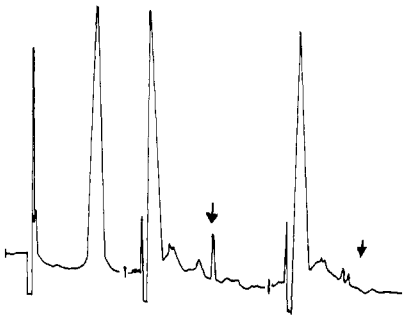

Figure 2. Typical chromatograms for the analysis of carbofuran as 7-phenol in soil: (A) 300 ng of 7-phenol; (B) check soil fortified with 0.1 ppm carbofuran, 782 mg injected (equivalent to 0.081 ppm carbofuran); (C) check soil, 653 mg injected.

One cautionary statement should be noted. The final acetonitrile extracts of soil samples are apparently unstable at room temperature. A sample quantitated directly after completion of workup and requantitated 1 week after storage at −10 °C in the dark gave comparable values. However, if the extract was allowed to stand at room temperature in a lighted environment for longer than 24 h, diminished recovery was noted. Therefore, the final acetonitrile extract should be quantitated immediately after workup or be stored at −10 °C in the dark if immediate analysis is impossible.

CONCLUSION

The method as presented is a relatively simple and efficient method for determining carbofuran in soil and water. Investigations toward extending the scope of this method are currently in progress.

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Residues of Mancozeb, 2-Imidazoline, and Ethyleneurea in Tomato and Potato Crops after Field Treatment with Mancozeb

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Mancozeb, 2-imidazoline, and ethyleneurea residues were determined in tomato and potato crops at various intervals for 28 days after field treatment with mancozeb. On tomato fruit mancozeb persisted for 28 days while no residues were found in potato tubers. Insignificant residues of 2-imidazoline were found in either crop, and low levels (<0.04 ppm) of ethyleneurea were present in tomatoes up to 28 days after spraying. Ethyleneurea was not detectable in potato tubers.

Several compounds have been identified as a result of studies to determine the fate of ethylenebis(dithiocarbamate) (EBDC) fungicides on plants. Lyman (1971) reported that ethyleneurea, ethylenediamine, and 2-imidazoline were major decomposition products occurring on leafy plants 2 weeks after the application of [^{3}H]-mancozeb outdoors. Lesser amounts of ethylenebis(isothiocyanate) sulfide (EBIS) and ethylenethiourea (ETU) were also found. Relatively large amounts of ethyleneurea, accompanied by several unknown metabolites, have been found in soybeans grown on soil treated with [^{14}C]maneb (Nash, 1976). In greenhouse experiments with [^{14}C]zineb on lettuce, Vonk (1976) found that while zineb and its degradation products ETU and EBIS decreased with time, ethyleneurea and 2-imidazoline were formed gradually and persisted for at least 3 weeks.

The dynamics of various EBDC's and some of their decomposition products have been examined in a field study with tomatoes (Newsome, 1976), where it was observed that ETU, EBIS, and ethylenebis(isothiocyanate) declined with time after spraying and that none of the residues constituted more than 0.5% of the parent EBDC. Since it remained to be determined what levels of the desulfurated residues ethyleneurea and 2-imidazoline could occur on food crops treated in the field with EBDC's, the present experiment was initiated. Tomatoes and potatoes were selected as examples of crops which receive widespread application of EBDC's in Canada.

EXPERIMENTAL SECTION

Field Experiment. Studies were conducted during the summer of 1978 at the Ottawa Research Station, Canada Department of Agriculture. Three plots of tomatoes (Ottawa 78 variety) were grown, one of which served as a control, while the others were treated at rates of 1.5 and 3.0 lb of mancozeb formulation (80% active ingredi-

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Table I. Residues[a] of Mancozeb on Tomato Fruit and Potato Tubers at Various Intervals after Treatment

time after spraying days	tomato		potato	
	1.5 lb/acre	3.0 lb/acre	1.0 lb/acre	2.0 lb/acre
0	2.4 ± 0.18	4.8 ± 0.39	0.08[b]	0.08
1	1.5 ± 0.09	3.6 ± 0.22	na[c]	na
2	1.5 ± 0.20	3.3 ± 0.38	na	na
5	1.1 ± 0.09	2.9 ± 0.23	na	na
7	0.54 ± 0.05	1.1 ± 0.08	0.08	0.08
9	0.78 ± 0.15	1.5 ± 0.05	na	na
14	0.36 ± 0.07	0.69 ± 0.14	0.08	0.08
21	0.43 ± 0.06	0.82 ± 0.06	na	na
28	0.37 ± 0.09	0.53 ± 0.09	0.08	0.08
control	0.014 ± 0.003			

[a] Values in ppm are the means of determinations on four samples ± standard error. [b] Minimum detectable limit for potatoes was estimated to be 0.08 ppm. [c] Not analyzed.

ent)/acre. Each plot of tomatoes contained two 30-ft rows of ten plants each. Potatoes (Katahdin variety) were also grown in three plots, each containing two 30-ft rows. Two plots were sprayed at rates of 1.0 or 2.0 lb/acre of the mancozeb formulation while the other was an untreated control.

For each treatment, the fungicide was applied in a volume of 4 L using a Chapin No. 135 hand sprayer. Each crop received seven treatments at intervals of 7 days. Tops of unharvested potatoes were killed 7 days after the final application of mancozeb by spraying with diquat (0.75 lb/acre as the dibromide). Total rainfall over the experimental period was 14.7 cm for tomatoes and 14.9 cm for potatoes.

Sampling. Sampling of tomato fruit or potato tubers was commenced as soon as the final spray of mancozeb had dried on the plants. Samples were removed from each half of a row, providing four samples from each plot at each time interval. Tomatoes were placed in plastic bags and transported to the laboratory where they were homogenized immediately without prior washing and frozen at −10 °C pending analysis. Potatoes were treated similarly but were washed by brushing in a stream of water before homogenizing and freezing.

Analytical Methods. Mancozeb was analyzed by hydrolysis with 1 N HCl containing stannous chloride and subsequent determination of the resulting ethylenediamine by gas–liquid chromatography as previously described (Newsome, 1974).

2-Imidazoline was determined by high-pressure liquid chromatography of the *p*-nitrobenzoyl derivative (Newsome and Panopio, 1978). The method was modified by reduction in the sample size to 2 g from 10 g and by the use of ice-cold 0.1 N HCl to extract the samples. These measures avoided the difficulty encountered in filtering the samples due to the large amount of starch in potatoes. A tenfold improvement in detectability was achieved by use of a Waters Model 440 detector coupled to a 1-mV recorder. Recoveries of imidazoline added to control samples at 0.02 and 0.10 ppm were 85 and 87%, respectively.

Ethyleneurea was extracted with acetone, derivatized with pentafluorobenzoyl chloride, and the derivative determined by gas–liquid chromatography of the fraction obtained by high-pressure liquid chromatography exactly as described (Newsome, 1978).

RESULTS AND DISCUSSION

The residues of mancozeb found on tomatoes in this study are similar to the 3-year average of EBDC residues found on commercially treated field tomatoes by the CS_2 evolution technique (Ripley and Cox, 1978). As shown by the data in Table I, treatment at one-half the maximum recommended rate of 3 lb/acre results in approximately half the residue. Analysis of treated tomatoes did not reveal free ethylenediamine at levels above the minimum detectable limit of 0.08 ppm. No compounds which yield ethylenediamine on acid hydrolysis in the presence of a reducing agent were found in potatoes. This result is in contrast to the finding of an average of 0.17 ppm (maximum 0.78 ppm) of acid-released ethylenediamine in potatoes treated with mancozeb (Lyman, 1971).

Data on the analyses for 2-imidazoline and ethyleneurea are given in Tables II and III, respectively. Neither to-

Table II. Analysis[a] of 2-Imidazoline in Tomato Fruit and Potato Tubers at Various Intervals after Treatment with Mancozeb

time after spraying, days	tomato		potato	
	1.5 lb/acre	3.0 lb/acre	1.0 lb/acre	2.0 lb/acre
0	0.014 ± 0.003	0.024 ± 0.004	0.011 ± 0.002	0.010 ± 0.002
7	0.006 ± 0.003	0.015 ± 0.001	0.006 ± 0.004	0.016 ± 0.003
14	0.007 ± 0.001	0.010 ± 0.001	0.011 ± 0.002	0.018 ± 0.003
21	0.005 ± 0.002	0.004 ± 0.001	0.017 ± 0.005	0.025 ± 0.007
	control	0.006 ± 0.003	control	0.011 ± 0.001

[a] Values, in ppm, are the means of determinations on four samples ± standard error.

Table III. Ethyleneurea Residues[a] in Tomato Fruit and Potato Tubers at Various Intervals after Treatment with Mancozeb

time after spraying, days	tomato		potato	
	1.5 lb/acre	3.0 lb/acre	1.0 lb/acre	2.0 lb/acre
0	0.020 ± 0.002	0.027 ± 0.001	0.001 ± 0	0.004 ± 0
1	0.009 ± 0.001	0.023 ± 0	0.001	0.001
2	0.010 ± 0	0.021 ± 0.001	na[b]	na
5	0.016 ± 0.004	0.039 ± 0.002	na	na
7	0.032 ± 0.002	0.035 ± 0.005	0.001 ± 0	0.002 ± 0
9	0.016 ± 0.003	0.024 ± 0.002	na	na
14	0.023 ± 0.003	0.028 ± 0.004	0.001 ± 0	0.001 ± 0
21	0.031 ± 0.002	0.028 ± 0.001	0.001 ± 0	0.002 ± 0
28	0.035 ± 0.004	0.031 ± 0.002	0.001 ± 0	0.002 ± 0
	control	0.004 ± 0.002	control	0.002 ± 0.001

[a] Values are the means of determinations on four samples ± standard error. [b] Not analyzed.

matoes nor potatoes contained significant amounts of 2-imidazoline. In contrast, Vonk (1976) found approximately 8% of applied zineb occurred as 2-imidazoline 2 weeks after treatment. The difference between the present result and the positive findings obtained with [^{14}C]zineb on lettuce (Vonk, 1976) may be attributable to differences in plant species or, more likely, due to removal of any imidazoline by rain since the residue is readily removed by washing. Some leaf surfaces apparently retain imidazoline and approximately 8% of the total EBDC residue can occur as imidazoline 2 weeks after the application of [^{3}H]mancozeb (Lyman, 1971).

Small amounts of ethyleneurea were present in tomatoes while none was detectable in potato tubers (Table III). The residues in tomato were confirmed by high-resolution mass spectrometry with single ion monitoring as previously described (Newsome, 1978). The levels in tomatoes were similar to those observed in an earlier survey of commercial products (Newsome, 1978) and appeared independent of the application rate of mancozeb. Much less ethyleneurea was found than was anticipated from other studies (Lyman, 1971; Vonk, 1976) where levels ranged from 10 to 17% of the total residue.

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Metabolism, Tissue Distribution, and Elimination of *cis*-[^{14}C]Chlordane in the Tropical Freshwater Fish *Cichlasoma* sp.

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Tropical freshwater cichlids, *Cichlasoma* sp., weighing 300 g each, were individually placed in 16 L of water with 80 μg of *cis*-[^{14}C]chlordane for 72 h. The fish rapidly absorbed the compound, accumulating 2.8 ppm in visceral fat, 1.2 ppm in bile, and 1.1 ppm in gall bladder. Recovery of the radioactivity added to water at the end of 72 h was estimated to be 64.8 and 6.1% in fish and exposure water, respectively. The rate of elimination of the absorbed radioactivity was about 2.9% per week. Dichlorochlordene, oxychlordane, chlordene chlorohydrin, dihydroxyheptachlor, dihydroxydihydrochlordene plus four unidentified compounds accounted for 12.5% of the radiocarbon recovered from fish and exposure water. The remainder was determined to be unchanged *cis*-chlordane.

Though extensively used for over two decades, work on the metabolism of chlordanes in comparison with other cyclodienes as aldrin, dieldrin, and heptachlor has been slow. Poonawalla and Korte (1964, 1971) showed that *trans*-chlordane administered to rats and rabbits was rapidly metabolized and eliminated. Identification, characterization, and probable pathway of formation of oxychlordane from chlordanes (Schwemmer et al., 1970; Lawrence et al., 1970; Polen et al., 1971; Street and Blau, 1972) was another important development in the toxicology of chlordanes. Detailed investigation of the in vivo metabolism of chlordanes was carried out by Barnett and Dorough (1974). Two other reports on the metabolism of chlordanes in the rat (Tashiro and Matsumura, 1977; Brimfield et al., 1978) have appeared more recently.

As stated, most of the work on chlordanes yielding important information has so far centered around mammalian species. The fate of these chemicals in other organisms is not well understood. In this area, aquatic organisms require special attention because of their importance in food chain and because contamination of aquatic environments with chlordane residues has been reported. For instance, of 546 samples of surface waters from various parts of the United States during 1964–1968, Lichtenberg et al. (1970) found 1% samples positive for chlordane residues. A survey for pesticide residues in Oahu, Hawaii, in 1970–1971 (Bevenue et al., 1972) showed chlordane residues ranging from 0.004 to 0.009 ppb in nonpotable waters and 190 to 378 ppb in the sediments. A later report from Hawaii Kai Marina (Tanita et al., 1976) indicated widespread occurrence of chlordanes in aquatic environments of Hawaii. Law and Goerlitz (1974) reported very high incidence (92% of the samples) of chlordane residues from sediments of tributaries to San Francisco Bay. Similarly Burns et al. (1975) detected high levels of contamination of water and sediments of Habitant Creek (Nova Scotia, Canada) with chlordanes. Some aquatic environments of Southern Florida were also reported

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