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TERMINAL RESIDUES OF ORGANOPHOSPHORUS PESTICIDES

Prepared for publication by

R. GREENHALGH

Chemistry and Biology Research Institute,
Ontario, Canada

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TERMINAL RESIDUES OF ORGANOPHOSPHORUS PESTICIDES

R. Greenhalgh

Chemistry and Biology Research Institute, Agriculture Canada
Ottawa, Ontario, K1A 0C6, Canada

Current data about the degradation of organophosphorus pesticides in animal, plant and soil substrates and the environment are summarized in this report. Comparative metabolic and chemical studies with racemic and optically active enantiomers are reviewed. These reactions provide an understanding of the stereochemistry involved in the oxidation of the P=S moiety, which is the main in-vivo activating reaction for organophosphorus insecticides.

ANIMALS

The metabolism of ^{14}C-triazophos [0,0-diethyl 0-(1-phenyl-1,2,4-triazol-3-yl) phosphorothioate] was studied in rats after single and repeated oral doses. Four days after the single dose, 76.3% of the total radioactivity had been excreted in the urine and 21.0% in the feces. After the daily application of 12 doses, 69-83.4% of the labeled material was eliminated in the urine and 30.9-18.1% in the feces by day 16. Elimination was markedly slower following prolonged application of the insecticide. Unchanged triazophos and 1-phenyl-1,2,4-triazol-3-ol were found in the feces, whilst the urine contained urea (ca. 85% of the urinary activity), the glucuronides of 1-phenyl-1,2,4-triazol-3-ol (ca. 3%), 1-phenylsemicarbazide (ca. 5%) and semicarbazide (ca. 5%) and two unidentified metabolites (1).

Rats were treated with 5 daily doses of ^{14}C-phenoxy- or ^{14}C-phenyl-leptofos [O-(4-bromo-2,5-dichlorophenyl) O-methyl phenylphosphonothioate]. The cumulative radioactivity excreted with the ^{14}C-phenyl isomer was 80.9% in the urine, and 24.98% in the feces after 11 days by male rats. ^{14}C-Phenoxy-leptofos gave levels of 94.03% in urine and 14.01% in feces for the same time interval. In both cases, the total ^{14}C recovered from female rats was less. No detectable residues of leptofos were found in urine. The ^{14}C-phenoxy labeled material was excreted in the urine almost quantitatively as 4-bromo-2,5-dichlorophenol (more than 98%), whereas the ^{14}C-phenyl isomer gave O-methyl phenylphosphonothioate. The continuous exposure of rats to leptofos did not limit the capacity of these animals to excrete the substance efficiently (2).

Phosfolan (0,0-diethyl 1,3-dithiolan-2-ylidenephosphoramidate) was administered to rats (1 mg/kg) in a single oral dose. Of the total ^{14}C activity, 18.2% was in the respiratory gases, 31.6 and 13.3% excreted in the urine and feces, respectively, and 33.1% retained in the carcass after 144 hours. At twice the dose, the elimination pattern was similar except that tissue levels were reduced to 13%. After 24 h the residual radioactivity in the tissues was highest in kidney (1.36 ppm) followed by liver (1.14 ppm) and blood (1.14 ppm). Thiocyanate ion was the main metabolite comprising 99% of the extractable tissue radioactivity and 76% of the urinary radioactivity (3).

Cis- and *trans*-dioxathion [S,S'-1,4-dioxane-2,3-diyl 0,0,0',0'-tetraethyl phosphorodithioate] were readily metabolized by rat liver microsomes in the presence of NADPH. The more toxic *cis*-dioxathion gave higher yields of the dioxaoxons (dioxadioxon and dioxeneoxon) than the *trans*-isomer. Two unknown ester metabolites from *trans*- and four from *cis*-dioxathion possibly arose from O-deethylation or mono-dephosphorylation of dioxathion. Two acid metabolites, diethyl hydrogen phosphorothioate (ESOP) and diethyl hydrogen phosphate (EOOP), comprised up to 64% of the original radioactivity, and small amounts of diethyl hydrogen phosphorodithioate (ESSP) were also detected.

In rats, most of the ^{14}C activity was excreted within 96 h, and mainly in the urine (80-87%). Unmetabolized *cis*-dioxathion appeared in the feces, but not urine. ESSP, ESOP and EOOP were identified as urinary metabolites, with

smaller amounts of ESOP and EOOP in the feces. In all, at least seven metabolites from both cis- and trans-dioxathion were formed (4).

Male and female rats dosed with the experimental isecticide N-2530 [O-ethyl S-(4-chlorophenyl) ethylphosphonodithioate] showed similar excretion patterns. After 48 h, 80% of the applied ^{14}C was detected in the urine and another 10% in feces. Three ether soluble urinary metabolites were identified: 4-chlorophenyl methyl sulfoxide, 4-chlorophenyl methyl sulfone and 4-chloro-3-hydroxyphenyl methyl sulfone. They were formed from 4-chlorothiophenol via S-methylation followed by oxidation of the sulfur and hydroxylation of the ring. The amount of polar metabolites increased with time and represented 96% of the urinary fraction after 72 h. They were present as both glucuronide and sulfate conjugates, which on hydrolysis gave 4-chloro-3-hydroxyphenyl methyl sulfone, and 4-chlorophenyl sulfonic acid. No N-2530 or oxon were detected in the urine. The level of insecticide found in fat after 96 h was 0.21 ppm, with less in the tissues and blood. The total ^{14}C had decreased to negligible levels after 8 days (5).

Caged chickens were sprayed with Ronnel (0,0-diethyl 0-trichlorophenyl phosphorothioate) for the control of ectoparasites. Residues in skin and fat of 1.2 and 0.86 ppm, respectively, were found after 1 day. The maximum residues in eggs (0.019 ppm) occurred 7 days post treatment. No tissue residues were detected after 21 days except for skin. Following a second spray 28 days after the first treatment residues were no higher than recorded for the first treatment. However, there were residues in the skin and muscle of 0.008 and 0.003 ppm, respectively, 28 days after the second treatment. No oxygen analog was detected in the tissues (6).

Phenthoate [0,0-dimethyl S-[α-(ethoxycarbonyl)benzyl] phosphorothioate] was metabolized rapidly by white mice and houseflies. The major portion of the ^{14}C in mice was excreted in the urine within 24 h. The predominant degradative reactions were hydrolysis of the ethoxycarbonyl moiety and cleavage of the P-O bond. Demethylphenthoate was the main urinary metabolite with smaller amounts of phenthoate acid, demethylphenthoate acid, demethylphenthoateoxon acid and mandelic acid. These metabolites were also found in houseflies together with more phenthoateoxon. The difference in toxicity of phenthoate towards insects and mammals was attributed to this latter fact (7).

Metabolic studies with fonofos [O-ethyl S-phenyl ethylphosphonothioate], carbophenotion [S-(4-chlorophenylthiomethyl) 0,0-diethyl phosphonodithioate] and R-14805 [0,0-diethyl 0-[4-[1-(methylaminocarbonyloxyimino)ethyl] phenyl] phosphorothioate were carried out in rats. Metabolites were formed by oxidation reactions of the P=S, thioether and oxime groups. All three insecticides were rapidly excreted in the urine without any bioaccumulation (8).

The racemic, (+)- and (-)- forms of ^{14}C-cyanofenphos [O-(4-cyanophenyl) O-ethyl phenylphosphonothioate] were administered orally to rats. The ^{14}C activity was almost completely eliminated after 3 days, mainly in the urine during the first 24 h. The major urinary metabolites were 4-cyanophenol, 4-cyanophenyl sulfate and small amounts of deethylcyanofenphos; no parent compound or oxon were detected. A marked difference in the proportion of the major urinary metabolites was found from the three forms of cyanofenophos, indicating stereo-selective metabolism. All three forms were metabolized at approximately the same rate by the rat liver microsome/NADPH system. It appears that (+)-cyanofenphos undergoes oxidation to the oxon and oxidative cleavage of the P-O-aryl linkage at equal rates in the microsome/NADPH system. (-)-Cyanofenphos is similarly oxidized to the oxon analog, which is rapidly hydrolyzed to 4-cyanophenol by a microsomal arylesterase type enzyme. The specificity of this enzyme accounts for the larger amounts of 4-cyanophenyl sulfate found in rats treated with the (-)-enantiomer. The optical isomers of cyanofenphos exhibited different toxicities to houseflies and rice stem borers, but were almost equally toxic to mice (9).

In vitro metabolism of the racemate, (-)- and (+)-enantiomers of S-2571 [O-ethyl O-(3-nitro-5-methylphenyl) N-isopropylphosphoramidate] by rabbit liver microsomes plus NADPH gave 6 metabolites from oxidative and hydrolytic reactions. The main oxidation products were S-2571-oxon, hydroxymethyl-S-2571 and hydroxymethyl-S-2571-oxon with smaller amounts of S-2571-aldehyde. The hydrolysis products were 2-nitro-5-methylphenol and 2-nitro-5-(hydroxymethyl)=phenol. Metabolism of (+)-S-2571 gave dextrorotary products while the (-)-enantiomer gave levorotary, indicating the reaction to be stereospecific involving retention of configuration at the phosphorus atom. Oxidation of S-2571, (+)- and (-)-enantiomers with m-chloroperoxybenzoic acid, or UV irra-

diation on silica gel also gave the oxon analogs with retention of configuration. These results together with those of the reaction with mixed function oxidase suggests that conversion of P=S to P=O proceeds by a similar mechanism (1o).

Oxidation of the chiral isomers of fonofos has also been studied. With m-chloroperoxybenzoic acid, $(S)_p$-fonofos gave predominantly $(R)_p$-fonofosoxon and some $(R)_p$-fonofos gave the $(S)_p$-products. Thus, oxidation of the P=S to the oxon proceeded with retention of configuration, but oxidation to the disulfide occurred with inversion at the phosphorus (11). In vitro studies involving mouse liver microsomal function oxidases and the fonofos enantiomers gave imilar results, proceeding with ca. 7o% retention of configuration at phosphorus (12).

The effect of chirality at the α-carbon of the ethoxycarbonylbenzyl moiety of Papthion [O,O-dimethyl S-[α-(ethoxycarbonyl)benzyl] phosphorodithioate] was studied on the toxicity to houseflies, mosquitoes, rice stem borers, diamond backmoths and white mice. In general, the (+)-enantiomer was more toxic than the (-)-form to most organisms except houseflies. Housefly head acetylcholine esterase (ChE) differed from bovine erythroxyte ChE and rice stem borer enzyme in sensitivity towards the enantiomers of papthion and papoxon (13).

SOILS

The degradation of some organophosphorus insecticides in the soil and by soil microorganism has recently been reviewed (14). Data were collected from both field and laboratory studies for parathion, methylparathion, fenitrothion, malathion, diazinon, thionazin, phorate, terbufos and disulfoton, chlorfenvinphos, dichlorvos and mevinphos and fonofos.

Bayer NTN93o6 [O-ethyl O-[4-(methylthio)phenyl] S-propyl phosphorodithioate] degraded in Lufkin fine sandy loam and sand to give the same products as found in plants. These were the NTN93o6 sulfoxide and sulfone with minor amounts of NTN93o6-oxon sulfone. The conversion of NTN93o6 to its sulfoxide was greater in loam than in sand. Solvent extraction did not remove all the radioactivity in the soil and for this acid extraction was required. The latter extract contained mainly 4-(methylsulfinyl)phenol (46%) with some NTN93o6 (2.1%), NTN93o6 sulfoxide (3.7%), 4-(methylthio)phenol (o.7%) and the remainder unidentified (15).

Fonofos and phorate were degraded rapidly in sub-tropical soils with $t_{1/2}$ less than 2 months and o.5 months, respectively. Six weeks after initial treatment of fonofos, only 36% remained with 8% after 12 weeks. No fonofosoxon was detected in soil. Only 4% of the phorate applied was recovered after 6 weeks and o.4% were recovered 12 weeks later. The rapid decrease in phorate is accompanied by a concommitant increase in phorate sulfoxide and sulfone representing 18 and 74%, respectively, of total metabolites after 6 weeks and 6.7 and 92% at harvest. In summer months, the degradation was more rapid and only the sulfoxide and sulfone were recovered, but no oxon. Residue levels of o.1 ppm fonofos and o.o1 ppm oxon were found in white potatoes grown in treated silty loam soil, and o.o4 ppm phorate sulfoxide was found in sweet and white potatoes (16).

Degradation of S-(2-diisopropylaminoethyl) O-ethyl methylphosphonothioate in humic sand, humic loam and clayey peat was studied under laboratory conditions. 9o% had disappeared in 2 days and after 3 weeks only o.1% was detected by gas chromatography. The initial fast rate of disappearance was ascribed to chemisorption. Degradation took place most rapidly in humic loam, followed by clayey and humic sand. Ethyl hydrogen methylphosphonate and methyl dihydrogen phosphate were the only phosphorus-containing metabolites detected (17).

The degradation of disulfoton [O,O-diethyl S-[2-(ethylthio)ethyl] phosphorodthioate] in Portneuf silty loam soil approximated first order with $t_{1/2}$ being about 3.5 days. The only metabolites isolated were disulfoton sulfoxide and sulfone; no significant amounts of oxon were found. The sulfoxide persisted for 32 days, and during this time the amount of sulfone increased and then remained constant for a further 32 days. The effect of low soil temperature and low moisture content was to increase the rate of degradation of disulfoton (18).

The metabolism and movement of Kitazin P (S-benzyl 0,0-diisopropyl phosphorothioate) and edifenphos (0-ethyl S,S-diphenyl phosphorodithioate) were examined in three types of soil. The order of mobility was sandy loam > alluvial clay loam > volcanic ash loam. Edifenphos was less persistent and was degraded initially to S,S,S-triphenyl phosphorotrithioate, 0,0-diethyl S-phenyl phosphorothioate, S-phenyl dihydrogen phosphorothioate and diphenyl disulfide. The latter was also formed and converted to sulfuric acid via benzenesulfonic acid. Kitazin P formed 0,0-diisopropyl hydrogen phosphorothioate in all three soils. The sulfur moiety of Kitazin was degraded to dibenzyl disulfide and then to toluenesulfonic acid and sulfuric acid (19).

PLANTS

The metabolism of ethephon [(2-chloroethyl)phosphonic acid] was reviewed last year (2o). Studies of the translocation and metabolism of ^{14}C-ethephon showed rapid translocation from leaves to fruit in walnut (21), apple and cherry (22) and olives (23), but reported no metabolism. The labelled residues (less than 3% of applied dose) of treated peach fruit, however, contained 7 unidentified ^{14}C-compounds, one conjugated with a sugar (24).

Between 4o.2 and 62.4% of the ^{14}C-ethephon applied to Calimyrna figs was converted to $^{14}CO_2$ in four days, o.2% of the activity remained in the fruit at harvest. Of the latter, 22-2o% had been converted to compounds other than ethephon, although it was thought not to result from active metabolic processes (25).

More recent work showed that detached leaves of Hevea brasiliensis converted labeled ethephon to at least twelve non-volatile acids, one being conjugated ethephon. A major product was 2-(hydroxyethyl)phosphonic acid, which was itself metabolized further. Application of ethephon to the bark gave only a single conjugate (26).

Ethephon residues in apples, 1o days after field treatment of the trees at the recommended level (3oo ppm), averaged 0.65 ppm ($t_{1/2}$ ca. 3 days). This was reduced by 5o-6o% on washing. Storage of the apples in the dark at 0°C resulted in little loss of the ethephon residues after 4 months. No metabolites were detected by GC after derivatization (27).

Half the amount of NTN93o6 applied to the leaves of cotton plants was absorbed during the first day, although residues of the insecticide were detected up to 8 days. Compounds recovered from the leaf surface included NTN93o6, its sulfoxide and sulfone analogs, small amounts of NTN93o6-oxone sulfone, 4-(methylsulfinyl)phenol, 4-(methylsulfonyl)phenol and two unknowns. Neither NTN93o6-oxon nor its sulfoxide analog were detected in leaf extracts. After 4 days, the majority of the radioactivity was in the water-soluble fraction of plant extract, mainly as 4-(methylthio)phenyl ß-D-glucoside. Hydrolysis of this fraction gave 4-(methylthio)phenol (23%), 4-(methylsulfinyl)phenol (41%) and 4-(methylsulfonyl)phenol (36%) (15).

PHOTODEGRADATION

The photodegradation of azinphosmethyl [S-(3,4-dihydro-4-oxobenzo[d][1,2,3]-triazin-3-ylmethyl) 0,0-dimethyl phosphorodithioate] was examined on the surface of a glass plate, three soils, bean and corn leaves. Most degradation occurred on the glass plate and the least in soils. The benzene-soluble photoproducts due to sunlight amounted to 46.7% of applied radioactivity on glass, 73.3, 83.5 and 91.1%, respectively, on sandy loam and muck soils. Increased soil moisture content increased the amount of degradation occurring with UV light but not sunlight. The amount of bound residues formed on irradiation was directly related to the organic matter of the soil and its moisture content. In sunlight, 86 and 72% of the total ^{14}C was recovered from corn and bean leaves, respectively. Methylazinphos, N-methylbenzazimide sulfide, methylazinphos-oxon and benzazimide residues were common to both plants. In addition, N-methylbenzazimide was detected only on corn leaves (28).

The volatility $t_{1/2}$ for cis- and trans-dioxathion from glass plates was about 32 days, but less than 3o min for dioxenethion. Sunlight irradiation on silica gel plates for 27 h gave dioxaoxon (5-1o%), dioxathion and dioxeneoxon from dioxenethion. ESSP was a major metabolite from trans-dioxathion and dioxenethion but minor from cis-dioxathion. EOOP, EOOP and ethylene

glycol were also detected. Cis- and trans-dioxathion were more stable than dioxenethion on bean leaves, but underwent some loss by volatilization and slow oxidation to the same products as were obtained on a glass plate. More oxons and dioxons were obtained with the cis-isomer than with trans-dioxathion (4).

NTN9306 was rapidly degraded on exposure to sunlight on glass plates, the $t_{1/2}$ was less than 9 days, but in water it was less than 0.5 days. The products found on glass were NTN9306 sulfoxide and sulfone, NTN9306-oxon sulfone, 4-(methylthio)phenol, 4-(methylsulfinyl)phenol; the phenols did not accumulate. In water (0.5 ppm), the main product was the sulfoxide (15%). Some sulfone (0.5%) was also detected after 0.5 day and then disappeared. The other main product formed after 1 day was 4-(methylsulfinyl)= phenol (10.6%), but it also disappeared. Seven unknowns were found (15).

ENVIRONMENT

A simple computer model based on laboratory experiments was developed to simulate the dynamic process involved in the degradation of methylparathion in eutrophic ponds and lakes, oligotrophic lakes and streams. The data obtained suggest that biodegradation is the major degradative path in eutrophic systems, whereas absorption, photolysis and hydrolysis are more important in oligotrophic systems. Volatilization of dissolved methylparathion is not a significant process. Demethyl(methyl)parathion and p-nitrophenol were the major products of neutral catalyzed and base catalyzed hydrolysis, respectively, and on photohydrolysis of methylparathion. Biodegradation gave p-nitrophenol. No oxon was observed (29).

Studies of a simulated pond, involving ^{14}C-NTN9306, showed the radioactivity to disappear slowly from water, 80.4% remaining after 16 days and 50% after 75 days. After 2 h only half the radioactivity was in the form of the insecticide, the remainder having been oxidized to the sulfoxide analog. The principal metabolite was 4-(methylsulfonyl)phenol (15).

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