Method for Congener-Specific Determination of Dioxin-like PCBs in Biota and Soil/Sediments.

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Abstract

PCBs have historically been analyzed and reported as total PCBs, characterized as Aroclors. Limitations of these characterization-based analyses have been documented in studies which found that original components and their relative concentrations change as a function of time and composition[1,2]. Congener analysis allows for a more accurate determination of what is present in environmental samples. Conventional methods analyze anywhere from 5 to all 209 congeners, although about 20 congeners are common to most methods. The TEF scheme as presented by World Health Organization (WHO), identifies TEFs for 12 PCBs (DLPCBs) that exhibit dioxin-like toxicity (AhR mediated responses).The toxicity of non-dioxin like PCBs have also been reported and are significant enough to warrant a parallel approach[3]. However, if DLPCBs are a critical contaminant, then variations found among components can be more accurately interpreted by applying the TEQ approach. In an effort to report a more uniform and accurate measure of dioxin-like toxicity for various environmental matrices, a congener specific method was developed to analyze for these 12 DLPCBs. The method was validated by replicate analyses, comparisons with certified reference material values and interlaboratory data comparison for the 12 DLPCBs.

Introduction

PCBs were produced and used world wide from the 1930's to the early 1970's as dielectric fluids, in paints, inks, sealants and numerous other materials. A number of different technical mixtures (Aroclors) based on % chlorine content were manufactured. Significant concentrations of these mixtures have found their way into the environment due to poor handling practices. After a number of years in the environment, the composition of these mixture are no longer representative of the original mixture due to weathering. There are 209 possible PCB congeners of which about 130 are typically found in Aroclor mixtures. Of these 130 PCBs, 12 behave like 2,3,7,8-TCDD in biological organisms (bind to AH receptor) and have been identified as DLPCBs. McFarland et al[1] showed that these 12 congeners, when found in environmental samples, are often lower than other PCB congeners. Therefore in order to identify and quantify them, a congener specific isotope dilution HRMS method similar to that used for dioxins and furans is required. Determination of

TEQ values for the PCB component of the sample is a more uniform and accurate representation of the dioxin-like toxicity of the sample, especially for one that has undergone considerable weathering.

Experimental

Standards

DLPCB standards were purchased from Wellington Laboratories Inc. (Guelph, Ont). The calibration series included a 7 point curve with the 12 native compounds in this series of standards ranging from 0.1pg/μL to 800pg/μL and the 12 $^{13}C_{12}$ analogs were constant at 50pg/μL throughout the series.

Sample Preparation

Approximately 5g of sample were used for biota and soil/sediment samples. The samples were spiked with the 12 $^{13}C_{12}$ DLPCB surrogates prior to extraction. Soil samples were Soxhlet extracted using toluene for ~16 hours. Tissue samples were acid digested overnight in concentrated HCl. The digested tissue samples were then extracted with hexane via liquid-liquid extraction.

Cleanup

The list of DLPCBs includes 4 coplanar (BZ#: 77, 81, 126, 169) and 8 mono-ortho congeners (BZ#:105, 114, 118, 123, 156, 157, 167 and 189). The coplanar congeners were isolated with the dioxins and furans in a classical 3 column (silica/alumina/carbon) dioxin/furan cleanup method. The method was developed to force polychlorinated diphenyl ethers (PCDPEs) into the mono-ortho PCB fraction, away from the dioxin/furan fraction. PCDPEs interfere with polychlorinated dibenzofurans giving rise to biased high quantitative results for the furans, therefore a single fraction dioxin/furan/PCB method was not pursued.

For tissue samples, the extracts were filtered through a column containing anhydrous Na_2SO_4 and H_2SO_4 impregnated silica. Soil/sediment extracts were initially chromatographed using a multi-layered acid/base/$AgNO_3$ impregnated silica column. Both matrices were then chromatographed on a basic alumina column where some of the early eluting mono-ortho PCBs were isolated from the chromatographic fraction containing PCDD/Fs and coplanar PCBs. However, some late eluting mono-ortho DLPCBs also end up in the PCDD/F fraction at this stage of the cleanup. In order to isolate these mono-ortho DLPCBs from the PCDD/F fraction, the extract is further separated using a 5% Amoco PX21 carbon/silica (w/w) column. Both the late eluting mono-ortho PCBs and chlorinated diphenylethers are eluted with 25% dichloromethane/hexane mixture and combined with the other mono-ortho congener fraction. The carbon column was then inverted and the PCDD/Fs and coplanar PCBs were eluted with toluene.

Instrumentation

All analyses were performed on a Micromass Autospec GC-HRMS. An HP6890 *Plus* gas chromatograph was interfaced to the mass spectrometer. Chromatographic separations were carried out on a 60m DB-5 column with an internal diameter of 0.25mm and a stationary phase film thickness of 0.25μm (J&W Scientific, USA). Ultrahigh purity helium was used as the carrier gas (Matheson Gas Products, Canada). The splitless injector temperature was set at 280°C with a

column head pressure of 43 psi. The GC oven temperature program was: Initial temperature 150°C held for 1 minute, 1st temperature ramp of 5°C/min until 200°C, 2nd temperature ramp of 3°C/min until 235°C with a hold time of 10 minutes followed by a final ramp of 12°C/min until 300°C, and a final hold time of 1 minute.

The GC-HRMS system was tuned to 10,000+ RP. The coplanar DLPCBs were analyzed with the PCDD/PCDF fraction. The conventional PCDD/F MS experiment was adapted to include appropriate ions for monitoring coplanar-DLPCBs , associated surrogates and the instrument recovery standards. The mono ortho DLPCBs were analyzed as a separate fraction, with a 4 function MS experiment, each function also monitors for potential contributions due to fragmentation from congeners with higher degrees of chlorination.

Results and Discussion

The 7 point calibration curves from this series of standards have consistenly generated RRFs with between 2 and 11 %RSD. Table I shows Method Detection Limits (MDLs) for biota and sediment matrices, calculated according to USEPA protocol[4]. The calculated MDL values for congeners 105 and 118 were elevated relative to other congeners . These elevated levels were attributed to background contamination and correlate with typical lab background amounts reported elsewhere[5]. Alaskan Pollock tissue was chosen because it had lower background levels.

Table I

DLPCB METHOD DETECTION LIMITS in pg/g		
	FISH	**SOIL**
PCB 81	3	1
PCB 71	3	8
PCB 123	3	5
PCB 118	61	20
PCB 114	3	3
PCB 105	22	9
PCB 126	2	4
PCB 167	2	4
PCB 156	6	2
PCB 157	3	3
PCB 169	2	5
PCB 189	<1	2

Biota MDLs were calculated from analyses of 10 fortified matrix blanks. The background contamination levels of PCB 118 and 105 elevated the biota MDLs relative to soil MDLs. For the purpose of the soil/sediment MDLs Ottawa Sand was used as a matrix. These MDLs were based on 6 degrees of freedom and a 98% confidence interval.

Table II – Interlaboratory Data Comparison of Reference Materials in pg/g

	EC-3			MOE REF. SED.		NRC CARP		
	CERTIFIED VALUES	WELL N=3	MOE N=2	WELL N=2	MOE N=2	CERTIFIED VALUES	MOE N=1	WELL N=4
PCB # 81		678	165	158	45		140	127
PCB # 77		7033	5900	1095	1400		1300	1363
PCB # 123		1530	1450	307	335		4400	3465
PCB # 118	28500	30164	34500	6985	7650	132000	112000	126250
PCB # 114		773	395	162	77		5800	5033
PCB # 105	13000	13701	15000	3395	3400	54000	42000	51475
PCB # 126		237	230	70	83		360	324
PCB # 167		5219	1250	1325	345		2800	4223
PCB # 156		2227	2600	595	670		8500	8645
PCB # 157		740	815	154	175		1700	1533
PCB # 169		70	16	9	11		20	23
PCB # 189		343	370	79.5	90.5		940	926
TEQ		32	31	1.43	1.43		60	59

Table II reports concentrations and TEQs for reference materials as analyzed by two different laboratories. Results between labs show excellent correlation of calculated TEQs for both soil/sediment and biota.

References

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