Endocrine effects of a Polychlorinated Biphenyl mixture (Aroclor 1254): Repressed sex accessory glands in hypothyroid rats with normal levels of serum and testicular testosterone.

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Polychlorinated biphenyls (PCBs) are a group of chlorinated hydrocarbons that are widespread in the environment. PCBs are fat soluble, extremely stable, and bioaccumulate in the ecological food chain[1]. In addition, low levels of PCBs are found in some human tissues and milk[2,3]. PCBs have been shown to have a number of adverse effects on mammalian and avian reproductive and endocrine function[4,5]. For example, in the mouse, administration of Aroclor 1254 in the diet for 15 d reduces testicular sperm content without affecting testis weight[6], but seminal vesicle weight, an androgen-dependent sex accessory gland, is reduced[7,8]. These alterations have generally been attributed to increased testosterone (T) turnover due to the induction of hepatic enzyme systems[9]. For example, hepatic cytochrome P-450 content, and liver weight are increased, along with a decrease in the dry weight of the seminal vesicles when castrated, testosterone-treated mice are treated with PCBs. However, there are also reports that PCBs directly inhibit both the synthesis of testosterone (T) by the Leydig cell in vitro[10] , and the in vitro binding of steroid hormones to their intracellular receptors[11]. In addition, adverse endocrine effects may also result from Ah receptor mediated-mechanisms, resulting in alterations of hormone receptor levels like those seen with 2,3,7,8 -tetrachloro-dibenzo-p-dioxin (TCDD)[12]. Acute administration with μg quantities of TCDD dramatically decreases plasma T, dihydrotestosterone (DHT), thyroxine (T4), and seminal vesicle and testicular weights.

However, in spite of the number of studies that suggest that serum T levels are altered as a consequence of PCB-treatment, the single study that examined the effects of PCBs on serum T levels[13] in mice found that the PCB (6-CB) did not influence serum T or the biosynthesis of T in vitro. These authors reported that the lack of effect on serum T could be due, in part, to the fact that the serum T values are extremely variable in the mouse, and are not normally distributed. Serum and gonadal T levels have not been reported in any studies of the reproductive toxicity of PCBs in the male rat[14,15]. Due to the lack of endocrine data from PCB-exposed male rats, the present study was designed to determine the effects of subchronic

administration of the PCB mixture Arochlor 1254 on seminal vesicle weight and testicular function in the male rat.

Young (31 d old) male Fischer (F344) rats were administered 0, 0.1, 1, 10, or 25 mg/kg/d Aroclor 1254 by gavage for 5, 10 or 15 weeks and necropsied. The hormones testosterone and thyroxine were measured in the serum, and body weight and weights of the liver, kidney, adrenals, testes, seminal vesicle plus coagulating gland, cauda epididymides and pituitary were taken. At 10 and 15 weeks, testicular interstitial fluid was collected and testosterone concentration was determined. The in vitro production of testosterone by the testes was also measured at these times. The serum was also analyzed for T4 because PCBs are known to cause hypothyroidism at low doses, and this hormone, along with T and a number of other extragonadal hormones, is necessary for normal sex accessory gland function[16]. Sperm motility was measured from a caudal epididymal sperm sample and sperm numbers in the testis and cauda epididymis were determined. In addition, tissues were examined for histopathological alterations.

Body, seminal vesicle, cauda epididymal, and pituitary weights were depressed in the high dose group at 10 and 15 weeks. In contrast, liver weights were significantly increased, being significant at 10 and 25 mg/kg/d at all ages, while at 15 weeks males in the 1.0 mg/kg/d dosage group also had larger livers (Table 1).

Testis and adrenal weights were similar to controls, in spite of the fairly large inhibition of growth. Sperm motility, and testicular sperm count, were also unaffected by PCB administration as were serum and testicular testosterone levels. Furthermore, the weight of the testicular IF, the concentration of T in the IF and the total T in the IF compartment of the testis were not reduced by PCB administration. In fact, the IF parameters tended to increase, rather than decrease, but these effects were not significant. In treated rats, Leydig cell testosterone production in vitro was significantly increased after 2 hr of hCG stimulation, whereas the basal level of T production prior to hCG treatment was not affected. Consistent with the lack of effect of PCBs on testicular T levels, testicular histology was not affected. In fact, no compound-related lesions were observed in the epididymis, adrenal, thyroid or pituitary gland at any of the three intervals in the study. PCB-induced histological lesions were detected in the liver (hepatocellular hypertrophy and vacuolar degeneration) at the 10 and 15 week intervals. in the two highest dose groups, while at the 15 week interval, compound-related liver lesions were associated with the three highest dose groups.

In contrast to the minimal alterations of testicular function observed in the current study, serum T4 levels were undetectable in most rats in the 10 and 25 mg/kg/d dosage groups. Serum T4 was also significantly reduced at weeks 5 and 15 at 1.0 mg/kg/d and at week 15 at 0.1 mg/kg/d. The reduction in serum T4 seen in the present investigation is very similar to that reported by Byrne et al.,[17]. They found that five months of treatment with PCBs (Aroclor 1254) or PBBs in the diet at low doses (1 and 5 ppm) resulted in hypothyroidism in female Sprague-Dawley rats. Their lowest dose of 1 ppm (0.08 mg/kg/d), which lowered serum T4 levels, is very similar to our findings at 0.1 mg/kg/d.

Table 1. Effects of administration of Aroclor 1254 for 15 weeks in the male rat. Means with asterisks differ from control values (a = $p < 0.05$; b = $p < 0.01$).

DOSE OF AROCLOR 1254 (MG/KG/DAY)					
	0	0.1	1.0	10	25
Body Weight	319 g	320	320	302	246b
Liver Weight	11.7 g	12.6	13.1a	16.3b	15.6b
Seminal Vesicle Weight	1.17 g	1.07	1.05	1.05	0.79b
Testis Weight	1.48 g	1.53	1.52	1.54	1.49
Serum T (ng/ml)	2.23	3.19	2.38	2.38	2.39
Testicular IF T (ng/ml)	194	198	230	242	292
Cauda Epididymal Sperm Count (x 10^6)	106	103	109	100	84b
Serum T4 (μg/dl)	5.47	3.72b	2.58b	0.81b	0.72b

In conclusion, adverse effects on serum T4 levels are seen in rats at low exposure levels when PCBs are administered by gavage (0.1 mg/kg/d) or in the diet (1 ppm). Future studies should be designed to determine if adverse neonatal outcomes from PCB exposure are related to altered thyroid function. Although clinical data is lacking, McKinney[18] calculated that PCB concentrations normally found in humans can affect free T4 levels producing hypothyroidism. The changes seen in seminal vesicle weight and epididymal sperm content of developing F344 male rats in this study cannot be attributed to reduced levels of serum testosterone. It is important to point out that somatic growth was retarded concurrent with these reproductive alterations. In addition, Andrews[19], and Smialowicz et al.[20] found that liver, immune, and kidney functions were also altered in these males. The effect on serum T4 stands out from all the other effects, being altered at all doses, while a dose 100 fold higher was required to alter immune or kidney function and a dose 250 fold higher than this failed to alter testicular and serum T levels.

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