declined to low levels following 10 days of aerobic conditions. The instability of carbofuran phenol under aerobic conditions was confirmed in a repeat experiment when carbofuran phenol (5.8×10^4 cpm/20 g of soil) formed from carbofuran at the end of 20-day anaerobic cycle declined to negligible levels (0.1×10^4 cpm/20 g of soil) after 20 days of subsequent incubation under aerobic conditions as compared to a value of 9.0×10^4 cpm/20 g of soil under 40 days of continued anaerobiosis. These studies indicated that carbofuran was more rapidly hydrolyzed under anaerobic conditions than under aerobic conditions; but its hydrolysis products, carbofuran phenol and 3-hydroxycarbofuran, which resisted further degradation under continued anaerobiosis, were rapidly transformed, perhaps to carbon dioxide when the anaerobic system was returned to aerobic conditions. Alternate anaerobic and aerobic conditions generated by intermittent flooding and drying cycles in rice fields may thus assist in more intensive, but less extensive, degradation of carbofuran than in either system alone.

ACKNOWLEDGMENT

We thank H. K. Pande (Director) for critical comments and A. Anjaneyulu (virologist) for providing soil samples from his field experiment. Labeled carbofuran and technical grade carbofuran and carbofuran phenol were gifted by FMC Corporation, Middleport, N.Y.

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Received for review November 21, 1977. Accepted June 2, 1978. The study was financed, in part, by funds from the International Atomic Energy Agency, Vienna, Austria (Contract No. 2089/SD), Department of Science and Technology, Government of India and Indian Council of Agricultural Research.

Degradation of Pentachloronitrobenzene (PCNB) in Anaerobic Soils

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Anaerobic degradation of ^{14}C-labeled pentachloronitrobenzene (PCNB) was examined in flooded and moist Hagerstown silty clay loam, with and without cellulose amendments. All treatments were aerated with nitrogen in a continuous flow-through system which permitted trapping of CO_2 and volatilized products. PCNB enhanced soil respiration in all treatments. Total CO_2 production was greater in moist than in flooded soils. Essentially no $^{14}CO_2$ was evolved from any treatments. PCNB volatilized from all treatments, but volatilization was reduced by cellulose amendments. Although extractable radioactivity was the same (70%) from all treatments at the conclusion of the 40-day incubation period, differences were observed in the relative distribution of PCNB and its degradation products. Product identification was by thin-layer and gas–liquid chromatographic comparison with authentic standards. Pentachloroaniline (PCA) was the principal degradation product. Pentachlorothioanisole (PCTA) was more abundant in moist (5.8–8.2%) than in flooded soil (0.3–0.5%). Pentachlorophenol was also detected as a degradation product. Further degradation of PCA and PCTA were examined in similar anaerobic soils. PCTA did not significantly alter soil respiration, whereas PCA provided slight inhibition in cellulose amended soil.

Pentachloronitrobenzene (PCNB) is used as a seed dressing and as a soil treatment to control several soilborne plant pathogens. Application rates have sometimes been as high as 200 lb/acre (Sharvelle, 1961; Thomson, 1967). Considerably lower rates of application are now more commonly used, however. Its degradation in soil (Caseley, 1968; Bauser and Bosshardt, 1975; Ko and Farley, 1969; Wang and Broadbent, 1972, 1973; De Vos et al., 1974; Beck and Hansen, 1974; Nakanishi, 1972; Nakanishi and Oku, 1969; Chacko et al., 1966; U.S. EPA, 1976) has been examined. Generally, PCNB is degraded in soil more rapidly under anaerobic or flooded conditions than in moist aerobic soil. The principal degradation products, pentachloroaniline (PCA) and pentachlorothioanisole (PCTA), have been identified in both soil and microbial systems.

The significance of PCNB residues in the environment is not fully understood. Gorback and Wagner (1967) investigated PCNB residues in potatoes grown in PCNB-treated soil and detected PCNB, PCA, and one unidentified metabolite in the potato peel. Both PCA and the unidentified metabolite, but not PCNB, were also found in the inner potato tissue. The unidentified metabolite was later described by Kuchar et al. (1969) as having an identical gas chromatographic retention time as PCTA. Kuchar et al. (1969) found PCNB, PCA, and PCTA, in addition to several PCNB impurities (pentachlorobenzene, hexachlorobenzene, and 2,3,4,5-tetra-

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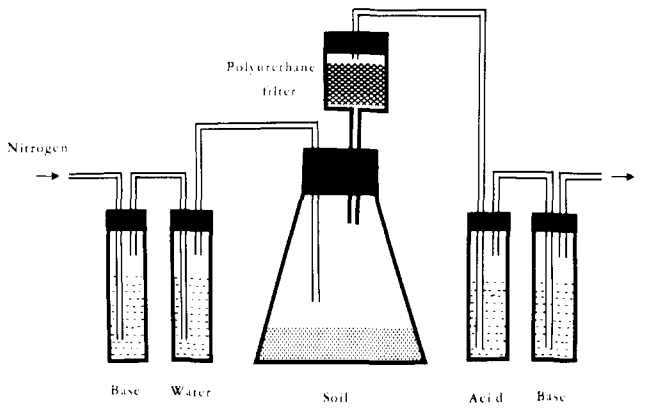

Figure 1. Apparatus used to study the degradation of ^{14}C-labeled pesticides in an anaerobic environment.

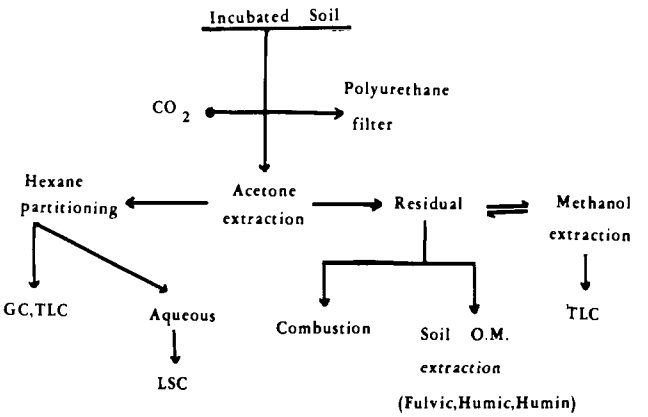

Figure 2. Extraction scheme used for ^{14}C-labeled pesticide treated soils.

chloronitrobenzene), in cotton plants grown for 2 weeks in soil containing 300 ppm of PCNB. Leistra and Smelt (1974) found PCNB at 40–70 cm depths in soil. They believed that soil tillage accounted for PCNB distribution to depths of 40 cm, whereas they attributed diffusion and leaching to PCNB movement into the 40–70 cm depths. Caseley (1968) observed an 80% loss of PCNB from soil after 10 months of which chemical and biodegradation accounted for only 18%. Most of the PCNB was lost by volatilization and recovered in trapping solutions.

None of the preceding investigations were performed with ^{14}C-labeled PCNB. The purpose of this investigation was to examine the degradation of [^{14}C]PCNB in anaerobic soils under conditions permitting a ^{14}C balance.

MATERIALS AND METHODS

A manifold assembly system was used to study the degradation of pentachloronitrobenzene (PCNB) in anaerobic environments (Figure 1). Each experimental unit consisted of two scrubbing tubes, a soil incubation flask equipped with a polyurethane filter to trap volatilized products (Kearney and Kontson, 1976), followed by two additional scrubbing tubes. The initial scrubbing tubes contained, respectively, 1.0 N NaOH to remove CO_2 from the incoming gas and water to maintain a saturated atmosphere. The final scrubbing tubes contained, respectively, 0.1 N HCl to trap evolved NH_3 and 0.1 N NaOH to trap evolved CO_2. While no attempt was made to monitor the actual gas flow (volume), needle valves were used in conjunction with each incubation unit to assure a uniform distribution (bubbling rate) through each unit.

Data on volatilization, NH_3, total CO_2, and $^{14}CO_2$ evolution were obtained at 4-day intervals throughout the 40-day incubation period. Volatilization of ^{14}C products was determined by extracting the polyurethane filters with 25 mL of hexane and quantitating the ^{14}C content by liquid scintillation counting. The volatilized products were identified by gas–liquid chromatographic (GLC) analyses. Ammonia determinations were made by titration of the acid trap contents with 0.1 N NaOH. Total CO_2 determinations were made by titration of aliquots of the base solution with 0.1 N HCl after $BaCl_2$ addition and precipitation of the resultant $BaCO_3$. Two 1-mL aliquots of the basic trapping solution were analyzed for $^{14}CO_2$ content by liquid scintillation counting.

Each incubation flask contained 100 g (air-dry weight) of a Hagerstown silty clay loam: pH, 7.5; sand, 17.0%; silt, 50.6%; clay, 32.4%; organic matter, 2.3%; cation-exchange capacity, 8.8 (mequiv/100 g); and 21.1% water at $^1/_3$ bar moisture content. The soil was treated with ^{14}C-labeled PCNB (sp. act. 14.5 mCi/mM) in benzene to a final concentration of 10 ppm. Microcrystalline cellulose powder

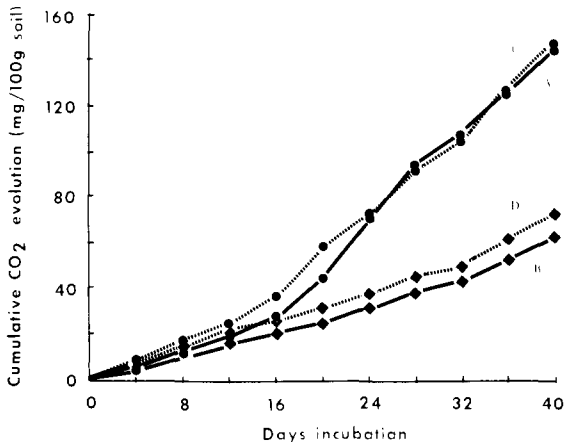

Figure 3. Cumulative CO_2 evolution from PCNB treated anaerobic flooded soil: (A) control, flooded and with cellulose; (B) control, flooded; (C) PCNB, flooded and with cellulose; and (D) PCNB, flooded.

was added at the 1% level to one-half of the PCNB-treated and to one-half of the control flasks. The cellulose was added as a microbial substrate to ensure a greater degree of anaerobicity. After mixing the chemicals into the soil by stirring and agitation, one set of flasks was watered to 75% of $^1/_3$ bar soil moisture content. The second set of flasks were flooded with water to a depth of approximately 1 cm over the soil surface. Each experimental unit was then attached to a manifold and aerated with N_2 to further insure anaerobic conditions. Duplicate flasks were used for each treatment.

At the end of the 40-day incubation period the soils were extracted twice with 100 mL of acetone and once with 100 mL of methanol with the aid of a polytron homogenizer (Figure 2). Extractable radioactivity was determined by volumetizing the solutions and liquid scintillation counting of 1-mL aliquots of the extracts. The acetone extracts were combined and partitioned with 50 mL of hexane. After a Florisil column cleanup, the hexane extracts were analyzed by GLC for PCNB and degradation products. A Hewlett-Packard Model 7600A instrument with a ^{63}Ni electron-capture detector was used for GLC analyses. The 6 ft × $^1/_4$ in. o.d. glass column was packed with 10% DC-200 on 80 to 100 mesh Gas-Chrom Q. GLC conditions were: a 95% argon–5% methane mixture at 120 mL/min flow rate; column, inlet, and detector temperatures were 190, 250, and 300 °C, respectively.

Unextractable, or bound residues, were determined by both combustion and a soil organic matter fractionation procedure (U.S. EPA, 1975).

Table I. Percent ^{14}C Recovered from PCNB Treated Anaerobic Soil

	% ^{14}C recovered as					
	volatiles			solvent		
treatment	$^{14}CO_2$	filter	aqueous	extractable	residual	total
flooded						
cellulose	0	0.3	0.3	69.4	20.7	90.7
no cellulose	0	0.5	0.4	69.9	19.5	90.3
moist						
cellulose	0	0.6		71.6	18.4	90.6
no cellulose	0	1.7		71.2	12.9	85.8

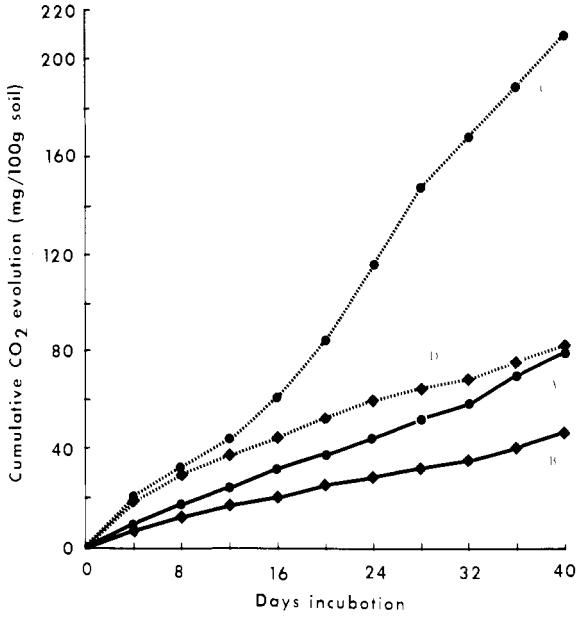

Figure 4. Cumulative CO_2 evolution from PCNB treated anaerobic moist soil: (A) control, with cellulose; (B) control; (C) PCNB, with cellulose; and (D) PCNB.

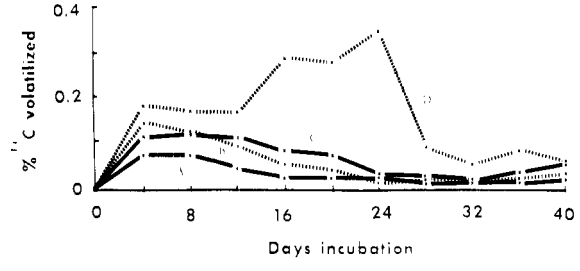

Figure 5. Volatilized ^{14}C trapped at indicated sampling periods: (A) PCNB, flooded and with cellulose; (B) PCNB, flooded; (C) PCNB, moist, with cellulose; and (D) PCNB, moist.

RESULTS AND DISCUSSION

PCNB and cellulose generally enhanced total CO_2 production in both flooded and moist soil treatments (Figures 3 and 4). Stimulation generally was less in flooded soils, possibly due to limitations imposed by the water barrier on CO_2 diffusion. Total CO_2 evolution in control flooded and moist soils, however, was quite similar. Stimulation of CO_2 production was maximum in moist soils. Enhancement of CO_2 production by PCNB was much greater in moist soil than in flooded soil. Actual differences between moist and flooded soils, however, may be more due to CO_2 solubility in water and/or the water acting as a physical barrier to CO_2 diffusion.

Loss of radioactivity from the soil by volatilization was minimal (0.3–1.7%) (Figures 5 and 6, and Table I). GLC analyses of hexane extracts of the polyurethane filters showed the presence of only PCNB. Volatilization was greater during the initial stages of incubation, but decreased with time (Figure 5). Cellulose ammendments decreased volatilization losses, presumably by providing more adsorption sites for the ^{14}C products. Volatilization losses were greater in moist than in flooded soils (Figure 6), presumably due to the limitations imposed by the water barrier over the flooded soils.

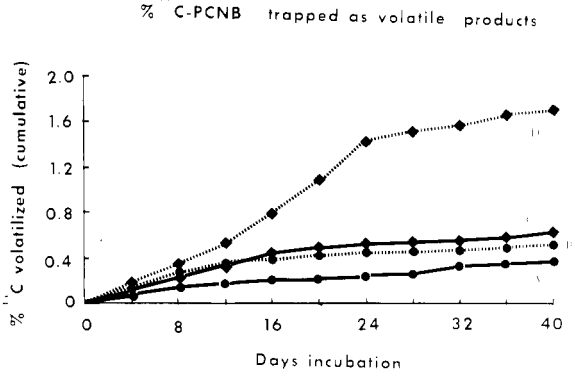

Figure 6. Cumulative volatilization of ^{14}C from treated soil: (A) PCNB, flooded and with cellulose; (B) PCNB, flooded; (C) PCNB, moist, with cellulose; and (D) PCNB, moist.

Very little information is available regarding the loss of PCNB from soil by volatilization. Most soil degradation investigations of PCNB have ignored this mechanism of PCNB dissipation from soils. PCNB has sufficient vapor pressure (1.13×10^{-4} at 24 °C) (Goring, 1967) to expect that some diffusion through soil could occur. Although Munnecke et al. (1962) were unable to show that soil treated with PCNB contained fungitoxic vapors, Richardson and Munnecke (1964) used a more sensitive bioassay method and were able to demonstrate fungitoxic concentrations of PCNB in the soil air. Caseley (1968) observed losses of up to 62% by volatilization from moist soil. Losses of PCNB by volatilization increased as soil moisture content increased, presumably due to its displacement from adsorption sites by water. The greatest PCNB losses recorded by Caseley (1968) were observed in soil at the 50% (w/w) level, which is far in excess of the moisture levels used in our investigation. Smaller, but still substantial PCNB losses were observed at a 20% (w/w) moisture level, which is more nearly comparable with our soil moisture content. Caseley's investigations, however, were conducted in presumably aerobic soil, whereas ours were conducted in anaerobic soils. In other investigations to be reported later, we have observed somewhat higher PCNB losses from the same soil at the same moisture content used herein, but incubated aerobically. Thus, it would seem apparent that the degree of aerobicity or anaerobicity may have some influence on volatilization of PCNB from soil.

Radioactivity in the water covering flooded soils was less than 0.5% of the isotope originally added (Table I). The combined (acetone and methanol) organic solvent extractable radioactivity was high (ca. 70%) and essentially the same from all treatments. Approximately 56–58% of

Table II. Percent Distribution of PCNB and Its Degradation Products in Hexane Extracts

treatment	% of products present as[a] PCNB	PCA	PCTA
flooded			
cellulose	13.5	86.5	0.5
no cellulose	12.1	87.6	0.3
moist			
cellulose	13.2	81.0	5.8
no cellulose	36.0	55.8	8.2

[a] PCNB, pentachloronitrobenzene; PCA, pentachloronitroaniline; PCTA, pentachlorothioanisole.

the original ^{14}C activity was recovered from all treatments with the initial acetone extraction. An additional 10–12% was recovered with the methanol extraction. When the acetone extracts were made up to a standard volume with water and partitioned with hexane, 84–92% of the extracted ^{14}C activity was partitioned into the hexane and 8–16% remained in the aqueous phase. Although the extractable radioactivity was nearly equal in all treatments, differences were observed in the distribution of ^{14}C in PCNB and its degradation products (Table II) in the hexane fraction. ^{14}C products partitioned into hexane were identified by GLC comparisons with authentic standards. Their GLC retention times were: PCNB, 2.7 min; pentachloroaniline (PCA), 3.5 min; and pentachlorothioanisole (PCTA), 4.9 min. More PCNB remained in the moist soil without cellulose than any of the other three treated soils. Less PCA was produced in moist soil without cellulose than in any of the other three treatments. More PCTA was produced in both moist soils than in either flooded soils. These results reflect the degree of anaerobicity that would have existed in these soils, i.e., the flooded soils were more anaerobic than the moist soils, and the moist soil with cellulose would be more anaerobic than the moist soil without cellulose. Thin-layer chromatography of these fractions with a hexane solvent system generally confirmed these results.

Unfortunately, after sampling for residual ^{14}C activity, we discarded the aqueous phase remaining after partitioning with hexane. In a supplemental investigation, however, the aqueous phase was acidified to pH 1 and again partitioned with hexane and submitted to further analysis. Both GLC and TLC analysis of methylated and unmethylated portions of this extract revealed that much of it was composed of pentachlorophenol (PCP). The GLC retention time of the pentachloroanisole derivative of PCP was 6.9 min under identical GLC conditions as described herein, except that a column temperature of 160 °C was used. On silica gel HF254 TLC plates, the unmethylated ^{14}C activity was chromatographically identical with standard PCP in the following solvent systems: R_f 0.16 in hexane–benzene–acetone (7:3:1) and R_f 0.61 in benzene–dioxane–acetic acid (90:25:4).

The methanol extracts were concentrated on a rotary evaporator and submitted to TLC analyses in the same solvent systems and exposed to no-screen X-ray film. We detected only two spots containing ^{14}C activity on hexane–benzene–acetone developed plates, and three spots on benzene–dioxane–acetic acid plates. Nearly all of the ^{14}C activity on both plates was concentrated in one spot chromatographically identical with standard PCA at R_f 0.50 in hexane–benzene–acetone and R_f 0.65 in benzene–dioxane–acetic acid. Further confirmation was obtained by a positive reaction (bright purple color) to a spray (1% *N*-1-naphthylethylenediamine dihydrochloride in ethanol) after exposure of the developed TLC plates to

Table III. Percent ^{14}C Recovered in Organic Matter Fractions of PCNB Treated Anaerobic Soil

treatment	% ^{14}C in soil organic matter fraction[a] fulvic	humic	humin
flooded			
cellulose	4.9 (1.0)	12.8 (2.7)	82.3 (17.0)
no cellulose	4.7 (0.9)	10.4 (2.0)	84.9 (16.6)
moist			
cellulose	4.5 (0.8)	10.7 (2.0)	84.6 (15.6)
no cellulose	6.6 (0.8)	9.7 (1.2)	83.7 (10.9)

[a] Numbers in parentheses equal percent of original application.

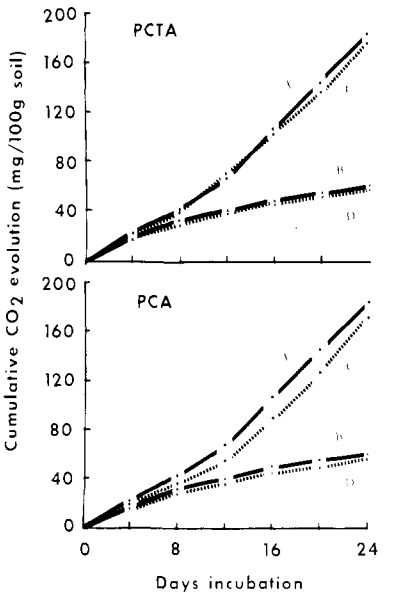

Figure 7. Cumulative CO_2 evolution from PCA and PCTA treated anaerobic moist soil: (A) control with cellulose; (B) control with cellulose; (C) PCA or PCTA with cellulose; and (D) PCA or PCTA only.

nitrous oxide fumes generated by reacting concentrated H_2SO_4 and $NaNO_2$ in a closed container. This system is extremely sensitive and generally specific for detecting aromatic amines. A second more polar spot containing ^{14}C activity (1%) on the benzene–dioxane–acetic acid plates was detected at R_f 0.31. The remaining spots on both plates (1%) were located on the origin. We made no further effort to characterize any of these latter compounds.

Residual radioactivity was greater in the flooded soils and the moist cellulose treated soils than in the moist soil without cellulose. Extraction and fractionation of the soil organic matter from these treated soils revealed that most of the ^{14}C activity was associated with the humin fraction (Table III). There seemed to be little influence of soil treatment on the distribution of the bound residue.

Further degradation of PCP, PCA, and PCTA was examined in both anaerobic and aerobic soils. In these investigations, [^{14}C]PCP and unlabeled PCA and PCTA were incubated for 24 days in anaerobic moist soil without the cellulose amendment. The results of the [^{14}C]PCP investigations are reported elsewhere (Murthy and Kaufman, 1978). The effects of these treatments on total CO_2 production in PCA- and PCTA-treated anaerobic soils are shown in Figure 7. PCTA did not significantly alter soil respiration in comparison with controls. A slight

Table IV. Percent PCA and PCTA Recovered from Soil after 24 Days Incubation as Determined by GLC Analysis

treatment	anaerobic	aerobic
PCA only	40.5	69.3
PCA + cellulose	53.5	
PCTA only	63.3	85.0
PCTA + cellulose	57.2	

inhibition of soil respiration by PCA was also observed in cellulose-amended soil. This observation agreed with that of Ko and Farley (1969), who found PCA somewhat inhibitory to several isolated soil microorganisms.

Because nonradioactive PCA and PCTA were used in these investigations, only the organic solvent-extractable-product distribution was examined. Less PCA and PCTA were recovered from anaerobic soils than from aerobic soils (Table IV). More PCA was recovered from cellulose treated anaerobic soil than from the moist anaerobic soil. Conversely, more PCTA was recovered from moist anaerobic soil without cellulose than from the moist anaerobic soil with cellulose.

Significantly lower product recoveries were obtained in these experiments (Table IV) than were obtained in experiments with PCNB (Table I), in spite of the shorter incubation period. These results would suggest either further more rapid conversion of the PCA and PCTA to other products, greater adsorption, and thus greater bound residual levels of these compounds, substantial losses through volatilization, or some combination of these possibilities. Although preliminary experiments indicated some losses of PCA and PCTA by volatilization from aerobically incubated soils, these loss rates were not sufficient to account for the low recoveries of PCA and PCTA in the experiments reported here. Also, no losses of PCA or PCTA by volatilization were observed in the [^{14}C]PCNB experiments reported herein. Without [^{14}C]PCA and [^{14}C]PCTA the significance of adsorption to soil particles can not be adequately determined in this investigation. Aromatic amines are known to be readily adsorbed in soil, however. PCTA, per se, could also be expected to strongly adsorb because of the electron density surrounding the S atom.

In our PCP investigations (Murthy and Kaufman, 1978), however, we observed that pentachloroanisole was readily reduced to pentachlorophenol in anaerobic soils. Conceivably, therefore, pentachlorothiophenol could be a product of PCTA in soil. The adsorptive capacity of the pentachlorothiophenol would be considerably greater than PCTA. The formation of pentachlorophenyl disulfide might also be expected. Sulfur compounds are also frequently metabolized to sulfoxides and sulfones. Steric hinderance by the bulky chlorine substituents, however, may preclude formation of the sulfoxide and sulfone compounds during degradation of PCTA. Both GLC and TLC analyses of the acetone–hexane extracts from PCA- and PCTA-treated soils revealed no additional degradation products. Thus, based on the results obtained in this investigation it would appear that PCA and PCTA are end products in the degradation of PCNB in soil.

Throughout these investigations, the extracts analyzed were also examined for the presence of hexachlorobenzene (HCB), pentachlorobenzene (PCB), and 2,3,5,6- and 2,3,4,5-tetrachloronitrobenzene (TCNB). None of these products were detectable in the [^{14}C]PCNB used herein. A small amount of HCB was the only impurity detected in the unlabeled PCNB used. No HCB was detected in the volatile fractions emanating from the anaerobic soils. This contrasts with the work of Beall (1976) who concluded that, while HCB was persistent when incorporated in soil,

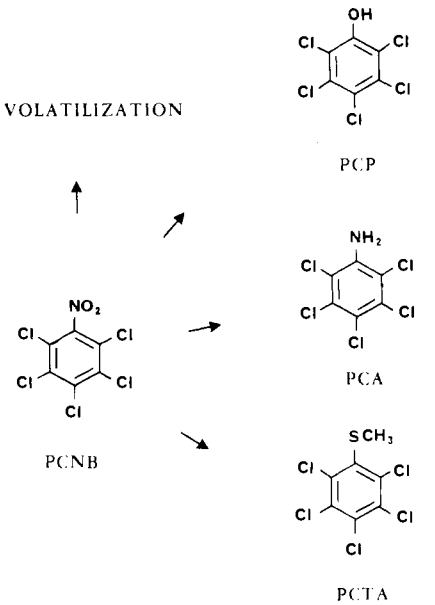

Figure 8. Pathways for loss of PCNB from anaerobic soil.

it did volatilize from soil surfaces. His investigations were performed in aerobic soils, however. In our investigations, recoveries of the HCB contaminant were greater from flooded soils than moist soils and from cellulose amended than from unamended soil. No other degradation products attributable to HCB were detected.

2,3,4,5-TCNB has been detected as an impurity in PCNB samples (Kuchar et al., 1969). Loss of 2,3,5,6-TCNB from soil is much more rapid than that of PCNB (Caseley, 1968). According to Caseley, 2,3,5,6-TCNB is about four times as volatile as PCNB, which accounted for the increased rate of loss. Neither 2,3,4,5- nor 2,3,5,6-TCNB were detected in any of the samples analyzed in our investigation, nor were they detectable in the starting materials. De Vos et al. (1974) isolated a compound from PCNB-treated greenhouse soils which had the same GLC retention time as 2,3,5,6-TCNB, but lacked a standard sample of 2,3,4,5-TCNB for chromatographic comparisons. They also identified a tetrachloroaniline and a tetrachlorothioanisole, but did not indicate which isomer. Presumably, these were simply the reduced or substituted degradation products of the tetrachloronitrobenzene impurity in the PCNB originally applied. The absence of any tetrachloro products in our investigations is actually somewhat surprising in view of our results obtained with PCP degradation under identical experimental conditions (Murthy and Kaufman, 1978). In those investigations the 2,3,4,5- and 2,3,5,6-tetrachlorophenol were isolated as the principal degradation products of PCP in anaerobic soils. The results obtained in this investigation would suggest that the presence of an amine, nitro, or methylthio group on the chlorobenzene ring confers a greater degree of stability than the hydroxyl group does.

Based on the results obtained in this and other investigations, the pathway illustrated in Figure 8 was proposed to account for dissipation of PCNB from anaerobic soils. Some loss by photodecomposition (Crosby and Hamadmad, 1971) might also be expected in aqueous systems or on soil surfaces exposed to light. Loss of PCNB in soil occurs principally by conversion to PCA with some loss by volatilization and conversion to PCTA and PCP. Both PCA and PCTA appear to be end products with no further degradation. These products would presumably remain in the soil as residues, unless absorbed by plants (Gorback and Wagner, 1967; Kuchar et al., 1969). Conversion to

PCP with subsequent further degradation of PCP (Murthy and Kaufman, 1978) would seem to be the only pathway by which PCNB is more completely degraded. Further work with PCA and PCTA is in progress, however, to verify this conclusion.

ACKNOWLEDGMENT

This work was completed during the tenure of the senior author as an International Atomic Energy Agency Fellowship recipient. The authors are indebted to E. J. Kuchar and T. O. Evrard of Agricultural Products Division of Olin for samples of pentachloronitrobenzene, pentachlorobenzene, hexachlorobenzene, pentachloroaniline, pentachlorothioanisole, and 2,3,4,5- and 2,3,5,6-tetrachloronitrobenzene.

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Received for review October 31, 1977. Accepted May 25, 1978. Presented at the 173rd National Meeting of the American Chemical Society, Pesticide Division, New Orleans, La., March 1977.

Physicochemical Characteristics of Bound Dinitroaniline Herbicides in Soils

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Soil bound residues of six [*phenyl*-^{14}C]dinitroaniline herbicides constituted 7–21% of the original ^{14}C added to aerobically incubated Matapeake soil. Bound residues of butralin [4-(1,1-dimethylethyl)-*N*-(1-methylpropyl)-2,6-dinitrobenzeneamine] from Chillum soil were 3 and 13% after aerobic and anaerobic incubation, respectively. Prolonged benzene–methanol extraction gave stable residue values. The extracts contained unidentified radioactive components, chromatographically separable from the parent compounds. Ultrasonic dispersion in certain alkaline solvents increased extractability of anaerobic butralin residues, probably because organic matter was solubilized. Distribution of residues in fulvic acid, humic acid, and humin was (a) butralin (aerobic Chillum soil), 51, 7, and 42%; (b) butralin (anerobic Chillum), 14, 40, and 46%; and (c) 6 herbicides (aerobic Matapeake soil), 50, 15–20, and 25–35%. When we used a milder extraction procedure, anaerobic bound butralin was widely distributed among soil organic and organo/mineral fractions, especially in humic acid and humin. The six dinitroaniline residues were thermally degraded between ca. 250 and 500 °C: anaerobic butralin was lost at 300–375 °C. This corresponds to oxidation of phenolic hydroxyl (and perhaps carboxyl) groups in soil organic matter, but tends to negate the occurrence of bound ^{14}C in clay interlayers or as carbonate.

Using radioisotopes as tracers within pesticide molecules has been of inestimable value in understanding their behavior in living systems, including the soil. It has also made us aware that unextractable residues may accumulate, since combustion of the extracted soil (when ^{14}C was used) yields $^{14}CO_2$. These residues are now termed "soil bound residues", defined as "that unextractable and chemically unidentifiable pesticide residue remaining in fulvic acid, humic acid, and humin fractions after exhaustive sequential extraction with nonpolar organic and polar solvents" (U.S. Environmental Protection Agency, 1975). Bound residues have been addressed as a possible concern for pesticide registration.

Bound and conjugated residues in soils and plants were the focal point of a recent conference (Kaufman et al., 1976). Kearney's (1976) final summary reflected the uncertainties of the bound residue question. Chemical identification of the bound entities is rare (Booth et al., 1976; Hill, 1976), although this is a primary research objective. Usually, measurement is of the total residue present and its distribution among three rather arbitrarily

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