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Polychlorinated Biphenyls: Phytotoxicity, Absorption and Translocation by Plants, and Inactivation by Activated Carbon

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A recent spillage of polychlorinated biphenyls (PCBs) along 210 miles of North Carolina highways stimulated this investigation of the effects of these substances on the growth of several plant species and the use of activated carbon to decrease their activity and bioavailability in the environment. LICHTENSTEIN et al. (1969) reported on the use of activated carbon to reduce the plant uptake of chlorinated hydrocarbon insecticides.

PCBs have been employed as dielectric fluids in transformers and for other assorted uses. These synthetic organics are oily, viscous liquids, with low water solubilities (0.01 to 0.08 ppm) and moderate vapor pressures (10^{-3} to 10^{-6} mm Hg at 25°C in pure form). A summary of the chemistry and physical properties of PCBs and some of their biological effects has been published by HUTZINGER et al. (1974). Recent studies show that PCBs and polybrominated biphenyls (PBBs) are absorbed and translocated by some crop plants (IWATA et al. 1974, JACOBS et al. 1976), estuarine and fresh water marsh plants (WALSH et al. 1974, MOSA et al. 1974, 1976, 1977), aquatic plants (MAHANTY 1975, MAHANTY and McWHA 1976, MAHANTY and FINERAN 1976), and phytoplankton (SINCLAIR et al. 1977). Recent studies by FURUKAWA and MATSUMURA (1976) showed that PCBs, including a highly chlorinated pentachlorobiphenyl, were degraded by bacteria. Recently conducted biotoxicity and epidemiology studies showed PCBs to be effective in inducing levels of the mixed-function oxidase (MFO) enzymes in animals and that the chemical has been found worldwide in the environment (CORDLE and KOLBYE 1978).

The purpose of these studies was to: 1) examine the effects of soil-applied PCB on soybean and fescue growth, 2) determine the amount of PCB absorbed and translocated by the two plant species, and 3) determine whether or not activated carbon would inactivate PCB and prevent its uptake by growing plants.

MATERIALS AND METHODS

Plant Growth Studies. Analytical grade PCB (Aroclor 1254), obtained from the Monsanto Company, in ethanol, was thoroughly mixed with 300 g of Lakeland sand (Typic quartzipsamments; siliceous, thermic, pH 4.7, 1% organic matter, 5% clay, 6% silt) in styrofoam pots at rates 0, 1, 10, 100, and 1000 ppm. Activated carbon (the same water treatment grade Nuchar SA carbon from Westvaco that was applied to the PCB spills along North Carolina highways) was thoroughly mixed with the soil in half of the pots

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at a rate of 3.7 t/ha (3333 ppm) (approximately half the rate applied to the PCB spills). Three replications were used. The pots were placed in a hood for 48 hours. After the ethanol had evaporated, the pots were seeded with six soybean seed [*Glycine max* (L.) Merr., "Ransom"] or 1 cc of fescue (*Fescue arundinacea* Schrib., "Kentucky 31") seed. In the fescue study, the ethanol was allowed to evaporate from the soil before the activated carbon was added.

Ten milliliters of nutrient solution (WEBER 1977) and 20 ml of water were added and the pots placed into growth chambers (200 hlx, 16 hrs day, 30°C day, 18°C night) and watered to 80% of soil field capacity whenever soil moisture dropped to 30% of field capacity. Pots planted with soybean were thinned to 4 plants per pot when the plants were 4 cm tall. Pots were weighed on a daily basis and daily water usage recorded (corrected for evaporation losses from pots with no plants). At the end of 26 days for soybean, and 42 days for fescue, plant height and fresh top and root weights were made.

^{14}C-PCB Studies. ^{14}C-Aroclor 1254 (specific activity = 31.3 mCi/mmole), obtained from New England Nuclear (Lot No. 872-193), average molecular weight = 326.25), was mixed with Aroclor 1254 in ethanol and thoroughly mixed with 300 g of Lakeland soil in styrofoam pots at rates of 0 and 20 ppm (0.5 μCi/pot). The ^{14}C-Aroclor 1254 has been found to more closely resemble Aroclor 1248, but contains lower chlorinated, more volatile congeners than Aroclor 1254.[1] Two replications were used. Activated carbon was thoroughly mixed with the soil in half of the pots at a rate of 3.7 t/ha. After the ethanol evaporated, the pots were seeded with 4 soybeans and 1 cc of fescue seed; nutrient solution was added and the pots were placed in a growth chamber and maintained under the same conditions as described previously. Soybean plants were harvested at 16 days after planting and fescue at 50 days after planting. ^{14}C-PCB was extracted from the plants by twice homogenizing with 25 ml of 1:1 hexane:acetone, combining the extract, centrifuging, pouring the supernatant into a beaker and evaporating to near-dryness. Five milliliters of hexane were added to dissolve the residue, after which three 1.0 ml aliquots were removed and placed into individual counting vials containing 0.5 ml of scintillation cocktail {5.0 g of 2,4-diphenyloxazole (PPO), 0.1 g of 1,4-bis[2-(4-methyl-5-phenyloxazolyl)]benzene (POPOP), 1000 ml of Triton X-100, and 1000 ml of toluene} amd 3 drops of clorox. After the chlorophyll had been bleached away (2-3 minutes), 15 ml of scintillation cocktail was added to the vials and the samples counted for three 10-minute periods in a liquid scintillation spectrometer. Extraction efficiency ranged from 86 to 99% and counting efficiency ranged from 70 to 80%. To test extraction efficiency, ^{14}C-residue remaining in extracted plant matter was analyzed by placing dried plant material on black paper, igniting in a Schoniger combustion flask, trapping $^{14}CO_2$

[1]Personal communications. Rogert G. Lewis, Environmental Protection Agency, Research Triangle Park, North Carolina, reported that the chromatographic isomer distribution more closely resembled Aroclor 1248, but the average chlorine content reported by New England Nuclear is 54%, therefore, the compound is designated Aroclor 1254.

in 15 ml of phenethylamine, and radioassaying a 2 ml aliquot. Counting efficiency ranged from 66 to 85%.

^{14}C-Aroclor was applied to the center leaflet of the first trifoliate leaf of 18 day old soybean plants at rates of 0.026 μCi per leaf. Twelve days later, the soybean plants were harvested and sectioned into the following parts: treated leaflet, adjacent untreated leaflets and petiole, plant portion acropetal to the treated leaf, plant portion basipetal to the treated leaf, and roots.

^{14}C-PCB was extracted from the various plant materials and radioassayed according to the previously described procedures.

RESULTS AND DISCUSSION

Plant Growth Studies. PCB applied to the soil significantly inhibited height and fresh top weight of soybean plants at the high rates of application (Table 1). Malformation (twisting and curling) of newly developing leaves was also observed on plants growing in the 1000 ppm PCB treated soils. Low rates of PCB were also inhibitory but the plant measurement values were not significantly different from the untreated controls. Activated carbon added to the soil removed the inhibitory effects of PCB at all rates of application. Root growth of soybean plants was inhibited to some degree (differences significant at 10% level) by both PCB and activated carbon (data not included). Root growth inhibition from PCB increased with increasing PCB rate, probably because of the phytotoxic action of the chemical. Plants growing in pots treated with carbon had smaller root systems because the carbon, which adsorbed applied nutrients, became localized in the upper 5 cm of the soil.

TABLE 1

Growth measurements of soybean and fescue plants after 26 and 42 days, respectively, exposure to PCB (Aroclor 1254) and activated carbon applied to Lakeland sand.

Application rate		Soybean				Fescue	
PCB	Carbon	Height		Fresh top weight		Fresh top weight	
(ppm)	(t/ha)	(cm)	(% I)	(g)	(% I)	(g)	(% I)
0	0	47	-	9.3	-	9.5	-
1	0	44	6	9.0	3	10.1	+6
10	0	43	9	8.3	11	10.3	+8
100	0	36	23	6.8	27	9.8	+3
1000	0	40	15	7.3	22	8.0	16
0	3.7	48	+2	10.2	+10	9.5	0
1	3.7	47	0	9.7	+4	9.8	+3
10	3.7	47	0	9.2	1	10.5	+11
100	3.7	49	+4	10.4	+12	9.8	+3
1000	3.7	47	0	10.1	+9	9.6	+1
LSD (.05)		6	12	1.3	14	1.0	11

On the 21st day of the study, it became apparent that soybean plants growing in the PCB treated soils were taking up less water than the control plants (Table 2). Inhibition of water uptake caused by PCB ranged from 12 to 52%, but it was not clear whether the effect of PCB was direct, indirect, or a combination of effects. The appearance of malformations on newly developing leaves, which later disappeared, suggested that PCB did affect plant metabolism at least to some degree. Additions of activated carbon to the soil completely removed the inhibitory effect of PCB on water uptake. Data in Table 2 illustrate that every level of PCB applied had an inhibitory effect on the accumulated water uptake by soybean plants over the 5-day period in which water measurements were recorded. Activated carbon additions prevented the inhibitory effects of PCB on water uptake.

TABLE 2

Five-day accumulated water uptake (21st to 25th day) for soybean plants exposed to PCB (Aroclor 1254) and activated carbon applied to Lakeland sand.

Application rate		Five-day accumulated water usage	
PCB (ppm)	Carbon (t/ha)	(cc)	% Inhibition
0	0	129	-
1	0	113	12
10	0	102	21
100	0	71	45
1000	0	62	52
0	3.7	138	+7
1	3.7	129	0
10	3.7	128	1
100	3.7	124	4
1000	3.7	127	2
LSD (.05)		7.4	6

PCB applied to the soil was also inhibitory to the fresh top weight of fescue, but only at the highest level of application (Table 1). Lower rates of PCB had no significant effect on fescue growth. Root weights of fescue were not significantly affected by PCB. Activated carbon applied to the soil removed the inhibitory effect of PCB on fescue growth and plants growing in soils treated with both PCB and activated carbon grew as though PCB were not present.

^{14}C-PCB Studies. Soybeans growing in ^{14}C-PCB treated Lakeland soil for 16 days absorbed and translocated very small amounts (163 dpm) of ^{14}C-PCB into the tops (Table 3). Fescue growing for 50 days, however, absorbed and translocated approximately 10x (1700 dpm) as much ^{14}C-PCB as soybean. Activated carbon added to the soils reduced uptake of ^{14}C-PCB by both soybean and fescue essentially to zero, 13 and 0 dpm, respectively. The studies confirm the inactivating effect of activated carbon on PCB availability to growing plants.

TABLE 3

Uptake by soybean and fescue of ^{14}C-PCB from Lakeland sand treated with ^{14}C-PCB (Aroclor 1254)[1] and activated carbon.

Application rate		^{14}C-PCB in plant tops[3]			
^{14}C-PCB	Carbon	Soybean[4]		Fescue[5]	
(dpm)[2]	(t/ha)	(dpm)	% of applied	(dpm)	% of applied
1×10^6	0	163	0.016	1700	0.17
1×10^6	3.7	13	0.001	0	0

[1] Chromatographic pattern more closely resembled Aroclor 1248

[2] 0.5 µCi/pot (20 ppm).

[3] Corrected for background of 32 dpm. Mean values for two replications.

[4] Total for two plants (7.3 g fresh weight) harvested at 16 days after treatment.

[5] Total of 5.5 g/pot fresh top weight harvested at 50 days after treatment.

Of the ^{14}C-PCB applied to the center leaflet of the first trifoliate leaf of soybean, only 6.7% was recovered 12 days after application (Table 4). Apparently 93.3% of the applied ^{14}C-PCB volatilized or somehow left the site of application. This is in agreement with MOSA et al. (1976) who, after 6 weeks, recovered only 3.7% of the ^{14}C-PCB applied to the leaf of a marsh plant. It is also known that the ^{14}C-Aroclor 1254 used in these studies more closely resembled Aroclor 1248 and contained isomers which were relatively volatile.[1]

TABLE 4

Distribution of ^{14}C in soybean plants 12 days after ^{14}C-PCB (Aroclor 1254)[1] applied to center leaflet of first trifoliate leaf.

Plant portion[2]	^{14}C in plant tissues[3]		^{14}C-PCB distribution in plant (%)
	(dpm)	% of applied	
Treated leaflet	2707	5.11	76
Attached leaflets & Petiole	77	0.14	2
Plant parts acropetal to treated leaf	35	0.07	1
Plant parts basipetal to treated leaf	683	1.29	19
Roots	64	0.12	2
Total	3566	6.73	100

[1] ^{14}Chromatographic pattern more closely resembled Aroclor 1248.

[2] Two plants per pot harvested. Average weight was 4.4 g/plant.

[3] 53,000 dpm applied. Mean values for two replications.

The majority (76%) of applied ^{14}C-PCB was found in the treated leaflet. Approximately 2% was found in the adjacent leaflets and petiole and a similar amount found in the roots. A lesser amount (1%) was found in plant parts acropetal to the treated leaf. The highest amount (19%), outside of the treated leaf, was found in the plant parts basipetal to the treated leaf. It appeared that most of the ^{14}C-PCB applied to the soybean leaf evaporated or remained at the treated site, and that very small amounts were translocated to other parts of the plants, primarily downward into the lower plant parts.

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