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Residues of Ethylenebisdithiocarbamates on Field-Treated Fruits and Vegetables

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The ethylenebisdithiocarbamates (EBDC) are an agriculturally important and widely used group of broad spectrum fungicides. Currently these fungicides are recommended by the Ontario Ministry of Agriculture & Food (O.M.A.F.) for use on many fruits and vegetables (O.M.A.F. PUBLICATION 360; 363; 367). A recent report in the FEDERAL REGISTER (1977), on the EPA rebuttable presumption against registration of the EBDC, documents the health hazard associated with these compounds because of their degradation to ethylene thiourea (ETU).

There is considerable concern in the agricultural industry that several crops would have no recommended alternative fungicides if the EBDCs were restricted. There was an obvious need to confirm current O.M.A.F. recommendations on rate of application and days to harvest, to ensure that residues conformed with current Canadian pesticide residue tolerances on food products, and to compare Ontario residue data with those reported by other investigators. Previous studies in Ontario have been carried out on EBDC and ETU residues on pears (RIPLEY and SIMPSON 1977), on grapes (RIPLEY et al. 1978) and on tomatoes (RIPLEY and COX 1978). This paper presents data on EBDC residues determined on several fruits and vegetables following application of maneb (manganese ethylenebisdithiocarbamate) or zineb (zinc ethylenebisdithiocarbamate).

MATERIALS AND METHODS

The EBDC were determined using the standard CS_2 evolution technique incorporating the modifications outlined by RIPLEY and SIMPSON (1977). The apparatus was set-up as described by PEASE (1957). The first absorbance trap contained 10 ml of 6.5 N NaOH covered with 5 ml of benzene, and the second trap contained 12.5 ml Cullen's reagent (CULLEN 1964). A 1 g leaf sample or 100 g sample of crop macerate was placed in the three-necked reaction flask, and 200 ml of 4N HCl and 5 ml of 40% $SnCl_2$ was added. The central neck was connected to the reflux condenser, another neck was connected to the air intake tube while the third neck was stoppered. When reflux commenced, the air intake was adjusted using house vacuum so that bubbles in the second trap were approximately 3 cm above the glass beads. After 35 min of reflux, the vacuum was disconnected and the chromogenic reagent was drained with ethanol rinsings into a 25 or 50 ml volumetric flask. The absorbance was determined at 435 nm in a one-cm cell against a reference mixture of 1:1 Cullen's reagent and ethanol.

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Commercial formulations (Rohm and Haas, Co.) of Dithane M-22 (maneb) or Dithane Z-78 (zineb) were applied to plots of vegetables or fruit using a 9-liter hand sprayer; mushroom casing soil was dusted with zineb. Although some of the crops did not have recommendations for these specific EBDCs, it was felt that the residue data would be indicative for the purpose of this study. Rates of application to the various crops are given in the text. To prevent any problems associated with storage of EBDC-treated crops (HOWARD and YIP 1971), the samples were analyzed on the day of sampling. The crops were not analyzed for ETU or other EBDC degradation products.

RESULTS AND DISCUSSION

EBDC are determined by boiling in dilute mineral acid to release CS_2 which is trapped in a chromogenic reagent (CLARKE et al. 1951). Various modifications in the method have been discussed (LOWEN 1951, LOWEN 1953, PEASE 1957, CULLEN 1964, LOWEN and PEASE 1964, GORDON et al. 1967, KEPPEL 1969, KEPPEL 1971). These modifications were examined and the described method was found to be satisfactory for routine determination of EBDC in a variety of crops. Although the method is non-specific, since all dithiocarbamates and thiram (tetramethylthiuram disulfide) react similarly, the residue results may be expressed as the zineb equivalent residue, thus establishing a common basis for comparing EBDC data and preventing possible error through calculation if the identity of the residue is unknown.

Quantitative recoveries using the described method were determined by comparison of various dithiocarbamate formulations against CS_2 standards. Because of possible errors in weighing the CS_2, standard curves were determined using a known assayed zineb formulation dissolved in Na_4EDTA (PEASE 1957). After several hundred standard analyses, it was found that there were slight daily variations in the absorbance values, but a mean calibration curve demonstrated reproducible results. Coefficients of variation in absorbance values were 11% for 0-150 μg (in a final 25-ml volume) and 4.8% for 50-500 μg (in a final 50-ml volume). Quantitative recoveries ($\pm$ 10%) were obtained, in all crops examined, after fortification of substrate with EBDC standards dissolved in Na_4EDTA prior to addition of the acid and subsequent digestion. As noted by KEPPEL (1971), some crop blanks showed apparent residues of 10-20 μg (<0.2 ppm), but this did not appear to be a function of the NaOH-benzene absorption mixture in the H_2S trap. After repeated use this solution became highly colored and was changed as required.

Most investigators that have used Na_4EDTA to dissolve the EBDC standard note that the standard solutions must be prepared daily. CULLEN (1964) observed that many dithiocarbamates decompose upon contact with slightly polar solvents. LYMAN and LACOSTE (1975) examined the hydrolysis of M-45 in water at various pHs and found a half-life of less than one day. It was felt, therefore, that it would be informative to examine the stability of maneb (M-22) in Na_4EDTA at room temperature. At daily intervals, the 20 and 400 ppm solutions were analyzed for CS_2 content. From the

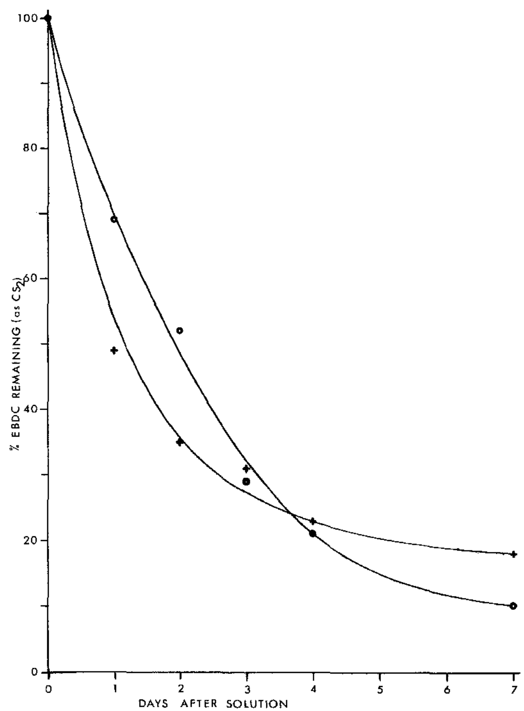

Figure 1. Decomposition of M-22 in Na_4EDTA solution. —o——o—20 ppm. —+——+— 400 ppm.

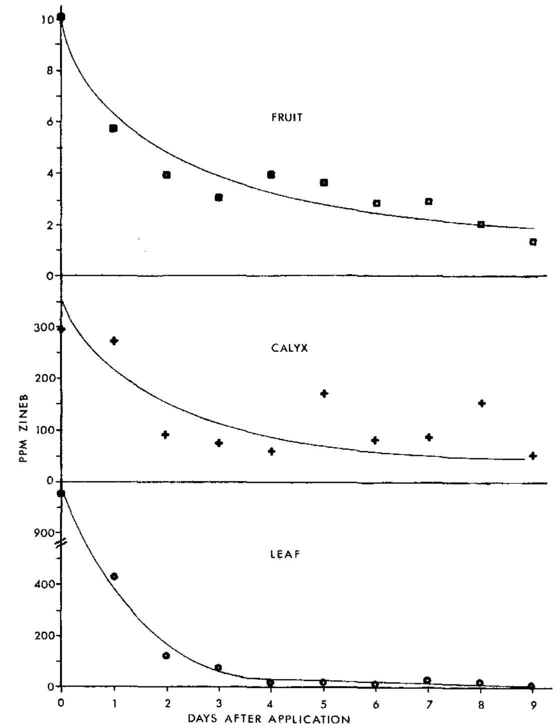

Figure 2. Dissipation of zineb on various parts of strawberry plants after treatment.

TABLE 1

EBDC Residues on Leaves of Field-Treated Vegetables.

Days after application	ppm zineb in,				
	Green Peppers	Broccoli	Cabbage	Squash	Cucumber
0	515	180	110	320	
1	285	190		650	500
2	650	25			230
3	400	60			240
4		20			235
5					125
6	610	70			
7	270	40			260
14	40				9

TABLE 2

EBDC Residues on Field-Treated Vegetables.

Days after application	ppm zineb in,					
	Green Peppers	Brussel Sprouts	Broccoli	Cauli-flower	Squash	Cucumber
0	1.4	8.0	23.		ND(<0.2)	0.8
1	1.2		20.			1.3
2	2.3		12.	6.7		1.1
3	2.0	4.8	9.2			1.1
4	0.8		10.			0.6
5	1.5		8.5	8.0		0.5
6	0.5		5.2			0.6
7	0.6	4.9	6.4			0.6
14	0.8		0.6			0.4
Control	ND(<0.2)	1.9				

results (Figure 1), there appears to be a slightly faster decomposition at the lower concentration although after 3 days the rates appeared to be reversed. The half-life is about 2 days. These observations underline the necessity of preparing fresh solutions of EBDC daily.

In July 1976, at the Cambridge Research Station, Cambridge, Ontario, a 13 sq m plot containing 3 rows of 5 strawberry plants was sprayed with an aqueous suspension of 75% Dithane Z-78 at 4.03 kg a.i./ha. Fresh fruit, calyx, and leaves were sampled at daily intervals after treatment. A heavy rainfall occurred on the evening of day 0 and therefore the trial was repeated 18 days later. No apparent removal of residue by the rain was evident except on the leaves where 90% of the zineb was lost. Except for this anomaly the residue data for both trials were apparently the same. The dissipation of EBDC residue (Figure 2) is typical of that found by YIP et al. (1971) and NEWSOME et al. (1975). As observed with zineb on pears (RIPLEY and SIMPSON 1977) there appears to be a rapid initial decline of residue concentration in the first 3-5 days followed by a slight dissipation. Prior to the second spray (day 18) EBDC was determined at 1.0, 36, and 7.5 ppm on the fruit, calyx and leaf, respectively. Zineb is no longer registered in Canada for use on strawberries, but the dissipation results are indicative of EBDC decay and are informative in relation to other EBDC residue data.

During the summers of 1975-77 at the Cambridge Research Station, broccoli, brussel sprouts, cabbage, cauliflower, green peppers and lettuce were planted in one large plot; each vegetable was placed in 7.5 m rows spaced 1 m apart. Controls were obtained prior to application of an aqueous suspension of 80% Dithane M-22 at 2.58 kg a.i./ha. Each crop received one spray in August or September when the vegetables were almost ready for harvest. Occasionally a second spray was applied to a vegetable 7-10 days later for replication.

The EBDC residues (as zineb equivalent) on the vegetable foliage and fruit are shown in Tables 1 and 2, respectively. Although the residues were variable on the leaves, a dissipation was evident on the fruit. These data agree with those results previously reported (LOWEN 1951, LOWEN 1953, PEASE and HOLT 1977). The O.M.A.F. recommendation for days to harvest is 7 days for these crops and the Canadian tolerance is 7 ppm for all these vegetables, with the exception of cucumbers at 4 ppm. Although tolerance was met under these conditions, it appears that problems could occur with the cole crops such as brussel sprouts, broccoli and cauliflower. An extention of the post spray interval to 14 days would probably alleviate any problem of high EBDC residue.

Residues on the inner and outer portions of the head crops are shown in Table 3. As noted by STEURBAUT et al. (1973) and DEJONCKHEERE et al. (1974) there is a large difference in the concentration of EBDC residue on the inner and outer leaves. Usual market practice involves stripping off the outer leaves of lettuce and cabbage, but obviously the degree of leaf removal will determine the terminal residue level. O.M.A.F. recommended days to harvest is 7-10 days and the Canadian tolerance is 7 ppm. From the weights of each portion of head, it was calculated that 15 ppm zineb was present on the whole cabbage 7 days after treatment.

At the O.M.A.F. Muck Research Station, Bradford, Ontario, a 33.5 sq m plot of celery was treated over the 1977 growing season with an aqueous suspension of 80% Dithane M-22 at 2.69 kg a.i./ha. Samples were taken at intervals after the terminal application of fungicide before harvest. The results (Table 4) show the residues

TABLE 3

EBDC Residues on Field-Treated Cabbage and Lettuce.

Days after application	Head part	ppm zineb in,	
		cabbage	lettuce
0	inner	1.0	
0	outer	65.	
2	inner	0.7	
2	outer	60.	
4	inner	1.0	
4	outer	43.	
5	inner	1.8	
7	inner	1.2	0.8
7	outer	33.	25.
Control		0.15	

TABLE 4

EBDC Residues on Field-Treated Celery.

Days after application	ppm zineb in,	
	whole celery	market-cut celery
0-pre	6.1[a]	1.7[b]
2	7.5	
3		2.4
5	7.9	
6		2.7
7	9.2	
8		3.1
9	6.2	
12	3.3	
13	2.1	1.3
Control		ND(<0.1)

a/ 9 days after last treatment.

b/ 10 days after last treatment.

TABLE 5

Zineb Residues on Mushrooms and Mushroom Casing Soil.

Break	ppm zineb[a] in,	
	mushrooms	casing soil
1st	0.33	10
2nd	ND(<0.2)	11
3rd	0.40	
4th	ND(<0.2)	
5th	ND(<0.2)	16

a/ mean of 2 samples

in whole celery plants and in market-cut samples. As observed previously (CLARKE et al. 1951, LOWEN 1951, LOWEN 1953) most of the residue is deposited on the top leaves, and trimming the celery for market reduces the EBDC residue. Following the O.M.A.F. recommendation of 14 days to harvest (7 days for Polyram), the EBDC residues would be below the Canadian tolerance of 5 ppm.

At the O.M.A.F. Horticulture Research Station, Vineland, Ontario, mushroom casing soil was dusted with 15% Zineb at 0.113 kg a.i./93 sq m two times per week in the pre-production period and three times during the production period immediately after the "break" (O.M.A.F. PUBLICATION 367). Samples of mushrooms were taken at each "break" as per commercial practice. Zineb is normally dusted after the completion of a "break" and never actually to the mushrooms; O.M.A.F. recommends that zineb not be applied within 2 days of harvest. Zineb residues on the mushrooms and the casing material (Table 5) show that although 10-16 ppm was present in the soil, only a trace of zineb (<0.5 ppm) was present on the mushrooms. Most of this residue could be attributed to adhering casing soil; duplicates and washing of the mushrooms showed no detectable zineb residue. The Canadian tolerance is 7 ppm.

In summary, it appears that fruits and vegetables sprayed according to O.M.A.F. recommendations contain EBDC residues at the normal spray-harvest interval that are below current Canadian tolerances. Further investigations should be conducted on some cole crops that showed residues near the tolerance level. Since these residues were determined on produce direct from the field, they are probably elevated with respect to those found in market-basket samples (CORNELIUSSEN 1970, 1972, DUGGAN and CORNELIUSSEN 1972). Although these crops were not analyzed for ETU residues, results from previous studies would indicate that ETU residues would be less than 0.1 ppm.

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