Improvements to Environmental Sample Cleanup for the Analysis of Dioxins and Dibenzofurans

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1. Introduction

Polychlorinated dibenzo-p-dioxins and dibenzofurans (PCDD/PCDF) in environmental samples have become the focus of great concern due to their extremely high toxicity [(1)]. The need to isolate PCDD/PCDF from a myriad of other organic contaminants in environmental samples has led to a lengthy and time-consuming cleanup process involving multiple gravity flow cleanup columns, after which analysis by gas chromatography/high resolution mass spectrometry (GC/HRMS) is necessary to obtain part-per-trillion detection limits. In our laboratory, cleanup columns used to prepare environmental sample extracts for PCDD/PCDF analysis include an acid/base silica column, a basic alumina column[(2)], and a carbon/celite column[(3)] as generally described in EPA Method 8290[(4)]. A preliminary evaluation of new materials for use in these cleanup columns was conducted in our laboratory. These materials included: 1) a new basic alumina to replace a basic alumina no longer manufactured; and 2) prepacked silica and carbon columns to eliminate the time required to prepare these columns in our laboratory.

2. Approach/Methodology

New Alumina

Our laboratory was routinely using BioRad AG-10 basic alumina for PCDD/PCDF sample extract cleanup. This material became unavailable from chemical suppliers. The new alumina considered to replace the BioRad basic alumina was Sigma WB-2 basic alumina. Key characteristics of these two aluminas are:

Characteristic	Bio Rad AG-10	Sigma WB-2
Activity Grade	I	I
Particle Size	45-75 um	100 um
Pore Size	30 angstrom	58 angstrom
Mesh	100-200	150

Prior to use in this evaluation, both types of alumina were heated to 300° C under purified nitrogen purge for 1 hour, cooled and rinsed with 350 mL of methylene chloride, dried under nitrogen purge at 50° C for 20 minutes, heated under nitrogen purge to 600° C for 24

hours, cooled, and stored at 130 °C. Alumina columns were made by cutting the tops from 10-mL serological pipettes, plugging with glass wool, adding 5 g of alumina, and topping with about 1/2 in. sodium sulfate.

To demonstrate the Sigma WB-2 alumina for PCDD/PCDF extract cleanup, four replicate aliquots of a single sediment sample were prepared and analyzed. All phases of sample preparation and analysis were identical for these four samples except for the alumina column cleanup. For this demonstration, one sample extract was processed through an alumina column made with the BioRad AG-10 basic alumina prepared as described above; two sample extracts were processed through alumina columns made with the Sigma WB-2 basic alumina prepared as described above; and one sample extract was processed through an alumina column made with the Sigma WB-2 basic alumina prepared only by baking at 130 °C prior to use. After application of the sample extracts to the alumina columns, all columns were rinsed with 15 mL of 3% methylene chloride/hexane which was collected and stored, then 40 mL of 50% methylene chloride/hexane which was further processed for analysis.

Prepacked Silica and Carbon Columns

Prepacked acid/base silica and carbon/celite columns from a commercial supplier were compared to acid/base silica and carbon/celite columns made in our laboratory for interference removal and recovery efficiency. For this comparison, duplicate soil samples were prepared for analysis according to general Method 8290 procedures. One sample was processed through the prepacked acid/base silica and carbon/celite columns and the second sample was processed through acid/base silica and carbon/celite columns prepared in our laboratory. All other aspects of the sample preparation and analysis, including extraction, alumina column cleanup, and GC/HRMS quantification were identical for the two samples.

The prepacked acid/base silica columns were made from Supelcosil A (Supelco, 60 angstrom, 75-150 micron) in 22 mm O.D. x 200 mm glass tubes. The prepacked 8% carbon/celite columns were prepared according to Method 8290, including the addition of a celite 545 plug at the top and bottom of the carbon/celite packed bed. The carbon used to make the prepacked carbon/celite columns was an experimental variety (non-porous, 90 m^2/g surface area). This new carbon was being tested by the manufacturer to determine its suitability as a replacement for Anderson Developing Co. AX-21 carbon which is specified in Method 8290.

Acid/base silica columns were prepared in our laboratory as follows. ICN silica gel (60 angstroms, 100-200 mesh) was activated by washing with 350 mL methanol and 350 mL methylene chloride, drying at 50° C for 20 minutes, baking at 150° C for 90 minutes, cooling under nitrogen to room temperature, extracting in Soxhlet apparatus overnight with methylene chloride, drying at 50° C for 20 minutes, then baking at 150° C for 90 minutes. Acid/base silica columns were prepared by cutting the tops from 25-mL serological pipettes and packing with: 1 g of silica, 2 g of basic silica (35 g silica + 17 mL 20% KOH), 1 g of silica, 4 g of acid silica gel (35 g silica + 15 mL concentrated H_2SO_4), and 1/2 of sodium sulfate. After application of the sample extract to the top of the column, the columns were eluted with 75 mL of hexane.

Carbon/celite columns were prepared in our laboratory as follows. An 8% mixture of AX-21 carbon (Anderson Developing Co.)/celite 545 (Supelco) was prepared, mixed well in a glass jar, and activated overnight in a 130° C oven. The carbon/celite columns were prepared by cutting both ends from 10-mL serological pipettes, fire-polishing both ends, plugging one end with glass wool, adding 1 g of the carbon/celite mixture, and plugging with glass wool. After applying the samples and rinsing the columns with a series of increasing polarity solvents, the columns were inverted and eluted with 30 mL of toluene.

3. Results and Conclusions

These tests were of a limited nature and do not represent a statistically designed study. The results obtained from these tests provide only an indication of each of these materials' utility for cleanup of certain environmental samples for PCDD/PCDF analyses.

New Alumina

The concentration of PCDD/PCDF in the four samples processed in the alumina evaluation are shown in Table 1. Recoveries for $^{13}C_{12}$-labelled PCDD/PCDF internal standards were above 50 percent for all four samples which validates the data quality. Results for the three samples processed through the Sigma basic alumina columns are comparable to results obtained for the one sample processed through a BioRad basic alumina column. Based on these results, our laboratory has switched from BioRad alumina to Sigma alumina for basic alumina columns used in PCDD/PCDF sample extract cleanup.

Prepacked Silica and Carbon Columns

The results of the comparison of commercially-supplied prepacked acid/base and carbon columns with laboratory-prepared columns are shown in Table 2. The laboratory method blank processed with these two samples showed no positive response for PCDD/PCDF analytes. The PCDD/PCDF concentrations for the sample processed through laboratory-prepared columns (Sample 1) compare well with the concentrations for the sample processed through prepacked columns (Sample 2) for some isomers (i.e. 1,2,3,6,7,8-HxCDD). However, for the majority of the PCDD/PCDF isomers, the concentrations in Sample 1 and 2 differ. Loss of PCDD/PCDF analytes was not a problem since internal standard recoveries for both samples were very good, ranging from 72% to 131% for Sample 1 and from 62% to 109% for Sample 2. Instead, the poor chromatography obtained in the analysis of Sample 2 suggests that the prepacked columns did not isolate the 2,3,7,8-substituted PCDD/PCDF isomers from other interfering organic contaminants to allow meaningful quantitation.

According to the supplier, the prepacked column most likely to exhibit this problem would be the carbon/celite, since the carbon used in the prepacked column was very different from AX-21 carbon. Most noteably, AX-21 carbon is very finely ground and disperses very thoroughly in the celite. The experimental carbon used in the prepacked column is much larger and non-porous. Further work on the prepacked columns will focus on testing the acid/base silica column independently from the carbon column.

4. References

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(4) USEPA (1987); Method 8290. Analytical Procedures and Quality Assurance for Multimedia Analysis of Polychlorinated Dibenzo-p-dioxins and Polychlorinated Dibenzofurans by High Resolution Gas Chromatography/High Resolution Mass Spectrometry. Environmental Monitoring Systems Laboratory, Las Vegas, Nevada, June 1987.

Table 1. Alumina Test Data

Analyte	Concentration Found (ng/Kg)					
	Sample 1[a]	Sample 2[b]	Sample 3[b]	Sample 4[c]	Average (1-4)	Std Dev
2,3,7,8-TCDD	*	*	*	*	*	*
1,2,3,7,8-PeCDD	*	*	*	*	*	*
1,2,3,4,7,8-HxCDD	*	*	*	*	*	*
1,2,3,6,7,8-HxCDD	*	0.16	0.51	*	*	*
1,2,3,7,8,9-HxCDD	*	*	*	*	*	*
1,2,3,4,6,7,8-HpCDD	2.16	2.05	3.01	2.54	2.53	0.43
OCDD	22.04	19.12	22.12	23.27	21.50	1.77
2,3,7,8-TCDF	0.76	0.66	0.89	0.78	0.78	0.09
1,2,3,7,8-PeCDF	*	*	*	*	*	*
2,3,4,7,8-PeCDF	*	0.14	*	*	*	*
1,2,3,4,7,8-HxCDF	0.75	0.55	0.69	*	*	*
1,2,3,6,7,8-HxCDF	*	*	*	*	*	*
1,2,3,7,8,9-HxCDF	*	*	*	*	*	*
2,3,4,6,7,8-HxCDF	0.43	0.37	0.48	0.44	0.43	0.05
1,2,3,4,6,7,8-HpCDF	1.06	0.93	1.32	1.27	1.17	0.18
1,2,3,4,7,8,9-HpCDF	*	*	0.15	*	*	*
OCDF	2.17	1.72	2.09	2.53	2.11	0.33

* Not detected.
[a] BioRad alumina baked and rinsed.
[b] Sigma alumina baked and rinsed as for BioRad alumina.
[c] Sigma alumina baked at 130° C.

Table 2. Prepacked Column Test Data

Analyte	Concentration Found (ng/Kg)			
	Bgd Sample 1[a]	% IS Recovery	Sample 2[b]	% IS Recovery
2,3,7,8-TCDD	12.08	84	*	86
1,2,3,7,8-PeCDD	*	77	1.65	96
1,2,3,4,7,8-HxCDD	2.93	79	*	107
1,2,3,6,7,8-HxCDD	19.09	78	22.09	62
1,2,3,7,8,9-HxCDD	14.13	#	9.93	#
1,2,3,4,6,7,8-HpCDD	261.78	96	220.44	85
OCDD	2106.73	131	2190.94	75
2,3,7,8-TCDF	11.18	97	27.39	93
1,2,3,7,8-PeCDF	25.54	84	*	91
2,3,4,7,8-PeCDF	9.27	82	*	93
1,2,3,4,7,8-HxCDF	120.92	72	12.97	109
1,2,3,6,7,8-HxCDF	40.34	73	*	61
1,2,3,7,8,9-HxCDF	3.92	85	*	93
2,3,4,6,7,8-HxCDF	5.63	79	*	87
1,2,3,4,6,7,8-HpCDF	112.28	90	61.22	95
1,2,3,4,7,8,9-HpCDF	10.15	95	4.59	86
OCDF	163.72	#	163.55	#

* Not detected.
\# No internal standard available.
[a] All laboratory-made silica, alumina, and carbon columns used in column cleanup.
[b] Prepacked silica and carbon columns used in column cleanup.