Time Course of Congener Uptake and Elimination in Rats after Short-Term Inhalation Exposure to an Airborne Polychlorinated Biphenyl (PCB) Mixture

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Despite the continued presence of PCBs in indoor and ambient air, few studies have investigated the inhalation route of exposure. While dietary exposure has declined, inhalation of the semivolatile, lower-chlorinated PCBs has risen in importance. We measured the uptake, distribution, and time course of elimination of inhaled PCB congeners to characterize the pulmonary route after short-term exposure. Vapor-phase PCBs were generated from Aroclor 1242 to a nose-only exposure system and characterized for congener levels and profiles. Rats were exposed via inhalation acutely (2.4 mg/m^3 for 2 h) or subacutely (8.2 mg/m^3, 2 h $\times$ 10 days), after which pulmonary immune responses and PCB tissue levels were measured. Animals acutely exposed were euthanized at 0, 1, 3, 6, and 12 h post exposure to assess the time course of PCB uptake and elimination. Following rapid absorption and distribution, PCBs accumulated in adipose tissue but decayed in other tissues with half-lives increasing in liver (5.6 h) < lung (8.2 h) < brain (8.5 h) < blood (9.7 h). PCB levels were similar in lung, liver, and adipose tissue, lower in brain, and lowest in blood. Inhalation of the airborne PCB mixture contributed significantly to the body burden of lower-chlorinated congeners. Congeners detected in tissue were mostly *ortho*-substituted ranging from mono- to pentachlorobiphenyls. Selective uptake and elimination led to accumulation of a distinct congener spectrum in tissue. Minimal evidence of toxicity was observed.

Introduction

Polychlorinated biphenyls (PCBs) are a family of synthetic organic chemicals that contain 209 congeners with varying chemical structures. Their industrial usage spanned 50 years until they were banned in 1977 in the United States. PCBs are still being emitted from industrial facilities, poorly maintained waste sites, building demolition, incineration, caulking material in buildings, and dredging of waterways (*1*, *2*). PCBs are also produced and released through production and use of paint and colored products (*3*). Exposure to PCBs from ingestion of contaminated food has been well studied (*4*). Inhalation of PCBs in indoor air is found comparable to dietary exposure (*5*) and can even become a major route of exposure to children (*6*). The majority of this pollutant in the ambient air environment exits as a vapor mixture rather than associated with particles (*7*), and the magnitude of airborne concentration rises with population density (*8*). Indoor air exposures to PCBs may be more significant than ambient air exposure, with at least 10-fold and up to 100000-fold higher concentrations (*9*). The lack of temporal decline in indoor air exposures as compared to the significant downward trend in dietary contamination (*5*) makes inhalation an increasingly important route of PCB exposure.

The impact of inhalation on overall human exposure of PCBs is most evident for the lower-chlorinated PCBs due to their lower lipophilicity, greater volatility, and susceptibility to metabolism (*10*). Human studies revealed that exposure to contaminated indoor air increased the concentrations of lower-chlorinated congeners in plasma (*11*, *12*). The toxicological importance of these more volatile congeners has recently been recognized because evidence of their adverse health effects is mounting. Thyroid hormone elevations and histologic abnormalities were observed in rats exposed to Aroclor 1242 vapors for 30 days (*13*). The mutagenicity of PCB 3 and the tumor-initiating activities of several other lower molecular weight congeners (PCBs 15, 52, and 77) have been demonstrated in vivo (*14*, *15*). Lower-chlorinated PCBs and their metabolites have repeatedly been shown as estrogenic and capable of interfering with hormone homeostasis (*16*, *17*). The metabolites can be more efficacious than their parent compounds as receptor agonists (*18*) and cancer initiators for their reactivity toward cellular nucleophiles (*19*). The susceptibility to metabolic activation implies a larger risk of these congeners. However, the complexity of exposure mixtures in the environment hampers risk characterization. Information on how the relative concentration of congeners in exposure mixtures relate to blood and tissue uptake and how the concentrations in tissues change over time is very limited.

To our knowledge, this is the first investigation describing the rate and extent of adsorption, distribution, and elimination of individual PCB congeners in blood and tissue of laboratory animals exposed via inhalation. We have developed a dynamic nose-only exposure system exposing animals to PCB vapor mixtures from commercial preparations and have shown diversity in the extent of uptake and elimination among congeners. We also report the biological half-lives of individual congeners in organs or tissues, especially the lower-chlorinated PCBs and congeners detectable in environmental samples.

Experimental Section

Generation of PCB Atmosphere. IUPAC identities, numbered PCB 1 (monochlorobiphenyl) through PCB 209 (decachlorobiphenyl), are used for congener identification (*20*). PCBs and all other chemicals were obtained from commercial sources or synthesized in our laboratory (see Supporting Information).

Aroclor 1242 (Electrical grade, Monsanto Lot KB-05-415) was used as the source material to generate PCB exposure atmospheres. The solution was held in a custom built,

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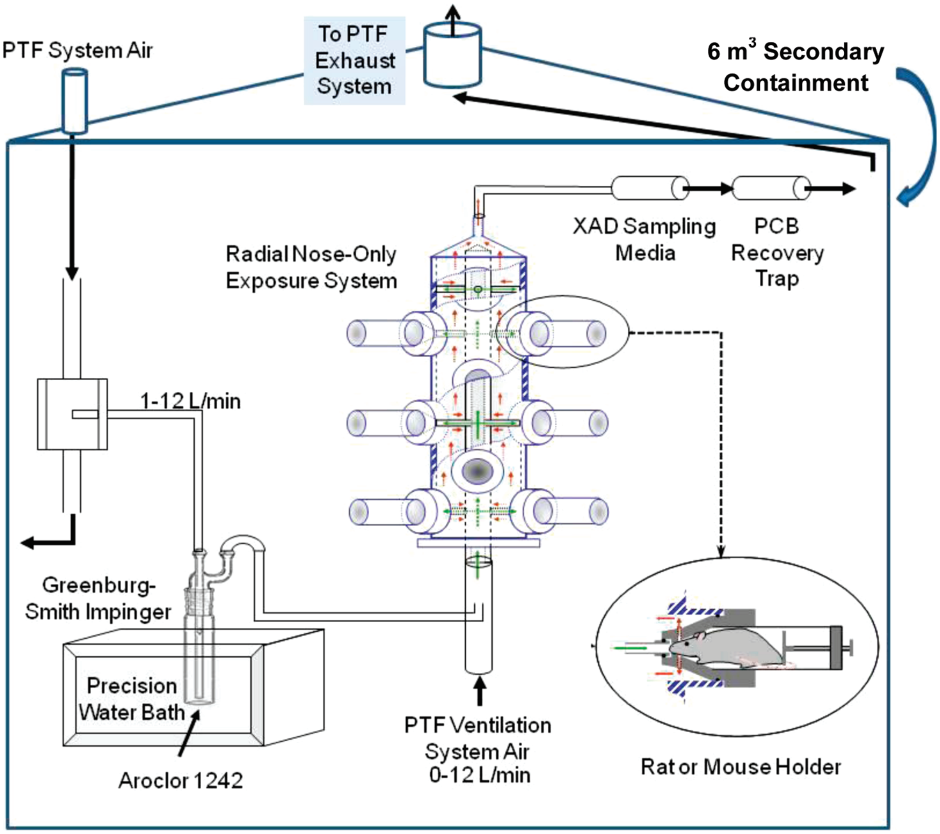

FIGURE 1. Diagram of the nose-only PCB inhalation exposure system in a secondary containment structure.

Greenburg–Smith-style impinger resting in a precision temperature-controlled water bath at 25 °C (Figure 1). All system components were constructed of glass. Pure air (4.0 L/min) was bubbled through the Aroclor liquid to facilitate PCB volatilization. The PCB vapor-laden air was then diluted and supplied to a nose-only exposure chamber (InTox, Inc., Albuguerque, NM) as described previously (*21*) at a flow rate of 11.0 L/min. A sampling cartridge filled with Amberlite XAD-2 polymeric absorbent resin (XAD, Supelco Analytical, Bellefonte, PA) captured the PCB atmosphere drawn out of the exposure apparatus. The XAD cartridge was collected each day over the exposure period and extracted for PCB atmosphere characterization. The PCB exposure system was placed in a 6 m^3 glass and stainless steel secondary containment structure for exhaust control and operated at negative pressure to ensure no leakage outward. A sham exposure nose-only system for control animals was located in an adjacent lab where no PCBs have ever been deliberately introduced.

Animal Protocol. Animal protocols were approved by the Institutional Animal Care and Use Committee, and animals were housed in our AAALAC-approved on-site vivarium with food and water provided ad libitum. Male Sprague–Dawley rats (Harlan, Inc., Indianapolis, IN) were exposed to the generated PCB atmosphere subacutely or acutely through a nose-only inhalation system (Figure 1), while sentinel rats were maintained in our vivarium for health surveillance. After exposure, the animals were euthanized with isoflurane followed by cervical dislocation. Whole blood was collected via cardiac puncture and serum samples were prepared. Lung, trachea, liver, and brain were excised and stored at −20 °C for PCB analysis. Adipose tissue was taken from the peritoneal fat surrounding the intestines.

In the subacute study, animals (187.9 ± 2.8 g) were exposed to either the generated PCB atmosphere ($n = 9$) or filtered lab air ($n = 9$) two h/day with two 1 h exposures separated by a 2 h break. Two rats in the PCB exposure group were exposed for 4 days, while 7 rats were exposed for 10 days during a two week period.

In the acute study, animals were randomly assigned into groups (239.9 ± 2.7 g). Five groups of PCB-exposed animals ($n = 3$/group) were exposed for a total of 2 h, with a 1 h break in between. They were serially euthanized at 0 h, 1 h, 3 h, 6 h, and 12 h post exposure. Two groups of sham animals ($n = 2$/group) exposed to filtered lab air were euthanized at 0 and 6 h post exposure. In both studies, animals were 8 weeks old at the time of euthanization.

Tissue and XAD Extraction for PCB Analysis. PCBs were extracted from lung + trachea, liver, brain (0.9−1.2 g each), and adipose tissue (0.3−0.4 g) samples using pressurized liquid extraction (ASE 200, Dionex, Sunnyvale, CA) as described earlier (*22*). Each set of tissue samples was accompanied by a method blank, a tissue blank, and an ongoing precision and recovery (OPR) spike sample. Each sample was spiked with a surrogate standard, including PCB 14 (100 ng) and deuterium-labeled PCB 65 (d-PCB 65, 100 ng) in hexane. The extracted solutions were concentrated to 0.75 mL (TurboVap II, Caliper Life Sciences, Inc., Hopkinton, MA), and cleanup was performed (*22*) for gas chromatography (GC) with mass selective detection (GC tandem mass spectrometry, GC/MS/MS) determination. PCBs were extracted from serum samples using the same protocol. Lipid content was determined by a standard gravimetric method (*22*). Total cholesterol and triglycerides in serum samples were determined using a commercial test kit (Trig/GB and Chol tests for Roche/Hitachi 917 system; Roche Diagnostics, Indianapolis, IN). Blood lipids were calculated using the formula (*23*)

$$\text{Total lipids} = 2.27 \times \text{Total cholesterol} + \text{Total triglycerides} + 62.3\,\text{mg/dL}$$

Each XAD cartridge was loaded with 10 g of pre-extracted XAD-2 resin packed with filters and cleaned glass wool. After

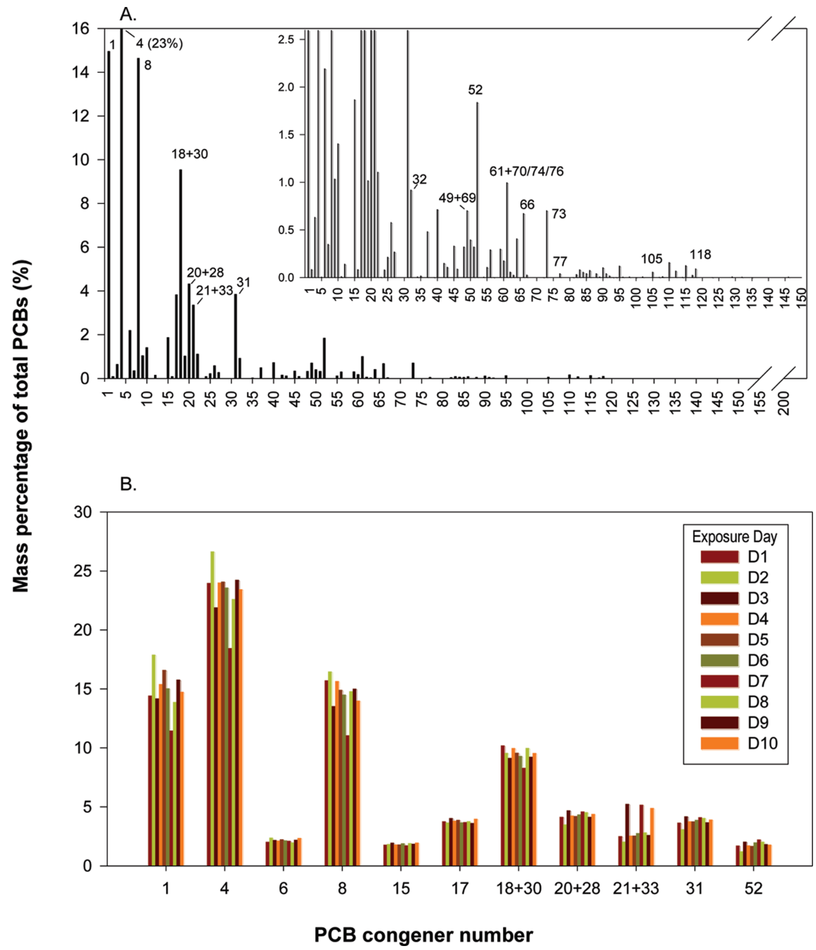

FIGURE 2. Average distribution profile of all congeners during 10 d exposure (A, inset plot showing 0−2.5% on *y* axis) and day-to-day profile of most prevalent congeners (B) in generated PCB vapor mixture.

collection, all samples were placed in sealed zip-lock bags, stored at 4 °C until analysis, and were later extracted using the same protocol as above.

PCB Analysis. XAD extracts were spiked with 100 ng of PCB 204 as internal standard prior to analysis. The tissue extract was concentrated to 100 μL for detection of low PCB levels and then spiked with 20 ng of PCB 204. PCB congeners were analyzed using GC/MS/MS modified from EPA method 1668A (*20*) as described previously (*2, 24*). Briefly, the quantification of PCB congeners used an Agilent 6890N GC with an Agilent 7683 series autosampler coupled to a Waters Micromass Quattro micro GC MS (Milford, MA), operating under electron impact positive mode at 70 eV and multiple reaction monitoring mode with a trap current of 200 μA. This method separated the 209 congeners into about 170 peaks. PCB congener concentrations were corrected for surrogate recoveries. Quality assurance measures are further described in the Supporting Information.

Toxicity Assessment. In the subacute experiment, bronchoalveolar lavage (BAL) fluid was collected, processed, and used for enumeration of total and differential cell counts and analysis of total protein, lactate dhydrogenase (LDH) activity, and cytokine levels as previously described (*25*).

Total protein was determined using the Bradford protein assay with bovine serum albumin as the standard (Bio-Rad Laboratories, Hercules, CA). LDH activity released from the cytosol of damaged cells was measured spectrophotometrically (Roche Diagnostics, Indianapolis, IN).

Cytokine assays were performed using a multiplex suspension array system (BioRad, Hercules, CA), including 10 selected cytokines (Invitrogen Corp., Carlsbad, CA). Cytokine concentrations below the valid range of the standard curve were assigned a value imputed from the lower limit of detection (LLOD) divided by $2^{1/2}$ (*26*).

Animals were decapitated, and the upper respiratory tracts were decalcified and trimmed for histologic evaluation (*27*). Lungs, livers, and thymuses were fixed and embedded in paraffin. Sections were stained with hematoxylin and eosin and evaluated by a certified veterinary pathologist (*28*).

Statistical Analysis. Statistical analyses were performed using SAS (version 9.2; SAS, Inc., Cary, NC). Summary data are generally expressed as the arithmetic mean and standard error. Two-way analysis of variance (ANOVA) with time as a repeated measure was used to determine differences in body weight gain between PCB-exposed and sham groups. Two sample t-tests for equal or unequal variances were used to compare other measurements between exposure and control groups. In all analyses, a p value < 0.05 was considered significant.

Results

Characterization of PCB Atmospheres. The performance of the generation system was evaluated by characterization of PCBs collected with XAD cartridges. The more volatile congeners were well represented in our generated vapor mixture, while hexa-, hepta-, octa-, and nonachlorobiphenyls were barely detected (Figure 2A). The lower-chlorinated congeners (mono-, di-, and trichlorobiphenyls) represented 90% of the total PCB load. PCB 1, 4, 6, 8, 15, 17, 18 + 30, 20 + 28, 21 + 33, 31, and 52 (+ indicates coelutions that are quantified as the sum of the congeners listed) were most abundant by mass, accounting for 83 ± 1% of total as compared to the much lower prevalence in the source material, Aroclor 1242 (41%). Our generation system showed

TABLE 1. Mean ± Standard Error of Total PCB Levels (ng/g lipid weight) in Liver, Lung, and Blood from Rats after Subacute Inhalation Exposure[a]

total PCBs	sham $n = 9$	10 day exposure $n = 7$	4 day exposure $n = 2$
liver	44.1 ± 24.7	6681.3 ± 324.7***	5965.2
lung	9.1 ± 2.4	6714.2 ± 1122.3**	1190.7
blood	0.21 ± 0.03	18.0 ± 2.5***	18.9

[a] Levels in the 10 day exposed group were significantly greater than the control group: ** $p < 0.01$ and *** $p < 0.001$ (t-test for unequal variances).

a high degree of consistency in total concentration and in profile distribution of congeners in airborne vapor production (Figure 2B).

Subacute Exposure. Rats exposed to a high concentration of PCB vapor mixture for 10 days experienced an average concentration of total PCBs of 8.2 ± 0.5 mg/m^3 (concentrations each day: 6.33, 5.46, 9.28, 7.31, 8.07, 8.53, 8.81, 9.90, 9.00, and 9.75 mg/m^3). Assuming a breathing frequency of 95 breaths/min, a tidal volume of 1.5 mL/breath, and complete uptake of inhaled PCBs, we would predict that each rat was nominally exposed to 1400 μg PCBs. Sham-exposed rats had low but detectable levels of PCBs in their tissues perhaps from dietary intake or low levels of PCBs in indoor air. However, exposed rats had PCB levels from 80-fold (blood) to 700-fold higher (lungs) than sham-exposed animals. Rats exposed for 4 days accumulated 18% as much in the lungs as 10 day exposed rats (Table 1), yet almost the same amount (89%) in livers as 10 day exposed rats. Much larger amounts of PCBs were found in liver and lung than in blood. PCB 20 + 28, 49 + 69, 52, 60, 61 + 70 + 74 + 76, 66, 83 + 99 + 112, 85 + 116 + 117, 90 + 101 + 113, 105, and 118 were leading congeners in the tissues (Table S1 of the Supporting Information). Toxic equivalency (TEQ) concentration, an estimate of the total 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD)-like activity, was calculated according to the re-evaluated toxic equivalency factors (TEF) (*29*). After exposure to the vapor mixture (TEQ = 3.7 ± 0.5 ng/m^3), the detected congeners with a TEF value included PCB 77, 81, 105, 114, 118, 123, and 126. The highest TEQ concentration was found in liver as 1464 pg/g lipid weight (l. w.) and much less in lung (11 pg/g l. w.) and blood (0.04 pg/g l. w.).

The average weight gain of the PCB-exposed animals was significantly lower than sham animals (Figure S1 of the Supporting Information, repeated measures ANOVA: $p < 0.01$, effect of time × treatment). However, BAL fluid macrophages, neutrophils, and lymphocytes were not significantly changed. Total protein and LDH activity measured did not show any significant difference (Table S5 of the Supporting Information), and this also applied to the measured cytokines (Table S6 of the Supporting Information). Histologic evaluation of the respiratory system and nonrespiratory tissue showed unremarkable or minimal changes that were not treatment related.

Time Course of PCB Distribution and Elimination in Rat Tissue. Following an acute exposure to PCB vapor mixture, we determined all congener level and distribution profiles at 5 postexposure time points. The total concentration of PCB congeners to which the animals were exposed was 2.4 mg/m^3. Each rat was estimated to inhale 40 μg PCBs. PCB concentrations were in the same range in lung, liver, and adipose tissue, lower in brain, and lowest in blood on the entire time course of 12 h (Figure 3). The sum of total PCB congeners (∑PCB) loaded in lung, liver, brain, and blood started decaying immediately after exposure in a first-order fashion, while PCBs in adipose tissue accumulated before reaching a relatively steady concentration at 3 h post exposure. The rate of elimination varied moderately among livers, lungs, brains, and blood, with biological half-lives of ∑PCB increasing in the order of liver (5.6 h) < lung (8.2 h) < brain (8.5 h) < blood (9.7 h).

Congeners that were found ≥10-fold higher in most exposed tissue samples compared to those of the background levels of sham/sentinel samples were considered to be reliably detected. Because of the relatively low concentrations in blood, congeners with ≥2-fold higher levels were also reported (Figures S1−S28 of the Supporting Information). Detected congeners included mostly PCBs with mono- or di-*ortho*-substitution, ranging from mono- to pentachlorobiphenyls (PCB 6, 8, 15, 16, 17, 18 + 30, 20 + 28, 21 + 33, 22, 24, 25, 26 + 29, 31, 32, 37, 49 + 69, 52, 59, 60, 61 + 70 + 74 + 76, 64, 66, 77, 83, 99, 105, 112, and 118), yet the majority fell into tri- and tetrachlorobiphenyl categories (Figure 4). PCB 20 + 28 (Figures 3 and S2D of the Supporting Information) was most abundant in every tissue. Other prevailing congeners included PCB 8 and 21 + 33 in liver, adipose tissue, and brain; PCB 15 in lung; PCB 31 and 66 in all tissues; PCB 49 + 69 in liver; and PCB 52 in adipose tissue (Figures 3 and S2 of the Supporting Information).

Biological half-lives were determined for every congener detected (Table S7 of the Supporting Information). It was apparent that the time course pattern of elimination depended on the individual congener as well as the nature of the target tissue. In liver, the levels of lower-chlorinated congeners decreased immediately after exposure with first-order kinetics as represented by PCB 21 + 33, while higher-chlorinated PCBs exhibited a peak at 1 h post exposure (PCB 60 and 105, Figure 3A), whereas in lung most congeners show a consistently decreasing trend for the entire time course (Figure 3B). Congeners behaved similarly in brain, yet some relatively higher-chlorinated congeners (PCB 52 and 118, Figure 3E) tended to show a slight increase within 6 h after exposure. Congeners in blood were generally eliminated less rapidly compared to the above tissues (Figure 3C). In adipose tissue, lower-chlorinated PCBs increased rapidly during the first hour post exposure before reaching a plateau, while higher-chlorinated congeners continued to show a slower increase after 1 h (Figure 3D). The only congeners detected that have a TEF value were PCB 77, 105, and 118. The highest TEQ concentration was found in liver, with a peak at 1 h after inhalation (Figure 5). TEQ concentration increased steadily in adipose tissue for the entire time course, suggesting that inhalation exposure produced a metabolic decrease of TEQ concentration in most tissues and sequestration into adipose tissue.

Discussion

As PCBs in the atmospheric environment contain predominantly the more volatile congeners, administration of commercial mixtures by feeding or injection cannot well represent realistic environmental exposures. We sought to create an inhalation exposure regimen that generates a complex mixture with a congener profile resembling the airshed of urban areas. We evaluated the efficacy of the system by establishing a model that predicts the theoretical concentration and distribution profile of the volatilized mixture (see Supporting Information). The measured concentration in our acute exposure experiment (2.4 mg/m^3) compared favorably with the model prediction of 2.6 mg/m^3. The congener profiles also had a high degree of agreement (data not shown), evidently showing that there was no aerosolization in the process.

In comparison to the air mixture, it is evident that via inhalation the majority of the material sequestered in the tissue shifted considerably from mono- and di- chlorinated PCBs to tri- and tetra- or even higher-chlorinated biphenyls

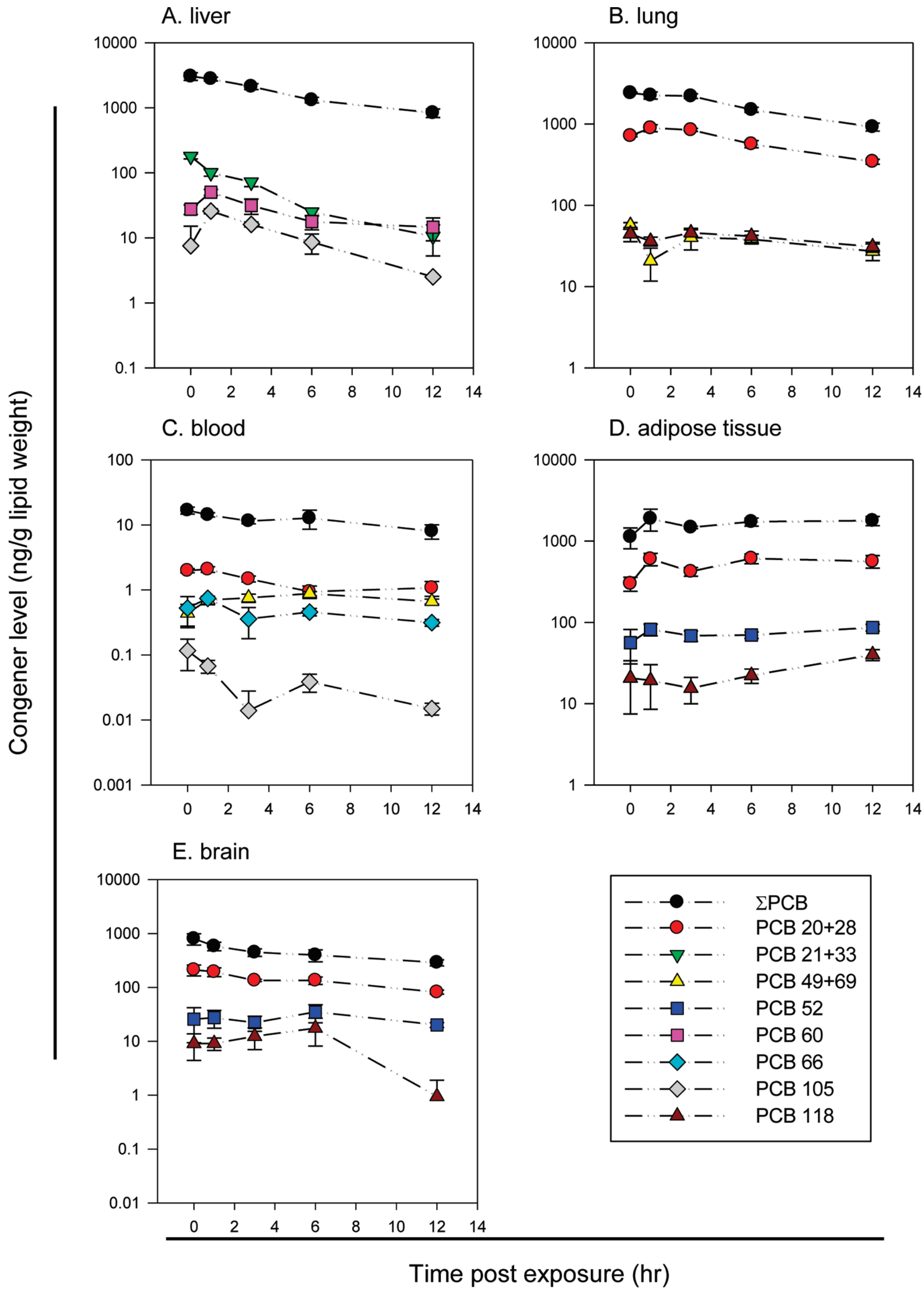

FIGURE 3. Time course of ΣPCB and selected congener concentration in rat liver (A), lung (B), blood (C), adipose tissue (D), and brain (E), following 2 h inhalation exposure.

(Figure 4). One explanation was that most of the lower-chlorinated compounds were rapidly metabolized and did not accumulate appreciably. Our time course study also showed this same shift; after exposure, the proportion of higher-chlorinated PCBs increased along with time in all tissue types, most markedly in organs with metabolic enzyme activity (i.e., liver, lung, and brain) (Figure 4). The time course change of individual congener levels showed clearly that the differential elimination/accumulation rates led to a characteristic congener spectrum.

Most congeners showed biological half-lives of hours in tissues other than adipose tissue, with only a few exceptions in brain (Table S7 of the Supporting Information). The half-lives reported in our study were shorter than those reported in the whole body of Wister rats dosed via the oral route (*30*). This is likely because of rapid uptake and distribution via inhalation. It has been recognized that PCB congeners with vicinal hydrogen substituents are subject to metabolism (*31*, *32*). For nondioxin like congeners, metabolic attack by cytochrome P450 proceeds faster if the open area is at adjacent *meta* and *para* positions (*32*). Therefore, those congeners generally have shorter half-lives. This structure−activity relationship could also be observed in our study, although the impact was less predominant because most lower-chlorinated congeners have open *m*, *p* positions. Congeners with both *para* positions substituted appeared more persistent than those with at least one open

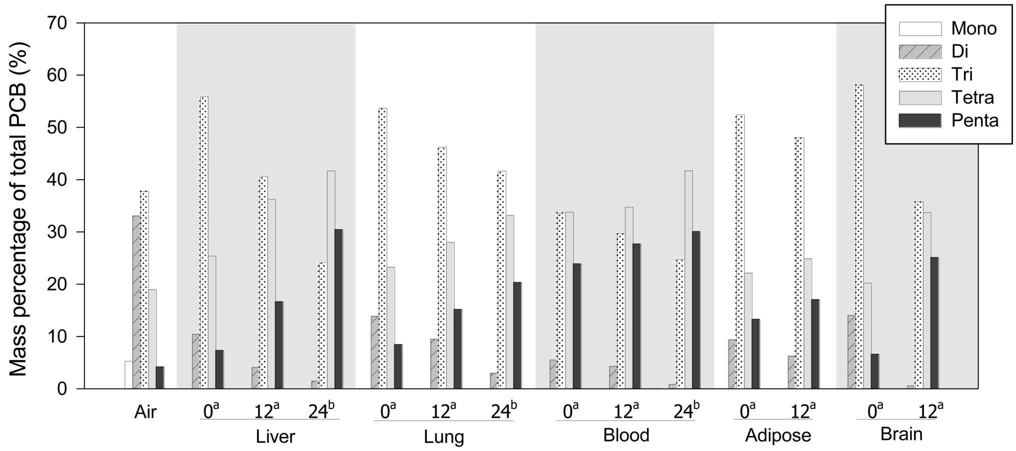

FIGURE 4. Comparison of congener distribution profiles in generated vapor mixture and tissue load at different time points post exposure as sorted by mono-, di, tri-, tetra-, and pentachlorobiphenyls. [a]Acute exposure (time course experiment), hours post exposure for tissue data. [b]Subacute exposure, hours post exposure for tissue data.

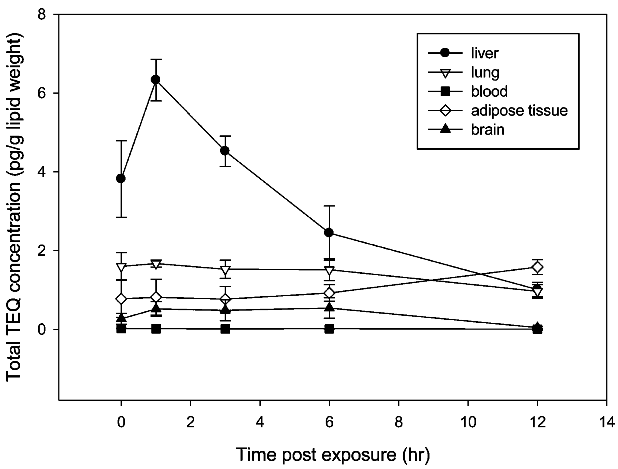

FIGURE 5. Time course of total TEQ concentration change in rat liver, lung, blood, adipose tissue, and brain, following 2 h inhalation exposure.

para position in most type of tissue (Figure S2 of the Supporting Information). However as an exception to the rule, PCB 52 was more persistent in brain than PCB 66, suggesting other factors besides metabolism contribute to the elimination of these compounds.

The less frequent detection and lower concentrations of lower-chlorinated congeners in human and animal tissues do not necessarily mean that the extent of exposure and uptake of these congeners are negligible as suggested by their particularly short half-lives. Nevertheless, many congeners detected in our study have been detected repeatedly among the general population in biomonitoring studies as well as populations exposed to elevated PCB levels: PCBs 49 + 69, 52, 60, 66, 74, 81, 87, 99, 101, 105, and 118 in adult plasma (*33*−*35*); PCBs 28, 31, 52, 60, 64, 70, 74, 99, 105, and 118 in human adipose tissue (*36*−*38*); and PCBs 28, 66, 74, 99, and 118 in human breast milk and infant cord serum (*39*). Yet our study also provides basic information for organs that are less accessible. For instance, PCB 15 was found in high concentrations in lung, and PCB 52 appeared particularly persistent in brain (Figure S2 of the Supporting Information).

Inhalation was a highly efficient route of exposure for uptake of atmospheric PCBs. The first-order kinetics of ∑PCB in most tissues indicates that PCBs gain rapid access to circulation via the lung and can be soon distributed into tissues. In contrast, oral administration is usually characterized by a delayed peak in tissue concentration hours after exposure (*40*). Following inhalation, the redistribution into adipose tissue and subsequent accumulation were observed. The slow rise in fatty tissue level was dwarfed by the rapid decline of PCBs in all other organs and blood, suggesting that metabolism rather than enrichment in adipose tissue contributed primarily to the clearance of the inhaled PCBs. It was noteworthy that the higher-chlorinated congeners accumulated in liver more than the same congeners in lung, whereas the levels of lower-chlorinated congeners were found consistently higher in the latter (Figure 4 and Table S4 of the Supporting Information). In fact, this discrepancy between profiles in lung and liver was already noticeable at 12 h post acute exposure even though they were very similar when exposure ended. Brain tissue was also preferentially taking up higher-chlorinated congeners. One possible explanation was that the higher-chlorinated congeners, which are generally more lipophilic, tend to stay in lipid-rich organs, the liver, brain, and adipose tissue being main sites of their metabolism and storage rather than lung, which was the route of entry.

The animals were exposed to a relatively high dose of PCBs, estimated from the inhaled concentration in air as 40 μg per rat. The amount of PCBs measured in the five tissues collected was 5 μg per rat. The calculation for the measured body burden (i.e., the sum of PCBs loaded at the end of exposure) based on the organ weights of lung, liver, and brain and the assumption that serum and adipose tissue respectively account for 3% of the total body mass (*41*) divided by the fractional body mass of these organs yields an estimated body burden of 33 μg per rat. The difference between the exposure and body burden can be ascribed to several plausible explanations: (1) PCBs likely do not distribute equally to all organs and tissues (*42*). (2) Excretion/metabolism during exposure time (3 h) could contribute considerably to the loss. (3) Exhalation of PCB vapor by animals was likely but not quantified.

TEQ concentrations were calculated to evaluate the potential toxicity of this inhalation exposure on the basis of accumulated parent congeners in tissue. Among dioxin-like congeners, only PCBs 77, 105, and 118 were detected after 2 h inhalation. As the exposure was prolonged, more dioxin-like congeners accumulated to reach the lower detection limit. The much higher TEQ concentration in liver than that in lung resulted primarily from the absence of PCB 126 in lung. A significant concentration change in liver was also seen shortly after acute exposure (Figure 5). We observed that these dioxin-like congeners accumulated in liver, which would thus be susceptible to toxic effects from inhaled PCBs. Measurement of metabolites of the dioxin-like congeners would help to elucidate the true burden of this exposure. In this study, we did not observe any significant toxicity, neither histologic abnormalities in respiratory system or in liver, or noninflammatory responses in the lung. However, it should be realized that the biological end points investigated in this study were relatively limited, and the exposure periods were short. As the toxicological potential of lower-chlorinated PCBs have been revealed and inhalation is shown to be a significant route of exposure to these compounds, new biological end points for inhalation exposure to airborne PCBs are needed as well as chronic exposure studies to address the question of disease from inhaled PCB mixtures.

Acknowledgments

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Appendix A

Abbreviations

ANOVA	analysis of variance
BAL	bronchoalveolar lavage
d-PCB 65	deuterium-labeled PCB 65
eV	electronvolt
GC	gas chromatography, gas chromatograph
LDH	lactate dehydrogenase
l. w.	lipid weight
MS	mass spectrometry
OPR	ongoing precision and recovery
PCB	polychlorinated biphenyl
TCDD	2,3,7,8-tetrachlorodibenzo-*p*-dioxin
∑PCB	sum of total PCB congeners, ng/g lipid weight

Note Added after ASAP Publication

This paper was published ASAP on August 10, 2010. Author affiliations for D.H. and K.C.H. were updated. The revised paper was reposted on August 30, 2010.

Supporting Information Available

Chemicals, extraction procedure, volatilization model, and quality control; tables for quality assurance measures, congener uptake, and immune response in subacutely exposed rat tissue, and biological half-lives of detected congener in acutely exposed rat tissue; and figures for rat weight gain and time course of congener concentration change in acutely exposed rat tissue. This material is available free of charge via the Internet at http://pubs.acs.org.

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