Humans and animals are exposed daily to a complex mixture of polyhalogenated aromatic hydrocarbons (PHAHs). Previous work has shown that exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) is associated with a dose-dependent increase in the incidence and severity of endometriosis in the rhesus monkey. Dioxin-like chemicals can also exert effects in combination with TCDD via the aryl hydrocarbon receptor. This study demonstrates that the serum levels of TCDD and specific dioxin-like PHAH congeners were increased in TCDD-treated animals with endometriosis 13 years after the TCDD exposure. Nine TCDD-exposed and 6 unexposed female rhesus monkeys were evaluated for serum content of relevant compounds and for endometriosis by surgical laparoscopy. Additional studies were done on 4 animals that died 7 to 11 years after exposure to TCDD and 4 lead-treated animals with no history of PHAH treatment. For TCDD-exposed and unexposed animals, TCDD exposure correlated with an increased serum TCDD concentration. Furthermore, TCDD exposure and an elevated serum TCDD concentration were associated with increased serum levels of triglycerides, 1,2,3,6,7,8-hexachlorodibenzo-furan, 3,3',4,4'-tetrachlorobiphenyl (TCB) and 3,3',4,4',5-pentachlorobiphenyl (PnCB). Importantly, the animals with elevated serum levels of 3,3',4,4'-TCB, 3,3',4,4',5-PnCB and an increased total serum TEQ had a high prevalence of endometriosis, and the severity of disease correlated with the serum concentration of 3,3',4,4'-TCB. Increased serum concentrations of coplanar PCBs were also present in lead-treated animals. Implications of these findings for human health and the prevalence of endometriosis in humans will be discussed.

Key Words: endometriosis; rhesus monkey; environmental toxicants; dioxin; TCDD; PCB; dioxin-like chemicals.

2,3,7,8-Tetrachlorodibenzo-p-dioxin (TCDD) and structurally related dioxin-like chemicals are industrial compounds or combustion byproducts, which are worldwide environmental contaminants. TCDD is a potent chemical toxicant, which has been used as a reference compound for polyhalogenated aromatic hydrocarbons (PHAHs) (Poland and Knutson, 1982). The biochemical and toxic effects of TCDD are mediated by binding to the aryl hydrocarbon receptor (AhR). The AhR acts as a signal transducer and transcription factor for target genes, which include cytochrome P450 and growth regulatory genes involved in proliferation, inflammation, and differentiation (Denison et al., 1996; Sutter et al., 1991; Whitlock, 1990). Other polychlorinated dibenzo-p-dioxins (PCDDs) and dibenzofurans (PCDFs) substituted in all 4 lateral positions also have high binding affinity for the AhR and elicit toxic effects similar to TCDD (Safe, 1990). Moreover, certain non-ortho coplanar polychlorinated biphenyls (PCBs) substituted in both para (4,4') and 2 or more meta (3,3',5,5') positions are AhR agonists. Differences in the potency between various chlorinated congeners of dioxins, furans, and biphenyals have been shown to relate to the affinity of individual congeners for the AhR (Safe, 1990).

Humans, wildlife, and laboratory animals are exposed daily to a complex mixture of TCDD and dioxin-like chemicals primarily via trace amounts present in food (DeVito et al., 1995; Vanden Heuvel et al., 1994). TCDD and related PHAH congeners are found in serum and accumulate in tissues of exposed individuals and the general population (DeVito et al., 1995; U. S. EPA, 1991; Safe, 1990; Schecter et al., 1994a,b; Vanden Heuvel et al., 1994). Although TCDD is a known carcinogen and teratogen in rodents (Kociba et al., 1978), its effect in humans is less clear. It has been postulated that increased concentrations of TCDD and dioxin-like chemicals in blood and tissues may disrupt endocrine and immune responses in susceptible humans and animals (Clark et al., 1992; DeVito et al., 1995). In addition, TCDD is associated with a dose-dependent increase in the incidence and severity of endometriosis in TCDD-exposed rhesus monkeys 10 years after termination of exposure (Rier et al., 1993). Endometriosis is a disease characterized by the growth of endometrial cells at...
extrauterine sites whose pathogenesis may involve immune and endocrine dysregulation (Rier and Yeaman, 1997).

In this study, serum levels of TCDD and dioxin-like PHAHs were analyzed in a group of rhesus monkeys 13 years after termination of exposure to TCDD. Serum concentrations of TCDD, PCDDs, PCDFs, and PCBs were measured, and the data were analyzed for an association with the presence and severity of endometriosis. Elevated serum levels of TCDD-related compounds correlated strongly with both TCDD exposure and endometriosis. Implications of these results are discussed.

**MATERIALS AND METHODS**

All experimental protocols using rhesus monkeys were performed in accordance with regulations in the "Guide for Care and Use of Laboratory Animals" and the amended Animal Welfare Act (7 USC 2131 et seq.). Animal protocols were approved by the Animal Review Committee of the University of Wisconsin. Peripheral blood was obtained from animals by femoral venipuncture from animals anesthetized with ketamine HCl (10mg/kg). To obtain the required amount of serum, blood was drawn prior to morning feeding on 3 separate occasions at 1-week intervals. A total volume of 10 ml serum was frozen at −70°C until use. Serum samples (1 ml) also were obtained from 4 of 5 TCDD-treated animals that died 7 to 11 years post-TCDD treatment. These samples were routinely obtained at autopsy and stored at −70°C.

**Rhesus study population and methods.** Detailed methods for TCDD treatment of rhesus monkeys were reported previously (Schantz et al., 1986; Schantz and Bowman, 1989; Bowman et al., 1989; Rier et al., 1993). In brief, 24 feral female rhesus monkeys (Macaca mulatta) approximately 6 to 10 years of age were obtained in 1977 (Hazleton Research Animals, Reston, VA). The animals were maintained under climate conditions that simulated their natural breeding season at University of Wisconsin Biotron. Monkeys were randomly assigned to 3 groups of 8 animals each. Control animals (0 PPT TCDD) were not exposed to TCDD, animals in the low-dose group were exposed to 5 parts per trillion (PPT) TCDD, and monkeys in the high-dose group were exposed to 25 PPT TCDD for approximately 4 years. The purity of the TCDD dosing solution (Dow Chemical, Midland, MI) was 98%. Dosing was by ingestion in the animal feed. Animals exposed to 0, 5, or 25 PPT TCDD were housed in a facility with its own air supply, which was separated from other animals at the time of surgery. The presence and severity of endometriosis was determined previously in these monkeys (Rier et al., 1993). In the 10 years between TCDD exposure and the present study on endometriosis in these animals (Rier et al., 1993), none of the TCDD-treated or 0 PPT animals had hysterotomies or gave birth by caesarian section. In addition, none of these animals underwent laparoscopy or laparotomy during this time period, with the exception of 3 animals in the 25 PPT group for diagnosis of severe endometriosis; 2 of these animals died due to intestinal obstruction by endometriosis.

Thirteen years after termination of TCDD treatment, serum concentrations of TCDD and dioxin-like PHAHs were determined in the 15 surviving animals from this colony (exposed, n = 9, and unexposed, n = 6). These 15 animals were grouped prospectively according to TCDD exposure: 0 PPT TCDD (n = 6), 5 PPT TCDD (n = 6), 25 PPT TCDD (n = 3) and ALL TCDD (n = 9). Animals were grouped retrospectively as follows: No TCDD and no endometriosis (−TCDD−EM, n = 4), no TCDD with endometriosis (−TCDD+/EM, n = 2), TCDD exposure and no endometriosis (+TCDD−EM, n = 2) and TCDD exposure with endometriosis (+TCDD+/EM, n = 7). Serum PHAH concentrations were also determined in 4 deceased TCDD-treated animals that died due to complications from endometriosis 7 (n = 1), 10 (n = 1) and 11 (n = 2) years after termination of exposure. A fifth TCDD-treated animal died during this time period postexposure; however, serum was not available from this animal. These TCDD-treated animals lived for a shorter period of time post-termination of TCDD exposure, therefore, these findings are reported separately from the 15 TCDD-exposed and unexposed animals referred to above.

Since exposure to environmental PHAHs may have occurred at Harlow Primate Laboratory via the feed or other environmental source, we sought to determine serum PHAH levels in other live, aged feral animals at this facility. Thus, serum PHAH concentrations were evaluated in 4 live, feral, lead-exposed rhesus monkeys of similar age (24 to 28 years) residing at Harlow Primate Laboratory for a similar period of time (12 to 18 years) as TCDD-treated animals (unexposed feral, live animals meeting age and residence criteria were not available). Results from lead-treated animals are reported separately from the 15 TCDD-exposed and unexposed animals referred to above. Lead was administered chronically in water at intervals over a period of 8 years and terminated 11 years prior to the initiation of this study. The postexposure diet of animals treated with lead or TCDD consisted of the same brand of monkey chow. TCDD-treated animals and lead-exposed monkeys participated in a series of similar studies on the effects of toxicant exposure on behavior and reproductive function (Franks et al., 1989; Laughlin et al., 1983; Laughlin, 1987; Schantz and Bowman, 1989; Schantz et al., 1986).

All live animals included in this study were feral monkeys of similar weight (data not shown). None of these animals experienced rapid weight changes in the 6 months preceding serum collection. The weight of TCDD-treated animals remained stable during dosing and for the past 13 years; animals treated with 25 PPT TCDD weighed 7.3 kg at the time of this study and 7.8 kg (median weights) at termination of TCDD exposure (Bowman et al., 1989). However, TCDD-treated animals that died 7 to 11 years postexposure exhibited symptoms of wasting related to endometriotic intestinal blockage prior to their death, and they weighed significantly less at autopsy than live animals (5.5 kg, median weight). In addition, the age of these animals was approximately 3 years younger than the live animals in these studies.

**Diagnostic laparoscopy.** The presence and severity of endometriosis was determined in 15 animals (0, 5, and 25 PPT TCDD groups) by diagnostic laparoscopy as described (Rier et al., 1993). In brief, surgery was carried out in the facilities of Harlow Primate Laboratory. While under general anesthesia, a 10 mm diagnostic laparoscope was used to inspect the pelvic organs, anterior peritoneum, visible bowel surfaces, the liver edge and the diaphragm. Clinical findings were recorded at the time of surgery. The presence of endometrial glands and stroma in endometrial lesions was confirmed by histological examination at autopsy performed the following year. The severity of endometriosis was determined according to criteria established for humans, using the revised American Fertility Society (rAFS) classification system (American Fertility Society, 1985). This system classifies disease severity according to the number, size, and location of endometriotic implants and the presence of adhesions. The stage of endometriosis is determined by the total number of points assigned to endometriotic lesions and adhesions. Although the severity of endometriosis was determined previously in these monkeys (Rier et al., 1993), it was reevaluated at the time of this study. Disease status was similar at first and second laparoscopy in TCDD-exposed and unexposed animals. Endometriosis was evaluated at surgical laparotomy in 4 lead-treated animals and at autopsy in 4 TCDD-treated animals that died at seven to 11 years post-TCDD exposure.

**Determination of serum levels of TCDD and related compounds.** Serum concentrations of TCDD and dioxin-like PCDD, PCDF, and PCB congeners were measured for an association with the presence and severity of endometriosis. Elevated serum levels of TCDD-related compounds correlated strongly with both TCDD exposure and endometriosis. Implications of these results are discussed.
were determined by high resolution gas chromatography and mass spectrometry, as previously described (DiPietro et al., 1997). Standards were obtained from Accustandard (NewHaven, CT) and Cambridge Isotope Labs, Inc. (Andover, MA). All serum analyte concentration measurements met quality assurance (QA) or quality control (QC) parameters. Serum lipid concentrations were quantified as described (DiPietro et al., 1997). Serum analyte values were not reported if QA or QC parameters were not met. Chlorinated dibenzo-dioxins (-CDD), dibenzo furans (-CDF), and biphenyl (-CB) congeners were quantified in rhesus serum (see Table 1).

TCDD and dioxin-like PHAH levels determined in serum and fat have been reported to be equivalent when the serum PHAH level is adjusted by the total lipid content in the serum (Patterson et al., 1988; Pirkle et al., 1989). However, TCDD exposure may be associated with increased serum lipids (Calvert et al., 1996). Thus, data is presented both on a parts-per-quadrillion (PPQ), whole-weight basis and on a PPT lipid-adjusted basis (PPT-Lipid, lipid-adjusted weight). Data were collected from TCDD-treated and untreated monkeys 13 years following the termination of treatment as previously described (Rier et al., 1993). Tables 2 and 3 show the serum concentration of TCDD and related compounds in both TCDD-exposed and unexposed animals; Table 2 presents PPQ and lipid-adjusted PPQ-Lipid values, and Table 3 presents the corollary TEQ and lipid-adjusted TEQ (TEQ-Lipid) values in TCDD equivalents. The TCDD-treated group includes animals dosed at either 5- or 25 PPT TCDD and is referred to as “all TCDD.” When appropriate, specific correlations were made for each group using their individual cumulative TCDD dose as described in the text and Figures 1 and 3.

A significant increase was observed in the serum concentration of TCDD in animals with TCDD exposure (4.6 PPQ, unexposed; 23.6 PPQ, exposed). For these animals, the cumulative TCDD dose correlated directly with the serum TCDD concentration ($p < 0.01$; data not shown). However, the change in the serum TCDD concentration was significant in the 25 but not the 5 PPT TCDD dosage group (Fig. 1A). In addition to TCDD, dioxin-like PCDDs and PCDFs were detected in both TCDD-treated and unexposed monkeys (Tables 2 and 3). However, the serum concentrations of total PCDD and PCDF compounds were not significantly different between unexposed and exposed animals (PCDD, 172 vs. 532 PPQ; PCDF, 55 vs. 97 PPQ). The most abundant PCDD congener in serum was 1,2,3,4,6,7,8 HxCDD (117 PPQ, unexposed; 165 PPQ, exposed). The most abundant PCDF congener was 2,3,7,8-TCDF, 2,3,4,7,8-PnCDF, and 1,2,3,4,7,8-HxCDF (12–17 PPQ, all animals). One furan PCDF congener, 1,2,3,6,7,8-HxCDF, was increased 2- to 3-fold in animals treated with 5 PPT TCDD (10.9 PPQ), 25 PPT TCDD (14.3 PPQ), and all TCDD (11.1 PPQ) relative to 4.4 PPQ in control animals (Fig. 1B and Table 2). Cumulative TCDD dose and serum TCDD level were directly correlated with an elevated serum level of 1,2,3,6,7,8 HxCDF ($p = 0.014$, $p = 0.001$, respectively; data not shown).

Coplanar PCBs were also detected in the serum of all animals (Tables 2 and 3). The total serum PCB concentration increased in TCDD-exposed animals (1961 PPQ, exposed; 147

## Table 1

<table>
<thead>
<tr>
<th>-CDD</th>
<th>-CDF</th>
<th>-CB</th>
</tr>
</thead>
<tbody>
<tr>
<td>2,3,7,8-tetra-CDD (TCDD)</td>
<td>2,3,7,8-tetra-CDF</td>
<td>3.3',4,4'-tetra-CB</td>
</tr>
<tr>
<td>1,2,3,7,8-penta-CDD</td>
<td>1,2,3,7,8-penta-CDF</td>
<td>3.4,4',5-tetra-CB</td>
</tr>
<tr>
<td>1,2,3,7,8,9-hexa-CDD</td>
<td>2,3,4,7,8-penta-CDF</td>
<td>3.3',4,4',5,5'-penta-CB</td>
</tr>
<tr>
<td>1,2,3,4,6,7,8-hepta-CDD</td>
<td>1,2,3,4,7,8-hexa-CDF</td>
<td>3.3',4,4',5,5'-hexa-CB</td>
</tr>
<tr>
<td>1,2,3,4,6,7,9-hepta-CDD</td>
<td>1,2,3,7,8,9-hexa-CDF</td>
<td>3.3',4,4',5,5'-hexa-CB</td>
</tr>
<tr>
<td>octa-CDD</td>
<td>2,3,4,6,7,8-hexa-CDF</td>
<td>3.3',4,4',5,5'-hexa-CB</td>
</tr>
<tr>
<td></td>
<td>1,2,3,4,6,7,8-hepta-CDF</td>
<td></td>
</tr>
<tr>
<td></td>
<td>1,2,3,4,7,8,9-hepta-CDF</td>
<td></td>
</tr>
<tr>
<td></td>
<td>octa-CDF</td>
<td></td>
</tr>
</tbody>
</table>
PPQ unexposed). The serum concentrations of 3,4,4',5-TCP and 3,3',4,4',5,5'-HxCB were not elevated in TCDD-exposed animals; in contrast, the serum concentration of 3,3',4,4'-TCP was elevated in TCDD-exposed animals. This dioxin-like PCB was not detected in serum from animals without previous TCDD exposure. In sharp contrast, a high level of 3,3',4,4'-TCP was found in TCDD-treated animals (5 PPT TCDD, 984 PPQ; 25 PPT TCDD, 1584 PPQ; see Fig. 1C). Moreover, the serum concentration of 3,3',4,4',5-PnCB was increased 3- to 5-fold in TCDD-treated animals in a dose-dependent manner (Fig. 1D). The cumulative dose of TCDD was directly correlated with the serum concentration of 3,3',4,4',5-PnCB ($p = 0.007$; data not shown). Furthermore, the serum TCDD level was directly correlated with the serum concentration of 3,3',4,4'-TCP and 3,3',4,4',5-PnCB ($p = 0.0024$, $p = 0.0004$, respectively; data not shown).

Different relative proportions of the PCDD, PCDF, and PCB congeners were present in exposed and unexposed animals (Table 2). In unexposed animals, 1,2,3,4,6,7,8-HpCDD, 3,3',4,4',5-PnCB and 3,4,4',5-TCP are the most abundant, accounting for 38%, 29%, and 7% of total serum PHAHs, respectively. In exposed animals, the congener 3,3',4,4'-PCB was the most abundant (57%) while 1,2,3,4,6,7,8-HpCDD, 3,3',4,4',5-PnCB and 3,4,4',5-TCP contribute 6%, 14% and 4%, respectively. Although the serum concentrations of TCDD and 1,2,3,6,7,8-HxCDF were increased in TCDD-exposed animals, each of these congeners comprise a small proportion (2% or less) of total serum PCB and PHAH compounds in TCDD-treated animals. PCB congeners comprise 77% of all PHAH compounds in TCDD-exposed animals.

### TABLE 2

<table>
<thead>
<tr>
<th>Congener</th>
<th>0 PPT TCDD</th>
<th>All TCDD</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n</td>
<td>PPQ</td>
</tr>
<tr>
<td>2,3,7,8-TCPD</td>
<td>6</td>
<td>6.2 (2.9)</td>
</tr>
<tr>
<td>1,2,3,7,8-PnCDD</td>
<td>4</td>
<td>9.9 (3.5)</td>
</tr>
<tr>
<td>1,2,3,7,8-HxCDD</td>
<td>6</td>
<td>ND$^b$</td>
</tr>
<tr>
<td>1,2,3,4,6,7,8-HxCB</td>
<td>6</td>
<td>129 (16)</td>
</tr>
<tr>
<td>1,2,3,4,6,7,9-HxCB</td>
<td>6</td>
<td>ND$^d$</td>
</tr>
<tr>
<td>OCDD</td>
<td>6</td>
<td>51.4 (51)</td>
</tr>
<tr>
<td>Total PCDDs</td>
<td>4</td>
<td>237 (87)</td>
</tr>
<tr>
<td>2,3,7,8-TCPD</td>
<td>5</td>
<td>12.1 (3.1)</td>
</tr>
<tr>
<td>1,2,3,7,8-PnCDD</td>
<td>4</td>
<td>2.3 (2.3)</td>
</tr>
<tr>
<td>1,2,3,4,7,8-HxCB</td>
<td>4</td>
<td>13.2 (1.6)</td>
</tr>
<tr>
<td>1,2,3,4,6,7,8-HxCB</td>
<td>4</td>
<td>14.8 (0.7)</td>
</tr>
<tr>
<td>Total coplanar PCBs</td>
<td>6</td>
<td>4.6 (2.1)</td>
</tr>
<tr>
<td>OCDF</td>
<td>6</td>
<td>ND$^d$</td>
</tr>
<tr>
<td>Total PCDFs</td>
<td>4</td>
<td>49 (14)</td>
</tr>
<tr>
<td>3,3',4',5'-TCP</td>
<td>6</td>
<td>ND$^d$</td>
</tr>
<tr>
<td>3,4',5'-TCB</td>
<td>6</td>
<td>38.3 (13)</td>
</tr>
<tr>
<td>3,3',4',5'-PnC</td>
<td>6</td>
<td>109 (9)</td>
</tr>
<tr>
<td>3,3',4',5,5'-HxCB</td>
<td>6</td>
<td>ND$^d$</td>
</tr>
<tr>
<td>Total coplanar PCBs</td>
<td>6</td>
<td>147 (20)</td>
</tr>
<tr>
<td>Total PCDD/PCDF/PCBs</td>
<td>4</td>
<td>426 (96)</td>
</tr>
</tbody>
</table>

$^a$ Note. Results are expressed as mean (SE) median serum analyte concentration in parts per quadrillion (fg/g) on whole weight basis (PPQ) or parts per trillion (pg/g) on a lipid-adjusted basis (PPT-Lipid). % Total PPQs equals the median fraction in percent an individual or group of serum analyte PPQs is relative to Total PHAH PPQs.

$^b$ Determined to be significantly different compared with corresponding value for 0 PPT TCDD group ($p < 0.05$).

$^c$ Serum analyte concentration below level of detection; median detection limits of 21.6 PPQ or 3.6 PPT-Lipid$^d$.

$^d$ For mean (SE) median calculations, 0 was used to designate ND values; median detection limits of 21.6 PPQ or 3.6 PPT-Lipid$^d$.

**TCDD Equivalents for PHAH Congeners in TCDD-Treated Monkeys**

The body burden of TCDD-exposed animals was also evaluated in TCDD equivalents (Table 3) using previously determined...
with an increased incidence and severity of endometriosis in
Serum Levels of TCDD and Other Dioxin-Like Chemicals
TEQ levels, respectively, accounting for similar percentages of
total untreated controls (43.3 total TEQ). PCDDs, PCDFs and PCBs
was elevated in TCDD-treated monkeys (92.5 PPQ) relative to
unexposed animals. Total serum TEQ from all PHAH compounds
2,3,7,8-PnCDF, had a TEQ value of 6 to 8 PPQ in exposed and
and 1,2,3,4,7,8-HxCDF. The most toxic of these congeners,
values were the furan congeners 2,3,7,8-TCDF, 2,3,4,7,8-PnCDF
(9.7 PPQ) while the furan TEQ was not elevated. Among dioxin-
of TCDD and 1,2,3,6,7,8-HxCDF were not significantly dif-
sults are summarized in Figures 1–3 or in the text. Serum levels
additional animals, were evaluated for endometriosis. The re-
of TCDD and unexposed animals (Table 3). TCDD-exposed
ferent in animals with endometriosis, relative to animals with-
observation. All 15 animals reported on above, as well as 8
were increased in TCDD-treated monkeys (92.5 PPQ) relative to
untreated controls (43.3 total TEQ). PCDDs, PCDFs and PCBs
comprised approximately 47–54%, 12–22% and 24–37% of total
TEQ levels, respectively, accounting for similar percentages of
total serum TEQs in TCDD-exposed and unexposed animals.

**Serum Levels of TCDD and Other Dioxin-Like Chemicals**

Previous work indicates that TCDD exposure is associated
with an increased incidence and severity of endometriosis in
the rhesus monkey (Rier et al., 1993). Evaluation of the ani-
mals in this study provides new information relevant to this
observation. All 15 animals reported on above, as well as 8 additional
animals, were evaluated for endometriosis. The results are sum-
arized in Figures 1–3 or in the text. Serum levels of TCDD and
1,2,3,6,7,8-HxCDF were not significantly different in animals with
endometriosis, relative to animals without disease. Thus, elevated serum concentrations of TCDD and
1,2,3,6,7,8-HxCDF resulting from TCDD exposure were not correlated with endometriosis (Figs. 1A and 1B). In contrast,
an increased serum level of the PCB congener 3,3',4,4'-TCB
was associated with the presence of endometriosis (Fig. 1C).

### TABLE 3

<table>
<thead>
<tr>
<th>Congener</th>
<th>TEF</th>
<th>TEQ</th>
<th>TEQ-Lipid</th>
<th>% Total TEQs</th>
<th>TEQ</th>
<th>TEQ-Lipid</th>
<th>% Total TEQs</th>
</tr>
</thead>
<tbody>
<tr>
<td>2,3,7,8-TCDD</td>
<td>1.0</td>
<td>6.2 (2.9)</td>
<td>4.6</td>
<td>1.1 (0.5) 0.8</td>
<td>21</td>
<td>101 (56)</td>
<td>23.6</td>
</tr>
<tr>
<td>1,2,3,7,8-PnCDF</td>
<td>1.0</td>
<td>9.9 (3.5)</td>
<td>12</td>
<td>1.7 (0.7) 1.9</td>
<td>27</td>
<td>16.7 (5.3)</td>
<td>12.9</td>
</tr>
<tr>
<td>1,2,3,7,8-HxCDD</td>
<td>0.1</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>0.6 (0.4) 0</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>1,2,3,4,6,7,8-HpCDF</td>
<td>0.01</td>
<td>1.3 (0.2)</td>
<td>1.2</td>
<td>0.2 (0.0) 0.2</td>
<td>4</td>
<td>2.3 (0.5)</td>
<td>1.6</td>
</tr>
<tr>
<td>1,2,3,4,6,7,9-HpCDF</td>
<td>0.0</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>OCDD</td>
<td>0.0001</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>Total PCDDs</td>
<td>20.5 (6.5)</td>
<td>26.2</td>
<td>3.6 (1.2)</td>
<td>4.1</td>
<td>54</td>
<td>135 (65)</td>
<td>43.3</td>
</tr>
<tr>
<td>2,3,7,8-TCDF</td>
<td>0.1</td>
<td>1.2 (0.3)</td>
<td>1.4</td>
<td>0.2 (0.1) 0.2</td>
<td>3</td>
<td>2.2 (0.9)</td>
<td>1.7</td>
</tr>
<tr>
<td>1,2,3,7,8-PnCDF</td>
<td>0.05</td>
<td>0.1 (0.1) 0</td>
<td>ND'</td>
<td>0</td>
<td>0.4 (0.2) 0.5</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>2,3,4,7,8-PnCDF</td>
<td>0.5</td>
<td>6.6 (0.8)</td>
<td>6.2</td>
<td>1.1 (0.2) 1.2</td>
<td>13</td>
<td>10.2 (2.7)</td>
<td>8.2</td>
</tr>
<tr>
<td>1,2,3,4,7,8-HxCDF</td>
<td>0.1</td>
<td>1.5 (0.1)</td>
<td>1.5</td>
<td>0.3 (0.0) 0.2</td>
<td>3</td>
<td>2.1 (0.6)</td>
<td>1.5</td>
</tr>
<tr>
<td>1,2,3,6,7,8-HxCDF</td>
<td>0.1</td>
<td>0.5 (0.2)</td>
<td>0.5</td>
<td>0.1 (0.0) 0.2</td>
<td>1</td>
<td>1.3 (0.3)</td>
<td>1.1</td>
</tr>
<tr>
<td>1,2,3,7,8,9-HxCDF</td>
<td>0.1</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>2,3,4,6,7,8-HxCDF</td>
<td>0.1</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>0.5 (0.2) 0</td>
<td>0 (1) 0</td>
<td>0</td>
</tr>
<tr>
<td>1,2,3,4,6,7,8-HpCDF</td>
<td>0.01</td>
<td>0.1 (0) 0</td>
<td>ND'</td>
<td>0</td>
<td>0.3 (0.1) 0.2</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>1,2,3,4,7,8,9-HpCDF</td>
<td>0.01</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>OCDF</td>
<td>0.001</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>Total PCDFs</td>
<td>9.5 (1.5)</td>
<td>10.5</td>
<td>1.7 (0.3)</td>
<td>1.8</td>
<td>22</td>
<td>16.9 (4.8)</td>
<td>13.7</td>
</tr>
<tr>
<td>3,3',4',5'-TCB</td>
<td>0.0001</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>0.2 (0.1) 0.2</td>
<td>ND'</td>
<td>&lt;1</td>
</tr>
<tr>
<td>3,3',4',5'-PnCB</td>
<td>0.0001</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
<td>ND'</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>3,3',4',5'-HxCB</td>
<td>0.1</td>
<td>10.9 (0.9)</td>
<td>9.7</td>
<td>1.8 (0.2) 1.6</td>
<td>24</td>
<td>43.3 (10.5)</td>
<td>29.6</td>
</tr>
<tr>
<td>Total coplanar PCBs</td>
<td>0.01</td>
<td>ND</td>
<td>ND</td>
<td>0</td>
<td>0.2 (0.2) 0</td>
<td>ND'</td>
<td>0</td>
</tr>
<tr>
<td>Total PCDD/PCDF/PCBs</td>
<td>40.8 (6.5)</td>
<td>43.3</td>
<td>7.1 (1.5)</td>
<td>6.8</td>
<td>24</td>
<td>43.8 (10.7)</td>
<td>29.8</td>
</tr>
<tr>
<td>199 (73) 92.5</td>
<td>19.6 (4.4)</td>
<td>10.0</td>
<td>10.0</td>
<td>10.0</td>
<td>24</td>
<td>199 (73) 92.5</td>
<td>19.6 (4.4)</td>
</tr>
</tbody>
</table>

* Results are expressed as mean (SE) median serum analyte TCDD equivalents (TEQs) in parts per quadrillion (fg/g) on whole weight basis (PPQ) or parts per trillion (pg/g) on a lipid-adjusted basis (PPT-Lipid). TEF, toxic equivalency factor according to values assigned by Van den Berg et al. 1998. % Total TEQs equals the median fraction in percent an individual serum analyte TEQ is relative to total PCDH TEQs.

a Determined to be significantly different when compared with corresponding value for the 0 PPT TCDD group; p < 0.05.

b Serum analyte concentration below level of detection. ND also designates serum levels less than 0.1 PPT-Lipid following TEQ calculations.
with (95.2 PPQ) and without disease (114 PPQ; Figure 1D). An increase in serum 3,3′,4,4′,5-PnCB concentration also was observed in TCDD-treated animals that did not have endometriosis (437 PPQ), therefore, the elevation in serum 3,3′,4,4′,5-PnCB correlated with TCDD exposure rather than disease. However, the animals with elevated serum concentrations 3,3′,4,4′-TCB and 3,3′,4,4′,5-PnCB had a high prevalence of endometriosis.

Increased serum concentrations of these coplanar PCBs were also present in TCDD-treated animals with endometriosis, which had died prior to the initiation of this study. The same increased concentrations occurred in lead-treated animals housed at the same facility with no previous PHAH treatment. TCDD-treated animals that died from complications due to endometriosis seven to 11 years postexposure had an elevated serum level of 3,3′,4,4′-TCB (332 PPQ, exposed; unexposed, not detectable) and 3,3′,4,4′,5-PnCB (1132 PPQ exposed; 96.7 PPQ unexposed). Lead-treated animals also had an elevated level of the same PCB compounds (3,3′,4,4′-TCB, 1248 PPQ; 3,3′,4,4′,5-PnCB, 475 PPQ; control values as above). Three of 4 of these lead-treated animals had endometriosis that was visualized at first surgical laparotomy (N. Laughlin, personal communication). Thus, for these, as well as the TCDD-treated animals, a correlation exists between the incidence of endometriosis and elevation of specific PCB congeners.

The total serum PCB concentration was higher in animals with TCDD exposure and endometriosis (2173 PPQ) than in unexposed animals with (115 PPQ) and without (186 PPQ) disease (Fig. 3A), primarily due to increased 3,3′,4,4′-TCB. The total PHAH level was also higher in TCDD-exposed animals with disease (2746 PPQ) than in unexposed controls without disease (334 PPQ). The total serum TEQ from PCB and PHAH compounds increased in TCDD-treated animals with endometriosis, due to the elevated serum level of 3,3′,4,4′,5-PnCB and TCDD (Fig. 3B). These toxic congeners contributed a nearly equal TEQ (22.1 PPQ, TCDD; 29.5 PPQ, 3,3′,4,4′,5-PnCB; the total TEQ derived from these congeners 475 PPQ unexposed). Three of 4 of these lead-treated animals had endometriosis that was visualized at first surgical laparotomy (N. Laughlin, personal communication). Thus, for these, as well as the TCDD-treated animals, a correlation exists between the incidence of endometriosis and elevation of specific PCB congeners.

**Serum Lipid Levels in TCDD-Treated Rhesus Monkeys**

Previous studies indicate that TCDD exposure may be associated with hyperlipidemia and hypertriglyceridemia in rodents (Zinkl et al., 1973) and elevated serum triglycerides in humans (Calvert et al., 1996). In this study, an elevated triglyceride level was found in the serum of TCDD-treated animals (Table 4). The serum triglyceride concentration was increased 3- to 5-fold in animals treated with TCDD (0, 5, and 25 PPT TCDD: 118, 337, and 497 mg/dl, respectively). For most animals, the serum triglyceride level was within the normal range for the female rhesus monkey older than 20 years (less than 415 mg/dl); however, 3 of the TCDD-exposed animals had an above normal triglyceride level (497, 578, and 1150 mg/dl). Cumulative TCDD dose and the serum concentration of TCDD were directly correlated with the increased serum lipid level ($p = 0.003$ and $p = 0.009$) and triglyceride level ($p = 0.001$; data not shown). The serum levels of 1,2,3,6,7,8-HxCDF, 3,3′,4,4′-TCB and 3,3′,4,4′,5-PnCB were also strongly associated with increased serum triglycerides ($p < 0.001$; $p = 0.03$; $p = 0.001$, respectively). In contrast, the serum level of cholesterol (total and free) and phospholipids was unaltered by TCDD exposure (Table 4). Serum triglycerides were 3- to 4-fold higher in TCDD-treated animals with and without endometriosis (323–423 mg/dl) relative to unexposed animals with and without disease (116–128 mg/dl). Thus, increased serum triglycerides correlated with TCDD treatment rather than endometriosis. However, the animals with an elevated serum triglyceride level had a high prevalence of disease.

**DISCUSSION**

The work described here demonstrates that TCDD exposure in the rhesus monkey correlates directly with a long-term
increased in the serum level of PHAH compounds including TCDD, 1,2,3,6,7,8-HxCDF, 3,3’,4,4’,5-PnCB. Importantly, the animals in this study with elevated levels of 3,3’,4,4’,-TCB, 3,3’,4,4’,5-PnCB and an increased total serum TEQ had a high prevalence of endometriosis, and the severity of the disease correlated with the serum concentration of 3,3’,4,4’,-TCB but not the serum TCDD level. The data suggest a potential involvement of an increased body burden of PCB compounds in the etiology of endometriosis in rhesus monkeys. Thus, it is important to consider the implications of this finding for human health, particularly with regard to the average human PHAH body burden and the prevalence of endometriosis in humans.

Humans are exposed daily to mixtures of PHAH chemicals, primarily through food, at estimated levels of 3–6 pg TEQ/kg/day in the U.S. (DeVito et al., 1995; Schecter et al., 1994a,b). TCDD and dioxin-like chemicals are highly stable and lipophilic, leading to bioaccumulation in target tissues and fluids and biomagnification at higher trophic levels of the food chain. Complex mixtures of PHAH congeners differentially bioaccumulate in serum, milk and tissues of animal and human populations (Safe, 1990, 1994; Schecter et al., 1994a,b).

The burden of PCDD, PCDF and total PHAH compounds is higher in the general human population than in the TCDD-treated rhesus monkeys in this study (20-, 5- and 4-fold higher, respectively). The magnitude of this difference is not precisely known, since the lipid-adjusted values used for comparing human and monkey PHAH levels may underestimate the PHAH concentration in the treated monkeys by as much as 2-fold. The following PHAH values evaluated in this study have been reported in blood samples from the general human population: 1361 PPT-Lipid, PCDD; 58 PPT-Lipid, PCDF; 101 PPT-Lipid, PCB; all PHAH, 1499 PPT-Lipid (Kahn et al., 1988; Schecter et al., 1994a,b). In contrast, the total PCB level in human blood is approximately 3-fold less than the level in TCDD-treated monkeys. However, a significant degree of variation is observed in the amounts of PCB compounds in human samples. The following ranges of values have been reported for the PCB congeners of interest in this study in human blood and milk: 3,3’,4,4’,-TCB, 8–490 PPT-Lipid; 3,3’,4,4’,5-PnCB, 40–80 PPT-Lipid; and 3,3’,4,4’,5’,P-HxCB, 40 PPT-Lipid. In human adipose tissue, concentrations of each of these PCB

![FIG. 3. Serum concentration (A) and TEQ (B) of PCDDs, PCDFs, and PCBs in TCDD-treated animals with endometriosis. Results are expressed as median serum PHAH levels. Significant differences between groups were determined using Wilcoxon’s rank sum test. Animal groups for Total PHAH, PCDD and PCDF PPQs (A) and TEQs (B): 0 PPT TCDD n = 4; 5 PPT TCDD n = 4; 25 PPT TCDD n = 3; ALL TCDD n = 7; –TCDD/–EM n = 3; –TCDD/+EM n = 1; +TCDD/+EM n = 2; +TCDD/+EM n = 5. Animal groups for Total PCB PPQ (A) and TEQ (B): 0 PPT TCDD n = 6; 5 PPT TCDD n = 6; 25 PPT TCDD n = 3; ALL TCDD n = 9; –TCDD/–EM n = 4; –TCDD/+EM n = 2; +TCDD/+EM n = 2; +TCDD/+EM n = 7. Indicated by (a): significantly different compared with corresponding value for 0 PPT TCDD controls; p < 0.05; (b): significantly different from corresponding value for –TCDD/–EM group; p < 0.05; (c): significantly different from corresponding value for –TCDD/+EM group; p < 0.05.](https://academic.oup.com/toxsci/article-abstract/59/1/147/1658860)

### TABLE 4

**Serum Lipid Levels in TCDD-Treated Rhesus Monkeys**

<table>
<thead>
<tr>
<th>Lipid</th>
<th>0 PPT TCDD</th>
<th>5 PPT TCDD</th>
<th>25 PPT TCDD</th>
<th>All TCDD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cholesterol, free</td>
<td>41.2 (5)</td>
<td>46.8 (10)</td>
<td>41.8 (4)</td>
<td>45.5 (9)</td>
</tr>
<tr>
<td>Cholesterol, total</td>
<td>164 (16)</td>
<td>166 (43)</td>
<td>183 (58)</td>
<td>172 (46)</td>
</tr>
<tr>
<td>Phospholipids</td>
<td>275 (44)</td>
<td>319 (46)</td>
<td>290 (43)</td>
<td>312 (44)</td>
</tr>
<tr>
<td>Triglycerides</td>
<td>112 (9)</td>
<td>340 (60)</td>
<td>607 (287)</td>
<td>429 (101)</td>
</tr>
<tr>
<td>Total lipids</td>
<td>634 (32)</td>
<td>906 (92)</td>
<td>1158 (330)</td>
<td>990 (120)</td>
</tr>
</tbody>
</table>

*Note. Results are expressed as mean (SE) median lipid concentrations per dl blood.*

* Determined to be significantly different as compared to the 0 PPT TCDD group; p < 0.01.

* Determined to be significantly different as compared to the 0 PPT TCDD group; p < 0.05.
compounds ranges from 30 to 800 PPT-Lipid (Ahlborg et al., 1994; Safe, 1994; Schecter et al., 1994a,b).

The TEQ (TCDD equivalents) for all PHAH compounds in human blood is also higher than in the TCDD-treated animals in this study (24 PPT-Lipid, human blood; 10 PPT-Lipid, TCDD-treated monkeys). In general, similar congeners are in both sources; however, a greater proportion of the human serum TEQ is derived from 1,2,3,7,8-PnCDD and 2,3,4,7,8-CDF (Calvert et al., 1996; Kahn et al., 1988; Schecter et al., 1994a,b). (Note that 1,2,3,6,7,8-HxCDD was not quantified in this study.)

The animals in this study were dosed with TCDD in feed over a period of approximately 4 years; unexposed and TCDD-treated animals were housed and fed under the same conditions before and after TCDD treatment. None of these animals had participated in other toxicant studies since their acquisition in 1977. Thus, the observation of elevated levels of 3,3',4,4',5'-TCB and 3,3',4,4',5-PnC3,3'-HxCB in TCDD-exposed animals is unexpected, and the cause of the elevated serum PCB level is unclear. Accumulation of PCBs in TCDD-treated animals may have resulted from PCB exposure during TCDD administration due to a contaminated TCDD solution or other inadvertent source. Alternatively, all animals in the study may have been exposed to PCBs in the feed or another environmental source, but differential metabolism of PCB congeners may have occurred in TCDD-treated animals or animals developing endometriosis. Consistent with this notion, previous studies with these animals and their offspring showed that 0 PPT and TCDD-treated chow contained 7.6 parts per billion total PCBs (Schantz and Bowman, 1989). In addition, it was established that other animals housed at the same facility over the same time period but with no known PHAH treatment have increased serum concentrations of these PCB congeners. This suggests that rhesus monkeys in this population were exposed to PCBs and that some of these animals retained 3,3',4,4',5'-TCB and 3,3',4,4',5-PnC2, total serum TEQ directly correlates with the severity of endometriosis (p < 0.05; total serum TEQ vs. rAFS point score; data not shown). These data suggest that an increased total serum TEQ may play a role in the pathogenesis of endometriosis.

The potential mechanism responsible for increased bioaccumulation of these coplanar PCB congeners in TCDD-treated animals is unclear. Although studies suggest that co-exposure of TCDD and PCBs would result in increased metabolism and excretion of 3,3',4,4'-TCB via synergistic activation of cytochrome P450s (Birnbaum, 1993), there is a paucity of data on bioaccumulation of specific congeners in humans and non-human primates following chronic exposure to complex mixtures of PHAHs. Trace amounts PCDDs and PCDFs in standard laboratory feed accumulate in the livers of control rats and mice with no intentional PHAH exposure (DeVito et al., 1998; Vanden Heuvel et al., 1994). Although TCDD and 2,3,4,7,8-PnCDF are not detected in feed, these chemicals with longer half-lives accrue in liver in the rat, accounting for 7% and 80% of PCDD/PCDF TEQs at 200 days of age, respectively. Recent rodent studies have shown that higher concentrations of certain coplanar and non-coplanar PCBs accumulate in liver tissue following co-treatment of specific PCB congeners alone or in combination with TCDD (De Jongh et al., 1993, 1994). McNulty reported that low concentrations of 3,3',4,4',5'-TCB accumulate in fat following chronic long-term treatment of rhesus monkeys with this congener (McNulty, 1985). Furthermore, serum levels of 3,3',4,4',5'-TCB were undetectable in animals without previous TCDD exposure or endometriosis, in the present study. These findings suggest that bioaccumulation of this congener may be low in healthy animals with no history of TCDD exposure or endometriosis. However, human serum, milk, and tissues contain concentrations of 3,3',4,4',5'-TCB and 3,3',4,4',5-PnC3,3'-HxCB are AhR agonists and contribute significantly to the toxicity of complex mixtures of environmental chemicals (Safe, 1994). These compounds elicit the same biochemical and toxic responses as TCDD, including induction of CYP1A1 and CYP1A2 gene expression, thymic atrophy, reproductive and developmental toxicity, immunotoxicity, altered lipid metabolism, and carcinogenicity. Moreover, certain PCB congeners modulate or disrupt the activity of certain steroid and sex hormones, including estradiol, vitamin A (retinoic acid), and thyroid hormone (Safe, 1994; Whitlock, 1994). The estrogenic activity of 3,3',4,4'-TCB was recently observed in vivo (SAFE et al., 1994a;b). (Note that 1,2,3,6,7,8-HxCDD was not quantified in this study.)

The potential association of an increased serum TEQ with endometriosis depends upon the TEF assigned to 3,3',4,4'-TCB. Recently the TEF for this congener was revised from 0.01 (Safe, 1990) to 0.0001 (Van den Berg et al., 1998) since it appears to be rapidly metabolized in vivo (McNulty, 1985). In the present study, total serum TEQ was increased in TCDD-treated animals with and without disease, using the 1998 TEF values for all PHAHs; therefore, this increase correlated with TCDD exposure rather than presence of disease. If bioaccumulation of 3,3',4,4'-TCB occurs in certain subjects or disease states, then the 0.01 TEF previously proposed for this toxic chemical may be more relevant in risk evaluation. When data are reevaluated using the 0.01 TEF value for 3,3',4,4'-TCB, total serum TEQ directly correlates with the severity of endometriosis (p < 0.05; total serum TEQ vs. rAFS point score; data not shown). These data suggest that an increased total serum TEQ may play a role in the pathogenesis of endometriosis.

The composition of PCBs in biological samples is highly variable and differs from commercial mixtures, due to differential species-specific metabolism and biodegradation of PCB congeners (Birnbaum, 1993; Safe, 1994). Structure-toxicity studies demonstrate that 3,3',4,4'-TCB, 3,3',4,4',5-PnC3,3'-HxCB, and 3,3',4,4',5,5'-HxCB are AhR agonists and contribute significantly to the toxicity of complex mixtures of environmental chemicals (Safe, 1994). These compounds elicit the same biochemical and toxic responses as TCDD, including induction of CYP1A1 and CYP1A2 gene expression, thymic atrophy, reproductive and developmental toxicity, immunotoxicity, altered lipid metabolism, and carcinogenicity. Moreover, certain PCB congeners modulate or disrupt the activity of certain steroid and sex hormones, including estradiol, vitamin A (retinoic acid), and thyroid hormone (Safe, 1994; Whitlock, 1994). The estrogenic activity of 3,3',4,4'-TCB was recently observed.
both in vitro and in vivo (Nesaretnam et al., 1996). Considering the critical roles of estradiol, retinoic acid, and immune cytokines in the regulation of normal uterine endometrial growth (Rier and Yeaman, 1997), immune-endocrine disruption by PCB congeners may have potential importance in the pathogenesis of endometriosis.

Rier et al. (1993) made the first report of a significant association between chronic TCDD exposure and increased endometriosis in the rhesus monkey, and subsequent animal and human studies have investigated the potential role of TCDD and dioxin-like chemicals in the pathogenesis of this disease. Recent work utilizing an estrogen-responsive rodent model of endometriosis (Cummins and Metcalf, 1995) shows that treatment with TCDD or the dioxin-like 2,3,4,7,8-PnCDF for 12 weeks produces a dose-dependent increase in the size of surgically induced endometrial lesions (Cummins et al., 1996; Johnson et al., 1997). In contrast, treatment of mice with nondioxin-like compounds has no effect on the growth of endometriotic sites, providing preliminary evidence that PHAH-promoted endometriosis may be AhR-mediated. The estrogen level in the animals at the time of TCDD exposure appears to be important to the PHAH-mediated effects in this model (Yang and Foster, 1997). Campbell et al. (1985) initially observed a possible link between aggressive endometriosis in the rhesus and PCB exposure. However, after final review of all autopsy data from this study, Arnold et al. (1996) found no evidence of an increased prevalence or severity of endometriosis in this colony of PCB-exposed rhesus monkeys 6 years after initiation of toxicant treatment. However, this study may have been terminated before endometriosis was clinically apparent in all PCB-treated animals, since disease appears to develop in the rhesus after a latency of approximately 7 years (Fanton and Golden, 1991; Rier et al., 1993; Wood et al., 1983).

Few studies have addressed the potential role of environmental toxicants in human endometriosis. In Belgium, Koninckx et al. (1994) noted that the incidence of endometriosis in women presenting at clinics with infertility is 60–80% and TCDD concentrations in breast milk are among the highest in the world (WHO environmental series, 1989). An association between exposure to PCBs and human endometriosis was suggested by Gerhard and Runnebaum (1992), who found a higher serum level of two non-coplanar PCB congeners in women with endometriosis than in healthy control women. However, this finding was not confirmed in a second report evaluating serum concentrations of non-coplanar PCBs in women with and without endometriosis (Lebel et al. 1998). Mayani et al. (1997) found that a higher proportion of infertile women with endometriosis had a detectable TCDD level in serum (18%) than infertile controls without disease (3%). Although previous studies suggest that TCDD exposure is strongly correlated with increased endometriosis in monkeys (Rier et al., 1993), in the current study the serum TCDD level was not related to the presence of endometriosis. These data suggest that TCDD exposure, rather than the serum level of this toxicant, may be associated with the development or progression of endometriosis. It is possible that increased environmental exposure to TCDD during a limited period of time may result in permanent disruption of physiologic processes which control the development of endometriosis in normal unexposed animals, and these changes may persist after the serum toxicant level returns to background. Pathogenic effects may be directly exerted by TCDD or indirectly via altered retention of or synergistic interactions with other environmental toxicants.

This study shows that the serum TCDD level in TCDD-exposed monkeys remains above the background level 13 years after termination of treatment. This result brings into question the half-life of TCDD in these exposed monkeys. Previous work suggests a TCDD half-life of 400 days in these animals (Bowman et al., 1989), and predicts that the serum TCDD concentration would have decreased to the background level within 10 years after treatment. It is possible that TCDD half-life is variable in the rhesus monkey population. This hypothesis is consistent with the fact that serum TCDD concentration covered a wide range in 9 TCDD-treated monkeys (not detectable to 472.9 PPQ). In addition, 2 animals that died 7 to 11 years post TCDD exposure had an unusually high serum TCDD level (2316 PPQ and 3189 PPQ; or 846 PPT-Lipid and 965 PPT-Lipid; data not shown). It is possible that this occurred as a consequence of cachexia and lipid mobilization accompanying endometriotic intestinal blockage, or these animals may have had a decreased ability to metabolize TCDD. Although the half-life of TCDD in humans is estimated to be 7 years, in 2 of 36 subjects the TCDD half-life was estimated to be 26 and 740 years and serum TCDD levels did not decrease over 5 years in 4 other subjects (Pirkle et al., 1989).

TCDD-exposed monkeys in this study demonstrated both elevated serum TCDD and elevated serum triglycerides. This result is consistent with earlier data in TCDD-treated animals (Brewster et al., 1988; Zinkl et al., 1973) and TCDD-exposed humans (Calvert et al., 1996; Martin, 1984; Pazderova-Vejlukova et al., 1981; Roegner et al., 1991). However, some studies in humans have found no association between TCDD exposure and increased serum triglycerides (Hoffman et al., 1986; May, 1982; Mocarelli et al., 1986; Moses et al., 1984; Suskind and Hertzberg, 1984; Webb et al., 1989). This study and other studies in humans (Calvert et al., 1996; Hoffman et al., 1986; May, 1982; Mocarelli et al., 1986; Moses et al., 1984; Pazderova- Vejlukova et al., 1981; Suskind and Hertzberg, 1984; Webb et al., 1989) find that serum cholesterol is not affected by TCDD exposure. However, increased serum cholesterol has been reported in TCDD-exposed animals (Schiller et al., 1985; Zinkl et al., 1973). The elevated serum triglyceride level observed in this report was associated with elevated serum concentrations of 1,2,3,6,7,8-HxCDF, 3,3′,4,4′-TCB and 3,3′,4,4′,5-PnCB. This result is consistent with the sug-
gestion that dioxin-like congeners, particularly PCBs, can alter lipid synthesis and metabolism (Safe, 1990, 1994).

Lipid adjustment of the PHAH level allows one to compare concentrations in adipose and other tissues (Kahn et al., 1988; Patterson et al., 1988; Pirkle et al., 1989). Lipid adjustment of the data presented here generally decreased the magnitude of differences in the serum PHAH level between animal groups; however, the significant differences calculated from whole weight values were mostly retained in lipid-adjusted values (Tables 2 and 3). Calvert et al. (1996) also found that lipid adjustment did not affect regression models in studies on TCDD-exposed humans. Increased serum concentrations of TCDD and other specific dioxin-like chemicals were also highly correlated with increased serum lipids in women with endometriosis, but not in fertile control women (Clark et al., manuscript submitted). Furthermore, a higher serum triglyceride level has been observed in young women with endometriosis relative to control women without disease (Crook et al., 1997). These findings suggest that a relationship between the serum level of specific PHAHs and lipids may exist in certain populations and that lipid adjustment of data may influence interpretation of results in various cohorts. In general, the strong association of elevated levels of serum triglycerides and TCDD suggests that both whole-weight and lipid-adjusted PHAH values should be calculated and evaluated carefully.

Recent advances in the detection and assay of individual PHAH congeners in biological samples have made it possible to assess total PHAH body burden in humans and animals. Future studies are expected to exploit this advance to assess the health impact of PHAH body burdens in both exposed individuals and the general population. The evidence presented in this report suggests that TCDD exposure and endometriosis may be associated with increased serum concentrations of 3',4,4',-TCB and 3,3',4,4',5-PnCB in the rhesus monkey. These findings may be relevant to human endometriosis, since the serum levels of these environmental chemicals in TCDD-exposed animals were similar to concentrations reported in serum, milk, and tissues from the general human population. Additional studies are warranted, particularly in human subjects, to explore the potential implications of these data.

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