EVALUATION OF POLYCHLORINATED BIPHENYL LEVELS IN WHOLE BLOOD SAMPLES FROM RESIDENTS OF CRYSTAL SPRINGS, MISSISSIPPI

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Introduction

As part of a much larger study, blood samples were taken from 121 individuals who resided proximate to a facility known to be contaminated with polychlorinated biphenyls (PCBs) and polychlorinated dioxins and furans. Many of these individuals reside on properties that are under current remediation for Aroclor 1260 contamination both inside and outside of their residences. While the larger study involves close to one thousand blood or serum samples from individuals and many dust and soil samples in this area, this report will deal only with the 121 individuals indicated above, whose blood was submitted to Midwest Research Institute (MRI) for analysis.

At the beginning of this study, it was important to define a background level of total PCBs in the sample population. It quickly became obvious that the background level was difficult to define because of the variety of reporting methods (e.g., serum vs. lipid basis), variation in analysis (e.g., electron capture detection, low resolution and high resolution mass spectrometry), and the observed trend that PCB body burden has been diminishing over recent years since the discontinued use of PCBs. Published literature values were time sensitive and extremely variable. When the program started two years ago, a value of 3 parts per billion (ppb) in serum was selected as being a background level that appeared realistic and could be measured using screening techniques. We now believe, however, that the normal background level could be as low as 1ppb in serum.

On January 31, 2003, the Centers for Disease Control published the Second National Report on Human Exposure to Environmental Chemicals[1]. This report provides information in terms of background in human blood samples for a select subset of PCBs. The report presents tables of statistics on the distribution of blood levels for PCB including geometric means and percentiles with confidence intervals. The objective of this paper is to provide a comparison of a select set of results to the published report.

Materials and Methods

Whole blood samples were collected from 121 individuals by medical technicians under the supervision of a registered phlebotomist. Six green-top vacutainers with sodium heparin were collected from each individual and were then shipped refrigerated to MRI. For each whole blood sample, five 10-mL tubes are combined, mixed, and weighed. Each sample was fortified with USEPA Method 1668A[2] ^{13}C-substituted surrogate solutions and allowed to equilibrate for 1 hr. Following equilibration, equal volumes of saturated ammonium sulfate and ethanol followed by

50-mL of hexane were added to each sample. Samples were rotary extracted for 30 minutes and the hexane layer was removed and passed through a sodium sulfate funnel into a KD receiver. Samples were extracted with two additional 50-mL volumes of hexane. The combined extracts were concentrated, quantitatively transferred to vials and evaporated to dryness for gravimetric lipid determination.

The lipid residues were dissolved in hexane and fortified with Method 1668A clean-up standards and then processed through silica, alumina, and AX-21 carbon columns using the Fluid Management Systems (FMS) Inc. Power Prep™. The PCB extracts were then concentrated and fortified with recovery standards and brought to a final volume of 20 microliters.

The extracts were analyzed by HRGC/HRMS on a DB-5MS column (60 meter, 0.25 mm ID, 0.25 μm film thickness) using a VG70-250S with acquisition performed at 10,000 resolution in the selected ion monitoring mode (SIM). A five-point calibration curve was performed and passed Method 1668A criteria. Daily calibration was performed using the midpoint of the curve in the beginning and end of each 12-hr run along with a solution containing all 209 possible congeners.

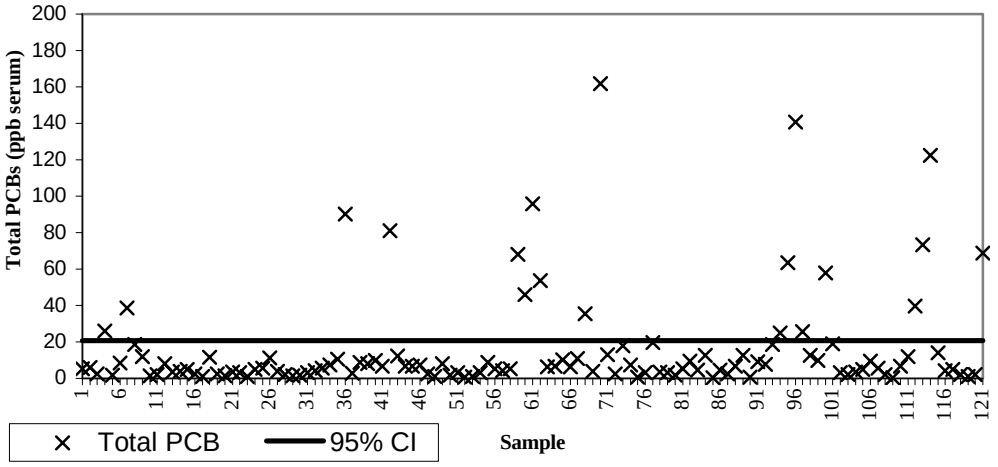
Initial workup and evaluation of the PCB data was based on results for total homologue by level of chlorination (LOC) and Toxic Equivalency Quotient (TEQ) for the 12 World Health Organization (WHO) toxic congeners. Total homolog values were reported on both a whole blood and lipid basis and were determined by summing responses for peaks within a LOC and using the average response factor for 1668A calibrated congeners within the LOC. WHO toxic congeners were also reported on a whole blood and lipid basis but these were determined using the response factor for the individual congener with non-detections reported to the minimum level as described in Method 1668A. After initial reporting of total homolog values, PCB congener data was calculated and reported as described in Method 1668A.

Results and Discussion

Total PCB results for 121 samples in units of nanograms per/gram (ng/g) or parts per billion (ppb) on a serum basis are presented in Figure 1. As discussed earlier, the screening level was established at 3ppb serum. Using this screening level, 90 individuals or roughly 75 percent exceeded the assumed background for PCBs indicating the background level is elevated. In order to assess the data set, a 95th percent confidence interval was calculated and determined to be 20.9 ppb based on the entire data set. As can be seen from this figure, there are several data points near or above the elevated confidence interval. Although any result greater than 3 ppb is considered significant, due to space limitations, only the 19 samples with the highest serum PCB level above the 95th percent confidence interval are discussed further below.

Inspection of the homolog contributions for the 19 highest samples indicated in all cases that the sum of the penta through octa homologs represented over 90 percent of the PCBs present in the samples with the hexa and hepta homologs representing the majority. This may be important from the perspective that the main contaminant in this case was Aroclor 1260. Although, as expected, the pattern found in these blood samples did not definitively match the congener patterns for Aroclor 1260 (Frame et. al 1996[3]), the elevation of the hexa and hepta congeners is unusual and additional evaluation of the PCB congener patterns found in relation to environmental weathering and human metabolism will be conducted.

Figure 1. Total PCB Concentrations in Serum Samples (ng/g)



Congener results for the 19 highest serum PCB samples were then compared against 95th percentile values presented in the NHANES report. The 95th percentile value was selected for comparison as the population with the highest levels of PCBs present. One problem, however, was that there are only 25 PCB congeners reported in the NHANES report and with differences in chromatographic systems (e.g., GC column phase and temperature programs), not all congeners are necessarily resolved in both data sources to the same extent to allow for a direct comparison.

Table 1 presents a comparison of determined congener values against the 95th percentile values presented in the NHANES report. Table 1 also presents a listing of total PCBs as well as total WHO Toxic PCBs in parts-per-billion lipid weight found in each of the blood samples. This comparison is for only eight of the possible 25 NHANES congeners. The coplanar PCBs (IUPAC 81, 126, and 169) were removed from the comparison as all determined values were less than the NHANES 95th percentile. Four congeners (IUPAC 28, 52, 66, and 74) were not included because this study focused on penta-octa substituted congeners. The remaining eight penta-hepta congeners shown in Table 1 are those that are not influenced by coelution with other congeners. Values greater than the NHANES 95th percentile value are shown in bold.

Based on the comparison shown in Table 1, it is observed that not only do several of the individuals have concentrations of total PCB concentrations in serum greater than “normal” background levels but they also have individual congeners in excess of the background population as defined by the NHANES report. It is also observed that concentrations of WHO toxic are a small fraction of the total PCBs. Therefore, if the blood samples were evaluated only on the toxic PCBs (TEQ basis), the full extent of contamination would not be known and as there is some indication of significant thyroid, developmental and reproductive toxicity for the presumably “non-toxic” congeners, a critical piece of the overall puzzle would be missed.

Table 1. Comparison of Congener Results to NHANES Values

	PCB IUPAC Number (ppb lipid)								Total PCB Lipid	Toxic PCB lipid	Total PCB Serum
Sample	**99**	**105**	**156**	**157**	**167**	**177**	**178**	**183**	**ppb**	**ppb**	**ppb**
NHANES	18.6	<6.4	16.5	<6.4	<6.4	<6.4	<6.4	<6.4	NA	NA	NA
B047	**23.0**	**18.0**	**28.4**	4.96	**18.8**	**89.6**	**42.1**	**101.1**	4000	159	25.8
B050	9.80	**22.9**	**91.3**	**20.2**	**23.8**	**84.6**	**81.4**	**47.5**	5750	312	38.7
B079	11.3	4.79	**60.8**	**6.72**	**21.2**	**125**	**117**	---	15800	149	90.2
B086	**69.6**	**11.3**	**39.3**	**8.40**	**20.6**	**94.1**	**53.0**	**50.2**	11100	158	80.9
B103	**42.6**	**20.7**	**22.8**	3.67	**13.5**	**60.3**	**32.7**	---	7950	157	67.9
B104	13.4	3.50	15.5	2.04	3.02	**28.7**	**26.6**	**43.6**	4860	43.8	45.9
B105	**36.6**	**12.7**	**37.1**	5.75	**17.1**	**93.1**	**62.7**	---	11100	145	95.8
B106	**48.2**	**28.3**	**52.6**	**6.84**	**16.6**	**50.4**	**36.8**	**106**	8140	238	53.8
B112	2.80	0.929	8.37	---	1.74	**40.9**	**15.8**	**70.0**	4440	19.0	35.6
B114	**32.0**	4.18	**37.3**	3.60	**12.6**	**123**	**85.7**	**330**	18300	93.4	162
B143	**23.2**	1.85	9.75	2.27	2.14	**7.70**	**6.50**	---	1960	27.5	25.0
B144	*	0.895	4.44	0.927	1.22	*	*	*	1660	12.9	63.6
B145	**92.4**	**19.5**	**26.8**	6.15	**12.2**	**75.7**	**27.7**	---	11000	159	141
B146	**20.6**	2.99	8.12	1.77	4.46	**29.2**	**11.3**	---	3150	34.5	25.7
B149	*	1.22	0.829	---	0.398	*	*	*	520	7.59	57.9
B162	*	2.01	3.28	0.735	1.02	*	*	*	680	14.6	39.5
B163	13.8	2.39	5.66	1.28	1.17	5.80	2.60	---	780	19.7	73.3
B164	*	1.39	2.91	0.820	0.791	*	*	*	340	12.4	123
B126	10.2	2.05	7.26	2.12	2.07	**7.30**	3.80	---	1360	28.8	68.7

--- Indicates result <LOD.
* Specific congener data not determined.

References

[1] USEPA Method 1668, Revision A: Chlorinated Biphenyl Congeners in Water, Soil, Sediment, and Tissue by HRGC/HRMS, December 1999.
[2] CDC, "Second National Report on Human Exposure to Environmental Chemicals" NHANES Report, January 2003.
[3] Frame, GM, Cochran, JW and Boewadt, SS, "Complete PCB congener distributions for 17 Aroclor mixtures determined by 3 HRGC systems optimized for comprehensive, quantitative, congener-specific analysis" J Hi Resol. Chromatogr., vol 19, 657-668(1996).

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