

Polychlorinated Biphenyl-induced Decrease in Liver Vitamin A in Japanese Quail and Rats

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Exposure of rats to DDT or polychlorinated biphenyls (PCBs) results in degenerative changes in liver tissue, an increase in liver size, and an increase in liver lipids (BENNETT et al. 1938, GRANT et al. 1971, LAUG et al. 1950, MILLER 1944, SARETT and JANDORF 1947, STREET et al. 1969). DDT also decreases utilization of dietary carotene and liver storage of vitamin A in rats and cattle (PHILLIPS 1963, PHILLIPS and HIDIROGLOU 1965). PHILLIPS (1965) has suggested that the decreased utilization of carotene and vitamin A is not a general effect of chlorinated hydrocarbon pesticides, but may be more or less specific for DDT. Recently, VILLENEUVE et al. (1971) reported that a polychlorinated biphenyl ('Aroclor 1254') reduced the liver concentration of vitamin A in pregnant rabbits, but total liver vitamin A was similar to controls, although 'Aroclor 1221' did not affect the concentration of vitamin A in the liver of pregnant rabbits.

There is little information on liver changes in birds fed PCB. Although a few reports do include liver weights of birds fed PCB, these findings have been inconsistent (McCUNE et al. 1962, PRESST et al. 1970, REHFELD et al. 1971, VOS and KOEMAN 1970). The present report compares liver changes in male and female Japanese quail (*Coturnix japonica*) and rats (*Rattus norvegicus*) after feeding either polychlorinated biphenyl ('Aroclor 1242', Monsanto) or DDT. Egg production, eggshell characteristics and body fat residues of quail fed PCB are also reported.

METHODS

Japanese quail (39 days old) or rats (21 days old) were fed a control diet or one containing 100 ppm p,p'-DDT or 100 ppm 'Aroclor 1242' for 2 months. The quail and rat diets contained 10,500 and 9,120 µg vitamin A per kg feed, respectively.

During the second month of feeding, quail eggs were collected, weighed and eggshell thickness measured with a micrometer at the equator, after removal of the membranes.

The animals were killed by decapitation and the liver quickly excised and weighed. Liver lipids were determined by extraction of the homogenized tissue with acetone:ethanol and liver vitamin A by the antimony trichloride colorimetric procedure (CARR and PRICE 1926) after the liver was digested in KOH and extracted with ethanol and ethyl ether.

RESULTS

Rat liver changes. We found both species and sex differences in untreated controls in liver weight, lipids and vitamin A content. The livers of 80 day old control male rats were twice as heavy and had a higher concentration of lipid than female rats ($P < 0.001$; Table 1). The body weights of male rats (320 g) were considerably higher than those of females (182 g). However, liver weights of males per unit body weight were still larger than the female ($P < 0.001$). Liver vitamin A concentrations of male and female rats were similar. The greater total vitamin A in male rats is a reflection of the larger liver.

Both 'Aroclor 1242' and p,p'-DDT increased liver weight and liver lipids in male and female rats (Table 1). Feeding DDT or PCB reduced liver vitamin A concentration in both male and female rats (Table 1). This lowered concentration was not a dilution caused by the increase in liver size that followed pesticide feeding as the vitamin A per total liver was also significantly reduced after DDT or PCB feeding. Total liver vitamin A of treated male or female rats was approximately 1/2 of the control value.

TABLE 1

Liver changes in male and female rats fed ad libitum for 2 months diets containing 100 p.p.m. p,p'-DDT or PCB ('Aroclor 1242').

Treatment	N	Liver Wt.	Liver Lipid	Vitamin A		
		g ± SE	% ± SE	μg/g Liver	mg/Liver	
Male Rats						
Control	10	12.2 ± .4	5.14 ± .12	564 ± 20	6.82 ± .22	
p,p'-DDT	10	14.1 ± .4 ^c	7.20 ± .26 ^e	298 ± 16 ^e	4.20 ± .26 ^e	
PCB-1242	8	13.0 ± .8	7.12 ± .18 ^e	270 ± 15 ^e	3.46 ± .21 ^e	
Female Rats						
Control	10	6.4 ± .2	3.15 ± .04	479 ± 23	4.54 ± .29	
p,p'-DDT	10	6.9 ± .1 ^b	3.48 ± .11 ^b	252 ± 15 ^e	2.84 ± .12 ^e	
PCB-1242	10	7.4 ± .3 ^c	3.46 ± .08 ^d	244 ± 15 ^e	2.25 ± .11 ^e	

Treated differs significantly from control ^a $p < 0.05$; ^b $p < 0.025$; ^c $p < 0.010$; ^d $p < 0.005$; ^e $p < 0.001$. Statistical comparisons were made using the Student's "t" test with correction for unequal group size.

Quail liver changes. The female quail liver was twice as large as the male liver and had more lipid per unit weight ($P < 0.001$; Table 2), although body weights were similar (female, 116 g; male, 108 g). This was a reversal of the condition in the rat. However, livers of male quail had a higher vitamin A, both on a concentration basis and in the total liver ($P < 0.001$). The decreased liver vitamin A in the female is probably related to egg laying since it is expected that greater amounts of vitamin A would be mobilized to provide vitamin A for yolk. The yolk of each egg contained approximately 72 μg of vitamin A.

In quail, only the liver of the male responded to the treatments, with 'Aroclor 1242' having a greater effect than p,p'-DDT (Table 2). PCB increased liver weight and decreased liver vitamin A by 25%. p,p'-DDT also reduced liver vitamin A in male quail. Neither treatment affected liver lipids. There was a great deal of individual variation in the female quail data, presumably due to the mobilization of liver constituents in formation of the yolk. Consequently, no effects of the treatment could be seen. In order to eliminate the effects of egg laying, a group of females were kept in the dark from the time they matured (39 days of age) and were fed the treatment diets for 2 months. Under these conditions egg laying is inhibited and cyclic mobilization of vitamin A for deposition in the yolk does not occur. In the birds kept in the dark, PCB and p,p'-DDT increased liver weight (Table 2). PCB decreased liver vitamin A concentration to 1/2 of the control level and the total liver vitamin A was reduced to 2/3 of the control level.

Ten samples of abdominal fat of female quail were analyzed for PCB content. After 2 months feeding 100 ppm Aroclor 1242, the residue was 279 $\mu\text{g/g}$ lipid or about 3 x dietary level.

Effects of PCB on egg production and eggshell characteristics of quail. Daily egg records were kept for the quail housed under standard lighting conditions (14 hr light:10 hr dark). During the first month of treatment no effects of PCB feeding were observed. Egg production was 83% for the 'Aroclor 1242' fed birds and the controls. Aroclor feeding caused a decline in egg production to 75% during the second month of feeding, while control production was 89% (Table 3). The quail fed 'Aroclor' also had a higher incidence of broken and membranous eggs. 'Aroclor 1242' had no effect on egg weight or eggshell thickness.

TABLE 2

Liver changes in male and female quail after feeding ad libitum for 2 months diets containing 100 p.p.m. p,p'-DDT or PCB ('Aroclor 1242').

Treatment	N	Liver Wt. g \pm SE	Liver lipid % \pm SE	Vitamin A	
				$\mu\text{g/g Liver}$	mg/Liver
Male Quail					
Control	15	1.6 \pm .09	8.4 \pm .4	1362 \pm 76	1.99 \pm .10
p,p'-DDT	10	1.5 \pm .05	7.8 \pm .1	1078 \pm 84 ^b	1.65 \pm .11 ^a
PCB-1242	8	2.4 \pm .07 ^e	8.7 \pm .5	623 \pm 38 ^e	1.48 \pm .09 ^d
Female Quail					
Control	20	4.0 \pm .2	12.5 \pm .62	309 \pm 39	1.16 \pm .12
p,p'-DDT	17	3.8 \pm .2	12.4 \pm .84	344 \pm 41	1.26 \pm .15
PCB-1242	14	4.2 \pm .1	11.0 \pm .56	299 \pm 42	1.53 \pm .26
Female Quail Kept in Dark					
Control	11	2.0 \pm .05	7.4 \pm .2	695 \pm 4	1.37 \pm .08
p,p'-DDT	17	2.3 \pm .07 ^c	8.8 \pm .7	727 \pm 4	1.61 \pm .08
PCB-1242	20	2.8 \pm .10 ^e	8.1 \pm .4	334 \pm 2 ^e	0.92 \pm .05 ^e

Treated differs significantly from control ^ap<0.05; ^bp<0.025; ^cp<0.010; ^dp<0.005; ^ep<0.001. Statistical comparisons were made using the Student's "t" test with correction for unequal group size.

TABLE 3

Changes in egg production and egg characteristics of Japanese quail during the second month of feeding 'Aroclor 1242'.

ppm 'Aroclor' 1242	Number of Eggs			Egg Produc- tion %	Egg- shell thick- ness mmx10 ⁻²	Egg weight g
	Total	Broken	Membranes			
0	471	4	2	89	16.5	9.31
100	325	6	4	75	16.9	9.75

DISCUSSION

Liver vitamin A. Neither the rats nor the quail in this study showed any toxic symptoms to the PCB or DDT treatment. Although the treatments reduced liver vitamin A storage by 20 to 50%, no symptoms of vitamin A deficiency were evident. In these experiments adequate levels of vitamin A were fed. However, in animals receiving a marginal level of vitamin A in their diet, PCB's could reduce liver vitamin A stores to such an extent that avitaminosis might occur.

PHILLIPS (1963) fed DDT to rats in rations deficient in vitamin A and then dosed the rats with either vitamin A or carotene 24 hours before killing. The DDT treated rats had less liver storage of vitamin A when either vitamin A or carotene was administered than the control rats. From this PHILLIPS (1963) concluded that the action of DDT is not in the conversion of carotene to vitamin A but is at some stage in their metabolism common to both. The exact mechanism by which vitamin A storage is affected is unknown. The decrease in liver vitamin A following DDT or PCB ingestion suggests that increased carotene and vitamin A destruction and an altered lipid metabolism are involved.

In addition to the rat and quail, PCBs have been shown to cause liver damage in monkeys (NISHIZUMI 1970) and the appearance of jaundice in industrial workers has been correlated with exposure to PCB (DRINKER et al. 1937).

Egg production and eggshell characteristics. In our study we found a reduction in egg production but no effect of 'Aroclor 1242' feeding on eggshell thickness in Japanese quail.

Published reports of the effects of PCBs on egg production and eggshell characteristics have been inconsistent. KEPLINGER et al. (1970) reported that feeding 100 ppm 'Aroclor 1242' to White Leghorn chickens reduced egg production and caused thinner eggshells.

Several workers, using other PCBs have noted an effect on egg production with no effect on eggshell characteristics. SCOTT et al. (1971) found that 10 and 20 ppm 'Aroclor 1248' reduced egg production but had no effect on egg breaking strength. DAHLGREN et al. (1972) reported reduced egg production in pheasants dosed with 'Aroclor 1254'. However, HEATH et al. (1970) found no changes in egg production, number of cracked eggs or eggshell thickness after feeding 25 ppm 'Aroclor 1254' to mallards and 50 ppm to bobwhite quail. PEAKALL (1971) also found no effects on eggshell weight of Ring Doves fed 100 ppm 'Aroclor 1254'.

With the exception of KEPLINGER et al. (1970), no other researchers have been able to demonstrate eggshell thinning during PCB feeding. Effects of PCBs on egg production have been inconsistent. PCBs have been shown to reduce egg production in the White Leghorn chicken, Japanese quail and pheasant. However, PCB ('Aroclor 1254') had no effect on egg production in the mallard and bobwhite.

SUMMARY

'Aroclor 1242' (PCB) or DDT increased liver weight and lipids and decreased liver vitamin A of male and female rats or male Japanese quail. Laying female quail did not show consistent changes, presumably due to mobilization of lipid and vitamin A for egg yolk. When egg laying was inhibited, PCB reduced liver vitamin A 50%. 'Aroclor 1242' reduced egg production, but had no effect on egg weight or eggshell thickness.

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