

INFLUENCE OF SOIL PROPERTIES ON THE UPTAKE OF HEXACHLOROBIPHENYLS BY RATS

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ABSTRACT

Uniformly ¹⁴C-labeled 2,4,5,2',4',5'- and 2,3,5,2',3',5'-hexachlorobiphenyls (HCB) were adsorbed to soils with high levels of sand, clay, or organic matter. The treated soils were added to normal rat diets at a 5% rate using HCBs added to soil-free diets as positive controls. Short-term balance studies were conducted at 5 days and 6 months after soils treatment. Quantities of ¹⁴C excreted in feces and concentrations of ¹⁴C in body fat were the criteria used for evaluating retention. About 75 to 80% of the ingested activity was retained in all experiments. No differences between soils and control, among soil types, between compounds, and between degrees of aging were statistically significant.

INTRODUCTION

Bioavailability is potentially an important factor in evaluating hazards associated with toxic chemical contamination of soil. Interest in bioavailability of contaminants in soil has increased recently in response to incidents that involved human and/or animal exposure to soil accidentally contaminated TCDD (2,3,7,8-tetrachloro-*p*-dioxin) and other persistent chemicals (2,5,8). Knowledge of bioavailability is also useful in assessing exposures associated with waste disposal methods that could involve movement of chemicals into human food chains (4,7).

Studies of bioavailability using laboratory animals suggest that the TCDD adsorbed to soil is only about 50% as available as TCDD in corn oil and other solvents (9,11,12). Aged residues of polychlorinated and polybrominated biphenyl in soil were 70 to 80% as available as the compounds added directly to feed of farm and laboratory animals (3,6). In contrast to the high values from "normal" soils, bioavailabilities of TCDD in soils from two industrial sites were in the range of 1 to 23% (14). This finding, and the fact that soils of different textures and organic matter contents differ widely in the capacity to adsorb chemicals (1), suggests that soil texture and composition may be important factors affecting bioavailability. Limited evidence also indicates that the length of time that a chemical is exposed to soil also influences availability (12).

This study was carried out to compare the effects of soil texture and organic matter content on the oral bioavailability of two persistent hexachlorobiphenyls. A secondary purpose was to examine the effects of residue aging on bioavailability.

MATERIALS AND METHODS

Soils. Three soils were selected to provide a wide range of sand, clay and organic matter contents. The soils were obtained from a single wetland location that provided soils with a variety of compositions and textures while

Table 1. Composition and characteristics of the test soils

Characteristic	Soil		
	Sand	Clay	Organic
Sand, %	71	43	63
Silt, %	24	30	17
Clay, %	5	27	20
Organic Matter, %	1.6	0.6	14.0
pH	4.9	4.7	4.2
Cation Exchange Capacity, meq/g	3.4	6.7	16.6

insuring that the underlying mineral compositions were similar. The soils were air dried and only the particles passing a 125 micron sieve were used. The major physical and chemical characteristics of the soils that might affect adsorption of chemicals are presented in Table 1.

Compounds. The experimental compounds were uniformly ^{14}C -labeled 2,4,5,2',4',5'- and 2,3,5,2',3',5'-hexachlorobiphenyls (HCB) with specific activities of about 14 mCi/mM (Pathfinder Laboratories, St. Louis, MO). The HCBs were purified by high pressure liquid chromatography, dissolved in hexane, and added to the soils at rates of 10 nCi/g. The soils were mixed thoroughly after the solvent evaporated.

Diets. The test diets consisted of 5% treated soils added to the meal form of a standard rat diet. The positive controls consisted of the equivalent amounts of ^{14}C -HCB added to soil-free diets.

Animals. Each treatment group within an experiment consisted of six male Sprague-Dawley rats weighing 250 to 300 g. Rats were housed individually in metabolism cages equipped for separate collection of urine and feces. The room was maintained at 21 C and 80% relative humidity with a 14/10 hr light/dark cycle. Feed and water were available continuously.

Feeding Experiments. Four separate experiments were conducted using the three soil types and the positive control. Each HCB was studied at 5 days and 6 months after the treated soils were prepared. The treated feeds were offered *ad libitum* for three days and the rats were then fed untreated feed for an additional seven days. Feed intake was measured daily throughout the study. Urine and feces were collected daily, stored under refrigeration, and composited by individual rats for the total experiment. The rats were sacrificed by CO_2 asphyxiation on the 10th day, and samples of omental fat were obtained.

Analyses. Total ^{14}C activity of feed, feces and urine was determined by combustion and counting the $^{14}\text{CO}_2$ by liquid scintillation. Replicate feed samples were analyzed for total activity to assure proper preparation and uniform mixing. Undried feces were pulverized in a blender using dry ice to maintain the frozen state until analysis of activity by combustion. Urine was freeze dried and counted after combustion because activity was too low for counting directly. Fatty tissue was rendered and expressed lipid was counted directly without combustion.

RESULTS AND DISCUSSION

The term bioavailability as used in this paper is defined as the fraction of an ingested chemical that is absorbed and available for metabolism, storage, or excretion by the animal. Concentrations of activity in body fat and the total activity excreted in feces were the two measurements made in this study that provided data useful for evaluating bioavailability. The values obtained for these two measurements are presented in Tables 2 and 3. As expected, excretion of activity in urine was negligible because metabolic conversion of these HCBs to water soluble metabolites would be unlikely (10).

Table 2. Mean concentrations (% dose/g) of hexachlorobiphenyls in omental fat of rats fed various laboratory contaminated soils*

Soil Type	5 Days		6 Months	
	2,4,5	2,3,5	2,4,5	2,3,5
Control	1.86	1.97	1.30	2.10
Sand	2.09	2.02	1.26	2.29
Clay	1.87	1.78	1.17	2.07
Organic	2.10	1.92	1.26	2.16
Standard Error, n=6	0.29	0.09	0.07	0.11

* 2,4,5 and 2,3,5 represent 2,4,5,2',4',5'- and 2,3,5,2',3',5'-hexachlorobiphenyl, respectively. The periods, 5 days and 6 months, indicate the time between treatment of the soils and conduct of the feeding experiments.

Table 3. Mean total excretion (% dose) of hexachlorobiphenyls in feces of rats fed various laboratory contaminated soils*

Soil Type	5 Days		6 Months	
	2,4,5	2,3,5	2,4,5	2,3,5
Control	22.1	22.7	18.1	21.8
Sand	23.5	25.3	14.0	21.2
Clay	22.2	23.7	16.9	21.8
Organic	26.5	21.3	15.2	15.7
Standard Error, n=6	0.9	0.7	1.0	1.2

* 2,4,5 and 2,3,5 represent 2,4,5,2',4',5'- and 2,3,5,2',3',5'-hexachlorobiphenyl, respectively. The periods, 5 days and 6 months, indicate the time between treatment of the soils and conduct of the feeding experiments.

There was no statistically significant differences among the soils, or between the soils and control, in the uptake of HCBs in body fat at either time period (Tables 2). Additionally, there were no consistent patterns in the nonsignificant differences that occurred in the four experiments. The concentration of activity in body fat of rats fed the soil treated with 2,4,5,2',4',5'-HCB for six months was much lower than the average concentrations found with the other three treatments. The body fat concentrations generated by retention of a given amount of compound will be inversely related to the body fat pool sizes of the experimental animals. Since each of the four individual experiments involved rats from a different populations, it is probable that the rats in the experiment with the 2,4,5,2',4',5'-HCB residue aged for six months had larger pools of body fat than the rats in the other experiments. Regardless of the reasons for the differences in overall average concentrations among experiments, there were no differences due to residue aging when the concentrations in the soil-fed rats were normalized to the concentrations in the controls.

The amount of activity excreted in feces (Table 3) supports the conclusions drawn from the fat accumulation data. The differences among soils and between compounds were not significant. In addition, aging the residue had no effect on the amounts excreted in feces relative to the amounts excreted by the controls. The activity excreted in feces would be indicative of the amount of unabsorbed compound. Since, however, recycling to the gastrointestinal tract is a route of HCB excretion, the total activity in feces will overestimate the amount of nonabsorbed HCB. Despite this limitation, the data indicate that 75 to 80% of the dose was absorbed from all treatments.

Previous work has demonstrated modest reductions in the uptake of persistent halogenated hydrocarbons contained in soil relative to the uptake of compounds in corn oil or a normal diet (3,9,11,12). In contrast, no effect of adsorption to soil was demonstrated in this study. The compounds selected for this study are highly nonpolar (13) and are expected to be readily adsorbed to the organic matter fraction of the soil (1). Adsorption of chemicals to soil often is thought of as binding to active sites. In the case of nonpolar compounds, however, a preferential distribution among solvents is probably the best model. Thus, it would not be surprising if desorption occurred readily under the chemical conditions of the gastrointestinal tract.

Small reductions in bioavailability are of some scientific interest. Major reductions in bioavailability are required before this factor will significantly impact a hazard assessment. As in this study, results of previous studies indicate little reduction in relative uptake nonpolar compounds contained in "normal" soils. The small reductions are not sufficient to impact the conclusions of hazard assessments. Significant reductions in uptake may occur, however, when soil contains unusual adjuncts from industrial activity (14) or when the contaminant is contained in such unusual matrixes as fly ash (6). These situations require case by case evaluations.

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