

SAVING TIME AND MONEY WITH AUTOMATION

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Introduction

During 2001, the two chemists working in the dioxin group left the EPA Region VII laboratory. The first was replaced by shifting another organic chemist with experience in GC/MS and organic extractions to dioxin analysis, the second was never replaced. In order to maintain the capacity to do dioxin samples, automated techniques were evaluated. Reports of an automated cleanup system were investigated¹⁻³ and resulted in the purchase of Fluid Management Systems Power PrepTM dioxin cleanup system. Evaluation of the Dionex accelerated solvent extraction system (ASE 200TM)⁴⁻⁶ resulted in the addition of this device to the laboratory. The FMS system was evaluated first, because it is used for all matrices. Then the ASE was evaluated and compared to results from Soxhlet extractions. The new procedure, using the ASE and FMS in tandem for solid samples, was compared to traditional Soxhlet extraction with manual cleanup, and was shown to improve recovery, reduce cost, and save substantial time over these traditional techniques.

Methods and Materials

Soxhlet extractions were performed using a 10.0 g solid sample, spiked with the 15 C₁₃ labeled recovery standards as outlined in EPA method 1613, rev. B⁷ and mixed with sodium sulfate and Ottawa sand until a homogenous mixture was formed. Matrix spikes were prepared by adding the ongoing precision and recovery standard (OPR⁷) to a replicate sample aliquot. The samples were extracted with a 20/80 acetone/hexane mixture for 16 hours. The extracts were concentrated and split for cleanup.

Accelerated solvent extractions (ASE) were performed according to EPA method SW846 3545A⁶ on the Dionex 200TM using toluene as the extraction solvent. The program used was: temperature 175°C, pressure 2000 psi, preheat time 1 minute, heat/equilibrium 8 minutes, static 10 minutes, flush volume 60%, purge 60 seconds, with three extraction cycles. The 10.0 g samples were spiked as described above and mixed with approximately 4 g of diatomaceous earth (DE) and 10 g of Ottawa sand. More or less DE was used, depending on the moisture content of the sample. Samples were transferred into 33 mL stainless steel extraction cells with cellulose filters on both ends, and loaded into position on the ASE. Extracts were collected in 60 mL vials. The extracts were dried with sodium sulfate and filtered through glass wool prior to concentration.

Manual cleanup was performed using multi-layered silica gel columns and basic alumina, as described in method 1613⁷. Automated cleanup was performed using the Fluid Management System (FMS) Power-prepTM, following the program prescribed by the manufacturer. Cleaned extracts were concentrated to dryness and diluted with internal standards and nonane to a final volume of 20 µL.

All samples were analyzed on a Micromass Autospec Ultima™ coupled with an Agilent™ 6890 gas chromatograph according to EPA method 1613⁷. A 60-m DB-5 column was used for separation, with a 2 µL split-less injection using an Agilent™ 7683 autosampler. The certified reference material (CRM) used was Wellington Laboratories WMS-01.

Table 1 RPD Split Samples

	#1	#2	#12	#2-MS
2378-TCDD				10
12378-PeCDD				2
123478-HxCDD				3
123678-HxCDD	8			2
123789-HxCDD	7			8
1234678-HpCDD	4	11	1	3
OCDD	8	11	7	3
2378-TCDF	54			15
12378-PeCDF				5
23478-PeCDF				0
123478-HxCDF				3
123678-HxCDF				13
234678-HxCDF				6
123789-HxCDF				2
1234678-HpCDF	8			2
1234789-HpCDF				2
OCDF	33		18	25

Results and Discussion

Data Quality: Initially, the FMS was compared to manual cleanup on Soxhlet extracts that had been split, with one half going through manual cleanup, and the other half run on the FMS. Table 1 shows the relative percent difference (RPD) for those compounds above the detection limit. The matrix spike (#2-MS) shows especially good comparison, with most RPDs less than 20%. An initial demonstration of capability (IDC) was performed on four laboratory control samples. The IDC results met the requirements of EPA method 1613⁷.

Once the cleanup protocol was established, the ASE was investigated. Table 2 presents a comparison of several soil samples extracted by the two different techniques. Each of the first five columns shows the relative percent difference between a sample extracted by Soxhlet and the same sample extracted by ASE, both using FMS cleanup. With one exception, the RPDs are less than 20%, indicating good correlation, especially for matrix spikes (6MS and 6MSD). The final two columns show the RPD of the MS/MSD pairs, which are identical samples spiked with OPR⁷. The Soxhlet MS/MSD column shows the RPD of the pair extracted by Soxhlet; the ASE MS/MSD column shows the RPD of the pair extracted by the ASE. Note the average RPD shows improved precision using the ASE.

Table 2: RPD

	#1	#4	#6	#6-MS	#6-MSD	Soxhlet MS/MSD	ASE MS/MSD
2378-TCDD				13	13.9	0	0.9
12378-PeCDD				14.3	12.2	3.8	1.7
123478-HxCDD				9.5	7.9	0	1.7
123678-HxCDD				11.8	6.3	2.8	2.6
123789-HxCDD		6.8		6.7	6.8	0.7	0.8
1234678-HpCDD	0.92	15.2	6.7	5.6	10.1	3.3	1.2
OCDD	0	13.7	16.5	2.4	9.1	17.6	3.2
2378-TCDF				15.2	14.5	2.4	1.7
12378-PeCDF				14.5	3.2	12.1	0.8
23478-PeCDF				12.5	6.3	2.8	3.4
123478-HxCDF				2.7	3.6	0.9	0
123678-HxCDF				12.4	9.8	0.9	1.7
234678-HxCDF				10.9	7.1	3.8	0
123789-HxCDF				4.6	7.2	0.9	3.5
1234678-HpCDF	7.3	11.4	5.9	4.2	9.3	2.6	2.4
1234789-HpCDF				12.5	7.7	6.4	1.7
OCDF	1.5	7.1	65.5	4.1	6.2	0.8	1.3
Ave RPD						3.6	1.9

The overall method was evaluated by comparing a CRM extracted once by Soxhlet with manual cleanup, and once by ASE with FMS cleanup. The results are presented in Table 3. The RPD shows excellent comparison except for three compounds. 1,2,3,7,8,9-HxCDF was an uncertified value reported barely above the detection limit of 1.4 ng/kg. OCDF and TCDD have higher RPDs, but note that the recoveries using the Soxhlet were 73% and 69%, respectively, while the ASE recoveries were both 94%.

Table 3

	Sox extraction		ASE extraction		RPD	CRM TRUE
	Manual cleanup	%R	FMS cleanup	%R		
	ng/kg	%R	ng/kg	%R		ng/kg
2378-TCDD	12.30	69	16.6	94	29.8	17.7
12378-PeCDD	6.65	84	7.13	90	7.0	7.96
123478-HxCDD	6.58	76	6.88	79	4.5	8.66
123678-HxCDD	17.60	85	18.4	88	4.4	20.8
123789-HxCDD	21.60	125	20.6	119	4.7	17.3
1234678-HpCDD	324.00	111	315	108	2.8	293
OCDD	2310.00	122	2150	113	7.2	1899
2378-TCDF	73.10	139	82.2	157	11.7	52.5
12378-PeCDF	11.60	92	13.1	104	12.1	12.6
23478-PeCDF	17.20	93	18.4	99	6.7	18.5
123478-HxCDF	72.60	108	71.8	107	1.1	67.3
123678-HxCDF	20.10	99	20.7	102	2.9	20.3
234678-HxCDF	13.00	81	14.3	89	9.5	16
123789-HxCDF	1.66	62	1.29	48	25.6	2.68
1234678-HpCDF	300.00	100	286	96	4.8	299
1234789-HpCDF	12.60	83	12.6	83	0.0	15.1
OCDF	370.00	73	476	94	25.1	509
Average %R		94		98		

Finally, an IDC was performed using the same CRM used above. The sample was extracted by ASE with FMS cleanup seven times, over a period of several weeks. The results presented in Table 4 show very low percent relative standard deviation (%RSD) and good percent recovery (%R). All %RSDs were below 17% with most less than 10%, indicating good reproducibility. All recoveries were within the CRM recovery windows (1,2,3,7,8,9-HxCDF was not a certified value). The TCDF was not confirmed, thus not presented with the recovery data.

Table 4: IDP for ASE with FMS

	%RSD	AVE%R
2378-TCDD	11.5	92.8
12378-PeCDD	8.7	91.9
123478-HxCDD	7.0	81.1
123678-HxCDD	7.2	100.5
123789-HxCDD	9.3	142.3
1234678-HpCDD	4.8	119.0
OCDD	6.3	122.3
2378-TCDF	8.5	
12378-PeCDF	7.7	108.7
23478-PeCDF	6.8	105.6
123478-HxCDF	6.0	98.1
123678-HxCDF	2.5	102.7
234678-HxCDF	6.1	90.3
123789-HxCDF	15.8	53.6
1234678-HpCDF	4.9	108.5
1234789-HpCDF	6.9	97.1
OCDF	16.1	99.7

Time savings: Using the FMS, the cleanup time for a set of five samples (including reagent preparation and glassware cleaning) has been reduced by at least one hour, while 3 ½ hours of bench time was saved. For a set of twenty samples, including at least 4 QC samples required by the laboratory, five FMS runs could be completed in 2 working days, with less than half requiring monitoring by an operator. The same number of samples using manual cleanup would require an additional day (almost all of which consists of glassware cleanup), with most of the additional time being spent in laborious bench work.

The laboratory has only enough glassware and space for 12 Soxhlet extraction set-ups. Considering the 4 required QC samples, only 8 "real" samples can be run in one batch. This results in the need for three extraction sets for 20 samples ($\{8+4\} + \{8+4\} + \{4+4\} = 32$). So, prior to the automation improvements discussed above, 20 solid samples would require three days

of extraction. After implementing improvements using the ASE, all 20 samples plus 4 QC could be loaded and run overnight. This resulted in a saving of 8 samples, one less analytical run, one less cleanup day, and a significant reduction in menial work such as cleaning glassware. It is estimated that at least 24 hours (three working days) are saved for the ASE extraction on a set of 20 solid samples.

Cost savings: By using both the ASE and FMS, a cost savings is realized. For the ASE, a savings of \$1.28/sample is due to reduction of 250 mL of solvent, \$60/sample is due to time savings (24 hours/20samples = 1.2 hours/sample @ \$50/hr estimated for senior chemist, including salary and benefits), while the extra cost for filters and seals is \$1/sample, resulting in a total savings of \$60/sample. For the FMS, an increased cost of \$46.75/sample is due to the disposable columns, \$3/sample is due to the filter used prior to cleanup, \$5/sample is due to increase in solvent and disposal, while \$35/sample is saved due to time savings (3.5hours/5samples = 0.7hours/sample @ \$50/hr), resulting in an increase of \$20/sample. Combining the two techniques, the savings are approximately \$40/sample.

Considering the cost of dioxin analysis and salary of a senior chemist, a laboratory can easily reclaim the cost of instrumentation to automate the extraction and cleanup of dioxin samples in one to two years. Automation frees the chemist to do other tasks, especially when performing both extraction and analysis. Automation has allowed the EPA Region VII laboratory to maintain the ability to perform dioxin analyses, while producing high-quality results even while faced with diminishing resources.

Acknowledgments

The author would like to thank the management and staff of the EPA Region VII laboratory for their assistance and encouragement.

Use of trade names is for identification only and does not constitute endorsement by the US EPA.

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