EXHIBIT B

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EXCRETION RATES OF PURE Di- AND TRICHLOROBIPHENYL-^{14}C IN THE RHESUS MONKEY

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The chemical properties of polychlorinated biphenyls (PCB's) which make them industrially useful are the same properties which cause them to persist in the environment. These include thermal stability, resistance to chemical oxidation and hydrolysis, water insolubility, solubility in organic solvents, and compatibility with many types of macromolecules. Accumulation in fatty tissues of animals and men has been extensively described, but little is known about the pharmacokinetics of PCB's. Body distribution has been studied in rats[1-3], quails[4], cockerels[5], hens[6], and boars[7] with commercial mixtures, e.g. Aroclor 1245 or 1242. Information upon excretion is available only from a few of those long term studies in which mostly high dose levels were investigated. Highly chlorinated biphenyls can be detected easily in natural samples. Residues of lower chlorinated mixtures, however, have not been reported in spite of their predominant industrial use. This may be due to more rapid metabolism or excretion of these compounds. The present study was designed to determine whether lower chlorinated biphenyls are stored in the body and how fast they are excreted when administered at low single doses to non-human primates.

The PCB's investigated were 2,4'-dichlorobiphenyl, a major constituent of Aroclor 1221 (13.6 %)[8], and 2,2',5-trichlorobiphenyl which is considered to be a major component of Aroclor 1232 and 1242. Both of these were ^{14}C-labelled compounds. The 2,4'-dichlorobiphenyl was synthesized in the German laboratory (spec. activity 5.1 mCi/mM) and the 2,2',5-trichlorobiphenyl was obtained from Mallinckrodt Chem. Corp. St. Louis (Lot Nr. 4449 a).

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Considering the levels of PCB's found in human fatty tissues (USA: 1 ppm, Germany: 5-6 ppm) and considering that the amounts of major components of PCB mixtures are about 10 % of the total weight of the mixture, doses corresponding to about 80 µg/kg body weight of both PCB-isomers as well as a high dose of 566 and a lower dose of about 17 µg/kg of the dichlorobiphenyl were administered. Since the fatty tissue accounts for 20 % of the total body weight of the monkey, the medium dose of the single PCB isomer corresponds, for example, to a value of approximately 4 ppm of the PCB mixture found in natural fat samples. Anticipating fatty tissues to be of special significance female monkeys were chosen for these studies because their abdominal fat allowed a much easier biopsy, although the actual difference in total amount of fat between males and females is very small. Four female animals weighing between 3.1 and 5.56 kg were housed in metabolic cages with free access to Purina monkey chow and water. The diet fed did not contain chlorinated hydrocarbons and all animals had no previous drug treatment so that enzyme induction or drug interaction could be excluded. Each animal was administered once intravenously in the left forearm vein. The carrier used consisted of 0.2 ml ethanol, 20 mg Tween 80, and 0.2 ml water. Urine and feces were collected daily and radioactivity was measured by liquid scintillation counting (Packard Model 3380) after addition of 0.5 ml urine to 10 ml of Patterson Greene[9] solution. Feces were homogenized in a mortar and an aliquot sample was burned in an automatic combustion apparatus (Oximat, Intertechnique) determining the $^{14}CO_2$ contents. Experimental data are shown in Table I.

	2,4-Dichlorobiphenyl			2,3',5-Trichlorobiphenyl
Monkey	I	II	III	IV
dose (µCi)	3.15	5.83	40.22	14.62
dose (µg)	94.19	254.45	1757.5	380.2
body weight (kg)	5.56	2.28	3.1	4.6
µg/kg body weight	16.8	77.6	566	82.6
excreted urine (ml)	1096	1245	1268	1337
excreted feces (g)	335.1	522.8	320.6	323.9
urinary excreted radioactivity (% of admin.)	44.2	50.19	59.48	42.32
fecal excreted radioactivity	20.27	25.84	20.64	39.79
totally excreted activity after 14 days (in % of admin.)	64.47	76.03	80.1	82.02
halftile (days)	2.9	1.1	1.8	2.1

Table I Exp. data on PCB application to monkeys

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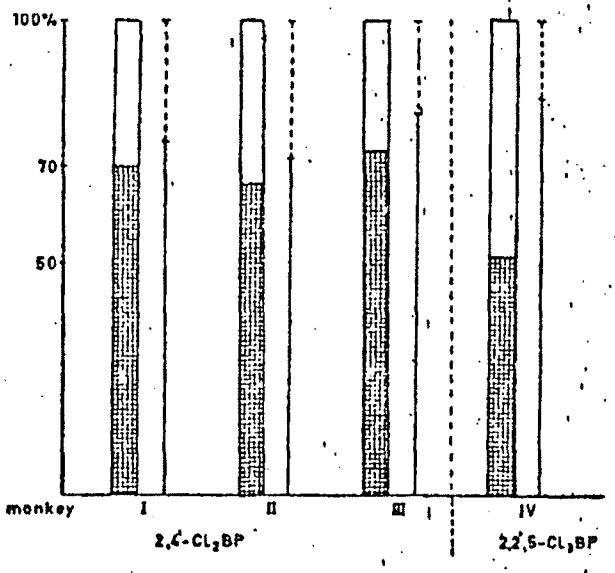

Fig. I: Rel. excretion of radioactivity in urine ▨ and feces □ and ratio urine | to feces |

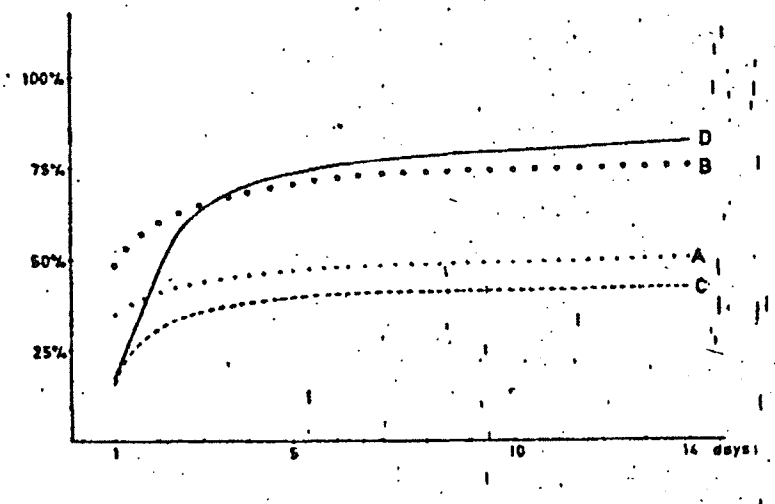

Fig. II: Excretion of radioactivity:
after 2,4'-dichlorobiphenyl appl., urinary A, total B
after 2,2',5 trichlorobiphenyl appl., urinary C, total D

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An average of 73,5 % of the administered radioactive 2,4'-dichlorobiphenyl was recovered during the first 14 days, of this 70 % was excreted in urine and 30 % via feces (Fig. I). The radioactivity excreted by either way corresponds to the excretions in ml or in grams. Although the dose level ranged from 1 to 33 times, there was no significant change in the total percentage eliminated. The dichlorobiphenyl was excreted very rapidly with the maximum amount of the first day after administration. The biological halflife differs within individual animals and with dose from 1.1 to 2.9 days. Continuation of the collection of urine and feces for another 7 days until day 21 resulted in an increase of eliminated radioactivity of only 0.58 % (0.35 % in urine, 0.23 % in feces). After 14 days about 7 % of the administered radioactivity could be detected in the fatty tissue, while the blood level was negligible.

A comparable medium dose of the 2,2',5-trichlorobiphenyl was given to one monkey and 82 % of the given radioactivity was recovered after 14 days. Fig. I shows that equal amounts of radioactivity were found in urine and feces, but the relative concentration in feces was much higher than in urine. Bilary excretion appeared to be more important for the elimination of the trichloro- than for the dichloro-compound. Excretion patterns (Fig. II) were also different: while the total excretion curve for the dichlorobiphenyl starts at a high level with slow increase, a high gradient exists for the trichlorobiphenyl curve. Radioactivity in feces reached high values on the second and third day. The halflife was 2.1 days in relation to 1.1 days for the comparable dose of the dichlorobiphenyl. The slower elimination of the trichlorobiphenyl from the body may be explained by a depot effect, a stronger plasma binding, or by the fact that the dichlorobiphenyl is more easily metabolized.

It is note worthy that different excretion rates already exist between these low chlorinated compounds, but with no sign of real storage as it is usually expected for PCB's. The general rapid excretion at these dose levels by the rhesus monkey might indicate that accumulation of these compounds in men will not occur. However, further investigations, especially elucidation and structural identification of metabolites, will be necessary to understand the toxicity of the PCB's.

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