

Intestinal Absorption and Distribution of Decachlorobiphenyl in Rats and Chicks*

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Polychlorobiphenyls (PCBs), widely used in industry and indispensable as electrically insulating liquids for transformers, have recently been found to be a global contaminant of oceans, seas and rivers (PEAKALL and LINCER, 1970). Several incidents in Japan and the USA (TAKESHITA and YOSHIDA, 1970; PLATONOW et al., 1971) indicated that PCBs may contaminate feeds and foods, and have alerted the attention of nutritionists to the problem of PCBs' toxicity. Pure isomers with up to six chlorine atoms were found to be absorbed up to 95% in rat intestine (ALBRO and FISHBEIN, 1972). Compounds with four or fewer chlorine atoms were metabolized and excreted more easily than the more chlorinated ones, and it was suggested that the use of the former is preferable (BENTHE and SCHMOLDT, 1973). It is clear, however, that metabolism of PCBs involves the drug-metabolizing system of the liver (JOHNSTONE et al., 1974) and may interfere with normal liver functions.

Finding compounds not absorbed by the intestine seems to be a better solution to the problem of safety, than using easily metabolized compounds. Since the highly chlorinated compounds are relatively inert, experiments were conducted with decachlorobiphenyl (DCB) administered to rats and chicks in the diet. DCB was not investigated by workers dealing with pure isomers (JOHNSTONE et al., 1974) due to its insolubility in the peanut oil used for injections.

The use of an inert marker is the most popular method for studying intestinal absorption. ^{91}Y was introduced successfully into the chick diet by HURWITZ and BAR (1965) to determine the intestinal absorption of calcium and subsequently also of other nutrients. It was used now as a marker for absorption study of DCB.

Methods

Diets. The animals were fed their usual commercial mash, containing DCB and ^{91}Y in the mineral premix. The DCB levels were 45 and 22 ppm in the mash of rats and chicks, respectively, and

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the corresponding ^{91}Y levels were about 0.2 and 0.1 $\mu\text{Ci/g}$.

Animals. Rats and chicks were fed the experimental diets for 6 days. Beginning from the 4th day feces were collected daily and freeze-dried. On the 7th day the animals were killed, the contents of the ileum were withdrawn by squeezing, and then freeze-dried. All the dried samples were analyzed for ^{91}Y and DCB.

In the study of DCB balance, the animals were given a measured amount of the DCB-containing mash for one day and killed on the morning of the second day. Feces, intestinal contents, blood, internal organs and carcass were analyzed for DCB and the DCB distribution was calculated as a fraction of the amount consumed.

Analytical procedures. A method was developed to determine DCB in feed and excreta. The method consisted of hot extraction with benzene (better results than with petrol ether) for 20 hours in a quick-extraction apparatus, or for 48 h in a conventional Soxhlet apparatus. For the determination of DCB in samples with high fat content (whole carcass), extraction was carried out with methanol to avoid subsequent time-consuming partitioning procedures. DCB was extractable at 95% with methanol.

The extracts were concentrated and cleaned by passing them through activated alumina columns (Merck, with 6% water added). DCB was quantitatively eluted from the column with petrol ether. After evaporation of the solvent, the contents were dissolved in benzene and determined by EC glc, in a Perkin-Elmer 801 apparatus. A coiled glass column (6 feet, $\frac{1}{4}$ " o.d.) was packed with 3% OV-1 coated on 80-100 mesh Gas Chrom.Q. Column temperature - 250°C , EC detector - 190°C , injector - 260°C , nitrogen flow - 60 ml/min.

^{91}Y was determined by liquid scintillation, on ashed samples (HURWITZ and BAR, 1965). The absorption of DCB was calculated from the equation: % net absorption = $100 \left(1 - (\text{DCB}/\text{Y})_{\text{s}} / (\text{DCB}/\text{Y})_{\text{f}} \right)$. The subindices s and f denote "segment" and "feed", respectively.

Results

Rats. The reliability of the method was tested by checking the dry matter absorption, using ^{91}Y levels in feces vs. the level in feed. The distribution of ^{91}Y in feces of the six rats (tested individually) was homogeneous, from $1.5 - 1.9 \cdot 10^6$ cpm/g. The average dry matter absorption was $77.9 \pm 2.3\%$, indicating small differences between the animals. However, the distribution of DCB in the feces was from 50 to 500 ppm and the calculated absorption values were from -45 to +45%.

The results, calculated from the ileal contents of the rats (Table 1), show the same trend of individual variation between the animals.

TABLE 1

DCB absorption calculated from the ileal contents of rats

Sample No.*	^{91}Y , cpm/g $\times 10^{-3}$	DCB	
		ppm	% absorption
12	548.3	87	43
22	520.0	78	47
32	432.0	170	-40
42	385.8	120	-10
52	646.4	121	34

* No intestinal contents were found in one experimental animal.

Chicks. When the combined excreta of four animals were examined, the average DCB absorption was about 20%. However, when the intestinal contents were tested individually, the results were as variable as in rats, with absorption varying from -43 to +65% (Table 2).

TABLE 2

DCB absorption calculated from the ileal content of chicks

Sample No.	^{91}Y , cpm/g $\times 10^{-3}$	DCB	
		ppm	% absorption
12	250	80	-30
22	275	30	55
42	266	93	-43
52	292	25	65

It was assumed that DCB may be absorbed and re-extracted through the intestine at different times after consumption. DCB was therefore determined in the blood and internal organs of rats and chicks given a single meal containing DCB.

The following DCB distribution was found in rats (average of three animals): the figures are percentage of the dose consumed \pm S.D.:

Liver	32.0 \pm 5
Carcass fat	20.0 \pm 4
Intestinal wall	11.0 \pm 2
Blood	2.0 \pm 0.5
Other organs together (kidney, spleen, brain)	2.0 \pm 0.4
Feces and intestinal contents	24.0 \pm 6

The total amount recovered was about 90%.

In the chicks most of the DCB was stored in carcass fat: 30±5%; 5-10% in the liver and 2-3% in other organs. About 30% was found in the feces and 5% in the intestinal tract. Total recovery of the DCB consumed was about 80%.

No additional peaks indicating metabolites of DCB were detected on the chromatograms.

It is concluded that about 60-70% of the DCB consumed is absorbed by the intestine of rats and chicks. It seems to be stored in fat in unchanged form. The liver levels of DCB in rats were several times higher than in chicks and it is probable that the transfer to adipose tissue is faster in chicks than in rats.

It is difficult to explain the great variability between DCB concentrations in the feces and intestinal contents of animals fed DCB continuously in the mash. It is possible that elimination from the body occurs through the intestine, not regularly, several days after consumption.

Since the assumption that DCB would pass the intestine without absorption was not confirmed, no further investigation was conducted.

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