	3052		34	

" * " = Calculated using average relative response factor.
" < " = Conservative value reported due to relative retention time difference (>5%) or co-elution with interference peak.
E = Estimated value from extrapolated standard data.
"-" = No peak detected at expected retention time.
INT = Chromatographic interference.

Note: Ion suppression was indicated for the interstitial water samples, reducing the internal standard responses to ~ 30% relative to those exhibited in standards, QC samples, and sediment samples.

Table 2. LAS / AES Results for Interstitial Water Samples

			UPSTREAM Concentration (µg/L)	DOWNSTREAM Concentration (µg/L)	Reagent Blank (µg/L)	Spiked Concentration Control Sample (µg/L)	Reagent Spike Recovery (%)	Estimated Detection Limit (ug/L)
AES	C12	EO 0	2.54	0.46	0.04 *	1.23	94	0.16
AES	C12	EO 2	1.12	0.26 *	-	0.71	127	1.42
AES	C12	EO 4	0.15	0.10 *	-	0.27	136	0.22
AES	C12	EO 8	-	-	-	0.04	180	0.03
AES	C13	EO 0	0.324 *	0.03 *	-	1.32	166	0.26
AES	C13	EO 2	0.031	-	-	0.53	146	0.21
AES	C13	EO 4	0.075 *	-	-	0.52	117	0.42
AES	C13	EO 8	<0.039 *	-	-	0.03	117	0.01
AES	C14	EO 0	0.324	0.117 *	-	0.94	140	0.38
AES	C14	EO 2	0.032	0.045 *	-	0.38	135	0.15
AES	C14	EO 4	0.024 *	0.028 *	-	0.20	123	0.08
AES	C14	EO 8	0.012 *	-	-	0.05	144	0.04
AES	C15	EO 0	0.107 *	-	-	0.77	140	0.15
AES	C15	EO 2	0.013 *	-	-	0.31	134	0.12
AES	C15	EO 4	0.005 *	-	-	0.12	148	0.05
AES	C15	EO 8	-	-	-	0.06	153	0.04
		TOTALS	4.80	1.04	0.04	7.48		3.74
LAS	C10	NA	1.1 *	0.55 *	0.36 *	38.2	111	2.2
LAS	C11	NA	2.5 *	1.9 *	-	100.6	128	1.9
LAS	C12	NA	1.8 *	2.3	0.59 *	100.1	111	1.9
LAS	C13	NA	0.46 *	0.66 *	0.20 *	16.3	94	2.2
LAS	C14	NA	-	-	-	16.3	94	1.3
		TOTALS	5.9	5.4	1.2	272		9.5

" * " = Calculated using average relative response factor.

" < " = Conservative value reported due to relative retention time difference (>5%) or co-elution with interference peak.

" - " = No peak detected at expected retention time.

NA = Not applicable.

Table 3. AE Results for Sediment Samples

Chain Length	EO Units	Average Concentration UPSTREAM (ng/g, dry weight)		Precision UPSTREAM (s, in ng/g)	Average Concentration DOWNSTREAM (ng/g, dry weight)		Precision DOWNSTREAM (s, in ng/g)	Estimated Detection Limit (ng/g)
12	0	4.6	±	2.8	9.3	±	2.2	0.4
12	1	0.6	±	0.3	2.4	±	1.3	0.3
12	2	-	±	2.1	3.3	±	5.7	0.9
12	3	INT <10.3	±	2.5	8.3	±	1.0	1.3
12	6	13.7	±	4.2	13.4	±	2.7	2.7
12	9	1.1 *	±	1.0	1.6	±	0.8	3.9
12	12	1.7	±	0.7	0.8	±	0.7	1.9
12	15	5.7	±	1.0	3.8	±	3.3	2.5
13	0	1.6	±	0.7	5.6	±	3.3	0.7
13	1	-	±	-	-	±	-	0.3
13	2	INT	±	-	-	±	-	13.5
13	6	0.2	±	0.4	1.2	±	2.0	0.5
13	9	-	±	-	-	±	-	0.6
13	15	-	±	-	-	±	-	2.6
14	0	2.7	±	0.4	8.2	±	7.1	0.3
14	1	-	±	-	4.7	±	4.1	0.5
14	2	-	±	-	6.6	±	11.5	0.8
14	6	-	±	-	-	±	-	0.4
14	9	-	±	-	-	±	-	0.6
14	15	-	±	-	-	±	-	2.1
15	0	0.3	±	0.5	51.0	±	46.5	0.5
15	1	-	±	-	0.8	±	1.3	0.5
15	2	12.6	±	21.2	32.3	±	7.5	0.6
15	6	-	±	-	-	±	-	0.4
15	9	-	±	-	-	±	-	0.6
15	15	-	±	-	-	±	-	1.8
16	0	10.4	±	18.0	29.1	±	4.9	0.2
16	1	-	±	-	-	±	0.1	0.1
16	2	0.6	±	0.7	3.0	±	1.3	0.2
16	6	-	±	-	1.4	±	2.4	0.3
16	9	-	±	-	3.5	±	13.5	0.5
16	15	-	±	-	1.8	±	3.1	2.2
18	0	63	±	58	47.7	±	82.7	0.3
18	1	-	±	-	-	±	-	0.1
18	2	7.6	±	13.1	8.4	±	14.5	0.4
18	6	1.2	±	2.0	1.4	±	2.4	0.3
18	9	-	±	-	-	±	-	0.5
18	15	-	±	-	-	±	-	2.5
TOTALS		138			250			49

s = standard deviation

" * " = Calculated using average relative response factor.

" < " = Conservative value reported due to relative retention time difference (>5%) or co-elution with interference peak.

- = No peak detected at expected retention time.

INT = Chromatographic interference.

Table 4. AE Results for Spiked Sediment

Chain Length	EO Units	Reagent Spike (ng/g) [10X nominal spike level]	Reagent Spike Recovery (%)	Sediment Spike Avg. Recovery (%)		Sediment Spike Precision (s)
12	0	16.4	85	88	±	7
12	1	1.9	93	122	±	50
12	2	3.4	74	114	±	110
12	3	5.0	102	82	±	6
12	6	10.2	115	112	±	10
12	9	14.5	99	48	±	0
12	12	14.3	105	38	±	1
12	15	9.3	108	28 *	±	4
13	0	16.6	86	91	±	2
13	1	1.9	48	27	±	1
13	2	3.3	INT	-	±	-
13	6	10.9	128	81	±	6
13	9	14.9	112	75	±	2
13	15	9.6	103	20 *	±	3
14	0	56.7	99	90	±	0
14	1	2.0	88	INT	±	-
14	2	3.1	77	70	±	5
14	6	7.6	106	98	±	3
14	9	12.0	291	225	±	5
14	15	7.7	85	24	±	1
15	0	116.1	97	84	±	7
15	1	1.8	95	59	±	9
15	2	2.1	101	INT	±	-
15	6	7.6	122	116	±	7
15	9	10.4	110	102	±	2
15	15	6.7	101	30 *	±	-
16	0	77.0	91	84	±	2
16	1	0.2	86	69	±	9
16	2	0.6	100	150	±	1
16	6	3.4	117	23	±	8
16	9	7.0	105	6	±	6
16	15	8.1	92	22	±	8
18	0	95.7	82	66	±	2
18	1	0.5	50	44	±	3
18	2	1.4	500 (a)	415 (a)	±	26
18	6	7.9	133	80	±	3
18	9	16.5	106	93	±	1
18	15	19.1	113	14	±	4

s = standard deviation

(a) Data verified. No obvious reason for high recovery.

(b) " * " = Calculated using average relative response factor.

"-" = Value not calculated.

INT = Chromatographic interference.

Table 5. LAS/AES Results for Sediment Samples

			Average UPSTREAM Co-entration (ng/g, dry wt.)	Average DOWNSTREAM Co-entration (ng/g, dry wt.)	Reagent Blank (ng/g)	Estimated Detection Limit (ng/g)	Spiked Sediment Co-entration at 10X (ng/g)	Spike Recovery (%)	Spiked Sediment Duplicate Recovery (%)	Spiked Sediment Avg. Recovery (%)	Spiked Sediment Precision (s, %)
AES	C12	EO 0	3.6 ±0.6 (s)	4.5±0.2 (s)	-	2.2	12.34	102	113	107	8
AES	C12	EO 2	-	-	-	18.9	7.08	131	156	143	18
AES	C12	EO 4	-	-	-	2.9	2.71	103	88	96	11
AES	C12	EO 8	-	-	-	0.4	0.41	53	32	42	15
AES	C13	EO 0	-	0.96±0.13 (s) *	-	3.5	13.20	84	91	88	5
AES	C13	EO 2	-	-	-	2.8	5.35	166	182	174	12
AES	C13	EO 4	-	-	-	5.5	5.20	117	118	118	1
AES	C13	EO 8	-	-	-	0.2	0.31	28	36	32	6
AES	C14	EO 0	-	3.1±0.4 (s) *	-	5.0	9.44	23	21	22	2
AES	C14	EO 2	-	-	-	2.0	3.76	117	117	117	0
AES	C14	EO 4	-	-	-	1.0	1.96	94	111	103	12
AES	C14	EO 8	-	-	-	0.5	0.52	75	88	81	9
AES	C15	EO 0	-	1.9±0.2 (s)*	-	2.0	7.68	31	25	28	4
AES	C15	EO 2	-	-	-	1.6	3.09	82	85	83	2
AES	C15	EO 4	-	-	-	0.6	1.22	124	133	128	7
AES	C15	EO 8	-	-	-	0.6	0.57	77	94	86	12
		TOTALS	3.6	10.5		50	75				
LAS	C10	NA	-	-	-	29.1	382	148	109	128	28
LAS	C11	NA	-	30 * (a)	-	25.5	1006	162	130	146	22
LAS	C12	NA	3.3±3 (s)	33±7 (s)	-	25.4	1001	133	102	118	22
LAS	C13	NA	31.9±0.6 (s)	56±3 (s)	-	29.0	163	105	83	94	15
LAS	C14	NA	-	-	-	17.6	33	138	109	124	20
		TOTALS	35.2	119		127	2585				

(a) Only 1 of 3 triplicate analyses showed presence of this homologue at 30 ng/g, dry weight.

" * " = Calculated using average relative response factor.

s = standard deviation

- = No peak detected at expected retention time.

Table 6. Results of AE Sediment Stablity Study.

Chain Length	EO Units	Nominal Spiked Co-'n (ng/g) [30X Spike Level]	Average Recovery Day 0 Samples (%)		Precision Day 0 Recovery (s)	Day 7 Average Recovery vs. Day 0 (%)	Day 14 Average Recovery vs. Day 0 (%)
12	0	48.5	41	±	6	141	135
12	1	5.7	77	±	6	125	106
12	2	10.1	53	±	2	127	90
12	3	14.9	15	±	4	221	151
12	6	30.2	49	±	10	124	81
12	9	43	50	±	6	123	89
12	12	42.4	30	±	0	101	87
12	15	27.7	12 *	±	2	92	112
13	0	49.3	61	±	11	115	118
13	1	5.7	39	±	3	112	88
13	2	9.8	INT	±	-	INT	-
13	6	32.4	57	±	13	129	97
13	9	44	52	±	7	129	113
13	15	28.4	4	±	0	94	142
14	0	168	56	±	5	120	103
14	1	5.9	23	±	9	192	19
14	2	9.1	72	±	22	117	INT
14	6	22.7	73	±	3	122	97
14	9	35.5	62	±	5	107	110
14	15	22.9	9	±	2	148	173
15	0	344	58	±	2	120	113
15	1	5.3	61	±	13	111	125
15	2	6.3	INT	±	-	INT	INT
15	6	22.7	69	±	16	132	113
15	9	30.9	67	±	6	124	106
15	15	19.9	12	±	1	106	121
16	0	229	55	±	3	117	98
16	1	0.6	49	±	14	109	95
16	2	1.7	INT	±	-	INT	INT
16	6	10	30	±	7	153	INT
16	9	20.8	23	±	16	183	82
16	15	24.1	18	±	1	72	13 *
18	0	284	58	±	3	119	96
18	1	1.4	46	±	8	100	85
18	2	4.2	166	±	92	108	131
18	6	23.5	59	±	11	116	111
18	9	49	64	±	11	111	107
18	15	56.6	19	±	1	110	131

s = standard deviation
" * " = Calculated using average relative response factor.
- = Value not calculated.
INT = Chromatographic interference.

Table 7. AE Laboratory Control Spikes Results from Stability Study

Chain Length	EO Units	Nominal Spiked Conc'n (ng/g) [Equivalent to 30X background)	Day 0 Recovery (%)	Day 7 Recovery (%)	Day 14 Recovery (%)	Average Recovery (%)		Precision (s, %)
12	0	49.1	82	83	93	86	±	6
12	1	5.8	101	108	101	103	±	4
12	2	10.2	71	76	93	80	±	11
12	3	15.1	10	40	38	29	±	17
12	6	30.6	70	74	71	72	±	2
12	9	43.5	93	86	86	89	±	4
12	12	42.8	96	90	97	94	±	4
12	15	28	92	84	92	89	±	4
13	0	49.8	78	82	87	82	±	5
13	1	5.8	84	89	55	76	±	18
13	2	9.9	74	81	69	74	±	6
13	6	32.7	100	103	102	102	±	2
13	9	44.6	100	100	106	102	±	3
13	15	28.7	90	93	94	92	±	2
14	0	170	86	84	90	87	±	3
14	1	6	77	98	83	86	±	11
14	2	9.2	67	96	86	83	±	15
14	6	22.9	103	114	109	109	±	5
14	9	35.9	98	103	104	102	±	3
14	15	23.2	87	99	102	96	±	8
15	0	348	86	89	89	88	±	1
15	1	5.3	91	107	96	98	±	8
15	2	6.3	90	107	104	100	±	9
15	6	22.9	109	112	117	113	±	4
15	9	31.3	95	95	103	98	±	5
15	15	20.2	92	99	99	97	±	4
16	0	231	86	78	86	83	±	5
16	1	0.6	88	75	73	79	±	8
16	2	1.8	77	79	79	78	±	1
16	6	10.1	91	101	101	98	±	5
16	9	21.1	81	92	98	90	±	9
16	15	24.4	79	96	98	91	±	10
18	0	287	83	83	83	83	±	0
18	1	1.4	58	61	50	56	±	6
18	2	4.2	423 (a)	448	427	433	±	14
18	6	23.8	102	108	99	103	±	4
18	9	49.6	85	91	89	88	±	3
18	15	57.2	91	106	106	101	±	9

(a) Data verified. No obvious reason for high recovery.
s = standard deviation

Note: Laboratory Control Spikes are spiked reagents only (no sediment) taken through extraction procedure.

Table 8. AE Reagent Blank Results from Stability Study

Chain Length	EO Units	Day 0 Reagent Blank (ng/g, equiv.)	Day 7 Reagent Blank (ng/g, equiv.)	Day 14 Reagent Blank (ng/g, equiv.)	Average Concentration for Reagent Blanks (ng/g, equiv.)	Estimated Detection Limit (ng/g)
12	0	2.2	-	-	<0.8	0.4
12	1	-	-	-	-	0.3
12	2	-	<1	-	<0.3	0.9
12	3	-	<21	<14	<11.6	1.3
12	6	-	<18	<16	<11.6	2.7
12	9	-	<2	<2	<1.3	3.9
12	12	-	<2	<2	<1.3	1.9
12	15	-	<4	<5	<3.1	2.5
13	0	1.0	1.0	1.0	1.0	0.7
13	1	-	-	-	-	0.3
13	2	INT	INT	INT	INT	13.5
13	6	<1.6	<2	<1	<1.3	0.5
13	9	-	-	-	-	0.6
13	15	-	-	-	-	2.6
14	0	0.34	0.38	0.49	0.40	0.3
14	1	-	-	-	-	0.5
14	2	-	-	<4	<1.4	0.8
14	6	-	-	-	-	0.4
14	9	-	-	-	-	0.6
14	15	-	-	-	-	2.1
15	0	-	-	-	-	0.5
15	1	-	-	-	-	0.5
15	2	-	-	-	-	0.6
15	6	-	-	-	-	0.4
15	9	-	-	-	-	0.6
15	15	-	-	-	-	1.8
16	0	-	-	-	-	0.2
16	1	-	-	-	-	0.1
16	2	-	-	-	-	0.2
16	6	-	-	-	-	0.3
16	9	-	-	-	-	0.5
16	15	-	-	-	-	2.2
18	0	-	-	-	-	0.3
18	1	-	-	-	-	0.1
18	2	-	-	-	-	0.4
18	6	-	-	-	-	0.3
18	9	-	-	-	-	0.5
18	15	-	-	-	-	2.5

" * " = Calculated using average relative response factor.
" < " = Conservative value reported due to relative retention time difference (>5%) or co-elution with interfere-e peak.
"-" = No peak detected at expected retention time.
INT = Chromatographic interference.

Table 9. LAS/AES Sediment Stability Results

			Day 0 Average Response	Day 0 Precision (RSD)	Day 7 Average Response	Day 7 Response vs. Day 0	Day 14 Average Response	Day 14 Response vs. Day 0
AES	C12	EO 0	0.0471	22	0.0428	91	0.0311	66
AES	C12	EO 2	0.0201	5	0.0169	84	0.0175	87
AES	C12	EO 4	0.0101	21	0.0083	82	0.0086	85
AES	C12	EO 8	0.0018	35	0.0020	113	0.0015	85
AES	C13	EO 0	0.0559	11	0.0427	76	0.0489	87
AES	C13	EO 2	0.0329	22	0.0259	79	0.0259	79
AES	C13	EO 4	0.0130	21	0.0101	77	0.0107	83
AES	C13	EO 8	0.0019	17	0.0017	90	0.0018	95
AES	C14	EO 0	0.0399	6	0.0394	99	0.0296	74
AES	C14	EO 2	0.0212	7	0.0184	87	0.0165	78
AES	C14	EO 4	0.0086	13	0.0083	97	0.0071	84
AES	C14	EO 8	0.0013	31	0.0012	93	0.0011	80
AES	C15	EO 0	0.0315	10	0.0245	78	0.0207	66
AES	C15	EO 2	0.0148	12	0.0135	91	0.0113	76
AES	C15	EO 4	0.0080	18	0.0065	82	0.0058	73
AES	C15	EO 8	0.0013	3	0.0009	71	0.0008	64
LAS	C10	NA	0.423	2	0.517	122	0.5809	137
LAS	C11	NA	1.14	7	1.31	115	1.3334	117
LAS	C12	NA	1.06	4	1.13	106	1.1310	107
LAS	C13	NA	0.151	10	0.162	107	0.1651	109
LAS	C14	NA	0.018	7	0.0237	132	0.0482	267

RSD = Relative Standard Deviation of average response.

Note: Downstream sediment, spiked at 30X estimated background, used for stability study.

Table 10. LAS/AES Spiked Sediment Recovery Results from Stability Study

			Theoretical Co-'n @ 30X Spike Level (ng/g, dry wt.)	Spiked Matrix Sample A Recovery (%)	Spiked Matrix Sample B (duplicate) Recovery (%)	Average Recovery @ 30X Spike Level (%)		Precision (RSD)	LCS Recovery (%)
AES	C12	EO 0	47..6	66	51	58	±	19	65
AES	C12	EO 2	27.3	78	74	76	±	4	105
AES	C12	EO 4	10.4	100	63	82	±	32	123
AES	C12	EO 8	1.54	90	38	64	±	57	121
AES	C13	EO 0	50.8	97	78	88	±	16	93
AES	C13	EO 2	20.6	127	93	110	±	22	118
AES	C13	EO 4	20.0	110	72	91	±	30	98
AES	C13	EO 8	1.20	67	39	53	±	38	119
AES	C14	EO 0	36.4	25	60	42	±	58	96
AES	C14	EO 2	14.5	88	79	83	±	7	103
AES	C14	EO 4	7.55	81	61	71	±	20	114
AES	C14	EO 8	1.98	84	41	62	±	49	119
AES	C15	EO 0	29.6	62	51	56	±	14	108
AES	C15	EO 2	11.9	66	53	60	±	16	109
AES	C15	EO 4	4.67	100	73	86	±	22	115
AES	C15	EO 8	2.15	78	72	75	±	6	152
LAS	C10	NA	1500	70	68	69	±	2	102
LAS	C11	NA	3940	77	84	80	±	7	108
LAS	C12	NA	3930	71	67	69	±	3	97
LAS	C13	NA	638	60	53	57	±	8	95
LAS	C14	NA	130	52	52	52	±	1	93

LCS = Laboratory control sample. Spiked solvent only--no sediment.

Note: Downstream sediment used for spiking experiment.

Table 11. LAS/AES Spiked Reagent Sample Results from Stability Study

			Spiked Co-'n (ng/g, equiv.)	Day 0 Recovery (%)	Day 7 Recovery (%)	Day 14 Recovery (%)	Average Recovery (%)		Precision (s, as %)
AES	C12	EO 0	12.34	65	72	70	69	±	4
AES	C12	EO 2	7.08	105	103	85	97	±	11
AES	C12	EO 4	2.71	123	117	126	122	±	5
AES	C12	EO 8	0.41	121	119	116	118	±	3
AES	C13	EO 0	13.20	93	119	109	107	±	13
AES	C13	EO 2	5.35	117	133	123	124	±	8
AES	C13	EO 4	5.20	98	126	124	116	±	16
AES	C13	EO 8	0.31	119	112	106	112	±	7
AES	C14	EO 0	9.44	96	121	87	101	±	17
AES	C14	EO 2	3.76	103	126	122	117	±	13
AES	C14	EO 4	1.96	114	133	117	121	±	10
AES	C14	EO 8	0.52	119	140	83	114	±	29
AES	C15	EO 0	7.68	108	122	100	110	±	11
AES	C15	EO 2	3.09	108	113	123	115	±	7
AES	C15	EO 4	1.22	115	137	139	131	±	13
AES	C15	EO 8	0.57	152	171	132	152	±	20
LAS	C10	NA	382.	102	90	101	98	±	7
LAS	C11	NA	1010	108	104	114	109	±	5
LAS	C12	NA	1001	96	92	105	98	±	7
LAS	C13	NA	163	94	100	102	99	±	4
LAS	C14	NA	33.1	93	101	105	100	±	6

LCS = Laboratory control sample. Spiked solvent only--no sediment.