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Metabolism and Biochemical Toxicity of PCBs and PBBs

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Effects of PCBs on Biochemical Functions

One of the major biochemical effects of PCBs is the induction of microsomal enzymes in the liver. Risebrough et al. (*1*) suggested that PCBs have the capability to induce the activities of microsomal enzymes. Subsequently, Street et al. (*2*), demonstrated the induction of liver enzymes in rats by PCBs. Since then, a great number of articles have been published on this subject.

The enzyme systems studied have included mainly hydroxylases, *N*- and *O*-demethylases and nitroreductases, and to a lesser extent nonspecific carboxylesterase, bromosulfophthalein-glutathione conjugating enzyme, *p*-nitrophenol UDP–glucuronyl transferase and EPN-detoxification systems.

The induction of microsomal enzymes by commercial PCBs has been demonstrated perorally in rabbits (*3*) rats (*4*) and primates (*5*) and via intraperitoneal injection (*6*) and skin application in rats (*7*). Values reported for the threshold of enzyme induction by PCBs vary between 0.5 and 25 ppm (*3*, *8*, *9*) (et al., 1974).

The time course of microsomal enzyme induction was studied by Litterst et al. in rats (*10*). They found that significant levels of enzyme induction occurred after 7 days of feeding PCBs in the diet. A single oral dose in rats resulted in maximum enzyme activities at 24 hr. Bickers et al. (*7*, *11*) have shown that maximum induction occurred within 2–6 days in rats after cutaneous exposure to Aroclor 1254 or microscope immersion oils containing 30–45% chlorine.

In order to study the effect of chlorine content of PCB's on enzyme induction, different commercial PCBs were investigated. Litterst et al. (*8*), studied PCBs with chlorine contents from 42 to 60%. In rats the maximum activity of demethylase was observed with the 54% chlorine PCBs; maximum activity for nitroreductase was obtained with the 60% chlorine PCBs. Chen et al. (*12*) reported that 54% chlorinated PCBs gave maximum response of demethylase in rats. Similar results were reported by intraperitoneal injection of various PCBs in rats (*13*). Goldstein et al. (*14*) and Iverson et al. (*15*) studied the effects of Aroclor 1016 and Aroclor 1242 on enzyme induction in rats. Both PCBs have approximately 42% chlorine, but the content of the penta-, hexa-, and heptachloro isomers of Aroclor 1016 is reduced to 10% of that in Aroclor 1242. Their results showed that Aroclor 1242 was a much more potent inducer than Aroclor 1016 in female rats. In comparing Aroclor 1232, Aroclor 1248, and Aroclor 1260, Schmoldt et al. (*16*) found that Aroclor 1260 was the most potent enzyme inducer in rats.

The synthetic and isomerically pure PCBs have been investigated for enzyme induction, Chen et al. (*12*) found that both 2,3,4,5- and 3,5,3′,5′-tetrachlorobiphenyls induced enzyme systems in male rats, but no significant differences were found

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between the two isomers. Johnstone et al. (*17*) and Ecobichon et al. (*19, 20*) compared the effects of position of chlorine atoms on the ring. They found that enhanced induction of monooxygenases was observed for PCBs having chlorine atoms substituted at the 4- and 4'- positions irrespective of chlorination at other positions. Substitution at the 2-position was next in importance followed by substitution at the 3-position. They concluded that the position of chlorination was as important as the degree of chlorination.

There are two types of enzyme inducers being reported in the literature. One group, to which phenobarbital belongs, resulted in increased cytochrome P-450 content, as well as increased benzopyrene hydroxylase and ethylmorphine demethylase activities in the liver. The second group includes polycyclic hydrocarbons. This group stimulates the formation of cytochrome P-448 and an increase in hydroxylation but not demethylation. Alvares et al. (*20*) reported that rats treated with PCBs produced an increase in cytochrome P-448, hydroxylase, as well as demethylase. Therefore, it appears that PCBs display induction behavior typical of both groups.

Most of the enzyme induction studies were evaluated *in vitro*. Several workers indeed have observed enzyme induction effects *in vivo* by demonstrating shortened barbiturate sleeping times (*6, 17, 21*). Commercial PCBs are known to contain small amounts of dibenzofurans (DBF). DBFs were found to be 170 times as potent as PCBs in inducing enzymes.

Alvares and Kappas (*22*) reported the induction of hydroxylase and demethylase in placenta, as well as in the fetus when pregnant rats were treated with PCBs. Vainio (*23*) reported that Clophen A50 enhanced hydroxylase 5-fold in the lung and 8-fold in the kidney microsomes, whereas the microsomes from duodenal mucosa exhibited no enhancement. Benzopyrene hydroxylase activity in skin was also increased by PCBs (*7*).

Vos and Koeman (*24*) found that several tissues of PCB-treated chickens were strongly fluorescent under ultraviolet light. They suggest that PCBs could induce chemical porphyria in chickens. Later, they demonstrated that mitochondrial ALA synthetase activity increased 20-fold and fecal porphyrin levels were significantly increased in Japanese quail treated with PCBs (*25, 26*). They suggested that the porphyria caused by PCBs is due to the increase of ALA synthetase, followed by overproduction of porphyrins. Similar results in rats were obtained by Goldstein et al. (*27*), and Bruckner et al. (*28*). Goldstein et al. (*27*) suggested that the induction of ALA synthetase is probably not the primary cause of PCB-induced porphyria because of the delayed onset of porphyria as compared with ALA synthetase induction. They suggest that the ALA synthetase in the liver may be related to the increase of cytochrome P-450 content. This idea was supported by Grote et al. (*29*). Aroclor 1242 was found to be a more potent inducer of liver porphyria than Aroclor 1016 (*14*). Both Aroclors have similar chlorine content, but Aroclor 1242 contains 9% homologs with five or more chlorines while Aroclor 1016 contains only 1% of such homologs. Sinclair and Granick (*30*) showed that cultured liver cells respond rapidly to PCBs in accumulation of uroporphyrin. Simon et al. (*31*) suggested that porphyria may be due to the destruction of the phospholipid structure of cell membrane, resulting in increased permeability of the substrate. Hirayama et al. (*32*), observed hypobilirubinemia in Yusho patients and suggested that the induction of bilirubin UDP-glucuronyltransferase may be responsible for the low plasma bilirubin content. However, recently Bastomsky et al. (*33*), showed that the lowering of serum bilirubin concentration by PCBs is due to the reduced binding of bilirubin to plasma protein rather than to the enzyme induction. Wit (*34*) has shown that PBBs which are similar to PCBs are potent heptoporphyrinogenic chemicals.

The chemical and structural similarity of DDT to PCBs led Jefferies et al. (*35*) to study the effects of PCBs on thyroid functions. They found an increase in thyroid weight in PCB-treated black-backed gulls as compared to controls. Hurst et al. (*36*) found no clear-cut effect of PCBs on thyroid gland size in quails. Byrne et al. (*37*) reported that PCBs increased thyroxine levels in the blood and also promoted peripheral degradation of thyroxine in female minks. Bastomsky (*38*) (1974) found a 4- to 5-fold increase of biliary excretion of thyroxine in rats treated with PCBs. PCBs also elevated iodine uptake by the thyroid and reduced serum PBI concentration. Recently Bastomsky and Wyse (*39*), showed similar results in rats due to either intraperitoneal injection or skin application of microscope immersion oil. Walker (*40*) found that inclusion of Aroclor 1254 in fish food resulted in an increase in fish thyroid activity.

The effects of PCBs on ATPase and oxidative phosphorylation have been examined by a number of investigators. Na^+K^+-ATPase and Mg^{++} ATPase from several fish tissues were found to be inhibited by PCBs (*41-44*). Further study showed that *in vitro* data were not in agreement with the *in vivo* data (*41*). The inhibition of ATPase in fishes by PCBs was also reported by Kinter et al. (*45*) and by Davis et al. (*46*).

Sivalingan et al. (*47*) investigated the effects of

PCB on the oxidative phosphorylation of rat liver mitochondria. They showed that the mode of inhibition appears to be different with PCBs of different chlorine content. PCBs with low chlorine content inhibited energy and electron transfer but PCBs with high chlorine content not only inhibited energy and electron transfer but also had an uncoupling effect. LaRocca and Carlson (*48*) showed that the administration of commercial PCBs, as well as purified isomers to rats inhibited the total ATPase activity in liver, kidney and brain tissues.

Pardini (*49*) has demonstrated that a marked inhibition of respiratory enzyme systems occurred when heavy beef heart mitochondria were exposed *in vitro* to numerous PCBs. Chesney and Allen (*50*) showed that the addition of PCBs to rat liver mitochondria *in vitro* caused an inhibition of oxidative phosphorylation and respiration. Feeding rats with PCBs at the 1000 ppm level increased the oxidative phosphorylation in liver mitochondria; however, no effect was seen at the 100 ppm level.

Sharp et al. (*51*) reported that the inhibition of beef brain and rabbit kidney Na^+K^+-ATPases can be reversed or prevented by phosphatidylserine or phosphatidylinositol, but not by phosphatidychloline and phosphatidylethanolamine. They suggested that acidic phospholipids are required to stabilize the enzyme and thereby overcome the effect of PCBs.

Risenbrough et al. (*1*) suggested that PCBs had the capacity to enhance steroid-hydroxylating enzyme activity, thus affecting estradiol metabolism. In addition, certain low chlorine containing PCBs have estrogenic activity which is reflected by an increase in glycogen content of the uterus of immature rats (*52*). Similar results were obtained by Fumiko (*53*) in rats and by Orberg and Kihlstrom in mice (*54*). Orberg et al. (*55*) found that the estrus cycle of the mouse increased in length after a single injection of PCBs. Lincer and Peakall (*56*) reported an increased rate of metabolism of estradiol *in vitro* in the liver of American kestrels after the feeding of PCBs. Similar results were also reported in White Leghorn cockerels and pullets (*57*). Platonow and Funnell (*58*) demonstrated that feeding PCBs to cockerels resulted in a decrease in testicular and comb growth. Decreased urinary excretions of estrogen and dehydroepiandrosterone were also observed in cockerels when PCBs were administered in high doses (*59*). Ecobichon and Mackenzie (*13*), studied the uterotropic activity of commercial and isomerically pure PCBs in rats. They found significant changes with a number of PCB mixtures, but experiments with the isomerically pure PCBs were inconclusive. Changes were observed with 2-chloro and 2,2-chlorobiphenyls. Nelson (*60*) reported that PCBs are effective inhibitors of the binding of ^3H-estradiol to the rat uterus cytosol fraction *in vitro*. Recent data showed that PCB feeding caused neither significant chromosomal damage nor arrest in the rate of spermatogenesis in male rats (*61*).

Flick et al. (*62*) (1965) noted enlarged adrenals and small spleens in PCB-treated White Leghorn cockerels. Wassermann et al. (*63, 64*) reported that rats receiving 200 ppm PCBs showed an increase in plasma corticosterone levels. These changes were concomitant with morphological changes in the zona fasciculata of the adrenal gland which indicated increased activity. Sanders et al. (*65*) have shown similar increases in serum corticosterone in mice fed 60 to 1000 ppm PCBs.

It is known that DDT decreases the utilization of dietary carotene and liver storage of vitamin A in rats and cattle (*66, 67*). Villeneuve et al. (*3*) reported that Aroclor 1254 but not Aroclor 1221 reduced the liver concentration of vitamin A in pregnant rabbits. Decreased liver vitamin A concentrations were found in male and female rats and also Japanese quail after PCB feeding (*68*). However, the laying female quail did not appear to be affected.

The effects of PCBs on vitamin E have been studied by Combs et al. (*69*). Their results indicated that dietary PCBs increased the incidence of exudative diathesis in chicks and that this increase can be overcome by increasing dietary vitamin E or selenium. The vitamin D mediated calcium metabolism was found to be altered by the oral administration of PCBs to chickens (*70*).

Comparative Biochemical Toxicity of Aroclor 1254 and FireMaster BP-6

PCBs have been reported to affect a large number of biochemical systems. To the extent that it has been reported, PBBs affect most of these same systems in a similar fashion. However, quantitative and possibly qualitative differences do not appear to exist in the response of some biological parameters. These include thyroid hyperplasia (*71*) and microsomal enzyme induction (*72*).

A preliminary study was conducted to compare the relative effect of Aroclor 1254 (Aro) with FireMaster BP-6 (FM) (*73*). This was set up as a broad survey to give an estimate of the relative biological activity of these chemicals in many different systems and pin point areas where further more definitive work should be done.

Table 1 summarizes the time sequence of the various parameters studied. Adult male rats were fed 0, 5, 50, or 500 ppm Aroclor 1254 or FireMaster BP-6, mixed in the diet, for 2, 3, or 5 weeks.

Table 1. Parameters evaluated in study.

Parameters	Exposure 2 weeks	Exposure 3 weeks	Exposure 5 weeks
Body weight	X	X	X
Liver weight	X	X	X
Other tissue weights		X	
Mitochondrial respiration	X		
Liver composition		X	
Liver protein synthesis		X	
Liver RNA synthesis		X	
Liver microsomal enzyme activities		X	
Liver intermediary metabolites		X	
Serum cholesterol		X	
Serum enzymes		X	X
Plasma corticosterone		X	
Plasma protein		X	
BUN		X	X
Gross pathology		X	X
Histopathology		X	X
Cytogenetic analysis			X

Due to the large number of parameters examined and the time restraints placed on the study, the experimental design did not permit a totally unambiguous comparison between all of the effects caused by Aroclor 1254 and FireMaster BP-6. Some apparent differences in biochemical responses determined at sacrifice may have been partly due to the fact that, for the three-week study, rats were exposed to FireMaster BP-6 for 48 hr longer than rats were exposed to Aroclor 1254. Although under the experimental conditions, this effect should be small, critical comparison of Aroclor 1254 and FireMaster toxicity should be considered tentative until similar experiments are conducted which can eliminate this difference in exposure time. The following evaluations were made based on the assumption that this two day difference had no effect.

Some parameters showed no consistent or dose-related change with either chemical at any level. These included relative kidney weight and testes weight, hematology, SGPT, SGOT, plasma BUN, plasma corticosterone, the rate of liver protein synthesis per gram tissue, liver dry weight, lyophilizable kidney, liver, or testes, the incidence of chromosome abnormalities, and the number of cells in mitosis from bone marrow of spermatogenial cells.

Growth and adipose tissue weight were equally depressed by Aroclor and FireMaster when fed over a 3-week period at 500 ppm. The growth effect appeared to be due primarily to decreased food efficiency.

Three parameters were more sensitive to Aroclor than to FireMaster: the liver protein and liver RNA per gram tissue and plasma glucose. On the other hand, six parameters were more sensitive to FireMaster: These included liver growth, liver lipid (total lipid, cholesterol, phospholipid, and neutral lipid), plasma cholesterol, microsomal enzyme activities, mitochondrial function, and liver RNA synthesis. Disruptions of the redox and energy states of the cell were produced by dietary exposure to Aroclor or FireMaster. These effects were indicated by studies with isolated mitochondria and frozen clamped liver. In the latter case the concentrations of adenine nucleotides, pyridine nucleotides, and glycolytic intermediates were determined. On a molar basis, hexabromobiphenyl was reported to be about five times more potent than Aroclor 1254 in causing some increased microsomal enzyme activities (*72*). A threefold difference was observed between FireMaster BP-6 and Aroclor 1254.

A pathology team autopsied rats after 3 and 5 weeks exposure to Aroclor 1254 and FireMaster BP-6 (*74*). Pathological analysis of the liver of rats fed Aroclor or FireMaster for 3 weeks showed no abnormalities on gross observation. No histopathological lesions were seen in controls, Aroclor groups, or the 5 and 50 ppm FireMaster groups after 3 weeks. In the 500 ppm FireMaster group, minimal vacuolation and focal hepatitis were each seen in one rat. After 5 weeks exposure, gross observation of the liver indicated an increasing incidence of enlargement, friability, and prominent lobular pattern with increasing dose of both chemicals. Microscopically the 5 ppb Aroclor group appeared normal while 2/7 rats fed 5 ppm FireMaster had minimal cytoplasmic degeneration. All rats had this lesion at 50 and 500 ppm Aroclor or FireMaster with increasing cellular lipid. This lesion was recent, centrilobular and coalescing with no change in fibrous tissue or glycogen. The histopathology was qualitatively similar but slightly more severe in rats fed FireMaster than in those fed Aroclor and is consistent with biochemical findings. The biochemical findings were all made after 3 weeks exposure at which time histopathological changes were not evident. Even at 5 weeks the histopathology was not severe enough to alter most of the clinical chemistry in the rat.

In most parameters evaluated, FireMaster BP-6 caused an equivalent effect at a lower dose level or in a shorter time or caused a greater effect at the same dose level (expressed as ppm in the diet) as Aroclor. When expressed on a molar basis, FireMaster becomes even more effective than PCB. Exceptions to this observation were noted at the 500 ppm dose level, where differential effects of Aroclor and FireMaster were often eliminated.

A summary of the major effects seen are found in Table 2. In all but one case the maximum response of a parameter occurred at the highest dose level.

Table 2. Relative biochemical response to Aroclor 1254 (Aro) and FireMaster PB-6 (FM) after dietary exposure.

Parameter	LEL, ppm[a]		Maximum effect, % control	
	Aro	FM	Aro	FM
	Two weeks exposure			
Liver weight/body weight	500	50	170	183
Mitochondrial respiration	500	500	152	192
	Three weeks exposure			
Body weight gain, g	50	500	52	52
Liver weight/body weight	50	50	203	214
Liver dry wt.	NE[b]	50	NE[b]	107
Liver lipid	50	5	138	156
Liver cholesterol	500	50	226	232
Liver protein	500	NE	84	NE
Liver RNA	5	50	79	84
Liver DNA	5	5	58	60
Liver RNA synthesis	500	5	78	86
p-Nitrobenzoate reductase activity	50	5	288[c]	520[c]
Plasma glucose	50	NE[b]	79	NE[b]
Plasma cholesterol	500	5	178	256
	Five weeks exposure			
Liver weight/body weight	500	50	171	216

[a] Lowest effective level that shows a dose-response relationship.
[b] No effect.
[c] Maximum effect occurred at 50 ppm dose.

Microsomal enzyme activities were, in general, greater at the 50 ppm level and the decline at 500 ppm relative to the 50 ppm level was more marked with FireMaster.

Metabolism of PCB and PBB Mixture

Generally, the tissues from animals and man containing PCB from environmental exposure have GLC patterns resembling those of PCB mixtures with more than 50% chlorination. This is in marked contrast to the major manufactured products that generally contain 42% or less chlorine. While one cannot rule out the possibility that differential uses favored introduction of more highly chlorinated materials in the environment, the observation has led to the general belief that the less chlorinated components are more readily metabolized.

The environmental observations have been confirmed in a large number of experimental feeding studies in a number of mammalian and avian species. The absence or diminished concentration of the early eluting peaks have been reported in the rat (*75, 76*), rabbit (*77*), cow (*78*), quail (*79*), and hen (*80*).

The peaks not present in tissues and products generally are the early eluting peaks that correspond to the PCBs with lower degrees of chlorination. The observations are consistent with the belief that the rate of metabolic attack on PCBs decreases with increasing chlorination. Studies of the single PCB homologs of various degrees of chlorination have shown that those with five or less chlorine atoms are more readily metabolized and excreted than the PCBs with higher chlorination (*81–83*). The position of chlorine substitution also affects the retention and elimination of single homologs (*84*). Metabolism of individual PCBs will be discussed more thoroughly in another section. Among the various food species there appears to be one exception to the general rule that the less chlorinated PCBs are hydroxylated. The PCB residues in fish closely resemble the PCB to which the fish was exposed. There was little change in the relative concentrations of the various homologs (*85*). Consistent with this observation it has been found that mono-, di-, and tetrachlorobiphenyls are not metabolized by trout (*82*).

Among the food producing animals (cattle, chickens) concentrations of PCB residues in milk and eggs reach a steady state after approximately 8 to 12 weeks of continuous intake (*78, 80*). The ratio of residue levels in food products to levels in the animal's diet are greatest with PCBs of 54% chlorination. The residue levels in milk are approximately four times the residue level in the cow's diet. In hens, residue levels in eggs are approximately equal to the diet while residue level in the body tissue (fat basis) is approximately six times the dietary level. The ratio of residue level in product and tissues to intake is lower with PCBs of greater than 54% chlorination. Since it is unlikely that there is greater metabolism of the most highly chlorinated PCBs, the results suggest that the absorption for the GI tract is lower for the more highly chlorinated PCBs (*86*). PCBs of four or fewer chlorines rarely occur in animal products when the animals are fed environmentally realistic levels. Much less is known about the metabolism of PBB than PCBs. However, the observations that have been reported tend to be consistent with the reports on PCB metabolism.

While the PBB of most immediate concern for human health (FireMaster BP-6, Michigan Chemical Corp.) is a multicomponent mixture like the PCBs, it is much simpler than the ordinary commercial PCB mixtures. The major component is a single hexabromobiphenyl accounting for over 60% of the total material. The only other component occurring in significant amounts is a single heptabromobiphenyl.

In general, the measures of retention in edible

tissues and excretion in edible products are similar for the hexabromobiphenyl and the hexachlorobiphenyls (*87, 88*). Heptabromobiphenyl retention and excretion is only 1/10 of that of the hexabromobiphenyl. In cattle that have been heavily contaminated with FireMaster BP-6, the heptabromobiphenyl was barely detectable (milk or tissues) 6 to 8 months after the time of ingestion, even though the hexabromobiphenyl level was over 1000 ppm (*89*). Since the heptabromobiphenyl is not more likely to be metabolized by the animal than the hexabromobiphenyl, it is reasonable to assume that its absorption is lower or its elimination through bile is greater than the hexabromobiphenyl component. Over 60% of octabromobiphenyl is excreted in the feces as the parent compound when it is fed to rats (*90*). This observation supports the conclusion that there is less absorption with increasing halogenation.

Metabolism of Individual PCBs

Quantitative studies of the distribution of PCBs indicate that most, if not all, PCBs which contain six or fewer chlorine atoms are efficiently absorbed from the gut of higher animals (*91–93*). Furthermore, only about one-tenth of the absorbed dose is excreted prior to metabolism to more polar compounds (*81, 92, 93*). A number of the less chlorinated PCBs (i.e., mono-, di-, tri-, and tetrachlorobiphenyls) are readily metabolized and excreted by birds and mammals. There is little evidence that fish can metabolize any PCBs.

It has been adequately demonstrated that a number of mammalian species and the pigeon can readily degrade and excrete monochlorobiphenyls, primarily 4-chlorobiphenyl (*82, 92, 94–97*). However, the rate of metabolism drops sharply as the number of chlorines per molecule increases. For example, the rate of metabolism and excretion of several PCB isomers has been shown to decrease as the number of chlorines increases in the following order: 4-chlorobiphenyl, 4,4′-dichlorobiphenyl 2,5,-2′,5′-tetrachlorobiphenyl 2,4,5,2′,5′-pentachlorobiphenyl (*92, 93, 98*). The rate of metabolism and excretion of 4,4′-dichlorobiphenyl is approximately one-half that of 4-chlorobiphenyl. In each case the PCBs are metabolized primarily by hydroxylation and conjugation with glucuronic acid (*81, 92*). The importance of the route of excretion, i.e., urine or feces, is determined primarily by the degree of chlorination. Approximately 60, 30, and 7% of a single dose of a mono-, a di-, and a pentachlorobiphenyl, respectively, were excreted in the urine of rats. The remainder of the dose was excreted in the feces (*92*).

Once the number of chlorine atoms on the biphenyl molecule reaches four, the position of the chlorine atoms becomes a more important factor in determining the rate of metabolism and excretion (*84, 92*). As was observed with the chlorinated benzenes (*99, 100*) those PCBs which have two adjacent unsubstituted carbon atoms are metabolized and excreted more rapidly than PCBs having the same degree of chlorination, but which do not have two adjacent unsubstituted carbon atoms (*92*). It is also likely that, as with the chlorinated benzenes, the absence of two adjacent unsubstituted carbon atoms has a greater effect as the degree of chlorination increases. For example, the initial half-life of 3,5,5′,5′-tetrachlorobiphenyl is only about twofold greater in the rat (*101*) than that of 2,5,2′,5′-tetrachlorobiphenyl in the same species (*93*). On the other hand, the initial half-life of 2,4,5,2′,4′,5′-hexachlorobiphenyl appears to be infinite, whereas the initial half-life of 2,4,5,2′,5′-pentachlorobiphenyl is only about 2 days (*92*). This particular hexachlorobiphenyl has been shown to be in the highest concentration of any PCB found in the adipose tissue of the Swedish population (*102*).

The importance of two adjacent unsubstituted carbon atoms to the metabolism of PCBs may be derived from their effect on the formation of arene oxide intermediates during the metabolism of these compounds by the hepatic mixed-function oxidases. Evidence for the formation of arene oxide intermediates in PCB metabolism was first provided by Gardner et al. (*103*) in their study of the metabolism of 2,5,2′,5′-tetrachlorobiphenyl and in a later study of 4-chlorobiphenyl metabolism by Safe et al. (*96*), which was specifically designed to provide evidence of an arene oxide intermediate. Metabolism of PCBs which do not have two adjacent unsubstituted carbon atoms by hepatic mixed-function oxidases, via direct hydroxylation of the biphenyl molecule and/or the formation of an arene oxide between a chlorinated and an unsubstituted carbon atom could occur, but each of these mechanisms would be expected to be slower than arene oxide formation between two unsubstituted carbon atoms (*104, 105*). The latter mechanism would be expected to result in dechlorination and/or shifting of a chlorine atom (*104*). The metabolism of selected hexachlorobiphenyls has been reported (*106-108*), and in two incidents (*108, 109*) dechlorination and/or chlorine shifting was observed, but in all cases the rate of metabolism was quite slow.

Thus, those PCBs which are more readily metabolized and excreted may also more readily form arene oxides. Certain arene oxides have been implicated as potential carcinogens (*104, 105*), as have certain PCB formulations (*110–112*), but there

is no published evidence which indicates that the less chlorinated PCB formulations have been so rigorously tested.

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