

Detection of Hydroxylated Polychlorinated Biphenyls (OH-PCBs) in the Abiotic Environment: Surface Water and Precipitation from Ontario, Canada

DAISUKE UENO,^{†,‡} COLIN DARLING,[‡]
MEHRAN ALAEE,[‡] LINDA CAMPBELL,[§]
GRAZINA PACEPAVICIUS,[‡]
CAMILLA TEIXEIRA,[‡] AND DEREK MUIR*^{†,‡}

Department of Applied Biological Sciences, Faculty of Agriculture, Saga University, Honjo 1, Saga, 840-8502, Japan, Aquatic Ecosystem Protection Research Division, Environment Canada, 867 Lakeshore Road, Burlington, ON, L7R 4A6, Canada, and School of Environmental Studies, Queen's University, Kingston, ON, K7L 3N6, Canada

Hydroxylated PCBs (OH-PCBs) are well-known metabolites of PCBs in organisms, but there has been no direct study of their presence in the abiotic environment. In this study, OH-PCBs were determined in samples of rain, snow, and surface waters from sites in Ontario, Canada. OH-PCBs were quantified by gas chromatography with high-resolution mass spectrometry (GC–HRMS) in order to provide complete characterization of all OH-PCB homolog groups. OH-PCBs and PCBs were detected in all the samples analyzed, although half of the Σ OH-PCBs could not be identified even with 71 individual congener standards. Total concentrations of OH-PCBs (Σ OH-PCBs) in water ranged from 0.87 to 130 pg/L and from 230 to 990 pg/g in particulate organic matter. Total fluxes of those compounds in snow and rain were from <1 to 100 pg/m² and from <1 to 44 pg/m²/day, respectively. Higher Σ OH-PCB fluxes in rain were found in southern Ontario than in a remote north-central Ontario site possibly reflecting greater sources of precursor PCBs near urban areas. Relatively higher Σ OH-PCB concentrations were found in surface waters from sites near sewage treatment plant (STP) outfalls in the cities of Toronto (130 pg/L) and Hamilton (35 pg/L) than in offshore samples from Lake Ontario (1.6 pg/L). The results indicate that STPs are one of the sources of OH-PCBs for lake waters in this region. Similar homolog and congener profiles in rain and offshore surface water samples suggest that atmospheric deposition is the predominant source at offshore sites. This is the first report to detect the OH-PCBs in the abiotic environment.

Introduction

The Great Lakes in North America are known as a region where polychlorinated biphenyls (PCBs) continue to be a

significant contamination issue particularly in air, water, sediments, and biota near urban areas (1). In the Great Lakes biota, thyroid dysfunction (large goiters and thyroid hyperplasia) has been reported in salmonids for more than 30 years (2). It has been suggested that some thyroidogenic contaminants such as metabolites of PCBs, hydroxylated PCBs (OH-PCBs), might be one of the causes of thyroid dysfunction in fish (2). OH-PCBs have demonstrated potential estrogenic activity (3–5) and especially thyroidogenic effects (6–8) in vitro and in vivo. These compounds can compete with thyroxin for binding sites on transthyretin, one of the three main thyroid hormone transport proteins in mammals (6). A number of OH-PCBs have been identified in the blood of humans and biota during the last 5–10 years (8); however, the identification of a full range of OH-PCB congeners has been limited by a lack of analytical standards.

It has been reported that OH-PCBs are formed biologically via cytochrome P450 mediated oxidation (9). OH-PCBs can also be formed abiotically by reaction of PCBs with OH radicals although evidence for this is limited. It has been suggested that gaseous PCBs in the environment could be removed by OH radicals to form OH-PCBs since an inverse relationship of PCB concentrations and OH radicals has been observed in the atmosphere (10, 11). Laboratory scale experiments demonstrated that small amounts of OH-PCBs were produced, alongside larger amounts of chlorinated benzoic acids, on reaction of gaseous PCBs with OH radicals generated by UV degradation of ozone (12). Those authors concluded that reaction with OH radicals was the major removal pathway for PCBs in the atmosphere (13). In addition, it has been reported that OH-PCBs could be formed in the aquatic environment through aerobic biodegradation (14) and the reaction of PCBs and OH radicals (15) which are produced by processes of excitation of organic chromophores and photolysis of nitrate ions in natural waters (16). If entering the aquatic environment in significant amounts, OH-PCBs would be expected to bioaccumulate since they are relatively hydrophobic. The replacement of an H- with an OH-group reduces the log K_{ow} by 0.67 according to the fragment constant method (17).

While reaction with OH radical has been thought to be the major removal pathway for PCBs in the atmosphere, to our knowledge, OH-PCBs have not been identified directly in abiotic environmental samples. Given their known toxicity and likely bioaccumulation potential, the full extent of OH-PCBs contamination in the abiotic environment needs to be elucidated in order to estimate exposure to wildlife and people.

There are 837 possible monohydroxylated PCB congeners compared to 209 PCB congeners; however, only 71 commercial standards of OH-PCBs are currently available. Thus, identification of specific OH-PCBs in environmental samples is challenging. This study has found gas chromatography with high-resolution mass spectrometry (GC–HRMS) provides a specific and complete characterization of all OH-PCB homolog groups and congeners by enabling detection of both identifiable and unidentifiable congeners (i.e., with unknown structure but having the same mass and fragmentation as OH-PCB standards). This study describes a preliminary investigation of the presence of OH-PCBs in abiotic samples and comparisons of congener patterns among various abiotic media.

Materials and Methods

Sample Collection. All the abiotic samples (snow, rain, and water) were collected in Ontario (Canada) from 2002 to 2004.

* Corresponding author phone: (905)319-6921; fax: (905)336-6430; e-mail: derek.muir@ec.gc.ca.

[†] Saga University.

[‡] Environment Canada.

[§] Queen's University.

TABLE 1. Sample Information of Snow, Rain, and Water for OH-PCBs and PCBs Analysis Collected in Ontario, Canada

ID	location	date	vol (L)	collected area (m ²)	method ^a	OH-PCBs	PCBs	note
Snow						Fluxes (pg/m ²)		
S1	Grimsby	Feb 20/03	44	8.0	d	56	27000	rural residential area
S2	Pinery, PP	Feb 26/03	98	5.1	d	45	54000	rural, provincial park
S3	Macgregor, PP	Feb 27/03	113	10.9	d	15	19000	rural, provincial park
S6	Turkey Watershed	Mar 4/03	88	4.0	d	100	24000	remote area
S7	Evansville	Mar 5/03	89	5.2	d	35	8900	residential area
S8	Dorset	Mar 6/03	95	6.0	d	34	13000	remote area
S10	Sibbald, PP	Mar 7/03	95	2.7	d	54	45000	remote area, provincial park
S11	Guelph Lake	Mar 12/03	116	5.8	d	48	17000	semiurban, park in residential area
S13	Pine Valley, GC	Mar 14/03	108	101.4	d	nd	1000	rural, golf course
Rain						Fluxes (pg/m ² /day)		
R2	Lake St. Clair	May 3–Jun 2/04	5	0.2	d	30	2000	rural agricultural area
R3	Lake St. Clair	Jun 3–Jun 30/04	2	0.2	d	22	2100	
R4	Grand Bend	Apr 1–May 4/04	7	0.2	d	nd	7300	rural agricultural area
R5	Grand Bend	May 4–Jun 3/04	5	0.2	d	39	2600	
R6	Grand Bend	Jun 2–Jun 30/04	2	0.2	d	44	2300	
R7	Rock Point	Mar 30–Apr 30/04	4	0.2	d	36	4300	rural agricultural area
R8	Rock Point	Apr 30–Jun 1/04	3	0.2	d	42	2700	
R9	Rock Point	Jun 1–Jun 29/04	5	0.2	d	31	2500	
R10	Turkey Lakes	Jun 5–July 2/04	5	0.2	d	4.9	1500	remote area
Water						Concentrations (pg/L)		
W1	Detroit river	Sept 1/02	80	ND ^b	c	0.87	ND ^b	near Peche Is., river, industrial area
W2	Detroit river	Sept 1/02	95	ND ^b	c	20	ND ^b	near Fright Is., river, industrial area
W3	Detroit river	Sept 2/02	95	ND ^b	c	14	ND ^b	Lake Erie, Detroit river outlet, rural urban area
W4	Detroit river	Sept 2/02	95	ND ^b	c	3.2	ND ^b	near Amherstburg, rural area
W5	Lake Ontario	Sept 17/02	87	ND ^b	c	10	ND ^b	Hamilton harbor, near STP, industrial area ^c
W6	Lake Ontario	Sept 22/02	95	ND ^b	c	4.6	ND ^b	Hamilton harbor, near steel factory, industrial area
W7	Lake Ontario	Oct 20/02	100	ND ^b	c	7.5	ND ^b	city of Toronto, offshore lake waters
W8	Lake Ontario	Oct 20/02	97	ND ^b	c	130	ND ^b	city of Toronto, lake waters near STP ^c
W11	Lake Ontario	Aug 21/03	96	ND ^b	c	16	ND ^b	city of Toronto, offshore water
W12	Lake Ontario	Oct 18/04	50	ND ^b	f	1.6	190	city of Hamilton/Burlington, offshore water
W13	Lake Ontario	Oct 27/04	91	ND ^b	f	35	980	Hamilton harbor, near STP, industrial area ^c
Suspended Solids						Concentrations (pg/g POC)		
SS1	Lake Ontario	Oct 18/04	0.95 ^d	ND ^b	f	990	290000	city of Hamilton/Burlington, offshore water
SS2	Lake Ontario	Oct 27/04	4.39 ^d	ND ^b	f	230	95000	Hamilton harbor, near STP, industrial area ^c

^a d: Direct extraction (no particle removal). c: Centrifuge particle removal before extraction (PCBs were not quantified due to contamination of the sampling system). f: Filtration before extraction (suspended solid on the filter was employed for chemical analysis). ^b "ND": not determined. ^c STP: sewage treatment plant. ^d Concentration of particulate organic carbon (mg POC/L).

Detailed sample information is shown in Table 1, and locations are shown in Figure 1.

Rain samples were collected monthly from April to June 2004 at three locations in southern Ontario and one location in north-central Ontario (Turkey Lakes) using an MIC-B automated wet-only collector ((MIC Co., Thornhill, ON) equipped with a 0.2 m² stainless steel funnel leading to a column of XAD-2 resin (no particle removal).

Large-volume water samples were collected from various sites in southern Ontario, with a Westfalia field centrifuge system (Westfalia-Separator AG, Oelde, Germany). Suspended particulates are retained in the centrifuge bowl while the centrifuged water was collected in precleaned stainless steel containers. The flow rate of lake water through the centrifuge was held at 4 L/min to allow for sufficient retention time of suspended particulates in the centrifuge bowl. Suspended particulates were removed from the centrifuge bowl in the laboratory and archived for future analyses. Approximately 100 L of centrifuged lake water was collected in precleaned stainless steel containers and pumped through 0.7 μm glass fiber filter (GFF) and XAD-2 resin columns in the field. PCBs were not determined in these samples because we had previously shown that the centrifuge system and steel containers introduced PCB contamination (C. Teixeira

and D. Muir, unpublished data, Environment Canada, Burlington, ON). Hamilton harbor and western Lake Ontario water samples in 2004 were collected by direct pumping through 0.7 μm GFF into an XAD-2 column (which was spiked with ¹³C-OH-PCBs prior to sampling). We had previously demonstrated very low blank concentrations of PCBs with this system. Particulate organic carbon (POC) on the GFF was stored in polyethylene "Ziplok" bags and frozen for later chemical analysis.

Large-volume snow samples were collected at various locations across Ontario from north of Sault Ste Marie to south of Hamilton in the end of February 2002 (Table 1). The entire snow column was collected to obtain samples representative of winter precipitation. Sampled areas (m²) were recorded, and the snow accumulation period was estimated as 3 months (Dec 1–Feb 28). Approximately 1–2 m³ of snow was collected using precleaned aluminum boxes lined with polypropylene autoclave bags, and those samples were transported to a laboratory clean room (HEPA- and carbon-filtered air). Snow samples were melted down in the aluminum boxes, and the meltwater was pumped through an XAD-2 resin column without filtration.

The XAD-2 resin used for all the sample collection was prepared in large batches by Soxhlet extraction with metha-

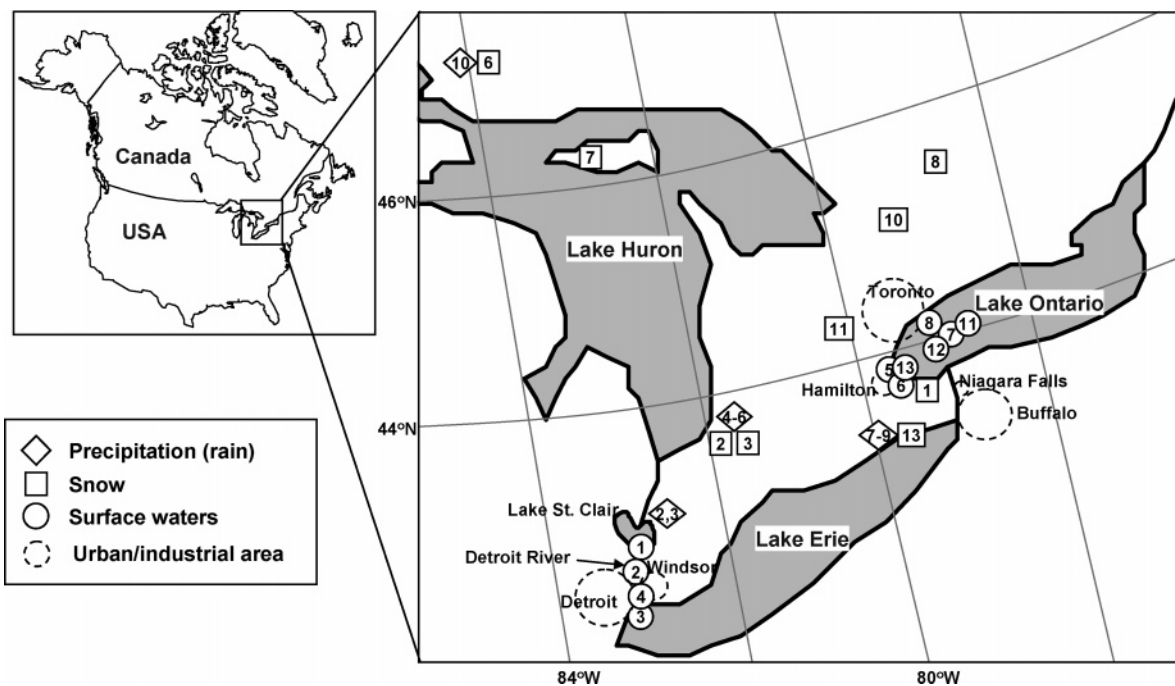


FIGURE 1. Sampling locations of water, snow, and rain samples collected in southern Ontario, Canada. The numbers in symbols indicate the sample identification (see Table 1).

nol, followed by acetone and dichloromethane for 4 days each. The precleaned XAD-2 resin was checked for PCB and OH-PCB contamination prior to use by extracting samples from each batch (as described below) and checking qualitatively using GC-ECD.

OH-PCBs Extraction and Cleanup. Extraction and isolation of OH-PCBs and PCBs was based on a method previously reported (18), and all the procedures were conducted in a clean room laboratory. The ^{13}C -OH-PCB standards {4'-OH-PCB 12 (di), 4'-OH-PCB 29 (tri), 4'-OH-PCB 61 (tetra), 4'-OH-PCB 120 (penta), 4'-OH-PCB 187 (hepta)} and PCB166 (Wellington Laboratories Inc., ON, Canada) were spiked onto the XAD-2 resin column for two water samples (W12 and W13) before field sampling, and for other water, snow, rain, and GFF samples after field sampling. XAD-2 resin was transferred to a 19 mm \times 420 mm glass column and eluted using 200 mL of methanol followed by 250 mL of dichloromethane, and the mixed solvent was washed with 200 mL of 3% NaCl solution. GFFs were extracted with dichloromethane using a Soxhlet apparatus. The eluate was dried with Na_2SO_4 and then extracted with 2-propanol and 1:1 methyl tert-butyl ether (MTBE)/hexane mixture after the addition of HCl to lower the pH. The organic layer was washed with a 3% KCl solution, and then the pH was raised using 1:1 1 M KOH solution/ethanol to force the OH-PCBs into an aqueous fraction (phenolic fraction) and the parent PCBs into the organic fraction (neutral/basic fraction). Concentrated H_2SO_4 was added to the aqueous fraction, and this solution was then extracted with 1:1 MTBE/hexane. This extract was evaporated to 1 mL, and the OH-PCBs were derivatized to methoxylated PCBs (MeO-PCBs) using diazomethane. The derivatized fraction containing MeO-PCBs was cleaned up using an acidified silica gel column. The neutral/basic fraction containing PCBs was also cleaned using an acidified silica gel column and then put through a nonacidified silica gel column for fractionation. The final solution was concentrated to 100 μL and 1 mL for the MeO-PCBs and PCBs, respectively.

Quantification of OH-PCBs. OH-PCBs were quantified as MeO-PCBs. The quantification of MeO-PCBs was carried out using gas chromatography (GC, Agilent 6890) coupled

with high-resolution mass spectrometry (HRMS, MicroMass Ultima). A 60 m DB5-MS (0.25 mm id, 0.25 μm film thickness, J&W Scientific) column was used for GC separations. Samples were analyzed in splitless mode with He carrier gas at a pressure of 1 kPa. The injector temperature and transfer line temperature were maintained at 280 and 250 $^\circ\text{C}$, respectively. The temperature program for the GC was controlled at 80 $^\circ\text{C}$ as the initial temperature for 1 min; ramped at 20 $^\circ\text{C}/\text{min}$ to 170 $^\circ\text{C}$; held at 170 $^\circ\text{C}$ for 15 min; ramped at 2 $^\circ\text{C}/\text{min}$ to 285 $^\circ\text{C}$; held at 285 $^\circ\text{C}$ for 24 min. Ionization was performed by electron ionization (EI) at an electron voltage of 30–40 keV depending on the optimization parameters of the instrument. The source temperature was 270 $^\circ\text{C}$, and the mass resolution of the analyzer was 10 000. The mass spectrometer was operated in selected-ion monitoring (SIM) mode.

Authentic OH-PCB (MeO-PCB) standards were purchased from Wellington Laboratories Inc. (ON, Canada) and Accustandard (CT). OH-PCBs were quantified by an isotope dilution method using ^{13}C -MeO-PCBs. The peaks that matched the retention times and isotopic ratio of primary and secondary ions of those compounds in the standards were quantified as “identified MeO-PCBs”. The higher selectivity of the HRMS (10 000 mass resolution) allows unknown peaks to be labeled as OH-PCBs. Unknown peaks that matched the isotopic ratio of the primary and secondary ions of those compounds in authentic standards were quantified as “unidentified OH-PCBs” using an average response factor for the same homolog group. Parent PCB congeners were determined by capillary GC-ECD as described previously (19).

Quantification and Quality Control. Recovery studies were conducted by spiking blank resin columns ($n = 5$) with the ^{13}C -labeled standards; 10 L of water (MilliQ grade) was pumped through. Percentage recoveries of ^{13}C -OH-PCBs for the recovery study averaged $98 \pm 18\%$. For the sample analysis, the percentage recovery of ^{13}C -OH-PCBs for the whole of the procedure (through field collection, extraction, and cleanup, $N = 47$) averaged $71 \pm 56\%$ for di–tetra ^{13}C -OH-PCBs and $104 \pm 56\%$ for penta–hepta ^{13}C -OH-PCBs. Detection limits (DLs) were defined as 3 times the signal-to-noise ratio of the HRMS. DLs of identified OH-PCBs ranged

TABLE 2. Fluxes and Concentrations of OH-PCBs in Snow, Rain, and Water Samples Collected from Southern Ontario, Canada^a

homologues/sample ID	fluxes (pg/m ²) snow (n = 9)		fluxes (pg/m ² /day) rain (n = 9)		concentrations (pg/L) water (n = 11)		concentrations (pg/g POC) POC (n = 2) ^b	
	mean	(min-max)	mean	(min-max)	mean	(min-max)	mean	(min-max)
mono OH-PCBs								
identified	nd	(<1-)	1.1	(<1-2.2)	2.7	(<0.05-14)	12	(<10-25)
unidentified	3.8	(<1-7.8)	0.13	(<1-1.2)	3.8	(<0.05-29)	nd	(<10-)
total	3.8	(<1-7.8)	1.2	(<1-3.1)	6.6	(0.054-37)	12	(<10-25)
di OH-PCBs								
identified	nd	(<2-)	1.7	(<2-3.4)	1.7	(<0.05-7.5)	nd	(<10-)
unidentified	11	(<2-17.3)	7.2	(<2-15)	6.3	(<0.05-54)	nd	(<10-)
total	11	(<2-17.3)	9.0	(<2-18)	8.0	(<0.05-62)	nd	(<10-)
tri OH-PCBs								
identified	nd	(<2-)	nd	(<2-)	0.66	(<0.05-4.5)	42	(<10-83)
unidentified	nd	(<2-)	7.3	(<2-13)	1.0	(<0.05-6.6)	100	(<10-200)
total	nd	(<2-)	7.3	(<2-13)	1.7	(<0.05-11)	140	(83-200)
tetra OH-PCBs								
identified	nd	(<2-)	nd	(<2-)	0.47	(<0.1-2.4)	nd	(<10-)
unidentified	2.4	(<2-4.5)	6.4	(<2-11)	1.0	(<0.1-3.4)	150	(55-250)
total	2.4	(<2-4.5)	6.4	(<2-11)	1.5	(0.13-3.9)	150	(55-250)
penta OH-PCBs								
identified	20	(<2-48)	1.7	(<2-7.2)	1.2	(<0.1-2.8)	43	(26-61)
unidentified	0.33	(<2-2.9)	1.9	(<2-4.8)	0.89	(<0.1-5.9)	35	(<10-69)
total	20	(<2-48)	3.6	(<2-9.2)	2.1	(<0.1-8.3)	78	(26-130)
hexa OH-PCBs								
identified	nd	(<2-)	nd	(<2-)	0.17	(<0.1-0.85)	120	(<10-250)
unidentified	nd	(<2-)	nd	(<2-)	0.075	(<0.1-0.23)	20	(<10-40)
total	nd	(<2-)	nd	(<2-)	0.25	(<0.1-1.1)	140	(40-250)
hepta OH-PCBs								
identified	5.6	(<2-26)	0.25	(<2-2.3)	0.30	(<0.1-1.1)	nd	(<50-)
unidentified	nd	(<2-)	nd	(<2-)	0.25	(<0.1-1.1)	49	(<50-98)
total	5.6	(<2-26)	0.25	(<2-2.3)	0.55	(<0.1-1.8)	49	(<50-98)
octa OH-PCBs								
identified	nd	(<2-)	nd	(<2-)	0.015	(<0.1-0.17)	33	(<50-67)
unidentified	nd	(<2-)	nd	(<2-)	1.7	(<0.1-15)	nd	(<50-)
total	nd	(<2-)	nd	(<2-)	1.7	(<0.1-15)	33	(<50-67)
nona OH-PCBs								
identified	nd	(<2-)	nd	(<2-)	nd	(<0.1-)	nd	(<50-)
unidentified	nd	(<2-)	nd	(<2-)	0.12	(<0.1-0.75)	nd	(<50-)
total	nd	(<2-)	nd	(<2-)	0.12	(<0.1-0.75)	nd	(<50-)
total identified OH-PCBs	25	(nd-74)	4.8	(nd-11)	7.3	(0.17-25)	250	(130-380)
total unidentified OH-PCBs	18	(nd-27)	23	(nd-40)	15	(nd-110)	360	(95-610)
total OH-PCBs	43	(nd-100)	28	(nd-44)	22	(0.87-130)	610	(230-990)
total PCBs ^c	23000	(1000-54000)	3000	(1500-7300)	590	(190-980)	190000	(95000-290000)
number of peaks	12	(0-19)	35	(0-61)	58	(7-95)	11	(9-12)
identified OH-PCBs (%) ^d	56	(40-75)	26	(0-100)	40	(6.8-100)	48	(38-58)
OH-PCBs/PCBs (%) ^e	0.2	(0.0-0.4)	1.1	(0.0-1.9)	2.1	(0.8-3.5)	0.3	(0.2-0.3)

^a Concentrations of individual identified congeners for each sample are shown in the Supporting Information. ^b POC: particulate organic carbon. ^c PCBs concentrations in water are available for two samples due to possible contamination of the continuous centrifugation sampling system. ^d Identified OH-PCBs (%): percentage of identified OH-PCBs to total OH-PCBs. ^e OH-PCBs/PCBs (%): percentage of total OH-PCBs to total PCBs.

from 1 to 2 pg/m² for snow samples, 1-2 pg/m²/day for rain samples, 0.05-0.1 pg/L for water samples, and 10-50 pg/g POC for POC samples.

Results and Discussion

Identification of OH-PCBs. The mean and range of the OH-PCB and PCB concentrations in abiotic samples (snow, rain, and water) analyzed in this study are summarized in Tables 1 and 2 (data on individual samples are available in the Supporting Information). OH-PCBs and PCBs were detected in all of the samples analyzed. The results in this study demonstrate the ubiquitous presence of OH-PCBs in abiotic media such as rain, snow, and water. To our knowledge, this is the first report to detect the OH-PCBs directly in the abiotic environment.

Total fluxes of Σ OH-PCBs in snow and rain were from <1 to 100 pg/m² and from <1 to 44 pg/m²/day, respectively (Table 2). A wide range of fluxes (differences of up to 2 orders

of magnitude) was observed. Concentrations of Σ OH-PCBs in water and particulate organic carbon (POC) ranged from 0.87 to 130 pg/L and from 230 to 990 pg/g POC, respectively. Σ PCBs fluxes in snow and rain were from 1000 to 54 000 pg/m² and from 1500 to 7300 pg/m²/day, respectively (Table 2). Where Σ PCBs were measured in water (dissolved phase), concentrations ranged from 190 to 980 pg/L, while concentrations on particulates ranged from 95 000 to 290 000 pg/g POC, respectively. The concentrations of Σ PCBs in offshore Lake Ontario surface waters (190 pg/L) were about 3-fold higher than observations for mid-Lake Ontario stations (59-92 pg/L) (20). The range of Σ PCBs fluxes in rain was in agreement with the data measured as the part of the Integrated Atmospheric Deposition Network (IADN) in the Great Lakes (21). Snow samples also showed a similar range of concentration for Σ PCBs with those of a previous report for Great Lakes snow (22).

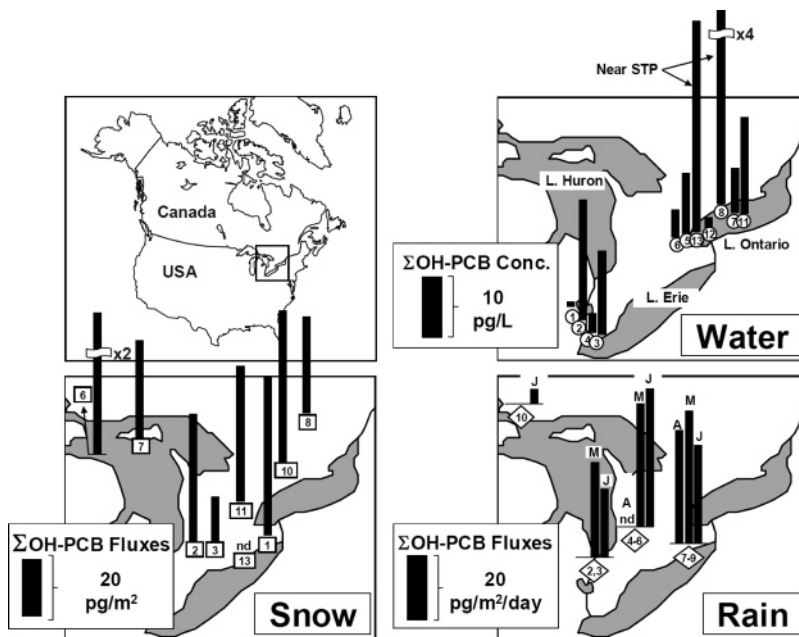


FIGURE 2. Geographical distribution and seasonal variation of total OH-PCB fluxes and concentrations in rain, snow, and water from Ontario, Canada. STP, A, M, J, and nd indicate sewage treatment plant, April, May, June, and less than detection limit, respectively.

A large number of unidentified OH-PCBs were detected. Some of the unidentified peaks were detected frequently, but no uniform pattern was observed. In order to evaluate the importance of “unidentified OH-PCBs”, the concentration/flux ratios between “identified” Σ OH-PCBs and “unidentified” were calculated. The values of the identified OH-PCB (%) to Σ OH-PCBs are shown in Table 2. Variations of this value ranged from 0 to 100%, and the average value among all of the media was almost 50%. Half of the total Σ OH-PCBs were not identified even with 71 congeners of authentic standards available. This result suggests that quantification of “unidentified OH-PCBs” is necessary to understand the full extent of OH-PCBs contamination in environment.

Since OH-PCBs are ionizable compounds, there is the possibility that the scavenging efficiency of these chemicals by rain and snow in the atmosphere could vary depending on the pH, as could collection efficiency by the XAD-2 resin. The pH in precipitation and lake water in this region is well documented by Environment Canada (23). The pH in rain is around 4–5 in the southern Ontario region. At this pH, most of the OH-PCBs in precipitation will be protonated, and thus amenable to extraction with XAD-2 resin.

Geographical Distribution of OH-PCBs. Figure 2 shows the geographical distributions of Σ OH-PCB concentrations and fluxes in abiotic samples (snow, rain, open lake, rivers, and harbor waters) from Ontario, Canada.

The distribution of Σ OH-PCB fluxes in snow samples was relatively uniform, and no clear trend was observed (Figure 2). The distribution of Σ PCB fluxes in snow samples was also uniform (Supporting Information, S1). The snow samples represented deposition over the entire winter because the entire snow column was collected toward the end of the season (late February and early March) to obtain samples representative of winter precipitation.

Higher Σ OH-PCB fluxes in rain were found at the three southern sampling locations than at the single north-central Ontario site (Figure 2). Σ PCB fluxes in rain samples also showed similar geographical distribution (Supporting Information, S2). This is generally consistent with trends for PCBs and PAHs between the upper Great lakes and Lake Ontario (21) and is likely due to the larger population and greater past use of PCBs in urban areas of the southern part

of Ontario than in northern areas (Figure 1). Σ OH-PCB fluxes did not show clear seasonal trends (Figure 2), but relatively higher fluxes were found in “May–June” (early summer) than “April” (end of winter). In addition, the opposite trend was found for Σ PCBs with “April” showing relatively higher concentration than “May–June” (Supporting Information, S2). Possible reasons for this difference will be discussed in the section on degradation ratios.

In water samples, relatively higher Σ OH-PCB concentrations were found in samples from near urban areas on Lake Ontario, such as east Toronto (W7, 8, 11; 7.5–130 pg/L) and Hamilton harbor (W5, 6, 13; 4.6–35 pg/L) (Figure 2; Supporting Information, S3). The distribution of Σ PCB concentrations in water samples also showed a higher concentration in Hamilton harbor (W13) than offshore water (W12) (Supporting Information, S3). Among those regions, the highest concentrations were found in samples collected within 0.5–2 km of a major sewage treatment plant (STP) in eastern Toronto (W7, 8), and the next highest was also collected near an STP flowing into Hamilton harbor (W13). The collection technique, i.e., pumping centrifuged water from stainless steel cans (W7, 8, 11) versus direct pumping from water (W12–13), did not influence the observed OH-PCB concentrations.

OH-PCBs could be formed in STP through microbial oxidation or reactions with OH radicals if ozone is used to treat the final effluent. The byproducts of industrial compound synthesis or chlorination/oxidation byproducts of biphenyl or biphenylol could also be potential sources of OH-PCBs. This result indicates that STPs are one of the sources of OH-PCBs into the aquatic environment. On the other hand, Σ OH-PCBs concentrations in water samples from the Detroit river (W1–W4) were relatively lower than those from Lake Ontario nearshore waters even though it is an industrialized and urban region (Figure 2). Those water samples were collected on the Canadian side of the Detroit river where the concentrations of PCBs and chlorinated dioxins in sediment were much lower than those in the American side (24). It may also be due to the fact that most of the water in the Detroit river originates from Lake Huron where there is relatively less anthropogenic activity compared with that of Lake Ontario.

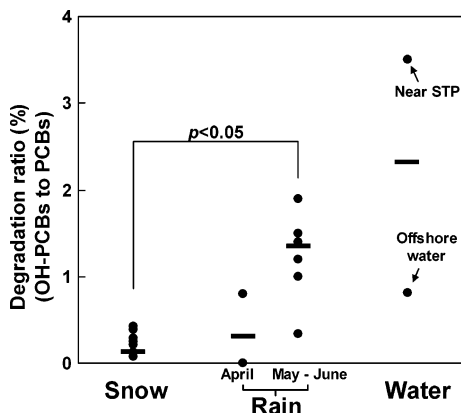


FIGURE 3. Degradation ratios (percentage contribution of total OH-PCBs to total PCBs) in snow, rain, and water from Ontario, Canada. STP indicates sewage treatment plant.

Degradation Ratios and Peak Number of OH-PCBs. In order to evaluate the degradation efficiency of PCBs to OH-PCBs, a “degradation ratio”, $\sum \text{OH-PCBs} / \sum \text{PCBs}$ (%), was calculated (Table 2 and Figure 3). The degradation ratios in rain (0–1.9%) were 2–10 times higher than those in snow (0–0.4%) (Table 2 and Figure 3). Those ratios in rain samples were divided into two groups, those collected in “April” (end of winter) and those from “May–June” (early summer), to compare seasonal differences (Figure 3). The degradation ratios in rain collected in “May–June” were relatively higher than those in “April”, and these values in “May–June” were significantly higher ($p < 0.05$, Mann Whitney’s *U* test) than those in snow (Figure 3). To provide additional evidence of degradation efficiency, the numbers of detected peaks in the GC–MS chromatogram were compared between rain and snow (Table 2 and Supporting Information, S1 and S2). A larger number of peaks was detected in rain samples (up to 61), and a much lower number was observed in snow (up to 19). The inverse relationship between PCBs and atmospheric OH radicals has been observed (10, 11). Sedlak and Andren (25) also concluded that oxidation by OH radicals in “cloudwater and raindrops” could be an important mechanism for PCB degradation. Higher temperature causes higher concentrations of parent PCBs in the gaseous phase (26), and stronger sunlight generates larger amounts of OH radicals by UV radiation (27). Higher degradation ratios and larger peak numbers of OH-PCBs in rain samples (May–June) than those in snow may be due to the combined effect of higher temperature and greater OH radical concentrations during summer.

It has been suggested that OH-PCBs could be formed in the aquatic environment through aerobic biodegradation (14). The reaction of PCBs with OH radical in surface water (15), produced by processes of excitation of organic chromophores and photolysis of nitrate ions in natural water (16), could also form OH-PCBs. In Figure 3, the highest degradation ratio (3.5%) among all of the media (snow, rain, and water) was found in water samples collected near STPs. In addition, OH-PCB peaks were much more numerous in water near STPs (73–87 peaks) (Table 2 and Supporting Information, S3). These results suggest that PCBs could be oxidized in the STP resulting in local emissions of OH-PCBs to nearby receiving waters. On the other hand, another water sample from offshore water showed a relatively lower degradation ratio (0.8%), that was similar to that of snow and rain (April) (Figure 3). The lower values in this offshore water sample suggest less influence of STP sources (15) (Figure 3).

Homolog Pattern of OH-PCBs. The homolog pattern of snow samples showed less variation among locations, and

the major components in snow were lower chlorinated homologs (mono- and di-) (Figure 4). In the rain samples, di- to tetrachlorinated homologs predominated, and their patterns were more highly chlorinated than those in snow samples. Regarding the seasonal variation of rain samples, a greater proportion of higher chlorinated homologs (tri- to tetra-) was detected in “May and June, early summer” and relatively lower proportions of higher chlorinated homologs were found in “April (R7), end of winter” (Figure 4). Those lower percentages of higher chlorinated homologs in rain samples in “April” were similar to those in snow samples. As noted previously, higher temperature and OH radical concentrations in “May and June” are probably responsible.

Clear differences in the homolog pattern were found among water samples (Figure 4). Samples collected in “urban and industrial areas” (W1–4, 6) showed a higher percentage of mono- to trichlorinated homologs than those in “offshore water” (W7, 11, 12). Much higher percentages of lower chlorinated homologs were found in samples from “urban area near STP” (W5, 8, 13) (Figure 4). The result of higher percentages of lower chlorinated OH-PCB homologs near STP (W5, 8, 13) may be due to the presence of lower chlorinated PCBs which, because of higher water solubility and lower Cl substitution, can be degraded by microbial oxidation or reactions with OH radical if ozone is used to treat the final effluent. On the other hand, samples collected from “offshore water” (W7, 11, 12) showed less percentage contribution of lower homologs, and the patterns were similar to those in rain samples (May and June; e.g., R2, 3, and 9) rather than other water samples. Thus, OH-PCBs in offshore waters are not affected by metropolitan and industrial effluents, but those may be related to precipitation inputs. Gas exchange and dryfall are other possible routes of entry of OH-PCBs to the aquatic environment.

Partition Coefficient between Water and Particulate Organic Carbon (K_{oc}) of OH-PCBs. The partition coefficient between particulate organic carbon–water (K_{oc}) was calculated using the data for OH-PCBs in Hamilton harbor POC that was collected by filtration (W13). To evaluate these empirical K_{oc} values, they were compared to those estimated from the octanol–water partition coefficient (K_{ow}) by the simple relationship (28): $K_{oc} = 0.41 K_{ow}$.

The K_{ow} of OH-PCBs to predict K_{oc} was estimated from PCBs with the same number of chlorines according to the fragment constant method (17) because there are no directly measured values to our knowledge. The observed and predicted K_{oc} values agreed within a factor of 0.5 log units for tetra–heptachlorinated OH-PCBs (Figure 5; individual data is available in the Supporting Information, S4). However, those values for mono- and trichlorinated congeners were higher than the predicted values. Ebner and Braselton (29) reported that the pK_a of some OH-PCB congeners varied from 6 to 10 among congeners and concluded that this could be explained by the chemical structure with *p*-hydroxylated PCBs with two adjacent chlorines having higher pK_a (e.g., 4-OH-3,4',5-PCB/4-OH-PCB 39, $pK_a = 6.8$). Those results indicate that some OH-PCB congeners having a pK_a around 7 could be 50% in the anion form at the pH of Lake Ontario and Hamilton harbor water (pH = 7.4–8.2), which could result in a lower observed K_{oc} for those congeners assuming the anionic form did not bind to POC. However, this was not observed. For example, 4-OH-2,2',3,3',5,6-PCB (4OH-PCB-134; K_{oc} = 5.0) which is *p*-hydroxylated with two adjacent chlorines had a similar K_{oc} to that of 4'-OH-2,2',4,5,5'-PCB (4'OH-PCB 101, K_{oc} = 5.1) which is not *p*-hydroxylated with two adjacent chlorines (Supporting Information, S4). The good agreement between the predicted and observed K_{oc} for tetra–heptachlorinated OH-PCBs suggests that the pK_a does not have much influence on congener binding under the natural conditions of pH and POC in lake water.

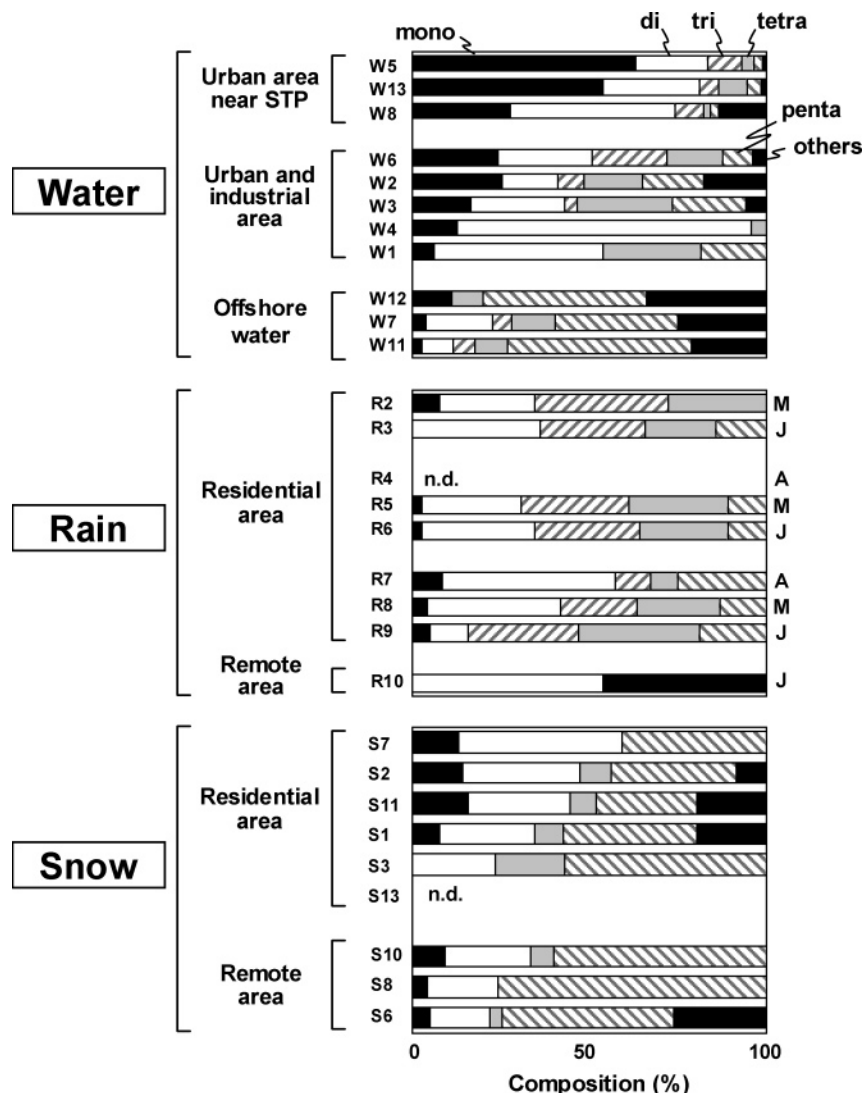


FIGURE 4. Percentage contribution of OH-PCB homologues detected in snow, rain, and water from Ontario, Canada. STP, A, M, and J indicate sewage treatment plant, April, May, and June, respectively.

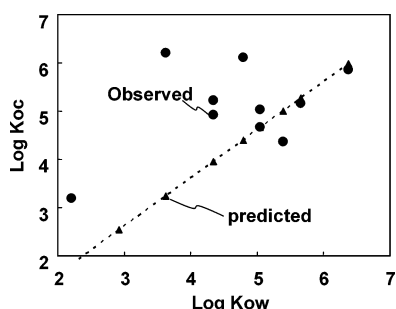


FIGURE 5. Comparison between observed and predicted values of partition coefficient (K_{oc}) for OH-PCBs and other phenolic compounds in the water sample from Hamilton harbor, Lake Ontario. The K_{oc} for triclosan (unpublished data) is included. Circles and triangles indicate observed and predicted values, respectively.

The reason the observed K_{oc} values were higher than the predicted values for lower chlorinated congeners is not clear. Lower recovery of lower chlorinated ^{13}C -OH-PCBs was observed (20–30%) during the analytical procedure for the POC fraction than the water fraction (50–80%), and also the relationship between the pH in water and the pK_a of OH-PCBs may cause variation of the collection efficiency on the XAD-2 resin and GFF. This makes the K_{oc} values for mono-

and trichlorinated congeners more uncertain especially after recovery correction. Additional studies are needed to examine the sources and environmental behavior of OH-PCBs in order to determine if concentrations in the abiotic environment represent a significant additional exposure route of fish and wildlife to PCBs.

Acknowledgments

This study was funded by the Environment Canada Great Lakes 2020 Fund and by the Natural Sciences and Engineering Research Council of Canada which provided a postdoctoral fellowship to Linda Campbell. We thank Robert McCrea of Environment Canada (Burlington ON) for providing the “POPCart” water sampling system used for Hamilton harbor and western Lake Ontario samples. We thank Sean Backus, Chris Spencer, Xiaowa Wang and Jay Guo (Water Science and Technology Branch, Environment Canada) for technical assistance, Rob Letcher (National Wildlife Research Centre, Environment Canada, Ottawa) for provision of analytical standards, and Shinsuke Tanabe (Ehime University, Japan) for providing valuable suggestions. We thank Grant-in-Aid for JSPS Fellows (15002714) from Japan Society for the Promotion of Science (JSPS) and Grant-in-Aid for Young Scientists (B), KAKENHI (18780245) from The Ministry of Education, Culture, Sports, Science and Technology (MEXT).

Supporting Information Available

Data on fluxes and concentrations of individual identified OH-PCBs in rain and snow (S1 and S2), concentrations in surface waters and suspended sediment (S3), organic carbon-water partition coefficients (S4), fluxes and relative retention times of all OH-PCBs in snow (S5) and rain (S6) as well as concentrations of all OH-PCBs in surface waters and suspended sediments (S7). This material is available free of charge via the Internet at <http://pubs.acs.org>.

Literature Cited

- (1) Hornbuckle, K. C.; Carlson, D. L.; Swackhamer, D. L.; Baker, J. E.; Eisenreich, S. J. Polychlorinated biphenyls in the Great Lake. In *Persistent Organic Pollutants in the Great Lakes, Handbook of Environmental Chemistry, 5N*; Hites, R. A., Ed.; Springer-Verlag: Berlin, 2006; pp 13–70.
- (2) Leatherland, J. F. Changes in thyroid hormone economy following consumption of environmentally contaminated Great Lakes fish. *Toxicol. Ind. Health* **1998**, *14*, 41–57.
- (3) Jansen, H. T.; Cooke, P. S.; Porcelli, J.; Liu, T.; Hansen, L. G. Estrogenic and antiestrogenic actions of PCBs in the female rat: in vitro and in vivo studies. *Reprod. Toxicol.* **1993**, *7*, 237–248.
- (4) Kramer, V. J.; Helferich, W. G.; Bergman, A.; Klasson-Wehler, E.; Giesy, J. P. Hydroxylated polychlorinated biphenyl metabolites are anti-estrogenic in a stably transfected human breast adenocarcinoma (MCF7) cell line. *Toxicol. Appl. Pharmacol.* **1997**, *144*, 363–376.
- (5) Andersson, P. L.; Blom, A.; Johannisson, A.; Pesonen, M.; Tysklind, M.; Berg, A. H.; Olsson, P.-E.; Norrgren, L. Assessment of PCBs and hydroxylated PCBs as potential xenoestrogens: in vitro studies based on MCF-7 cell proliferation and induction of vitellogenin in primary culture of rainbow trout hepatocytes. *Arch. Environ. Contam. Toxicol.* **1999**, *37*, 145–150.
- (6) Brouwer, A.; Morse, D. C.; Lans, M. C.; Schuur, A. G.; Murk, A. J.; Klasson-Wehler, E.; Bergman, A.; Visser, T. J. Interactions of persistent environmental organochlorines with the thyroid hormone system: mechanisms and possible consequences for animal and human health. *Toxicol. Ind. Health* **1998**, *14*, 59–84.
- (7) McKinney, J. D.; Waller, C. L. Polychlorinated biphenyls as hormonally active structural analogues. *Environ. Health Perspect.* **1994**, *102*, 290–297.
- (8) Letcher, R. J.; Klasson-Wehler, E.; Bergman, A. Methyl sulfone and hydroxylated metabolites of polychlorinated biphenyls. In *The Handbook of Environmental Chemistry Vol. 3 Part K, New Types of Persistent Halogenated Compounds*; Paasivirta, J., Ed.; Springer-Verlag: Berlin, 2000; pp 315–359.
- (9) Bergman, A.; Larsen, G. L.; Bakke, J. E. Biliary secretion, retention and excretion of five ¹⁴C-labelled polychlorinated biphenyls in the rat. *Chemosphere* **1982**, *11*, 249–253.
- (10) Totten, L. A.; Eisenreich, S. J.; Brunciak, P. A. Evidence for destruction of PCBs by the OH radical in urban atmospheres. *Chemosphere* **2002**, *47*, 735–746.
- (11) Mandalakis, M.; Berresheim, H.; Stephanou, E. G. Direct evidence for destruction of polychlorobiphenyls by OH radicals in the subtropical troposphere. *Environ. Sci. Technol.* **2003**, *37*, 542–547.
- (12) Brubaker, W. W., Jr.; Hites, R. A. Gas-phase oxidation products of biphenyl and polychlorinated biphenyls. *Environ. Sci. Technol.* **1998**, *32*, 3913–3918.
- (13) Anderson, P. N.; Hites, R. A. OH radical reactions: the major removal pathway for polychlorinated biphenyls from the atmosphere. *Environ. Sci. Technol.* **1996**, *30*, 1756–1763.
- (14) Flanagan, W. P.; May, R. J. Metabolite detection as evidence for naturally occurring aerobic PCB biodegradation in Hudson River sediment. *Environ. Sci. Technol.* **1993**, *27*, 2207–2212.
- (15) Sedlak, D. L.; Andren, A. W. The effect of sorption on the oxidation of polychlorinated biphenyls (PCBs) by hydroxyl radical. *Water Res.* **1994**, *28*, 1207–1215.
- (16) Haag, W. R.; Hoigne, J. Photo-sensitized oxidation in natural water via OH radicals. *Chemosphere* **1985**, *14*, 1659–1671.
- (17) Leo, A. Octanol/water partition coefficient. In *Handbook of Property Estimation Methods for Chemicals*; Boethling, R. S., Mackay, D., Eds.; CRC Press: London, 2000; pp 89–114.
- (18) Sandau, C. D.; Ayotte, P.; Dewailly, E.; Duffe, J.; Norstrom, R. J. Analysis of hydroxylated metabolites of PCBs (OH-PCBs) and other chlorinated phenolic compounds in whole blood from Canadian Inuit. *Environ. Health Perspect.* **2000**, *108*, 611–616.
- (19) Campbell, L. M.; Muir, D. C. G.; Whittle, D. M.; Backus, S.; Norstrom, R. J.; Fisk, A. T. Hydroxylated PCBs and other chlorinated phenolic compounds in lake trout (*Salvelinus namaycush*) blood plasma from the Great Lakes region. *Environ. Sci. Technol.* **2003**, *37*, 1720–1725.
- (20) USEPA. Great Lakes National Program Office (GLNPO), Chicago, IL. Unpublished PCB data in water, 2001, provided by M. Hulting, 2004.
- (21) Simcik, M. F.; Hoff, R. M.; Strachan, W. M. J.; Sweet, C. W.; Basu, I.; Hites, R. A. Temporal trends of semivolatile organic contaminants in Great Lakes precipitation. *Environ. Sci. Technol.* **2000**, *34*, 361–367.
- (22) Franz, T. P.; Eisenreich, S. J. Snow scavenging of polychlorinated biphenyls and polycyclic aromatic hydrocarbons in Minnesota. *Environ. Sci. Technol.* **1998**, *32*, 1771–1778.
- (23) Environment Canada. 2005. Acid rain and the facts. <http://www.ec.gc.ca/acidrain/acidwater.html> (accessed Nov 2006).
- (24) Marvin, C.; Alaei, M.; Painter, S.; Charlton, M.; Kauss, P.; Kolic, T.; MacPherson, K.; Takeuchi, D.; Reiner, E. Persistent organic pollutants in Detroit River suspended sediments: polychlorinated dibenzo-*p*-dioxins and dibenzofurans, dioxin-like polychlorinated biphenyls and polychlorinated naphthalenes. *Chemosphere* **2002**, *49*, 111–120.
- (25) Sedlak, D. L.; Andren, A. W. Aqueous-phase oxidation of polychlorinated biphenyls by hydroxyl radicals. *Environ. Sci. Technol.* **1991**, *25*, 1419–1427.
- (26) Brunciak, P. A.; Dachs, J.; Gigliotti, C. L.; Nelson, E. D.; Eisenreich, S. J. Atmospheric polychlorinated biphenyl concentrations and apparent degradation in coastal New Jersey. *Atmos. Environ.* **2001**, *35*, 3325–3339.
- (27) Seinfeld, J. H. Urban air pollution: state of the science. *Science* **1989**, *243*, 745–752.
- (28) Karickhoff, S. W. Semiempirical estimation of sorption of hydrophobic pollutants on natural sediments and soils. *Chemosphere* **1981**, *10*, 833–849.
- (29) Ebner, K. V.; Braselton, W. E., Jr. Structural and chemical requirements for hydroxychlorobiphenyls to uncouple rat liver mitochondria and potentiation of uncoupling with Aroclor 1254. *Chem. Biol. Interact.* **1987**, *63*, 139–155.

Received for review June 29, 2006. Revised manuscript received December 27, 2006. Accepted December 27, 2006.

ES061539L