

Biological Effects of Polychlorinated Biphenyls in Rats and Quail

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Polychlorinated biphenyls (PCBs) have been administered to rats and to quail to discover the nature of the biological effects which these environmental pollutants produce in mammals and birds. Biological effects evaluated were:

- (1) estrogenic activity of the PCBs in rats,
- (2) alterations of pentobarbital metabolism induced by the PCBs as judged by sleeping time of Japanese quail,
- (3) alterations of liver vitamin A, liver lipids and liver weight induced by Aroclor 1242 in rats and quail, and
- (4) egg production of Japanese quail.

Estrogenic Activity

Estrogenic activity was assessed by determining the stimulation of the uterine glycogen response of the immature rat uterus 18 hours after administration of the test compound(1). The potency of active compounds is reported in terms of the minimal subcutaneous dose which will increase glycogen to a level significantly different from control. In a series of 11 polychlorinated biphenyls and terphenyls, the compounds containing up to 48% chlorine were estrogenically active (Table 1). A polychlorinated terphenyl containing 42% chlorine was found to be 8 times more active. This is a low level of estrogenic activity and probably arises from hydroxylated metabolites during *in vivo* metabolism.

Effects on Pentobarbital Metabolism in Japanese Quail

The polychlorinated biphenyls have been demonstrated to stimulate liver enzyme systems which metabolize barbiturates in rats(2). We utilized a sleeping-time test with pentobarbital in quail as a means of determining effects of polychlorinated biphenyls and terphenyls on liver microsomal enzyme activity. The liver is the major site for the metabolism of foreign chemicals, and a standard dose of pentobarbital will produce a standard sleeping time. Stimulation of the liver enzymes which metabolize the barbiturate will be reflected in a shorter sleeping time, and conversely, any inhibition of the liver enzymes will be reflected in a longer duration of pentobarbital hypnosis(3).

The PCBs and PCTs were tested for their acute or short-term effects (48 hours) and for longer-term effects at 3 and 7 days. At appropriate times after administration of the PCBs or PCTs, sodium pentobarbital was injected intramuscularly at a dosage rate of 50 mg/Kg body weight. Birds were considered asleep as long as they could not right themselves when placed on their backs.

A single dose of Aroclor 1268 or Aroclor 5460 administered orally at a dosage level of 100 mg/Kg to mature male or female Japanese quail initially increased sleeping times (Fig. 1). At 18 hours, however, sleeping times were less than untreated control values in the male quail, and by 48 hours, the PCBs and PCTs had stimulated the liver enzymes which metabolize pentobarbital, so that sleeping times were only about 50% of control sedation times. In the female quail, there was little effect of the single oral dose after 5 hours. In both male and female quail, Aroclor 1268 appeared

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Table 1. Estrogenic Activity of Polychlorinated Biphenyl (PCB) and Terphenyl (PCT) Compounds.

Name	Minimum effective dose mg
PCB Aroclor 1221 21% Chlorine	8
PCB Aroclor 1232 32% Cl	8
PCB Aroclor 1242 42% Cl	8
PCB Aroclor 1248 48% Cl	8
PCB Aroclor 1254 54% Cl	Inactive
PCB Aroclor 1260 60% Cl	Inactive
PCB Aroclor 1262 62% Cl	Inactive
PCB Aroclor 1268 68% Cl	Inactive
PCB Aroclor 4465 60% PCB, 40% polychlorinated triphenyl (PCT), 65% Cl	Inactive
PCT Aroclor 5442 42% Cl	1
PCT Aroclor 5460 60% Cl	Inactive

to exert greater effects than Aroclor 5460. The diphasic pattern elicited suggests that the PCBs and PCTs first inhibited the enzymes which metabolize pentobarbital, but then, at later times, stimulate the induction of greater enzymic activity.

Continuous feeding for 3 and 7 days at levels of 100, 300, and 625 ppm ad libitum stimulated the metabolism of the standard dose of pentobarbital

PENTOBARBITAL SLEEPING TIME IN QUAIL

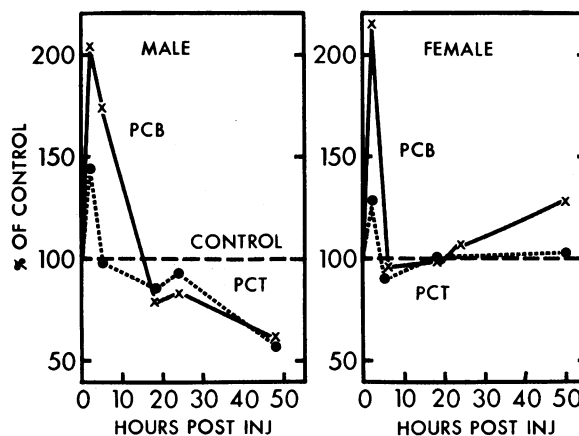


FIGURE 1. Sleeping times of quail fed PCB 1268 or PCB 5460. Each point represents six quail.

(Fig. 2). Sleeping times were about 50–70% of control levels, thus indicating greater metabolism of the barbiturate. Higher dose levels exerted greater effects and, again, Aroclor 1268 appeared to be more stimulatory than Aroclor 5460. Sleeping times were not decreased to as great an extent in the female quail as in the male.

The sleeping time pattern elicited by PCBs and PCTs is in marked contrast to that which the organochlorine pesticide analogs of DDT produce in quail(3). DDT and its analogs produce an

EFFECT OF AROCLORS ON SLEEPING TIME IN QUAIL

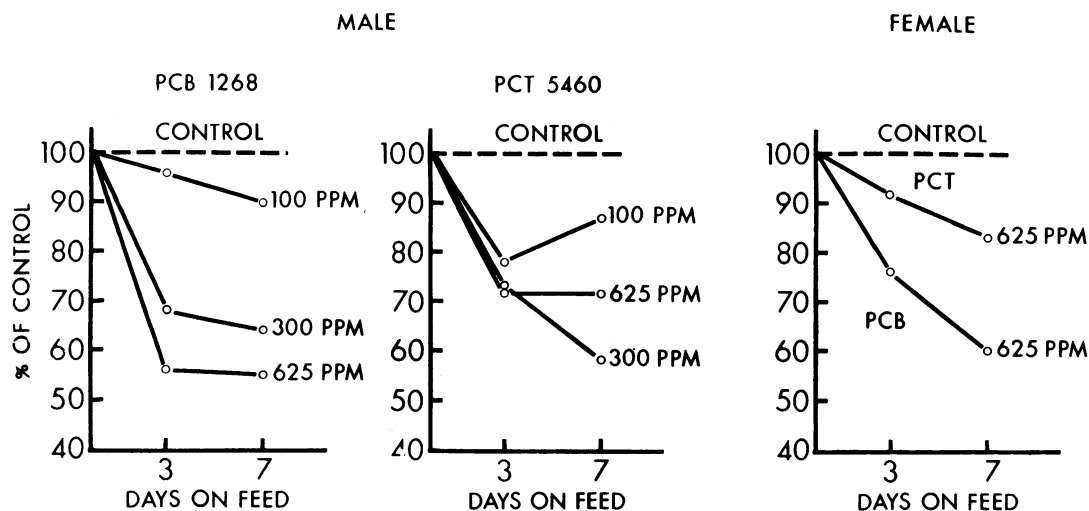


FIGURE 2. Sleeping times of quail fed PCB 1268 or PCB 5460. Each point represents six quail.

Table 2. Liver Changes in Male and Female Rats after Feeding Ad Libitum for Two Months Diets Containing 100 ppm Aroclor 1242.

Treatment	n	Liver wt. g ±SE	Liver lipid % ±SE	Vitamin A μg/g liver	Vitamin A mg/liver
Female rats					
Control	10	6.4 ± .2	3.15 ± .04	479 ± 23	4.54 ± .29
Aroclor 1242	10	7.4 ± .3 ^a	3.46 ± .08 ^b	244 ± 15 ^c	2.25 ± .11 ^c
Male rats					
Control	10	12.2 ± .4	5.14 ± .12	564 ± 20	6.82 ± .22
Aroclor 1242	8	13.0 ± .8	7.12 ± .18 ^c	270 ± 15 ^c	3.46 ± .21 ^c

^a p < 0.01.
^b p < 0.005.
^c p < 0.001.

initial inhibition of pentobarbital metabolism, and sleeping times are longer during the first week of feeding in males and during the first 3 weeks of feeding in female quail.

Effects on Liver Vitamin A in Rats and Quail

Repeated exposure of rats to DDT results in degenerative changes in liver tissue, an increase in liver size and an increase in liver lipids(4), and a decrease in liver storage of vitamin A (5). The

possibility that PCBs would produce similar effects was investigated in rats by feeding Aroclor 1242 for 2 months at a dietary level of 100 ppm (Table 2). In both male and female rats there was an increase in liver weight and liver lipids. There was a very striking decrease of about 50% in the liver concentration and content of vitamin A.

Aroclor 1242, fed to male Japanese quail for 2 months at a dietary level of 100 ppm, produced similar effects, an increase in liver weight and

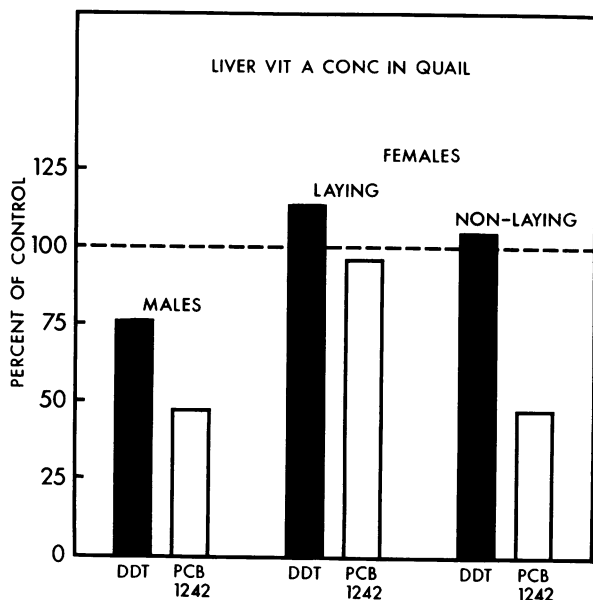


FIGURE 3. Liver vitamin A concentrations of quail fed 100 ppm DDT or PCB 1242 for two months. Groups contained 14 to 20 quail.

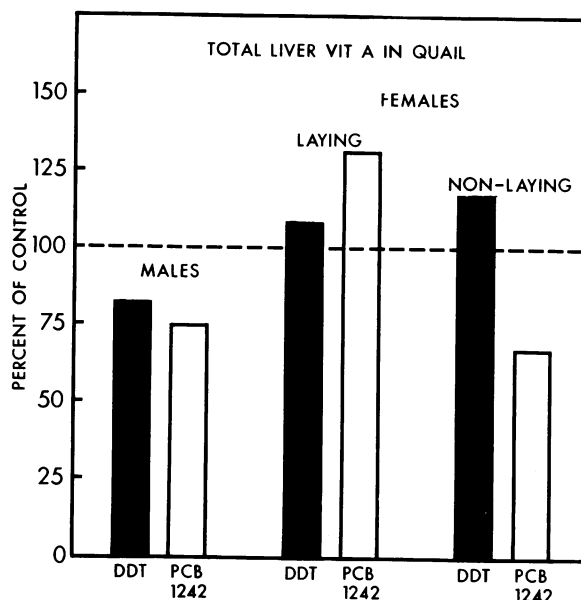


FIGURE 4. Total liver vitamin A of quail fed 100 ppm DDT or PCB 1242 for two months. Groups contained 14 to 20 quail.

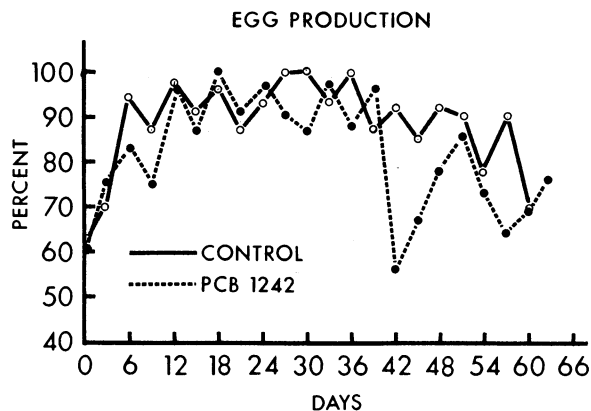


FIGURE 5. Egg production of Japanese quail fed PCB 1242. Average production for three day periods is plotted for groups of 16 quail.

lipids, and a large decrease in liver vitamin A (Fig. 3 and 4). Female quail that were laying did not show consistent changes. During the ovulatory cycle, large amounts of vitamin A stores are mobilized to provide the vitamin A for the yolk of the eggs. Because of these large, almost daily

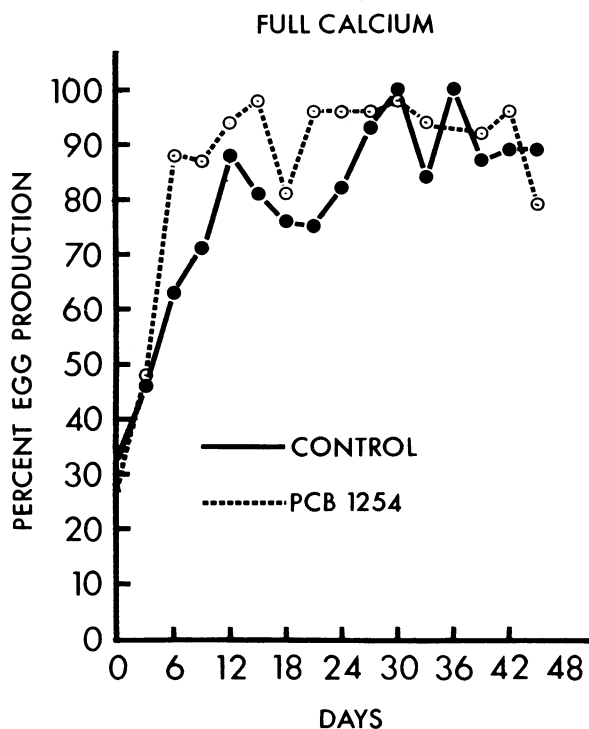


FIGURE 6. Egg production of Japanese quail fed PCB 1254 on a full calcium diet. Average production for three day periods is plotted for groups of 16 quail.

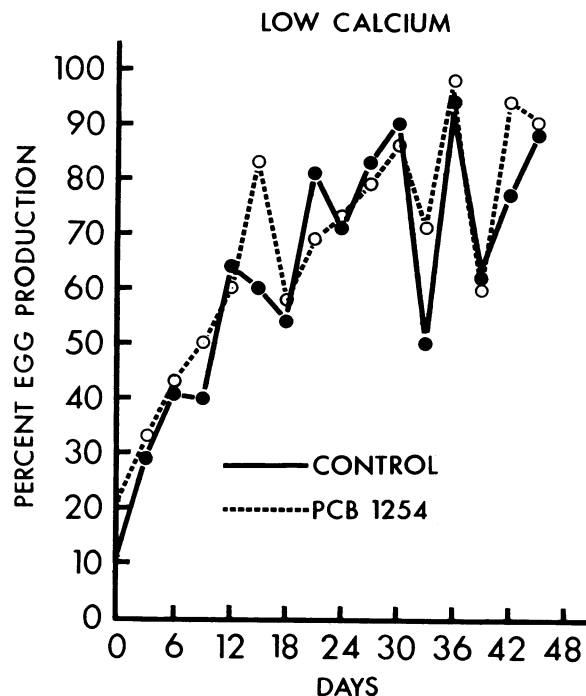


FIGURE 7. Egg production of Japanese quail fed PCB 1254 on a low calcium diet. Average production for three day periods is plotted for groups of 16 quail.

changes, variability in liver vitamin A levels is great. In order to determine whether PCBs exert similar effects in female quail, a group of young females were kept in the dark, from the time they matured (39 days of age), and fed Aroclor 1242 for 2 months. Under these conditions, egg-laying is inhibited, and the cyclic mobilization of vitamin A for deposition in the egg yolk does not occur. In the birds kept in the dark, Aroclor 1242 caused about a 50% reduction in liver vitamin A (Figs. 3 and 4).

Effects on Egg Production

The effects of Aroclor 1242 and Aroclor 1254 on egg production was studied in Japanese quail. Aroclor 1242 was incorporated into an adequate calcium laying mash at a level of 100 ppm for 60 days. Egg production (Fig. 5) was similar for controls and Aroclor 1242-fed birds during the first 40 days (>90%). Average egg production in the Aroclor 1242-fed quail declined to about 71% during the next 3 weeks. The number of membranous and broken eggs in the Aroclor 1242 group was over 2 times that of the control quail.

Aroclor 1254 was fed at a level of 100 ppm in both a full calcium (2.55%) and low calcium (0.66%) diet. Egg production was not suppressed by the PCBs in either case (Figs. 6 and 7), although there was a negative effect of the low calcium diet. There were no differences in the numbers of broken or membranous eggs attributable to Aroclor 1254.

A depression in egg production of White Leghorn hens fed Aroclor 1248 was recently reported by Scott et al.(6). Heath et al.(7), however, found little effect of Aroclor 1254 on eggshell thickness or reproduction in mallards or bobwhite quail. An effect upon egg production and reproduction of Aroclor 1242 and Aroclor 1254 has been previously reported in White Leghorn chickens(8). Recently, Peakall(9) found no effects upon eggshell weight in Ring doves fed 100 ppm Aroclor 1254. Thus, there is a notable lack of consistency in the published reports of the effects of PCBs on egg production and reproduction in birds.

Conclusion

A study was made of the effect of PCBs on estrogenic activity, liver barbiturate metabolism, liver vitamin A and egg production using a limited number of PCB materials. It appears that the biological effects of specific PCB compounds are different, as might be expected with this series of

materials containing compounds which differ so greatly in chlorine content and in chemical and physical properties. More extensive research on individual PCB compounds must be undertaken to assess fully the biological effects of polychlorinated biphenyls on mammals and birds.

REFERENCES

1. Bitman, J. and Cecil, H. C. 1970. Estrogenic activity of DDT analogs and polychlorinated biphenyls. *J. Agric. Food Chem.* 18: 1108.
2. Street, J. C. et al. 1969. 158th Fall Meeting American Chemical Society, New York City. Abstract. Sept. 7-12, 1969.
3. Bitman, J. et al. 1971. Comparison of DDT effect on pentobarbital metabolism in rats and quail. *J. Agric. Food Chem.* 19: 333.
4. Laug, E. P. et al. 1950. Liver cell alterations and DDT storage in the fat of the rat induced by dietary level of 1 to 50 ppm DDT. *J. Pharmacol. Exp. Ther.* 98: 268.
5. Phillips, W. E. J. 1963. DDT and the metabolism of Vitamin A and Carotene in the rat. *Can. J. Biochem. Physiol.* 41: 1793.
6. Scott, M. L. et al. 1971. Proc. 1971 Cornell Nutrition Conf. Feed Manufacturers, Buffalo, New York, November 2-4, 1971.
7. Heath, R. G. et al. 1970. Proc. XV Int. Ornithol. Cong., The Hague. Aug. 30-Sept. 5, 1970. E. J. Brill, Leiden.
8. Industrial BIO-TEST Laboratories, Inc. Rpt. No. J7300, June 4, 1970.
9. Peakall, D. B. 1971. Effect of polychlorinated biphenyls (PCBs) on the eggshells of Ring Doves. *Bull. Environ. Contamin. Toxicol.* 6: 100.