TRANSLOCATION OF BENOMYL IN ELM (*Ulmus americana* L.)

IV. UPTAKE AND MOVEMENT THROUGH THE FOLIAGE UNDER LABORATORY AND FIELD CONDITIONS

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Information Report CC-X-52

September 1973

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RÉSUMÉ ET CONCLUSIONS

Au moyen d'expériences effectuées en laboratoire et sur le terrain, on a étudié l'absorption et le transport du bénomyle et du MBC-C^{14} au niveau des feuilles de l'arbre. La pénétration et le déplacement de ces deux substances anti-champignons, à partir des feuilles, est lent et le transport s'effectue surtout par le courant causé par l'évaporation. Cependant, une rétention, une absorption et un transport appréciables semblent s'effectuer par l'écorce; on pourrait exploiter cette méthode de lutte à titre préventif. Une étude sur le terrain de la pulvérisation du bénomyle et du MBC-SO_4 sur le feuillage et l'écorce, étude qui a duér trois ans, montre que des mélanges, un moment et une fréquence de pulvérisations appropriés donneraient un bon espoir quant à la protection préventive des ormes qui autrement seraient condamnés.

INTRODUCTION

Considerable disagreement exists as to whether benomyl or benomyl-like compounds can be effectively used as foliar sprays for the control of systemic and vascular diseases. On one hand, foliar sprays with benomyl and the closely related compounds, MBC, MBC-Cl and thiophanates, have yielded adequate protection against Verticillium wilt of cotton (Buchenauer and Erwin 1971; Buchenauer, Erwin and Keen 1973), boll rot of cotton (Hine et al 1971), Verticillium wilt of chrysanthemum (Busch and Hall 1971), powdery mildews and scab of apples (Delp and Klopping 1968), citrus diseases (Hearn et al 1971, Solel et al 1972) brown rot of peaches (Kable 1970), leaf spots of peanuts (Smith and Crosby 1972) and various foliar diseases of roses and other ornamental, agricultural and horticultural crops (Delp and Klopping 1968). On the other hand, repeated applications of benomyl over the foliage of elm trees have produced erratic to good responses for control of the Dutch elm disease (Prasad 1971, Smalley 1972, Hart 1972, Stipes 1972, Biehn 1972). Whereas some investigators (Smalley 1973, Biehn 1972, Hart 1972) reported adequate protection of the elm trees following benomyl spraying, others (Stipes 1972, Prasad 1972) did not find any control whatsoever. Therefore, there was a need for a comprehensive investigation and the present report describes some factors that control the entry and movement of benomyl and MBC-C^{14} into the elm leaves both under laboratory and field conditions.

MATERIALS AND METHODS

A. Laboratory Experiments:

(a) Culture of Plants:

Elm seedlings were grown from seeds planted in sterilized soil in a greenhouse and when the plants were 4 months old, they were transferred to growth chambers set at 3000 ± 200 foot candles (16 hr. photoperiod), 76 ± 2^0F; and at 50 ± 10% relative humidity. The seedlings were regularly irrigated with half Hoagland solution and under these optimum conditions of nutrition and environment, they produced excellent growth of foliage.

(b) Application of benomyl and radioactive MBC:

Two series of experiments were carried out, one with cold benomyl suspension and the other with radioactive benomyl, (MBC-C^{14}). For experiments with MBC-C^{14}, 3 fully developed leaves were either painted with one millilitre of 1500 ppm solution on both sides or the pesticide was simply added to a lanolin well of known area (Fig. 1) on the lamina according to procedures of Prasad, Foy and Crafts (1967). Various adjuvants were added to benomyl and MBC-C^{14} formulations whose details are given in Appendix I.

Radioactive MBC was purchased from the International Nuclear Corporation, California (specific activity 500 mC/mM) and before use, its radiochemical purity was tested. MBC-C^{14} was first solubilized in a minimal of 44% lactic acid and then further dilutions were made from this stock solution. Normally the radioactivity was adjusted to a level (45 x 10^6 DPM per ml) whereby it produced sufficient counts in the leaf and stem samples.

(c) Determination of Fungicides:

The uptake and translocation of benomyl was estimated by the bioassay technique (Hock and Schreiber 1970, Prasad and Travnick

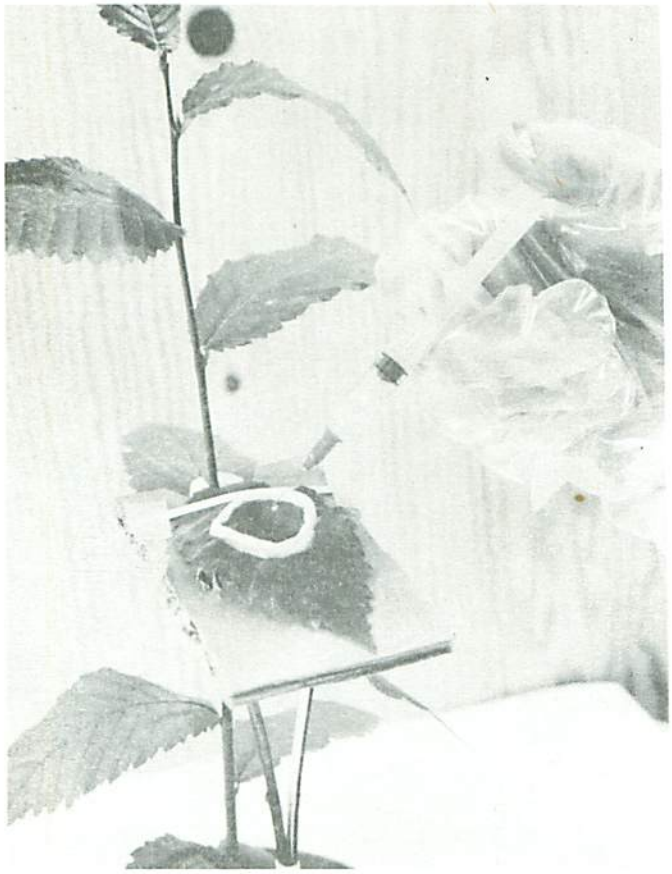


Fig. 1. Method of application of MBC-C^{14} to elm leaves. A known area of the lamina is confined with invert lanolin and a known amount of the fungicide together with appropriate adjuvants are filled in the well.

1972). Leaf disks (21 mm) and segments of elm twigs from the treated trees were excised and placed on the agar media seeded with Penicillium expansum. The zones of inhibition thus produced after 48 hours were measured and these reflected the concentrations of the fungitoxicants that were translocated (Fig. 2). In some cases, the movement of benomyl was studied with filter paper "leaves". A Whatman filter paper (15 cm) was cut into the size of a fully grown "leaf" having a wick, simulating the petiole (Figs. 3,4,5). This "petiole" was then dipped in a cencentration (1500 ppm) of benomyl for 6 hours and "leaf disks" (21 mm) were cut from this paper leaf for bioassay of the fungicide.

(d) Radiometric Determination:

The radioactivity of the plant samples was measured (i) qualitatively by gross autoradiography and (ii) quantitatively by liquid scintillation counting. The technique of extraction of radioactivity from the fed samples together with the details of counting and autoradiographic procedures are described in the first report of this series (Prasad 1972).

B. Field Experiments:

(a) Selection of Mature Trees:

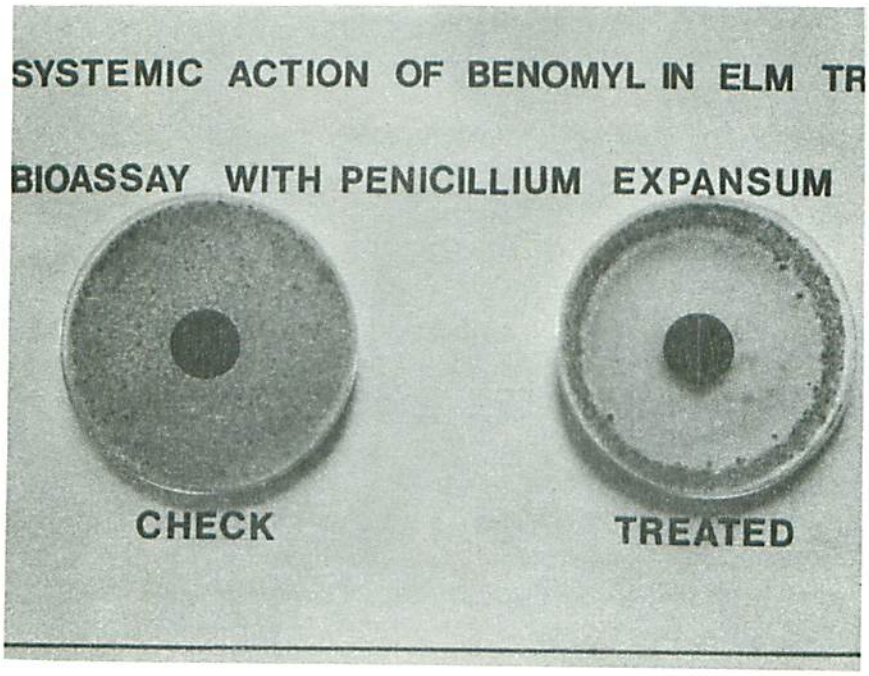
Trees healthy in appearance and 30-60 years old were selected at two different sites maintained by the National Capital Commission, (N.C.C.) Ottawa. One site had light loam soil and was located at the National Research Council's campus at the Montreal Road, while the other site was composed of silt, loam and gravel soil and was located at Shirley's Bay, just west of Ottawa.

About 200 trees were selected, marked and measured for the diameter at breast height (D.B.H.). All trees were tested for the presence of the Dutch elm disease using standard laboratory procedures and only those giving negative results were chosen. All trees at the NRC campus were previously sprayed with methoxychlor by the N.C.C. while those at Shirley's Bay had not been treated by any pesticides. The experiments were performed over a period of three years (1971,72 and 73) and each year new trees were chosen.

In one year (1972) application of benomyl and MBC-SO_4 (MBC-sulphate) at different stages of the leaf-flush was investigated. Accordingly, groups of trees were selected, (a) before leaf-flush (when buds were swollen and no leaves unfolded), (b) during leaf-flush (leaves just emerging from the buds, and (c) after leaf-flush (leaves fully expanded). For details see Fig. 6.

(b) Method of Spray Application:

The primary aim of the spraying experiments was to monitor the uptake and movement of benomyl and MBC-SO_4 in large trees so that after having established the concentrations of the fungitoxicants in the sap stream, one could inoculate these trees with the Dutch elm disease. Contrariwise (i.e. infecting the healthy trees and then spraying the benomyl and MBC-SO_4) would have killed many high value trees unnecessarily and the National Capital Commission would not have approved this. Therefore, to study foliar uptake and translocation under field conditions, the ground area under the canopy of each tree was covered with a Mylar (2 mm) plastic sheet (40 x 40 ft). This plastic was then held firmly to ground by means of wooden frames



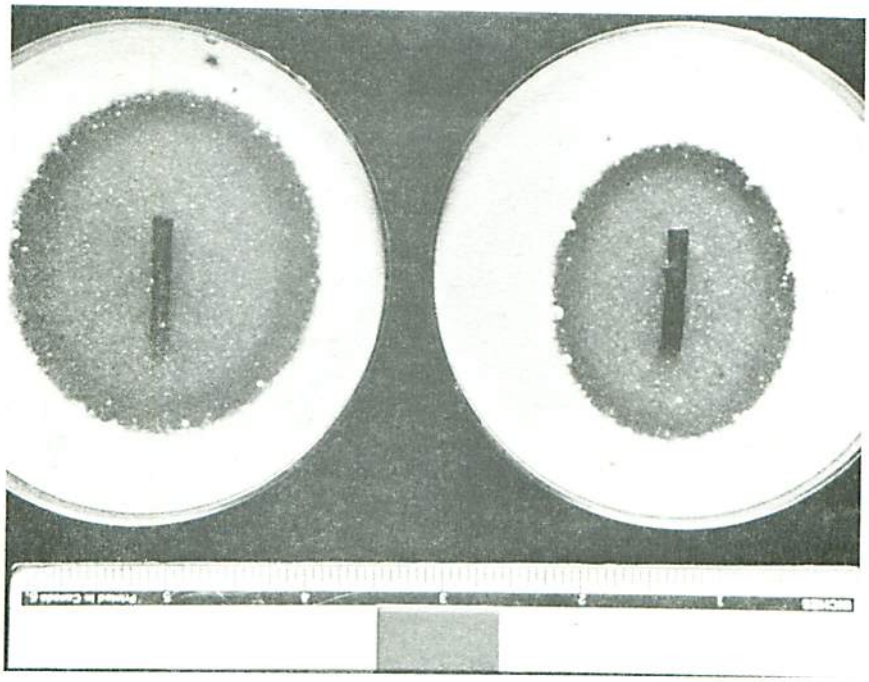


Fig. 2. Photographs showing zones of inhibition produced by treated and untreatd leaves (top) and two pieces of treated stem with benomyl (bottom).

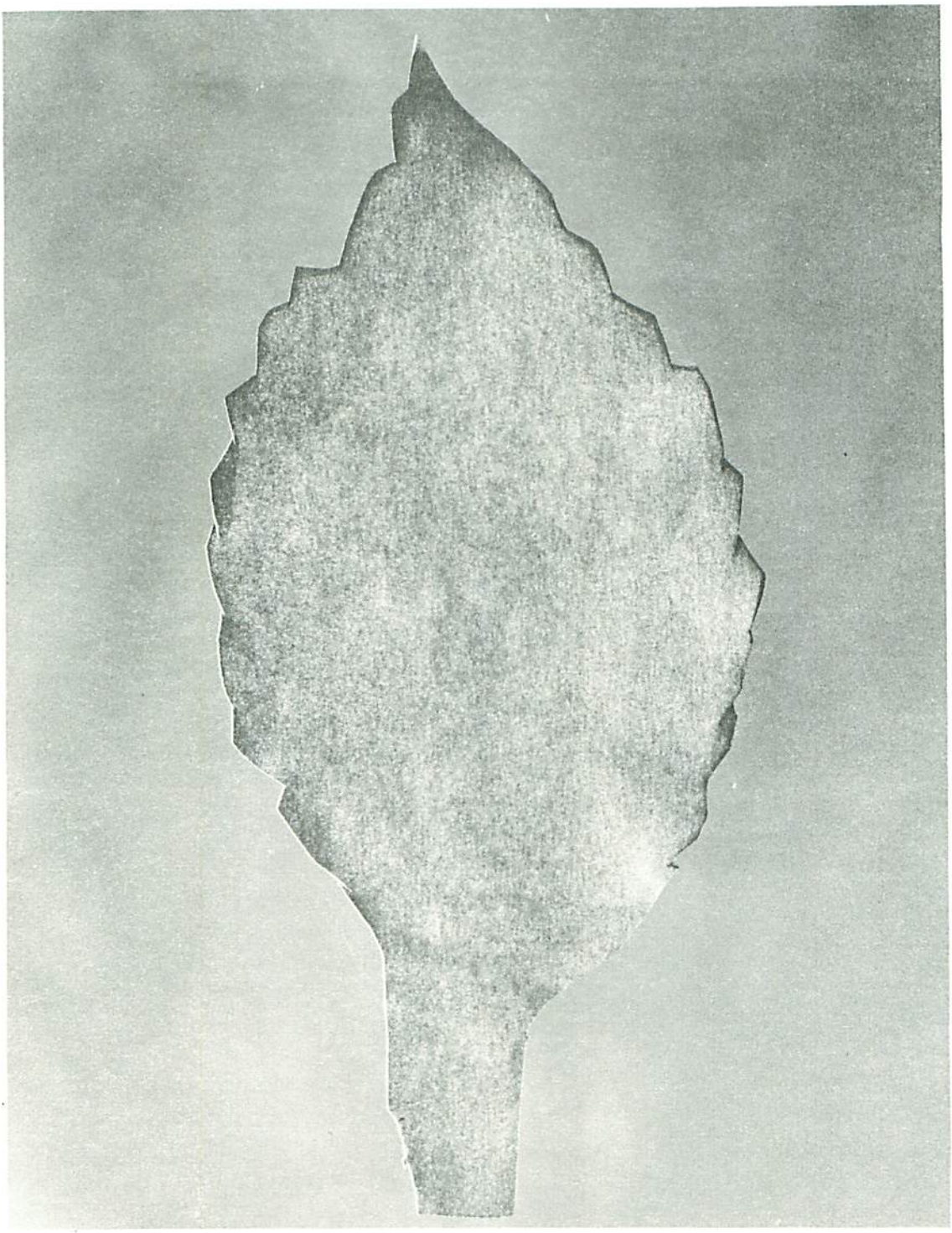


Fig. 3. Movement of dye (rhodamine o.1%) in filter paper "leaf". Note the concentration of the dye on the margins of the "leaf".

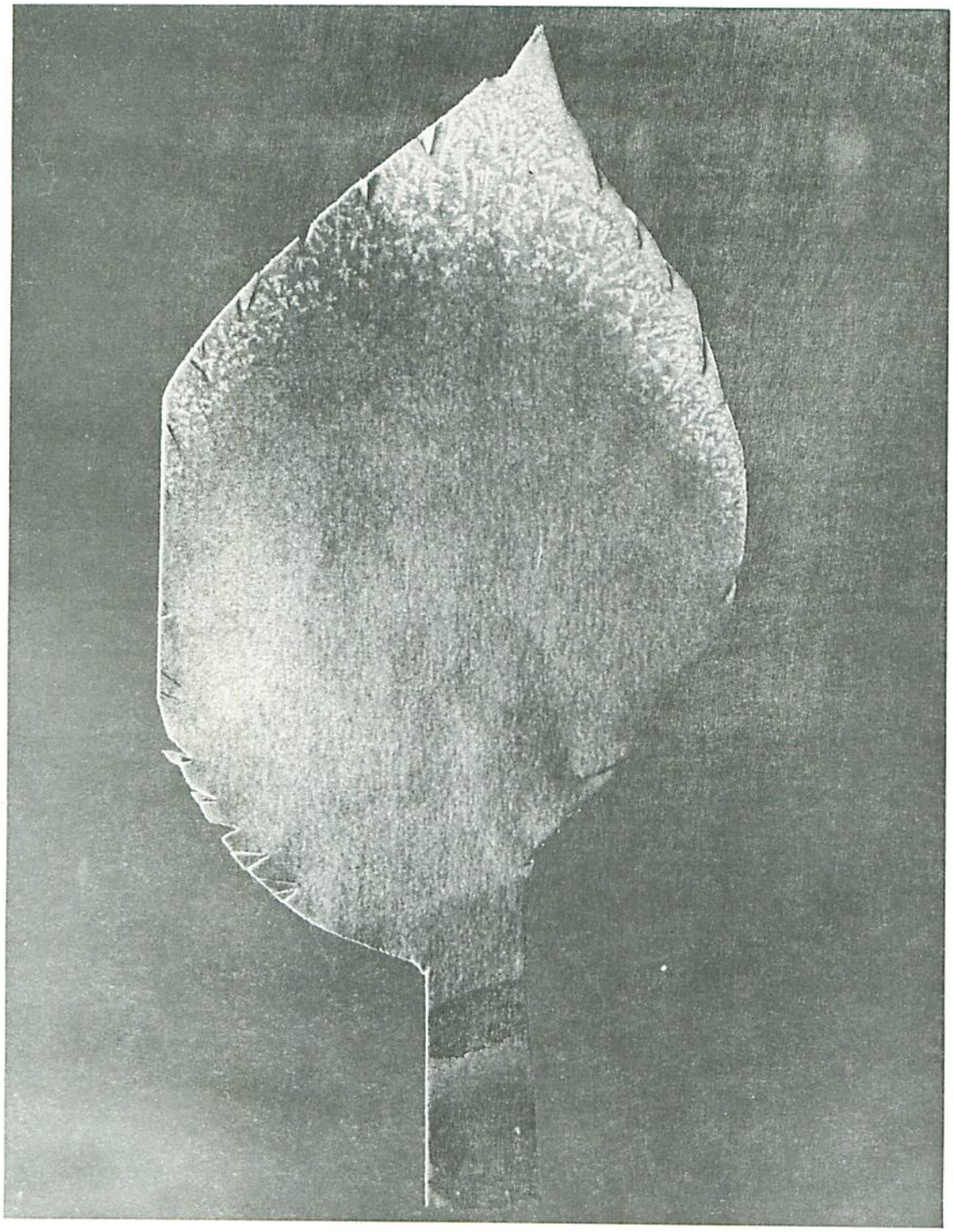


Fig. 4. Movement of benomyl in filter paper "leaf". Note the concentration of crystals in the apical regions.

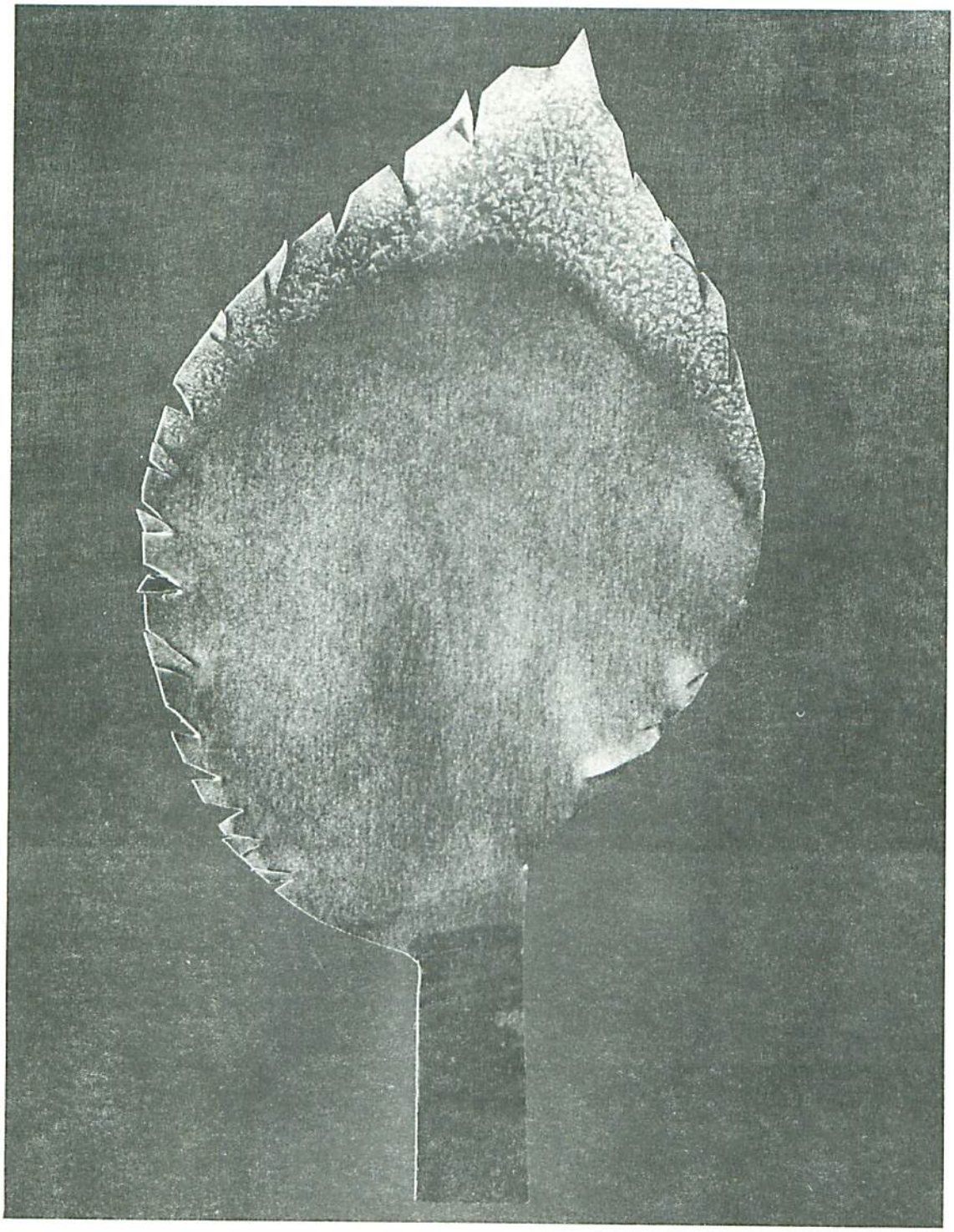


Fig 5. Movement of dye and benomyl in a filter paper "leaf".

and was finally sheltered from wind by a snow fence (Fig. 7). The fence and plastic sheet were left under the tree for 4 months after spraying so that no leachates from the sprayed leaves went down to the roots. After rainfall, the spray washings tended to create puddles on the plastic sheets (Fig. 8a) but these were quickly and periodically removed by hand so as not to contaminate the soil and roots.

All parts of the crown were sprayed with the fungicides except those branches which were enclosed within a plastic bag (6 ft. x 6 ft.) (Fig. 8b). Spraying was performed with a mist blower mounted on a cherry picker (Fig. 9a) or on a scaffolding (Fig. 9b). The dosage was @ 10 gallons (2500 ppm) of benomyl solution or suspension with or without adjuvants (0.1% Biofilm or Nufilm) per tree. Apart from some trees which were in pre-flush stages, all trees were sprayed in May-June. The pre-flush trees were however, sprayed late in April or very early in May depending upon the weather conditions. Leaf, twig, bark and core samples from the sprayed and unsprayed (bagged) regions of the trees were removed periodically and bioassayed with the P. expansum agar technique.

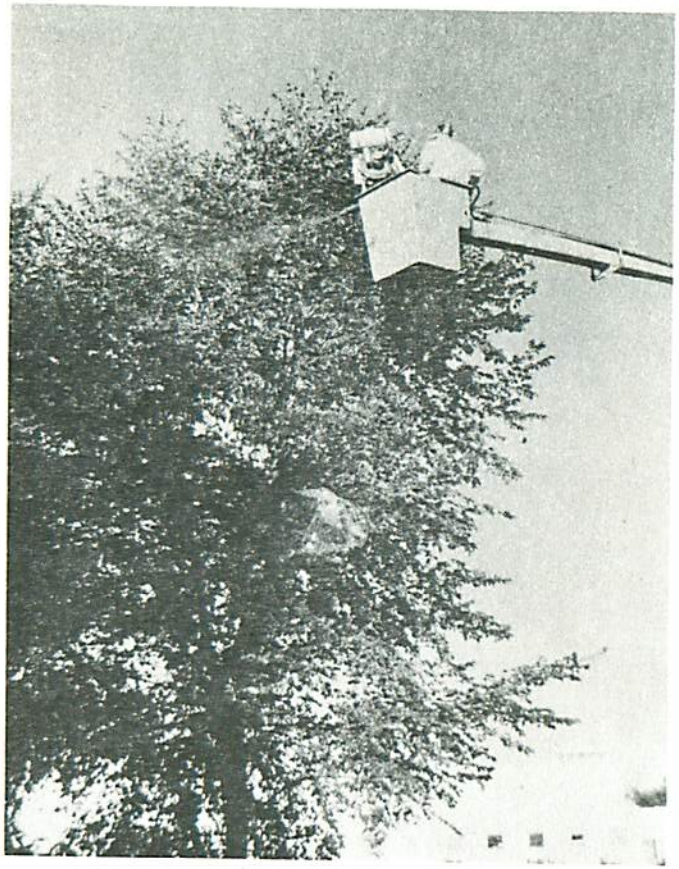
Fig. 6. Elm trees during and after the leaf-flush at the Shirley's Bay site, Ottawa, 1972

Fig. 7. Method of investigating foliar translocation of benomyl under field conditions; the tree base was covered with a plastic sheet and a snow fence was installed around the tree to reduce wind disturbing the plastic cover.

(a)

(b)

Fig. 8. Methods of studying foliar translocation of benomyl under field conditions (a) puddles of spray residue on the plastic sheet after rainfall (b) bagged unsprayed branches on a sprayed tree.



(a)

(b)

Fig. 9. Spraying of elm trees with a mistblower under field conditions by using (a) "cherry-picker" or (b) scaffolding.

RESULTS

A. Laboratory Experiments:

(a) Uptake and translocation from painted leaves:

The data in Table I clearly indicate that following a uniform coating of MBC-C^{14} on mature foliar surfaces, radioactivity begins to migrate to untreated parts of the seedlings but the major portion of the applied dosage remained in the treated leaf. Biofilm apparently enhanced the penetration and translocation by approximately twofold into unpainted regions of the seedling.

TABLE I

Distribution of MBC-C^{14} in Painted and Unpainted Area of Elm Seedlings Following Uptake from a Conc. of 1700 ppm for 6 Days.

Parts	MBC Alone		MBC + Biofilm	
	DPM/mg	% translocated	DPM/mg	% translocated
Treated Leaves	5×10^5	Not assayed	5×10^5	Not assayed
Untreated Leaves	31	0.09	45	0.13
Untreated Stem	15	0.04	31	0.09
Untreated Roots	29	0.08	76	0.22

Because of very poor uptake from the mature leaves, it was thought that penetration from a younger leaf might be faster. Accordingly, young leaves were painted near the apex and old leaves near the basal region of the tree and the results (Table II) indicate that there is slightly greater uptake and movement through the young foliage.

TABLE II

Distribution of Radioactivity in Painted and Unpainted Areas of Elm Seedlings Following Uptake Through Young and Old foliage*

Part	Young leaf		Old leaf	
	µgm/gm	Percent	µgm/gm	Percent
Treated leaves	74.210	91.50	58.291	98.65
Untreated leaves	0.464	0.57	0.283	0.48
Untreated stem	5.795	7.15	0.421	0.71
Untreated root	0.629	0.78	0.095	0.16
Total	81.098	100	59.090	100

* Conc. of MBC-C^{14} - 1500 ppm; pH - 3.5; uptake period - 6 days.

Again, much of the radioactivity seems to be confined to the treated leaves, both in the young as well as in the old leaves but it is clear that younger leaves possess more efficient mechanism for mobilization of MBC to stem and roots. Why there should be about a 10 fold translocation of activity within the stem of a young leaf-treated plant over that from an old leaf-treated plant is not clear.

Distribution of MBC-C^{14} within a single leaf:

Since a major part of the activity seemed to concentrate in the treated leaves - be it young or old (Table II), the question arose where most of the activity was being redistributed within the leaf itself. Therefore a single leaf after being fed with MBC-C^{14} (1700 ppm) by the lanolin ring technique (Fig. 1) was excised into three component parts: apical, central, and basal and the amount of radioactivity determined in each part. From Table III it is

evident that the transpiring regions (apical) are the primary site of concentration of activity. It should be noted that the actual treated area within the lanolin ring was discarded and therefore the contents in the central part of the leaf reflect distribution from the source of application.

TABLE III

Movement and Distribution of MBC-C^{14} Within a Single Leaf After Removal of the Treated Area (lanolin ring).

Leaf Parts	DPM	% Translocated
Apical	244	0.75
Central	61	0.21
Basal	109	0.33

To substantiate that movement follows the path of transpiration stream within a leaf, experiments were carried out with "leaves" made of filter paper as shown in Figs. 3,4,5. A rhodamine dye (0.1%) was incorporated with the benomyl solution (1500 ppm) and the "leaves" were allowed to take up the dye plus the pesticide for 24 hours. Disks (21 mm) were subsequently cut from the apical, central and basal parts of the filter paper "leaves" and bioassayed for benomyl with the standard technique. The data (Table IV) and Figs. (3,4,5) confirm that much of the movement is by diffusion and that fungicidal activity concentrates in the region of high evaporation similar to transpiration from an authentic leaf surface.

TABLE IV

Distribution Pattern of Benomyl in Filter Paper "Leaves" after Immersion of "Petiole" into a Suspension for 24 hours*

"Leaf" Parts	Zone of Inhibition (mm)
Apical	93
Central	68
Basal	69

* Conc. 2500 ppm; pH - 3.5;

Influence of Adjuvants and Metabolites on Foliar Penetration and Translocation

Having established the pattern of translocation by and within the leaf, the next logical step was to investigate what additives could enhance the uptake and movement. Accordingly a few spray adjuvants and two host metabolites (sucrose - 1% and urea - 0.1%) were mixed with the MBC-C^{14} solution or pure benomyl suspension and pipetted into the well of the lanolin ring. In some cases the leaf surface within the ring was ruptured with carborundum (2.5 gm/L). After the uptake period the lanolin ring was cut out and discarded and the fungicidal activity monitored in apical parts of the treated leaf (Table V) by the bioassay technique or autoradiographed as shown in Figs. 10,11,12. Inspection of Table V revealed that some adjuvants (biofilm) had significant effects on the foliar uptake. It seemed rupture of the leaf surface also promoted uptake. On the other hand, the addition of host metabolites neither augmented the rate of uptake significantly (see Figs. 11,12) nor did they appreciably accelerate

the translocation into the shoot and root. The metabolites, however, seemed to have had some effects on uniform distribution of the label within the treated leaves but no radioactivity appeared on the untreated shoot and root.

TABLE V

Effects of Spray Additives on Uptake into Elm Leaves Following Application by the Lanolin Ring Technique*

Treatments	Concentration of adjuvants	Zone of Inhibition (b) (mm)
Benomyl alone	-	11
Benomyl + Rewetting (a)	-	17
Benomyl + Regulaid	1.3 ml/L	18
Benomyl + Biofilm	0.65 ml/L	23
Benomyl + Carborundum	2.5 gms/L	25

* Conc. benomyl - 1500 ppm; uptake period - 14 days.

(a) Dried benomyl residues rewetted with distilled water every day.

(b) Only apical parts bioassayed.

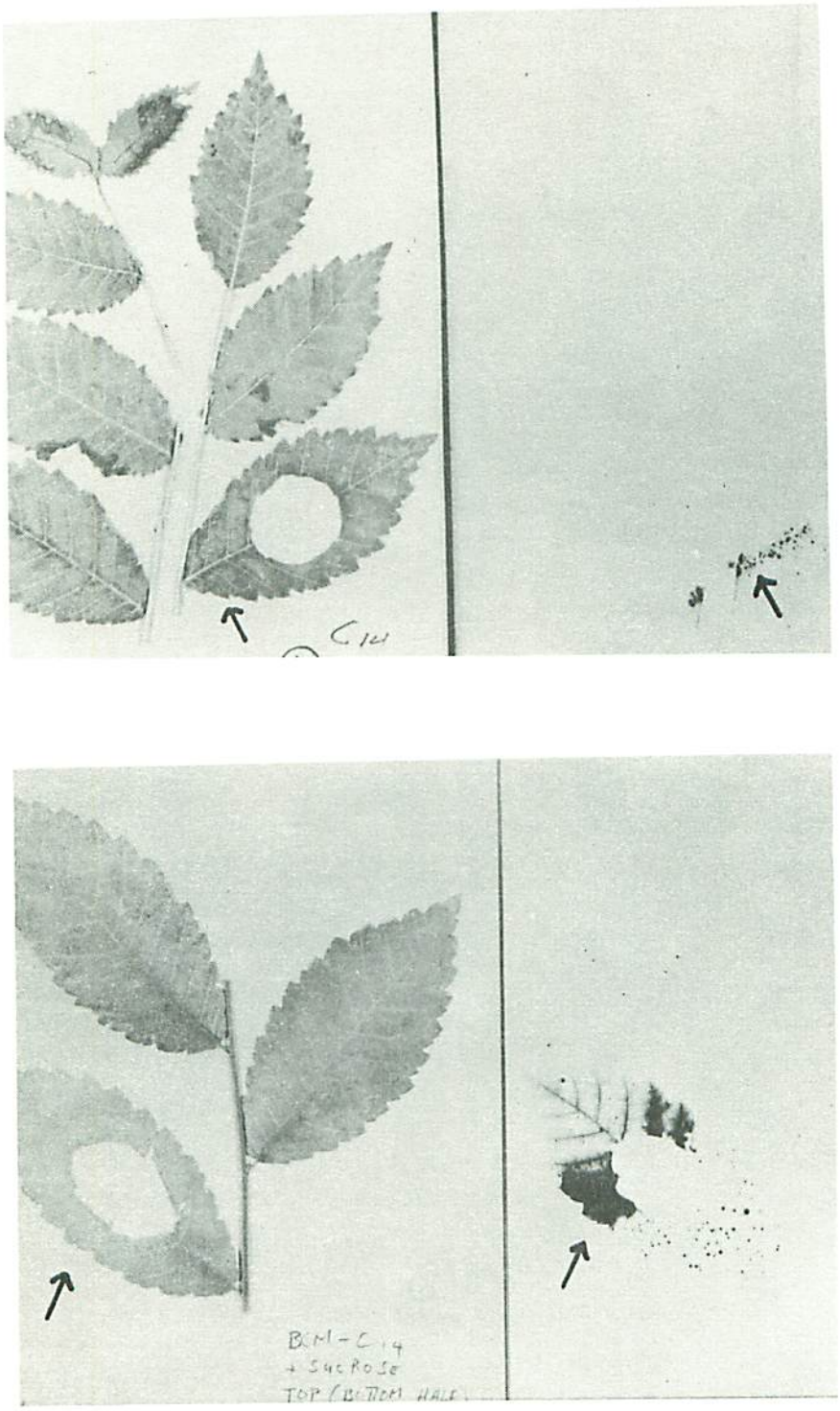


Fig. 10. Foliar uptake and translocation of MBC-C^{14} alone (top) and with added sucrose (bottom) in elm seedlings. Photograph of treated plant on left and autoradiograph of the same on right as indicated by arrows.

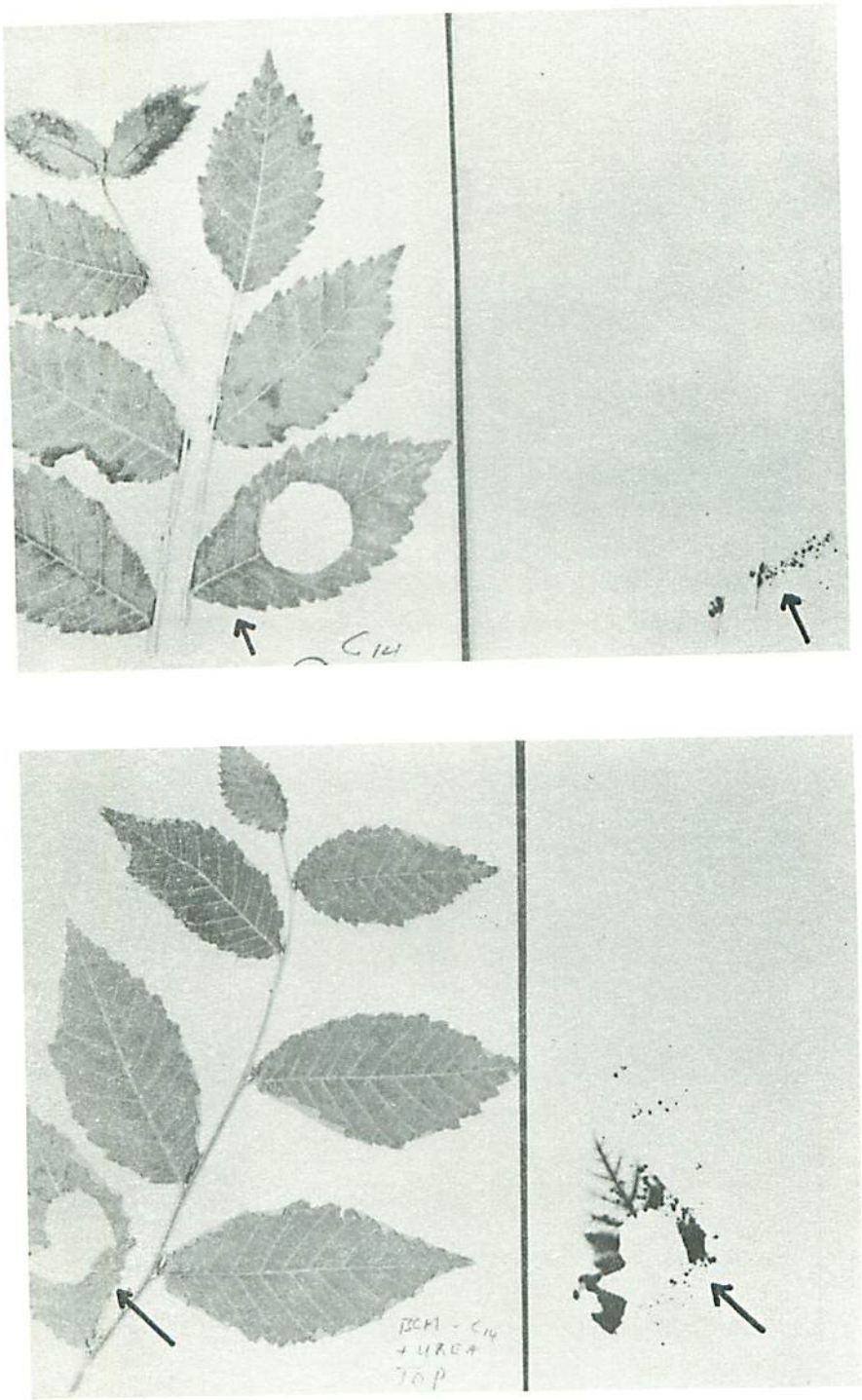
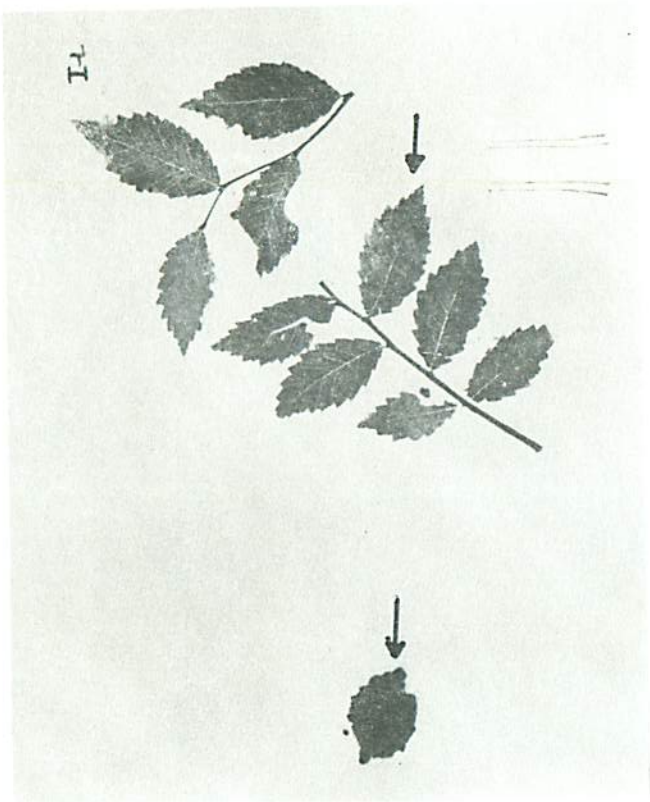
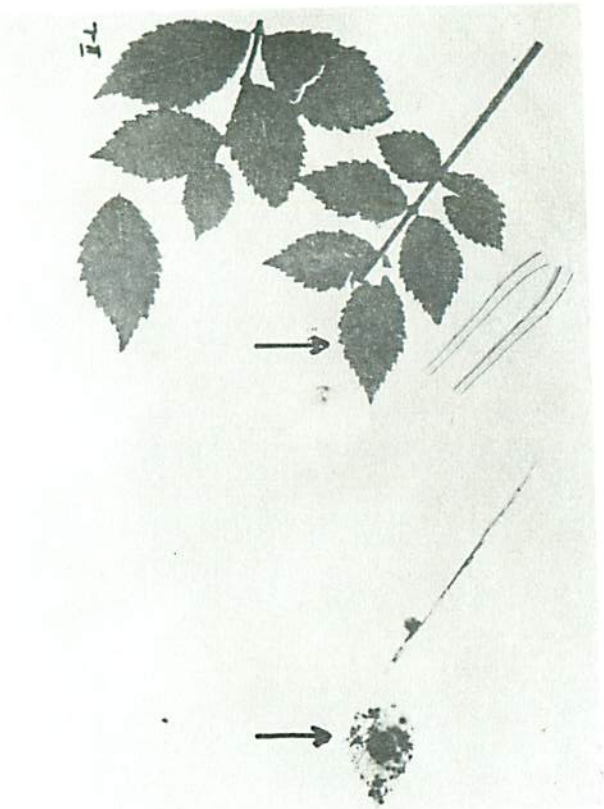


Fig. 11. Foliar uptake and translocation of MBC-C^{14} alone (top) and MBC-C^{14} + urea (bottom). Photograph of treated leaves on left and autoradiograph of the same on right as indicated by arrows.



(a)



(b)

Fig. 12. Foliar uptake and movement of MBC-C^{14} alone (a) and with biofilm (b). Autoradiograph of the treated leaf with lanolin smear on left and photograph of the same plant on the right as indicated by arrows. Note the greater movement of MBC-C^{14} in presence of biofilm.

B. Field Experiments:

The data from the field experiments consist largely of measurement of zones of inhibitions produced by benomyl or MBC-salts on the agar plates seeded with P. expansum. Zone of inhibition was taken to be a true measure of the presence of the pesticides into and onto a tissue. Table VI shows the distribution pattern in mature trees at the NRC site, Ottawa, following spraying of trees with benomyl and MBC-SO4.

TABLE VI

Zones of Inhibition Produced by Leaf, Twig, Bark and Wood Samples of Large Elm Trees Following Spray Application with MBC-SO4 and Biofilm* in 1971.

	Zones of Inhibition (mm)			
	Leaf	Twigs	Bark	Wood
Sprayed - open branch				
(a) MBC-SO4 alone	0.15	0.85	6.25	1.62
(b) MBC-SO4 + biofilm	1.62	0.68	26.13	3.01
Unsprayed - bagged branch				
(a) MBC-SO4 alone	0	0	3.2	0
(b) MBC-SO4 + biofilm	0.45	0.44	15.5	0

* Dosage 15 gallons (1000 ppm)/tree; biofilm 0.65 ml/L; sampled after 9 weeks.

Evidently there is little uptake and movement from

the sprayed regions into the unsprayed (bagged) branches. Biofilm has some positive effects and this might be due to its spreader-sticker action. Intermittent rainfall took place during the post-spray periods and much of the water soluble MBC-S04 was leached down to the ground level on the plastic covers.

Because laboratory experiments with MBC-C^{14} had demonstrated greater penetration through the young foliage, subsequent spraying operations were performed on trees (a) before, (b) during and (c) after leaf-flush in the spring and summer of 197?. Relatively small trees (see Fig. 6) were selected at Shirley's Bay, Ottawa, so that spraying and sampling operations could be carried out with convenience. The site was uneven and stony and thus the cherry picker could not be employed. Instead, an ordinary scaffolding and ladder were used. Table VII shows that there was no spectacular effect of spraying before the leaf-flush apart from the fact that the exposed branches (bark) received and retained somewhat higher amounts of the deposit. Further data on the uptake and retention of spray residues on trees treated during and after leaf-flush are given in Tables VIII, IX X.

TABLE VII

Uptake, Movement and Persistence of MBC-SO_4 on Elm Following Foliar Spraying of Mature Trees Before Leaf Flush in 1972

	Diameter of Zone of Inhibition (mm)											
	Leaf				Bark				Wood			
	Follow-up Sampling - Days											
	15	30	60	90	15	30	60	90	15	30	60	90
Sprayed (Open Branch)[1]												
(a) MBC-SO_4 Alone	0	0	0.2	2	25	6.3	4.8	4	5.7	0	0	0
(b) MBC-SO_4 + Biofilm	0	1.7	0	0	35.3	16	3.3	2	6.2	0	0	0
(c) MBC-SO_4 + Nufilm	0	0	0	2	26	18.7	2.8	2.7	3.0	0	0	0
Unsprayed (Bagged Branch)[2]												
(a) MBC-SO_4 Alone	0	0	0	-	0	0.7	1.7	0	0	0	0	0
(b) MBC-SO_4 Biofilm	0	0	0	-	0	2.2	0	7	0	0	0	1
(c) MBC-SO_4 Nufilm	0	0	0	0	0	0.5	1.7	0	0	0	0	0

[1] Sprayed branch represents retention and persistence.

[2] Unsprayed branch reflects true uptake and translocation.

TABLE VIII

Uptake, Movement and Persistence of MBC-SO_4 on Elm Trees Following Foliar Spraying with and without Adjuvants During Leaf Flush in 1972

	Diameter of Zone of Inhibition (mm)											
	Leaf				Bark				Wood			
	Follow-up Sampling - Days											
	15	30	60	90	15	30	60	90	15	30	60	90
Sprayed (Open Branch)[1]												
(a) MBC-SO_4 Alone	7.5	1.3	0	0.3	12.3	10.2	2.1	5.4	0	0	0	0.7
(b) MBC-SO_4 + Biofilm	22.3	0.8	0	0	34.5	10.8	4.7	6.4	8.5	0	0	0
(c) MBC-SO_4 + Nufilm	7.3	9.7	3.2	1.5	11.6	12.0	3.8	14.4	0	0	0	1.2
Unsprayed (Bagged Branch)[2]												
(a) MBC-SO_4 Alone	0	0	0	0	2.5	3.0	0.6	1.0	0	0	0	0
(b) MBC-SO_4 + Biofilm	1.3	0	0	0	0	11.0	1.3	6.5	0	0	0	0.2
(c) MBC-SO_4 + Nufilm	2.0	0	0	0	3	5.3	0.3	8.2	0	0	0	1.2

[1] Sprayed branch represents retention and persistence.

[2] Unsprayed branch reflects true uptake and translocation.

TABLE IX

Uptake, Movement and Persistence of MBC-SO_4 on Elm Trees Following Foliar Spray with and without Adjuvants After Leaf Flush in 1972

	Diameter of Zone of Inhibition (mm)											
	Leaf				Bark				Wood			
	Follow-Up Sampling - Days											
	15	30	60	90	15	30	60	90	15	30	60	90
Sprayed (Open Branch)[1]												
(a) MBC-SO_4 Alone	23	6.6	2.5	3.1	20	10.9	9.4	19.3	0.5	0	0	0
(b) MBC-SO_4 + Biofilm	19.2	17.6	3.1	11.4	16.9	17.2	18.6	28.7	0.8	0	4.9	4.8
(c) MBC-SO_4 + Nufilm	20.5	7.8	2.1	3.0	18.2	14.3	15.5	19.8	3.9	0.3	1.7	1.8
Unsprayed (Bagged Branch)[2]												
(a) MBC-SO_4 Alone	2.8	0	0	0.5	3.8	6.9	2.4	22.3	0.3	0	0.2	4.6
(b) MBC-SO_4 + Biofilm	0	2.5	0	--	2.2	8.1	16.5	22.5	0	0	2.9	0.4
(c) MBC-SO_4 + Nufilm	0	0.3	0	--	3.7	3.9	9.6	21.5	0	0	0	4.1

[1] Sprayed branch represents retention and persistence.

[2] Unsprayed branch reflects true uptake and translocation.

TABLE X

Effect of Repeated Foliar Application of MBC-SO_4 on Uptake, Movement and Persistence on Elm Trees with and without Adjuvants, After Leaf Flush in 1972

	Diameter of Zone of Inhibition (mm)											
	Leaf				Bark				Wood			
	Follow-up Sampling - Days											
	15	30	60	90	15	30	60	90	15	30	60	90
Sprayed (Open Branch)[1]												
(a) MBC-SO_4 Alone	19.1	7.3	16.9	5.5	25.2	34.1	23.9	23.9	3.0	0	2.2	2.8
(b) MBC-SO_4 + Biofilm	33.3	17.3	17.5	4.7	24.9	32	25.6	21.2	9.4	11.2	1.0	4.7
(c) MBC-SO_4 + Nufilm	29.6	19.9	18.2	14.2	28.8	33.6	25.7	31.1	10.9	9.5	3.8	8.7
Unsprayed (Bagged Branch)[2]												
(a) MBC-SO_4 Alone	0	0.8	1.7	--	9.8	20.3	8.2	18.3	0	0	0	0
(b) MBC-SO_4 + Biofilm	1.3	4.4	0	0	11.0	23.8	3.7	20.6	0.2	4.9	0	2.6
(c) MBC-SO_4 + Nufilm	0.5	2.6	0	--	7.7	28.3	14.7	25.4	0	3.8	2.5	2.2

[1] Sprayed branch represents retention and persistence.

[2] Unsprayed branch reflects true uptake and translocation.

Summarizing Tables VI, VII, VIII & IX, it is evident that there is little uptake and movement into the bagged (untreated) areas. Neither Biofilm nor Nufilm seem to enhance penetration to a great extent. The action of Biofilm even though small is consistent while the response of Nufilm is erratic. The persistence of the spray on the foliage and bark is of interest; there is no residue deposited on the leaves in trees sprayed before the leaf-flush whereas trees sprayed during and after the leaf-flush retained significant quantities on their leaves. The maximum amount of deposits were found on the bark tissues. Penetration into the wood was variable and usually increased 90 days after spraying. Greater deposits were seen on the foliar parts of trees treated after the leaf-flush and thus the weather conditions (frequency of rainfall) during the post flush period did not completely wash away the residues from the foliar surfaces. Considerable quantities of fungitoxicants were found on flowers and germinating seeds as a result of pre-flush sprays on the mature trees. Repeated spraying with the same dosage during the post leaf-flush stage did increase the level of residues on the tree and it appeared from Table IX that the uptake and translocation were also accelerated. After the double spray both Biofilm and Nufilm had a positive effect on the retention and absorption of benomyl into the tree parts. Therefore, during the 1973 field trials, only Biofilm was incorporated into the MBC-SO_4 formulations and again a double spraying with this formulation during the post-flush stage produced excellent penetration and translocation into the inner bark and wood (Table XI). Not only did MBC-SO_4 penetrate the wood but it seemed transported into the core samples of heartwood.

TABLE XI

Persistence of MBC-SO_4 on Elm Tree

Following Two Foliar Applications During July 1973

Diameter of Zone of Inhibition (mm)

Treatment	Leaf				Bark				Wood			
	15	30	60	90	15	30	60	90	15	30	60	90Days
MBC-SO_4 + Biofilm	10.2	6.8	10.5	2.8	22.2	20.8	23.7	20.2	2.8	3.7	5.1	5.6

DISCUSSION

Evidence collected from the laboratory and field experiments suggest that benomyl and MBC-SO_4 penetrate into the foliar, bark and wood tissues but in a very slow manner. Some adjuvants seem to accelerate the penetration and some do not exert any effect. Age of the leaf is not a critical factor in the uptake process even though younger foliage under laboratory conditions absorb more than the old leaves. But this situation becomes insignificant in the field because mature trees retain and absorb MBC-SO_4 foliarly more or less to the same extent before, during and after the full leaf-flush. However, what seems more important is the frequency of applications. Certainly, the persistence, uptake and translocation of MBC-SO_4 has been somewhat greater when the trees were sprayed twice after the leaf-flush in both years 1972 and 1973. This suggests that either the dosage applied in these experiments had been less or that a certain degree of saturation of foliar and bark tissues with MBC-SO_4 and benomyl is a prerequisite before significant uptake can take place. Upham & Delp (1973) reported about 20 fold greater penetration of benomyl than of MBC in herbaceous leaves. This may partly explain why MBC-C^{14} did not significantly move out of the foliar surfaces in the laboratory experiment. The autoradiographs do not demonstrate clear movement from the treated areas. On the other hand, there was appreciable penetration and translocation when leaves were treated with benomyl and various adjuvants (Table V). Why there should be a differential response, was not clearly elucidated by these workers, but it seems, on a priori ground, that the butyl carbomyl

moeity of benomyl and the added adjuvants in the 50% WP benomyl formulation were responsible for greater penetrability. This is further borne out by the data in Table V indicating that Biofilm, Nufilm or even Regulaid increased the foliar uptake. Another feature that emerges from these findings is that wettability of the leaf surface and formulation is important for prolonged uptake and translocation because whenever the dried pesticide was rewetted with water on the foliar surface, the penetration was increased. Perhaps this drying out of spray droplets under the field conditions might have contributed to erratic absorption and movement into the trees and repeat applications of MBC-SO_4 or benomyl might have rewetted the dried powder on the foliar and bark surfaces for longer periods. Since it is well known that plant leaves cannot absorb solid powder, the pesticide must be maintained in aqueous state for effective translocation to take place (Prasad, Foy & Crafts 1967).

After the initial absorption, the pattern of translocation appears to be largely apoplastic because the radioactivity seems to preferentially migrate into the regions of high transpiration (margins) of either live leaves or those made of the filter paper. But if some movement takes place downward via the phloem to the untreated part of the tree and roots, the translocation pattern has to be symplastic as well. These findings are somewhat similar to those reported elsewhere (Prasad 1973 and Solel & Edgington 1973) that even though the major part of the benomyl and MBC movement is in the transpiration stream, some fungitoxicity does leak to the phloem and consequently

the transport becomes symplastic. In this connection, it should be remembered that the pathogenesis brought about by the DED, occurs in the shoot and therefore it is imperative that the pesticide must migrate downward into the phloem after foliar sprayings so that applications can be of practical value in combatting the DED. Erratic protection of trees following foliar applications of benomyl might be thus related to its manner of movement into the trees. Usually an apoplastic type of pesticide cannot confer phytoprotection if applied from the foliage. Therefore, the claim of Hart (1972) and Smalley et al (1973) should be treated with caution. The fact that they obtained adequate protection from foliar sprays could be interpreted that appreciable uptake and movement was taking place from the bark and lenticels rather than from the foliar surfaces of elm (Prasad 1973). Therefore, unless bark and twigs are thoroughly sprayed and drenched with benomyl during the spraying operations, the authenticity of foliar treatment in ensuring tree protection is questionable. Because benomyl and MBC can persist on elm trees (bark and twigs) for over a year even after the leaf fall, chances are that these residues would aid in prophylactic protection of the trees (Prasad 1973).

Finally, the question remains whether foliar applications can be employed for curative treatment of elm trees infected with the DED? Scanty and erratic data on the foliar uptake and translocation suggest that there would not be sufficient symplastic movement to ensure eradication of the pathogen from the host. Therefore, the role of foliar application as a mass treatment for large number of elm trees can be considered only for preventative purposes.

According to Smalley et al (1973) elm trees already infected with DED by more than 20% in the crown and those infected through the root grafts cannot be protected by foliar sprays of benomyl. Side effects of benomyl and MBC are known now (Prasad 1974) and even though foliar sprays could eradicate earthworms under the tree, the benefits derived by saving the doomed elms are greater than being concerned about worms whose role beneath an elm tree is uncertain and unknown.

SUMMARY AND CONCLUSIONS

Using laboratory and field experiments, the foliar uptake and translocation of benomyl and MBC-C^{14} was investigated. Penetration and movement of both fungitoxicants is slow from the leaves and most of the translocation takes place in the transpiration stream. However, appreciable retention, uptake and translocation seem to take place through the bark and it is suggested that this method could be exploited for preventive control. A three year field study with foliar and bark spraying of benomyl and MBC-SO_4, indicate that appropriate formulations, time and frequency of spray applications would offer hope for prophylactic protection of otherwise doomed elms.

ACKNOWLEDGEMENTS

The authors thank P. Davis, R.M. Moody, and B. Limmer for technical assistance and Dr. Fettes, Director, Chemical Control Research Institute, for encouragement during the course of investi-

gation. They are indebted to the National Capital Commission (Messrs. McLaren and White) for providing part time student help and elm trees for experimentation. Dr. D.J. Durzan (Ecology Research Institute, C.F.S. Ottawa) very kindly provided the darkroom and Dr. Childs (Canada Dept. of Agriculture, Ottawa) the greenhouse facilities and the authors duly acknowledge their cooperation and assistance. Appreciation is expressed to Great Lakes Forest Research Centre, Sault Ste. Marie (Drs. Kondo, Thomas) and to University of Toronto (Profs. Roy and Jorgensen) for providing MBC-SO_4.

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APPENDIX I

Details of Spray Adjuvants Used in this Study

Common Name	Chemical Name	Source
Biofilm	Alkylarylpolyethoxy ethanol	Colloidal Prod. Corp. Petaluma, California.
Nufilm 17	di-1-p-methene	Miller Chemical & Fertilizer Corp., Hanover, Penn.
Regulaid	Polyoxyethylene polypropoxy-propanol. Alkyl 2-ethoxyethenol	Colloidal Prod. Corp. Petaluma, California.
Carborundum	Silicon carbide	Fisher Scientific Co. Ottawa, Ontario.
Urea	NH_2CONH_2	Fisher Scientific Co. Ottawa, Ontario.
Sugar	Sucrose ($C_{12}H_{22}O_{11}$)	Fisher Scientific Co. Ottawa, Ontario.