

# Uptake of Ethylenebis(dithiocarbamate) Fungicides and Ethylenethiourea by Soybeans

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Radioactivity from soil treatments of 10 to 100 ppm of  $^{14}\text{C}$ -labeled ethylenebis(dithiocarbamate) (EBDC) fungicides (nabam, zineb, and maneb) and 0.1 to 10 ppm of ethylenethiourea (ETU), a degradation product, is readily absorbed by soybeans and translocated throughout the plant. The presence of ETU on soybean leaves after EBDC foliar treatments decreased rapidly from <20 ppm after 1 day to <2 ppm after 15 days in the greenhouse. ETU applied to leaves of soybeans, which were kept outdoors, decreased from an initial level of 27 ppm to <0.04 ppm after 14 days and was below detectable levels by 24 days. ETU content in soybeans from EBDC fungicide soil treatments remained low (<0.2 ppm) throughout the 15-day monitoring period. Methanol extract from leaves contained degradation products (7 to 10) common to both ETU and the EBDC's, as determined by thin-layer chromatography. Ethyleneurea was the major degradation product.

Ethylenebis(dithiocarbamate) (EBDC) compounds, the most important fungicide class for protecting plants from destructive pathogens (Tweedy, 1973), have been extensively used since their introduction over 30 years ago. Without the EBDC fungicides, fruit and vegetable losses would be much higher. However, several toxicological studies have indicated that a degradation product, 2-imidazolidinethione (ethylenethiourea or ETU), was toxic to rats (Graham et al., 1973; Meland et al., 1972) and to mice (Innes et al., 1969; Khera, 1973). Significant levels of ETU in or on our harvested food crops should therefore be avoided.

ETU is present in formulated EBDC fungicides and additional amounts may form under some storage conditions (Bontoyan et al., 1972; Bontoyan and Looker, 1973; Czeglédi-Janko, 1967). Therefore, ETU on crops is possible through applying formulated EBDC materials and through subsequent degradation of the parent fungicide.

Blazquez (1973), Nash (1974), Newsome et al. (1975), and Yip et al. (1971) have observed that ETU disappears rapidly from plants and soil. Applying ETU to crops through the formulated fungicides probably is not as much of a threat to foods as the parent compounds which have a potential to degrade to ETU, and perhaps, more importantly, the EBDC fungicides degrading to ETU during cooking of fruits and vegetables (Blazquez, 1973; Newsome and Laver, 1973; Watts et al., 1974). However, fungicides on the food crops are not very persistent (Czeglédi-Janko, 1967).

## MATERIALS AND METHODS

The soil used throughout these experiments was Lakeland sandy loam: pH 5.6; cation exchange capacity, 3.0 mequiv/100 g; organic matter content, 0.9%; clay content, 12%; silt content, 16.4%; and 8.5% moisture content at  $1/3$  bar.

**Surface vs. Subsurface Treatment.** The method of Beall and Nash (1971) was used for distinguishing root uptake from soil vs. volatilized materials. Potted soybeans (*Glycine max* [L.] Merr. var. Cutler) were treated by injecting 0.1, 1.0, or 10.0 ppm (soil weight) of  $^{14}\text{C}$ -ethylene labeled ETU (>98% purity) or 100 ppm of nabam, zineb, or maneb (each of >95% purity) into the soil with a syringe (60 ml), or onto the soil surface with a pipet (20 ml). The  $^{14}\text{C}$ ETU was applied at 0.01  $\mu\text{Ci/g}$  of soil, and each  $^{14}\text{C}$ -labeled fungicide at 0.006  $\mu\text{Ci/g}$  of soil. Each treat-

ment was replicated four times with 1 plant/pot. The surface (250 g) and subsurface (1700 g) soil layers were separated with a plastic saucer, and the plants were caged in plastic film so that all aerial residues were from either root uptake and translocation from subsurface treatment, or volatilization from surface-treated soil followed by condensation on plant parts. No plant part touched the surface soil. Also, a combined surface-subsurface treatment and alkaline (1%  $\text{CaCO}_3$  added) surface and subsurface treatments were included with ETU. The plants were harvested 16 days after treatment, frozen, and stored for future analyses. At treatment the soybeans were 40-days old for ETU treatments and 70-days old for EBDC treatments.

**Soil pH Effects.** Actively growing (35 days old) soybeans were treated with 10 ppm of  $^{14}\text{C}$ ETU or  $^{14}\text{C}$ EBDC by soil injection (60 ml). Each treatment consisted of three replications with five plants in 2 kg of Lakeland soil with or without  $\text{CaCO}_3$ . The first and third trifoliolate leaves were harvested 1 day after treatment, the second trifoliolate leaf after 8 days ( $\pm 1$  day) and the remaining leaves and stem on day 15 ( $\pm 1$  day). The new leaves (<2 weeks old) and stem were frozen. Also, the terminal leaflet from each harvested trifoliolate leaf was frozen, whereas the remaining two freshly harvested lateral leaflets were extracted immediately for analysis.

**Foliar Treatment (Greenhouse).** Actively growing soybeans (40 days old) were treated by spraying 5 ml of a 2500-ppm aqueous solution or suspension of  $^{14}\text{C}$ ETU or  $^{14}\text{C}$ EBDC, similar to recommended solution levels for EBDC fungicides, onto selected leaves of 4 plants/pot and only the terminal leaflet of each trifoliolate leaf of the fifth plant. There were three replications/treatment. On days 1, 8 ( $\pm 1$ ), and 15 ( $\pm 1$ ), the first, second, and third trifoliolate leaves, respectively, of each plant were harvested and immediately analyzed, except for the fifth plant (terminal leaflet treatment) whose trifoliolate leaves were frozen and stored.

**Foliar Treatment (Outside).** Actively growing soybeans (45 days old) were treated by spraying 25 ml of a 25-ppm aqueous ETU solution onto the leaves of three plants/pot. The pots were placed outdoors on all clear days, except weekends. Immediately after treatment, one upper leaf and one lower trifoliolate leaf from each plant were harvested and bulked and their ETU content determined. Further upper and lower trifoliolate leaves were harvested at 1, 3, 8, 14, 24, and 35 days for ETU analysis.

**Analyses.** To aid in tracing the fate of the applied chemicals, radioactivity was monitored throughout the experiments and analyses. Total  $^{14}\text{C}$  content was de-

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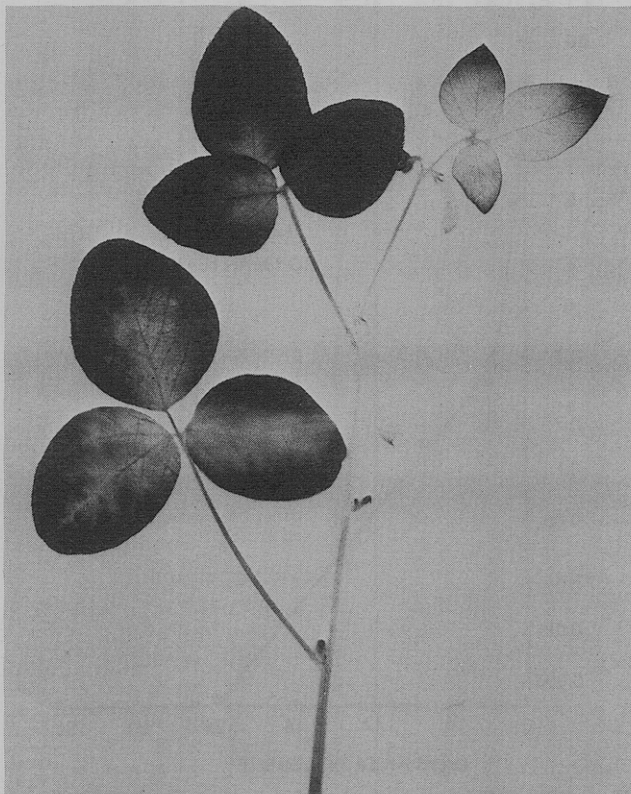


Figure 1. Radioautograph of first, third, and fourth trifoliolate soybean leaves showing distribution of translocated radioactivity from [ $^{14}\text{C}$ ]maneb applied to Lakeland sandy loam soil (pH 7.6) at 10 ppm (w/w). Fourth trifoliolate emerged after treatment. Harvested 15 days after treatment.  $^{14}\text{C}$  content greatest in dark areas.

terminated using a freeze-dried short stem section or whole leaf, for all experiments, except surface vs. subsurface ETU treatments for which a wedge was cut from each leaf. The freeze-dried plant part was dried in a 65 °C oven for 2 h, weighed, and combusted as previously described (Nash, 1974).

ETU was determined on frozen or fresh leaves by methanol extraction followed by formation of 2-(benzylthio)-1-(pentafluorobenzoyl)-2-imidazoline (Nash, 1974) and 2-(*o*-chlorobenzylthio)-1-(pentafluorobenzoyl)-2-imidazoline derivatives (Nash, 1975). The latter was used as a second ETU derivative. The ETU derivatives were analyzed by gas-liquid chromatography (GLC) using a  $^{63}\text{Ni}$  electron-capture detector, using two 1.8 m  $\times$  4 mm i.d. glass columns at 210 °C with methane-argon flow of 75 ml/min for the benzyl derivative: (1) 3% OV-17 on 100–120 mesh Gas-Chrom Q and (2) 3% OV-17 or 3% OV-1 on 80–100 mesh Chromosorb W (AW DMCS). A column with conditions common to both the benzyl and *o*-chlorobenzyl derivatives was 3% XE-60 on 80–100 mesh Chromosorb W (AW DMCS) at 220 °C with methane-argon flow of 50 ml/min.

$^{14}\text{C}$  content was determined in all extracts, nonextracted tissue, extracted tissue, and solutions of the final ETU derivatives. Also, normally discarded fractions of the ETU derivatization process were monitored for radioactivity. In extracted tissue,  $^{14}\text{C}$  was determined by combusting the dried extracted leaves, trapping the evolved  $^{14}\text{CO}_2$ , and counting by liquid scintillation. All  $^{14}\text{C}$  values are expressed as the parent  $^{14}\text{C}$ -labeled compound equivalent on a plant dry weight basis.

Translocation of  $^{14}\text{C}$ -labeled compounds was determined by radioautography with medical x-ray film on freeze-dried

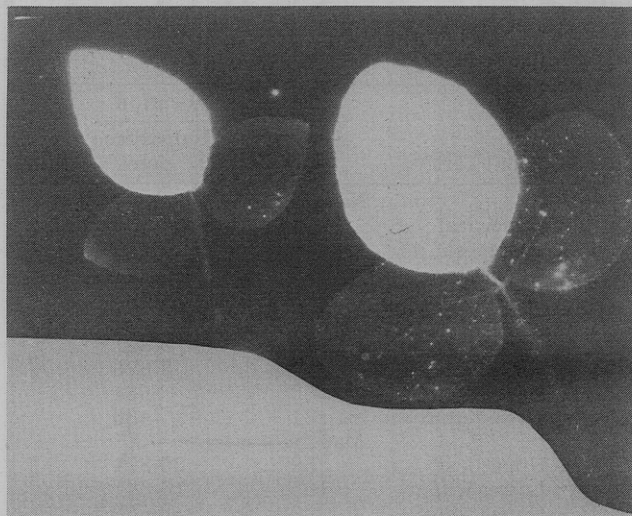


Figure 2. Radioactivity in and on soybean leaves from spray application of [ $^{14}\text{C}$ ]ETU to terminal leaflet only.  $^{14}\text{C}$  content greatest in light areas.

Table I. ETU Content (ppm)<sup>a</sup> of Soybeans 16 Days after a Surface or Subsurface Soil Treatment of [ $^{14}\text{C}$ ]ETU

Plant part	Treatment rate, ppm				
	0.1	1.0	10		
			No amend-ment	CaCO <sub>3</sub> amend-ment	Surface + sub-surface
		Surface			
Upper leaf	0.03	0.12	2.0	2.6	1881
Lower leaf	0.07	0.96	7.8	14.9	1594
Upper stem	0.01	0.05	1.1	2.5	97
Lower stem	0.01	0.07	0.7	2.0	120
		Subsurface			
Upper leaf <sup>b</sup>	1.24	25.8	1740	1992	
Lower leaf	0.37	13.4	1327	858	
Upper stem	0.17	3.7	93	186	
Lower stem	0.17	4.0	107	202	

<sup>a</sup> ETU equivalent based on  $^{14}\text{C}$  content. <sup>b</sup> Previously published (Nash, 1974).

upper plant portions. Degradation of  $^{14}\text{C}$ -labeled compounds was ascertained by thin-layer chromatography (TLC) and radioautographs of extracts (alumina, Woelm neutral, as adsorbate and developing 20 cm with 1-propanol-H<sub>2</sub>O, 85:15, v/v).

## RESULTS AND DISCUSSION

The data collected from the various experiments varied extremely and should be considered an indication of possible trends and not absolute repeatable values. This variation may be the result of nonuniform treatments, although each pot received identical quantities of ETU or EBDC, and variable rate of degradation within the soil-plant environment.

**Total Radioactive Residues.** Radioactivity was readily taken up and translocated throughout soybeans from [ $^{14}\text{C}$ ]ETU- and [ $^{14}\text{C}$ ]EBDC-treated soils (Figure 1). Smaller quantities of radioactivity were translocated to lateral leaflets from a terminal leaflet sprayed with the labeled compounds (Figure 2).

Leaves and stem contained radioactive residues from ETU or EBDC applied to either the surface or subsurface soil (Tables I, II, and III).

Subsurface treatment residues were one to five orders of magnitude greater than that from surface treatment. As expected, lower leaves from surface treatments con-

Table II. Fungicide Content of Soybeans 16 Days after 100-ppm Surface or Subsurface Soil Treatment of [<sup>14</sup>C]EBDC Fungicide

Plant part	Treatment location	
	Surface ppm	Subsurface ppm
	Nabam	
Upper leaf	25 <sup>a</sup>	5 995
Lower leaf	50	5 422
Upper stem	18	3 477
Lower stem	34	5 668
	Zineb	
Upper leaf	62	3 361
Lower leaf	112	3 626
Upper stem	50	554
Lower stem	81	486
	Maneb	
Upper leaf	59	25 871
Lower leaf	165	18 341
Upper stem	9	8 652
Lower stem	6	7 694

<sup>a</sup> Expressed as parent compound equivalent based on <sup>14</sup>C content.

tained more radioactivity than the upper leaves and stem. However, upper leaves contained more radioactivity, except for zineb, from subsurface treatments, which shows the residues' extreme mobility in plants.

#### Amount of Applied Radioactivity Found in Plants.

Leaf and stem residues from ETU-surface soil treatments increased as treatment rate increased, but residues from subsurface treatments increased disproportionately as treatment rate increased (Table I). However, for radioactivity (percent added to soil) found in or on plants, these differences for ETU decreased (Table IV). There were 250 g of soil (2.5 μCi) in the surface layer and 1700 g of soil (17 μCi) in the subsurface layer. The percentages (0.4%) of radioactivity trapped by the plants from surface volatilization were uniform for non-CaCO<sub>3</sub>-amended soil. The amended soil had twice as much (0.7%).

The higher (100 ppm) EBDC treatments (Table II) contained greater radioactive residues in soybeans than the lower (0.1 to 10 ppm) ETU treatments (Table I); however, the percent EBDC added (Table IV) was considerably less than that for ETU subsurface treatments. Plant uptake was probably dependent upon EBDC-

Table III. Radioactivity and ETU Content in Leaves of Soybeans Grown in Lakeland Soil at Two Different pH Values and Treated with [<sup>14</sup>C]ETU or [<sup>14</sup>C]EBDC Fungicide at 10 ppm

Days (± 1) after treatment	pH values <sup>a</sup>									
	Total		Extracted		Nonextracted		ETU derivative		ETU derivative, ppm, GLC value	
	5.6 ppm	7.6 ppm	5.6 ppm	7.6 ppm	5.6 ppm	7.6 ppm	5.6 ppm	7.6 ppm	5.6 ppm	7.6 ppm
	ETU									
1	430	475	368	394	83	113	49.5	38.2	8.16	3.67
8	1758	2676	1883	2515	555	1762	4.6	274	2.32 <sup>b</sup>	82.0
15	2070	1051	1902	1801	862	1476	1.9	44	1.145	55.4
	Nabam									
1	63	151	84	95	17	47	0.25	2.06	0.067	0.076
8	386	323	226	409	153	226	0.47	0.83	0.067	0.048
15	396	357	331	240	252	229	0.46	0.44	0.048 <sup>c</sup>	0.020 <sup>c</sup>
	Zineb									
1	73	69	40	76	15	31	0.30	0.47	<0.005	<0.005 <sup>d</sup>
8	390	489	307	369	118	179	0.55	0.60	<0.005	0.030
15	528	510	474	401	335	292	1.10	0.92	0.011 <sup>c</sup>	0.055
	Maneb									
1	54	91	28	94	11	30	0.17	0.67	<0.005	<0.005
8	149	356	119	275	43	130	0.21	0.80	<0.005	0.187
15	295	457	219	307	125	215	0.40	0.98	0.13	0.023 <sup>e</sup>

<sup>a</sup> Expressed as parent compound equivalent based on <sup>14</sup>C content. <sup>b</sup> Two values missing. <sup>c</sup> Two values <0.005. <sup>d</sup> One value missing. <sup>e</sup> One value <0.005.

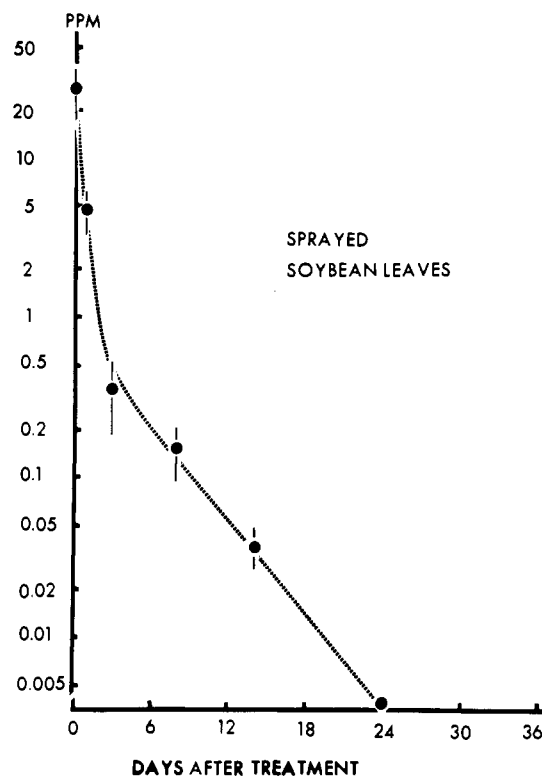


Figure 3. ETU lost with time from spray soybean leaf application of plants kept outside during working hours.

degradation products, whereas, ETU could be absorbed directly. Another reason possibly is because the EBDC part of the surface vs. subsurface treatments was applied 1 month later in the summer on older plants than the ETU part, and soybeans grew considerably less after this treatment.

Over 18% of the applied ETU radioactivity was absorbed by the soybeans from the subsurface treatments (Table IV). For surface plus subsurface treatment, the percentage was smaller than the subsurface treatments alone because it reflected the surface treatment, with its diluting effect.

**Effect of Time.** The amounts of radioactivity in the soybean tops within 24 h indicate the rapidity with which

Table IV. Radioactivity (Percent of That Added to Soil) in the Top of Soybeans after a Surface or Subsurface Soil Treatment of <sup>14</sup>C-Labeled ETU or EBDC

Compound	Treatment rate, ppm			
	0.1	1.0	10	100
Surface				
ETU	0.44	0.43	0.39	
ETU + CaCO <sub>3</sub>			0.73	
Nabam				0.06
Zineb				0.16
Maneb				0.13
Subsurface				
ETU	1.25	2.76	18.3	
ETU + CaCO <sub>3</sub>			18.1	
Nabam				1.29
Zineb				0.62
Maneb				4.21
Surface + Subsurface				
ETU			16.8	

ETU and EBDC can be taken up. Radioactive uptake considerably increased between the first and second harvests (Table III), with little or no increase between the second and third harvests.

Time had little or no effect on total leaf radioactive content (Table V), regardless of whether the fungicides were applied directly by spraying or translocated from the sprayed to nonsprayed leaves (Table VI, Figure 2). On the sprayed leaves (Table V), concentration of <sup>14</sup>C-labeled compounds remained about 2500 ppm for all compounds throughout the 15-day experiment, and the nonsprayed leaves (Table IV) likewise remained near 15 ppm throughout the 15 days. The ETU content on soybean leaves kept outside during working hours decreased rapidly with time (Figure 3). The initial concentration of 27 ppm dropped to <5 ppm after 24 h, and <0.4 ppm after 3 days. The ETU content was 0.04 ppm after 14 days with no ETU detected at the 24- or 35-day harvests. The ETU losses initially were greater than that which a first-order equation could predict, which is the case with most pesticides.

**Amount of ETU Detected and Soil-pH Effect.** No ETU was detectable by derivatization and GLC analysis from ETU surface or subsurface treatments of less than 10 ppm (Table VII). For the soil-pH effects experiment (Table III, last column), considerable quantities of ETU

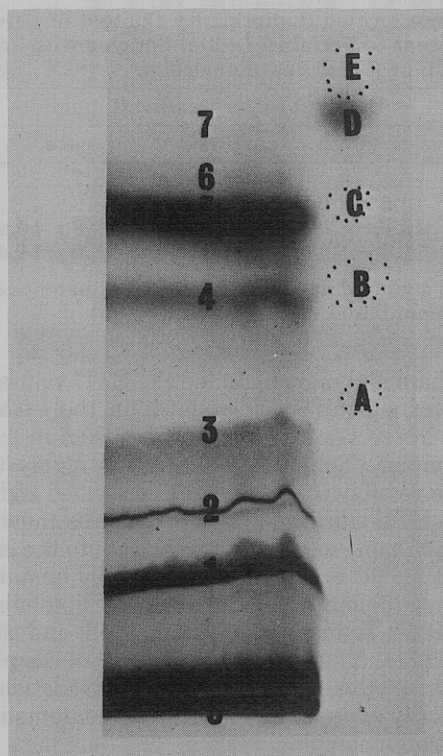


Figure 4. Thin-layer chromatogram of methanol extract from soybeans grown in soil treated with 100 ppm of maneb: (A) Jaffe's base; (B) hydantoin; (C) ethyl-eneurea; (D) ETU; and (E) *N,N*-dimethylene-5-imino-1,2-dithio-4-azolidine-3-thione (DIDAT). The Al<sub>2</sub>O<sub>3</sub> plates were developed with 1-propanol-water (85:15, v/v).

were found in both the acidic (1.2 to 8.2 ppm) and alkaline (3.7 to 82 ppm) ETU-treated soils. The contrast between the surface + subsurface and soil-pH effects experiments may have resulted from differences in handling the harvested plants. The leaves were frozen and stored 10 months before extraction for the surface + subsurface experiment, whereas for the soil-pH effects experiment the leaves were extracted immediately after harvesting and a stable ETU derivative was made from an aliquot of the

Table V. Radioactivity and ETU Content of Soybean Leaves Sprayed with 2500 ppm of [<sup>14</sup>C]ETU or [<sup>14</sup>C]EBDC Fungicides

Days (±1) after treatment	<sup>14</sup> C, ppm <sup>a</sup>						ETU, ppm GLC value <sup>b</sup>			
	Total	Extracted	Nonex- tracted	ETU derivative			ETU, ppm GLC value <sup>b</sup>			
				Benzyl	<i>o</i> -Chloro- benzyl	Mean	Benzyl	<i>o</i> -Chloro- benzyl	Mean	
ETU										
1	2036	2911	76	1271	1057	1164	354	271	313	
8	2537 <sup>c</sup>	3322	609	1014	955	984	215	204	210	
15	2219	2965	831	836	934	885	138	133	136	
Nabam										
1	2712 <sup>c</sup>	2306	670	367	425	396	18.8	20.1	19.5	
8	2128	1803	44	213	308	261	8.2	11.0	9.6	
15	1759	913	530	38	53	45	1.4	1.8	1.6	
Zineb										
1	2230 <sup>c</sup>	1042	1886	368	372	370	16.5	15.3	15.9	
8	4091	1286	713	109	159	135	2.6	3.5	3.1	
15	2998	966	644	37	54	46	0.7	1.1	0.9	
Maneb										
1	1752 <sup>c</sup>	3263	603	447	624	552	5.9	6.9	6.4	
8	3148	2520	565	131	136	134	0.8	1.1	0.9	
15	2915	2142	1141	14	13	13	0.2	0.2	0.2	

<sup>a</sup> Expressed as parent compound equivalent based on <sup>14</sup>C content. <sup>b</sup> ETU content of ETU samples may have been too high for derivatization method, though similarity between the benzyl and *o*-chlorobenzyl derivative values indicates that they may be real. <sup>c</sup> One value missing.

Table VI. Translocated Radioactivity Content of Lateral Leaflets Adjacent to Terminal Leaflet Sprayed with 2500 ppm [<sup>14</sup>C]ETU or [<sup>14</sup>C]EBDC Fungicides

Compd	Days (± 1) after treatment, ppm <sup>a</sup>		
	1	8	15
ETU	16.5 ± 3.0	19.6 ± 4.8	15.7 ± 3.8
Nabam	13.6 ± 3.9	18.6 ± 8.5	18.3 ± 4.9
Zineb	14.8 ± 6.6	12.2 ± 4.8	12.8 ± 10.2
Maneb	12.8 ± 10.2	16.3 ± 0.6	25.0 ± 11.4

<sup>a</sup> Mean and standard deviation expressed as parent compound equivalent based on <sup>14</sup>C content.

extract. Nabam and maneb, unlike the ETU values, had more ETU for subsurface treatments in the extracted (Table VII) frozen (18 months) samples than in freshly analyzed samples (Table III). Nabam is highly water soluble and is possibly absorbed and translocated and then degraded to ETU during storage. Maneb is much less soluble and probably would not translocate to the leaves. However, all EBDC fungicides are generally unstable in water (Nash, 1975) and biological systems (Sijpesteijn and Kaslander, 1964; Truhaut et al., 1973; Vonk and Sijpesteijn, 1970, 1971). Therefore, the presence of the parent fungicides during storage and subsequent degradation does not satisfactorily explain the higher ETU contents in the extracts of frozen samples.

**Balance Sheets for Radioactivity.** Partial balance sheets for radioactivity are given in Tables III, V, VII, and VIII. Generally, the amount extracted was about the same as that not extracted, except for the ETU soil surface treatments, where nonextracted was much larger than the

extracted radioactivity (Table VII).

Presumably, much of the nonextracted radioactivity, especially from subsurface treatments, entered into the plant metabolic system as suggested by Sijpesteijn and Kaslander (1964). Only a small part of the radioactivity extracted was ETU, especially for the fungicides. The maximum for ETU subsurface treatments was 3.25% after 8 days (Table III), and for foliar treatment, 10.75% after 1 day (Table V). Most of the methanol extractable radioactivity was insoluble in chloroform (Table VIII), which indicates quite polar products.

**Degradation Products.** The solution containing the final ETU derivative usually had a magnitude more radioactivity than was measured as ETU by GLC (Tables III, V, VII, and VIII), except for the ETU treatments at 15 days (Tables III and VII, subsurface + CaCO<sub>3</sub> treatment). The radioactivity was measured after a double derivative was formed and the final product cleaned up. This would indicate a fairly nonpolar compound(s) or possibly even a benzyl or pentafluorobenzoyl derivative that survived the ETU procedure and cleanup, which would necessarily be cationic in acid solution and neutral in base, as was the S-benzylated ETU (Newsome, 1972). Derivatized compounds, except ETU, were not readily observable on the electron-capture GLC, although pentafluorobenzoyl derivatives of two known degradation products, 2-imidazolidinone (EU) (mp 173 °C) and ethylenediamine (ED) have been formed and partially characterized on GLC.

The major conversion or degradation product of both ETU and the EBDC fungicides was EU, as indicated by TLC plates (Figure 4). The 2,4-imidazolidinedione

Table VII. Radioactivity and ETU Content in Leaves from Soybeans Grown 16 Days after a Soil Treatment of [<sup>14</sup>C]ETU

Treatment location	<sup>14</sup> C, ppm <sup>a</sup>				
	Total	Extracted	Nonextracted	ETU derivative	ETU derivative, ppm GLC values
Surface	0.05	0.1-ppm Treatment		nd	nd
Subsurface	0.81 <sup>c</sup>	nd <sup>b</sup>	0.020	nd	nd
Surface	0.54	1.0-ppm Treatment		nd	nd
Subsurface	19.6 <sup>c</sup>	0.001	0.134	0.001	nd
Surface	4.91	10.0-ppm Treatment		0.008	nd
Subsurface	1534 <sup>c</sup>	539	565	0.010	0.004
Surface + subsurface	1738	431	558	0.032	0.009
Surface + CaCO <sub>3</sub>	8.75	0.016	2.42	nd	nd
Subsurface + CaCO <sub>3</sub>	1425	612	766	0.025	0.025

<sup>a</sup> ETU equivalent based on <sup>14</sup>C content. <sup>b</sup> None detected. <sup>c</sup> Previously published (Nash, 1974).

Table VIII. Radioactivity and ETU Content in Leaves from Soybeans 16 Days after a 100-ppm Soil Treatment of [<sup>14</sup>C]EBDC Fungicides

Treatment location	<sup>14</sup> C, ppm <sup>a</sup>						ETU derivative, ppm GLC values
	Total	Extracted	Nonextracted	Acid solution <sup>b</sup>		ETU derivative	
				Chloroform soluble	Chloroform insoluble		
Surface	50	14	Nabam		0.20 <sup>c</sup>	0.036	
Subsurface	5 708	3282	18	139	3721	7.66	0.631
Surface	87	35	Zineb		0.27	0.003	
Subsurface	3 494	2010	28	158	3078	4.36	0.060
Surface	112	63	Maneb		0.36	0.045	
Subsurface	22 106	8641	24	397	9960 <sup>d</sup>	56.1	9.17 <sup>e</sup>

<sup>a</sup> Expressed as parent compound equivalent based on <sup>14</sup>C content of frozen plant material. <sup>b</sup> Solution normally discarded during derivatization of ETU for GLC analysis (Nash, 1974). <sup>c</sup> Values less than 1 are unreliable because of low <sup>14</sup>C content. <sup>d</sup> Two values missing. <sup>e</sup> One replication contained 25 ppm; a more realistic value might be 3.91.

(hydantoin) and 3-(2-imidazolin-2-yl)-2-imidazolidine-thione (Jaffe's base) seem to be present in several extracts, but further confirmation is necessary. At least seven and possibly 10 distinct radioactive areas have been observed on TLC plates from methanol extracts of ETU- and EBDC-fungicide treatments. The TLC pattern was similar, regardless whether ETU and EBDC fungicides were sprayed onto the soybean leaves or injected into the soil. The products were probably a mixture of those obtained by simply exposing the fungicides to air (Czegledi-Janko, 1967), metabolic degradation (Sijpesteijn and Kaslander, 1964; Vonk and Sijpesteijn, 1971), and possibly photodegradation (Cruikshank and Jarrow, 1973; Ross and Crosby, 1973). Engst and Schnaak (1974) have proposed a degradation pathway that results in five end products: 2-imidazoline, ethylenebis(isothiocyanate) sulfide (EBIS), ethylenebis(thiocyanate), EU, and ED. The 2-imidazoline is an end product from both the EBDC, with ETU as an intermediary, and ETU. Recently, Newsome et al. (1975) have found *N,N'*-dimethylene-5-imino-1,2-dithia-4-azolidine-3-thione (DIDAT) or 5,6-dihydro-3*H*-imidazo-[2,1-*c*]-1,2,4-dithiazole-3-thione (DIDT), formerly known as ethylenethiuram monosulfide (ETM), to be an important degradation product in addition to smaller amounts of ED.

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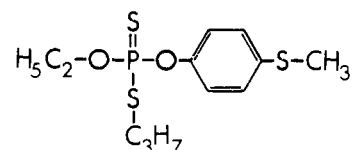
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## Fate of *O*-Ethyl *O*-[4-(Methylthio)phenyl] *S*-Propyl Phosphorodithioate (BAY NTN 9306) in Cotton Plants and Soil

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About half the dose of <sup>14</sup>C- or <sup>32</sup>P-labeled BAY NTN 9306 (*O*-ethyl *O*-[4-(methylthio)phenyl] *S*-propyl phosphorodithioate) applied to the leaves of cotton plants was absorbed during the first 24 h; surface and internal residues of the insecticide and its toxic derivatives diminished to insignificant levels after 8 and 32 days, respectively. The principal alteration products of BAY NTN 9306 found in different tests with plants, soil, and water were the toxic sulfoxide and sulfone derivatives formed by oxidation of the ethereal sulfur and the respective substituted phenols, both free and conjugated, produced by hydrolysis of the organophosphorus esters.

The experimental insecticide BAY NTN 9306 (*O*-ethyl *O*-[4-(methylthio)phenyl] *S*-propyl phosphorodithioate)



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is being developed for use in controlling phytophagous lepidopteran insects, especially *Heliothis* sp., that attack cotton. Previous reports by Bull and Ivie (1976) and Ivie et al. (1976) described in detail the metabolic fate of BAY NTN 9306 in white rats and a lactating dairy cow. The present paper reports additional studies of the fate of the