The Effect of a Polychlorinated Biphenyl (Aroclor® 1254)* on Liver Microsomal Enzymes in the Male Rat

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INTRODUCTION

Polychlorinated biphenyls (PCB) are mixtures of various chlorinated biphenyl molecules and recently they have been detected as environmental pollutants in wildlife (JENSEN, 1966; HOLMES et al., 1967; HOLDEN and MARSDEN, 1967).

RISEBROUGH et al. (1968) suggested that PCB had the ability to induce enzymes and STREET et al. (1969) showed that induction of liver microsomal enzymes occurred. The lowest dose level of PCB in the diet used by STREET et al. (1969) was 25 ppm. LITTERST et al. (1972), using four different Aroclors varying in chlorine content, fed rats for 4 weeks at varying dose levels (0.5, 5, 50 and 500 ppm) in the diet. They measured several microsomal parameters and found that disruptions of normal enzyme activity occurred even at dose levels that had no effect on liver weights or on liver : body weight ratios. The present study was designed to show the effect of dosing low levels of PCB for longer periods of time on a range of NADPH-dependent microsomal enzymes using type I and type II substrates and on the NADPH-independent microsomal esterase.

MATERIALS AND METHODS

Sources of materials

Aroclor 1254 was supplied by Gollin and Co. Wellington, New Zealand. Sources of other chemicals and purification of enzyme substrates have been previously described (TURNER and GREEN, in press).

Preparation of animals

Twenty-eight 5-week-old male rats of the Dunedin Wistar strain maintained on diet 86 (shown to have <0.05 ppm HCB, DDT and DDE) were divided into four equal groups. One group served as the control and the other three groups were fed for 12 weeks on diet 86 containing 0.1, 1.0 and 10.0 ppm of Aroclor 1254 respectively. The rat food was pelleted as described by GREEN and TURNER (in press).

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* Aroclor (R), Monsanto Co., U.S.A., registered trade name for polychlorinated biphenyls.

Bulletin of Environmental Contamination & Toxicology,
Vol. 12, No. 6 © 1974 by Springer-Verlag New York Inc.

TABLE 1

EFFECT OF PCB ON SOME HEPATIC MICROSOMAL PARAMETERS OF MALE RATS*

Dose rate in diet	1 /g liver	2 /g liver	2 /mg micros. protein	3 /g liver	3 /mg micros. protein	4 /g liver	4 /mg micros. protein	5 /mg micros. protein	6 /mg micros protein	7 /g liver	7 /mg micros. protein
Control	11.69 ±2.02	7.51 ±1.71	0.65 ±0.15	40.83 ±10.89	3.48 ±0.67	28.46 ±4.15	2.46 ±0.24	0.48 ±0.06	1.01 ±0.13	21.36 ±7.67	1.83 ±0.53
0.1 ppm	12.80 ±2.23	8.13 ±2.49	0.64 ±0.15	47.90 ±19.73	3.72 ±1.11	29.53 ±5.74	2.34 ±0.50	0.41 ±0.07	0.98 ±0.06	22.80 ±6.47	1.79 ±0.43
1.0 ppm	12.34 ±2.23	9.13 ±1.48	0.75 ±0.10	55.94 ±13.53	4.53 ±0.78	32.19 ±5.70	2.63 ±0.33	0.39[b] ±0.08	1.05 ±0.15	23.10 ±3.64	1.89 ±0.27
10.0 ppm	16.46[a] ±4.18	13.91[a] ±2.44	0.87[b] ±0.15	116.03[a] ±33.73	7.08[a] ±1.53	46.47[a] ±7.53	2.94 ±0.62	0.43 ±0.05	1.28[a] ±0.09	33.30[b] ±11.47	2.00 ±0.45

* Values represent mean of seven animals ± S.E.M.

1. Microsomal protein : milligram/g liver
2. Aniline hydroxylase : nanomoles 4-aminophenol formed/min
3. 4-Nitroanisole O-demethylase : nanomoles 4-nitrophenol formed/min
4. Biphenyl 4-hydroxylase : nanomoles 4-hydroxybiphenyl formed/min
5. Nanomoles cytochrome b_5
6. Nanomoles cytochrome P_{450}
7. Phenyl acetate esterase : micromoles phenol formed/min

Superscript a denotes significant difference from control at 1% level

Superscript b denotes significant difference from control at 5% level

Preparation of liver samples and assays

All rats were starved for 12 h, weighed and killed by cervical dislocation. The preparation of liver 12 000 g supernatants and microsomal pellets, and the assays were all carried out as described previously. (TURNER and GREEN, in press.)

The data in Table 1 were analysed using Duncan's Multiple Range test (DUNCAN, 1955).

RESULTS AND DISCUSSION

The results are summarised in Table 1. At the dose rates of 0.1 and 1.0 ppm PCB there were no significant alterations in liver enzyme activity compared to the controls except for a slightly reduced cytochrome b_5 level at the 1.0 ppm dose rate.

At the 10 ppm dose rate there was a significant increase on a per g liver basis in the microsomal protein, aniline (type II substrate) hydroxylase, 4-nitroanisole (type I substrate) O-demethylase, biphenyl (type I substrate) 4-hydroxylase and esterase. These increases probably reflect the increase in protein synthesis which occurs in the treated animals. There was also a significant increase in the level of cytochrome P_{450} but not cytochrome b_5 at this dose level. Expressed on a per mg protein basis the increase in the levels of 4-nitroanisole O-demethylase and aniline hydroxylase were significant.

The induction of mixed-function oxidases by PCB has been documented (STREET et al. 1969; LITTERST et al. 1972) but the present results indicate that concentrations of PCB in the diet as low as 10 ppm are sufficient to produce measurable changes in the microsomal enzyme system if the animal is exposed for a long enough period. The data also suggest that chronic exposure to low levels of PCB might have an effect in other species since, generally speaking, inducing agents, particularly of the organochlorine type, are not considered to be species specific in their action.

ACKNOWLEDGEMENTS

The authors thank Dr J. Revfeim, Biometrics Division, Ministry of Agriculture and Fisheries, New Zealand, for the statistical treatment of the results.

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