Measurement of Polychlorinated Dibenzo-p-Dioxins (PCDD), Polychlorinated Dibenzofurans (PCDF), Polychlorinated Biphenyls (PCB), Pesticides and Other Bioaccumulating Compounds in Fish, Mussel and Sediment Samples Using a Concerted Analytical Scheme

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Concerted analytical methods are the most rapid and cost effective means for surveying aquatic environments for a large number of polychlorinated aromatic compounds, pesticides and other bioaccumulating residues. Previous reports from our laboratory described the development of a single method for the analysis of the PCBs and 37 other bioaccumulating compounds in fish and sediment samples using high resolution GC and full-scan MS. In the present study these procedures have been adapted and extended to include 24 additional pesticides, as well as PCDD and PCDF analysis.

The analytical scheme which has been developed in the present study is illustrated schematically by the flow chart shown in Figure 1. An aliquot of the sample is mixed with sodium sulfate and extracted with 50% dichloromethane-in-hexane in a Soxhlet extraction apparatus. A portion of the extract is removed for PCDD/PCDF analysis using analytical procedures which have been described previously. After a second portion of the Soxhlet extract is removed to measure the lipid concentration, the analytes are separated from the sample matrix using a gel permeation chromatography (GPC) column packed with Biobeads S-X3. The portion of the 50% cyclopentane-in-dichloromethane GPC column eluate containing the analytes is concentrated and then split into three fractions. The analytes included in Set A (see Table 1 and Figure 1 for identification of the analytes in each set) are separated from the lipid material a second time by adding the fraction to a micro-scale silica column and eluting the analytes from the column with 15% methylene chloride-in-hexane. The analytes in Set B are measured in a portion of the sample extract which is not separated on the silica column because these analytes are irreversibly adsorbed by the column. The compounds in Set C are measured by reversed-phase HPLC using a UV detector.

A second aliquot of the tissue or sediment sample is extracted by mixing the sample with ethyl acetate using a high speed blender or a tissue homogenizer. The analytes are separated from co-extracted materials on the GPC column using the conditions described above. Following concentration of the GPC eluate, a portion of the extract is removed for GC/MS analysis of the compounds included in Set F. Derivatization of the analytes in Sets D and E with a solution of trifluoroacetic anhydride and trifluoroethanol is necessary because these compounds are not directly amenable to GC/MS analysis.

A solution containing three surrogate standards is added to each sample prior to extraction and a three-component internal standard is added to each prepared extract prior to GC/MS analysis. The instrumentation and analysis conditions used are: Gas Chromatograph: Hewlett-Packard 5890 GC with a 7673A Auto Sampler; Capillary Column: DB-5 (60 m, 0.32 mm I.D., 1.0 micron film thickness) split/splitless injection at 275°C using helium carrier gas; Temperature Program: 60°C for 1 minute, 15.0°C/ minute to 180°C, 5.0°C to 300°C, then hold for 30 minutes; Mass Spectrometer: Extrel ELQ-400 Quadrupole; Ionization Mode: Electron impact; Ions Monitored: Computer Controlled Full Scan Monitoring. The identification of the target analytes in the sample extracts is based on the GC retention time, the ratio of the quantitation ion to the confirmation ion, and the goodness of fit when the mass spectra of the analytes detected are compared with the spectra of standards resident in a computerized reference library. The concentrations of the target analytes in the samples and the recovery data for the spiked samples were calculated using a computer program developed in our laboratory.

Table 1 shows the percent recoveries of the Set A analytes from tissue samples spiked at two different concentrations, as well as results from the analysis of a batch of fish and mussel samples for these compounds. The concentrations of 2,3,7,8-TCDD and 2,3,7,8-TCDF found in these samples are shown in Table 2. The percent recoveries of the analytes in Sets B, D, E and F from fortified mussel tissue samples are shown in Table 3. These are generally within an acceptable range.

Table 2. Concentration of 2,3,7,8-TCDD and 2,3,7,8-TCDF in Fish and Mussel Tissue Samples

Sample Number	Conc. 2,3,7,8-TCDD (pg/g)	Conc. 2,3,7,8-TCDF (pg/g)
1	N.D.(0.46)	2.27
2	N.D.(0.25)	1.85
3	N.D.(0.95)	2.11
4	N.D.(0.41)	N.D.(0.97)
5	N.D.(0.48)	1.88
6	N.D.(0.27)	N.D.(0.71)
7	N.D.(0.26)	N.D.(0.72)
8	N.D.(0.50)	1.79
9	1.51	5.49
10	N.D.(0.81)	2.55
11	N.D.(0.28)	1.99
12	N.D.(0.58)	2.87
13	N.D.(0.36)	N.D.(0.51)
14	1.90	3.83

Table 3. Percent Recovery of Target Analytes in Sets B, D, E and F from Fortified Fish and Mussel Tissue Samples

Analytes	% Recovery	Analytes	% Recovery	Analytes	% Recovery
Set B		Set E		Set F	
Diazinon	71	Benzoic Acid	--	Alachlor	88
Disulfoton	51	2,4-D	38	Metolachlor	118
Set D		Dinoseb	140	Ethoprop	159
Endothal	50	Acifluorfen	111	Hexazinone	55
		Carbaryl	154	Acephate	--
		Picloram	71		

Table 1. Percent Recoveries of Set A Target Analytes from Fortified Tissue Samples and Concentrations in Environmental Biological Tissue Samples

	Percent Recovery of Target Analytes in Fortified Samples		Ranges of Concentrations of Target Analytes Found in 13 Fish and Mussel Samples in ng/g (Frequency Detected)	
	156 ng/g	5.0 ng/g		
1,3,5-Trichlorobenzene	23	20		
1,2,4-Trichlorobenzene	32	22		
1,2,3-Trichlorobenzene	34	24		
1,2,3,5-Tetrachlorobenzene	19	21		
1,2,4,5-Tetrachlorobenzene	20	20		
1,2,3,4-Tetrachlorobenzene	53	50		
Pentachlorobenzene	47	77		
Hexachlorobenzene	54	105	3.0	(1)
Hexachlorobutadiene	33	20		
Biphenyl	45	53		
Trifluralin	59	87	5.1	(1)
Alpha-BHC	56	53	3.0-28.3	(8)
Pentachloroanisole	40	102		
Lindane	108	70	4.1-17.0	(5)
Pentachloronitrobenzene	54	114	3.2	(1)
Diphenyldisulfide	34	88	3.5-48.2	(4)
Heptachlor	66	48	1.4-6.5	(4)
Chloropyrifos	45	491	3.0-10.9	(5)
Isopropalin	79	126		
Octachlorostyrene	62	98	3.0	(1)
Heptachlor Epoxide	29	93		
Oxychlordane	62	345	5.0-154	(6)
Butachlor	55	69	3.5-35.9	(2)
Trans-Chlordane	69	109	9.1	(1)
Cis-Chlordane	81	104	11.8	(1)
Trans-Nonachlor	43	109	11.3	(1)
Cis-Nonachlor	58	49		
p,p'-DDE	60	(a)	4.9-217	(10)
Dieldrin	141	(a)	3.6-869	(8)
Perthane	95	119	3.4-12.0	(2)
Nitrofen	57	62	11.0	(1)
Chlorobenzilate	70	145	5.7-6.0	(2)
Endrin	91	(a)	9.6-14.1	(2)
Triphenylphosphate	45	27		
Methoxychlor	44	119		
Dicofol	40	(a)		
Mirex	75	108		
Diethylphthalate		75		
Methyl parathion		121		
Di-n-butylphthalate		112		
PCBs		45	65-80	(2)
--Surrogate Standards--				
Iodobenzene Average	21	19		
Iodonaphthalene Average	54	59		
4,4'-diiodobiphenyl Average	96	90		

(a) target analyte suppressed by chemical interference

FIGURE 1. ANALYTICAL SCHEME FOR THE DETERMINATION OF PCDD, PCDF, PCB, AND PESTICIDES IN FISH AND OTHER AQUATIC BIOLOGICAL TISSUES AND RELATED SAMPLES