TRANSLOCATION OF BENOMYL IN ELM (ULMUS AMERICANA L.)

VIII Prevention of the Dutch Elm Disease, *Ceratocystis ulmi* (Buism), Moreau, in Mature Trees Following Pressurized Trunk Injections.

by

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TABLE OF CONTENTS

RÉSUMÉ

On a obtenu une forte atténuation de la maladie hollandaise de l'orme chez des ormes adultes (d.h.p. 7") par injection dans le tronc de bénomyl soluble (chlorure MBC, 1000 ppm). Tandis que chez 85% des arbres non-traités (témoins) la maladie se développait aprés inoculation de spores de la maladie, chez les arbres traités, les symptômes de la maladie étaient supprimés à 100%. Le coûtdu traitement était d'environ $25 l'arbre, et on propose d'effectuer des recherches plus approfondies pour améliorer cette technique et obtenir une certaine garantie des résultats.

INTRODUCTION

While investigating the rate and pattern of distribution of benomyl (MBC-C^{14} and MBC-Chloride) in seedlings and mature elms it was concluded that after trunk-injection, the material moved upward in the transpiration stream, arrived at the site of pathogenesis and finally accumulated in the leaves (Prasad 1972, Prasad & Travnick 1974). It was also shown that considerable radioactivity and fungitoxicity resided in the xylem and cambium regions of the tree following stem injections of benomyl. Both in juvenile and mature trees, the transport was, on the whole, uniform and apoplastic. However, occasionally the benomyl (Lignasan® and MBC-Chloride) showed erratic distribution across the crotches and new twigs. These studies on the manner and extent of transport were of considerable significance in relating the systemic behaviour of the pesticide in the tree but they could not focus on the degree of protection afforded to individual trees in the event DED infection took place. Thus there was a need to correlate translocation studies with disease protection under field conditions and experiments were, therefore, designed to assess any prevention and cure conferred by the benomyl treatments employing the trunk injection method. This preliminary report describes a one-year field study on the curtailment of disease symptoms in mature elms treated with MBC-Chloride by the trunk injection technique.

MATERIALS AND METHODS

(i) Selection of Trees: Twenty-two elms of uniform size and shape were selected at Deschenes, Quebec and were monitored for the absence of the

DED by the standard bioassay technique. Only healthy and vigorous individuals were tagged for injection and before start of the experiment, the diameter at breast height (d.b.h.), height and general appearance of growth were recorded. The site contained largely silt and loam with some gravel and stones which were subsequently removed so that the injection apparatus mounted on wheels could be transported easily. The trees were distributed in a 25 acre area and were interspersed by diseased and dead elms as well.

(ii) Method of Injection:- The standard procedure of trunk injection as described by Prasad & Travnick (1974) was employed. Briefly, 4-8 holes (3/16") were drilled in each tree and after the removal of wood shavings, high pressure injector screws were fitted into the holes in such a way that they remained in the sapwood-heartwood area. For the sake of efficient uptake, too deep penetration into the heartwood area was avoided and in order to avoid blow-outs, injectors were not attached near cracks and crevices in the bark.

After the injections, the holes were filled with a grafting wax. The control (check) trees were given a similar treatment save that no fungicide was injected.

(iii) Bioassay of Fungicide:- Samples of foliage, twigs and core were periodically removed from the treated and check trees for monitoring the movement of the fungitoxicant and for this the standard bioassay technique employing Penicillium expansum (Prasad & Travnick, 1973)was used. Two trees were felled after the injection and the systemic activity of MBC-chloride from sap and heartwood was monitored.

(iv) Determination of Disease Protection:- Following injections, the check and treated trees were allowed to mobilize the fungicides into the tree system for 2 weeks after which inoculations were made in each tree by the procedure of Santamour (1974). A virulent strain of the DED (Soo-strain) kindly supplied by Dr. Edward Kondo of Sault Ste. Marie Laboratory was first cultured and propogated in juvenile trees. Re-isolations were made from these diseased trees and only such a virulent strain culture was used for induction of disease in the check and treated trees. Inoculations were made in 3 top branches per tree by introducing spore suspension (0.5 ml) into the xylem sap stream by means of two knife wounds. These infected wounds were then covered with an adhesive tape to provide high humidity for germination of the conidia. This method was very effective as over 80% of the untreated trees showed severity of disease symptoms (flagging and wilting). Progression of the disease development in terms of percentage damage to the crown was further monitored over a period of three months.

Estimation of the Cost of Treatment

A record of all expenditures in terms of cost of material, labour, equipment and operation was maintained and from this an approximate cost of the treatment per tree was computed.

RESULTS

(i) Pattern of Distribution

Before assessing the degree of protection offered by the injection treatments, samples of twig, leaf and cores were collected from all trees and subjected to bioassay so that a correlation between the

dosage injected and the biological response could be established. Table I shows the description of tree samples as well as the dosage of MBC-chloride injected into each tree: as expected there is variability among the height and d.b.h.

Samples bioassayed 15, 30 and 60 days after the treatment (Table II) indicate that MBC-Chloride moved from the site of injection to upper part of the tree. Two opposite branches were sampled and usually the coverage was good, in as much as 70% of the twigs were completely covered.

TABLE I

Size and Injected Dosage of MBC-Chloride in Test Trees

Treated Tree No.	DBH(inches)	Height (feet)	Volume Injected (litres)	Dosage (gms)
42	8.0	33	7.5	7.5
43	4.5	16	4.0	4.0
44	7.5	23	5.5	5.5
45	6.1	30	5.0	5.0
46	7.2	31	5.0	5.0
47	7.5	27	4.5	4.5
48	4.5	18	4.0	4.0
49	7.5	25	5.5	5.5
50	4.5	19	4.0	4.0
51	5.1	22	5.0	5.0
Check Tree No.				
81	6.5	27	0	0
82	5.1	21	0	0
83	3.5	16	0	0
84	4.5	20	0	0
85	4.0	15	0	0
86	4.5	15	0	0
87	3.5	18	0	0
88	4.0	18	0	0
89	4.2	16	0	0
90	5.1	20	0	0

TABLE II

Distribution of MBC-Chloride in Elm Trees Following Trunk Injection after 15, 30 and 60 days[1)]

Tree Part	Zone of Inhibition (mm)		
	15	30	60 days
Wood	3.25	1.93	1.31
Leaf	4.41	3.35	4.90

1) Two opposite branches bioassayed on a tree.

There is a tendency for the fungitoxicant to concentrate in the leaves with the passage of time, and this confirms previous findings (Prasad & Travnick 1974) that it preferentially moves into the transpiration stream. As the fungitoxicant content in the wood declines, the amount in leaves increases until 60 days at least, thereafter the toxicant would dissipate into the soil through leaf fall.

Disease Protection

Because of a good coverage of MBC-Chloride in the injected trees, the degree of prevention of the DED was assessed in twenty trees. Ten check trees and ten injected trees were inoculated with the soo-strain and the progression of the disease symptoms was monitored after 30, 60 and 90 days, both in the local branch infected as well as in the rest of the crown. The disease began to appear in two weeks and after 3 months over 80% of the control trees were heavily diseased or dead whereas none of the treated trees showed any of the disease symptoms (Table III).

TABLE III

Observation of Disease Symptoms in Elm Trees After Inoculation with DED and Trunk Injection of MBC-Chloride[1)]

Tree No.	Disease Symptoms[2] in inoculated branches		Disease Symptoms in the Crown
	Branch I	Branch II	% Damage
Check 81	+	+	33
82	-	-	0
83	+	+	44
84	+	+	100
85	+	+	100
86	+	+	60
87	+	-	35
88	+	+	80
89	+	+	45
90	+	+	85
Treated			
42	-	-	0
43	-	-	0
44	-	-	0
45	-	-	0
46	-	-	0
47	-	-	0
48	-	-	0
49	-	-	0
50	-	-	0
51	-	-	0

1) Trees injected - 28th June
DED inoculated - 17th July

2) Disease developed, + +
No disease developed, - -

The inoculated branches of the check trees showed flagging and wilting first and subsequently the infection spread to rest of the crown. On the other hand, since there were no such symptoms in the injected trees the treatment must have had a pronounced effect on the curtailment of the disease. Two of the inoculated check trees did not express any visible symptoms of the disease and remained healthy, green and vigorous like the treated trees for at least 30 days. Thereafter disease surveillance was carried out at 60 and 90 days and even after the three months, the pattern of disease protection remained similar.

(a)

(b)

Fig. 1 Protection of Elm Trees by Trunk Injection of MBC-Chloride

(a) Check-inoculated (b) Treated - inoculated

Note the severity of infection in the check trees. Ribbons indicate the inoculated branches.

None of the injected trees contracted the disease whereas 85% of the control-inoculated had symptoms of severe infection. Again, at least one tree among the inoculated controls remained unaffected. This is not surprising since it is known that even under natural conditions, all infected trees do not exhibit obvious disease symptoms. During the experiments, photographs of protected, diseased and dying trees were taken and Fig. 1 (a) and (b) shows the striking difference in MBC-Chloride treated trees against the DED. Whether the same pattern of protection would hold on a large number of trees, remains to be ascertained but killing of too many trees by artificial inoculation is not permitted by the National Capital Commission, that maintain elm trees at the Deschenes site.

Cost of the Treatment

The following figures were obtained from the records and as may be seen from Table IV, the approximate cost for treating an average tree is $23.75.

TABLE IV

Cost Analysis of Treating 10 Elm Trees with MBC-Chloride

Cost Parameters	Quantity Used	Dollar Value	Cost/Tree (Dollars)
Materials	200 gms	17.50	
Labour	15 man hours	120.00	23.75
Machinery Rental	15 hours	100.00	
	Total	237.50	

Apparently, the labour and machinery rental costs accounted for most of the expense. A considerable reduction in man-hours could be effected by using power drills. Similarly the motor-pump could be replaced by gas (nitrogen) cylinders capable of generating adequate pressure to force solution into the trees. Thus it is possible to reduce these costs to a level which would be attractive to many for the protection of valuable trees.

DISCUSSION

The evidence presented by the experimental data clearly demonstrates that the trunk-injection method of administration of benomyl (MBC-Chloride) to healthy and mature trees is effective in suppressing the disease development. For example, none of the treated trees showed any signs of the disease following inoculation with the DED spores whereas the check trees succumbed to the disease after 2 weeks. The disease symptoms were most pronounced in the branches where inoculum was introduced. Subsequently the infection spread to other parts of the tree and after 3 months many of the inoculated check trees were completely dead or dying. However, in 2 check trees the progression of the infection was anamolous. One tree did not express the disease symptoms and the other showed only localized infection. Such a situation has been reported before (Kondo 1972, Sinclair & Brenner 1974) where all trees do not take complete infection. Under natural conditions many elm trees have been observed to escape the ravages of the DED even if the disease is prevalent in the vicinity. Perhaps there are differences in the ecotypes (physiology) of some trees that confer the disease resistance. That the trunk injection of fungicides can protect many horticultural and shade trees was recently

reported by several investigators. Shabi et al (1974) and Pinkas et al (1973) have infilterated pear trees with benzimidazole fungicides and Nyland (1972) and Hunt et al (1974) have injected oxytetracyclines and mycostatin into peach and palm trees and they have reported a high degree of remission of the vascular diseases. Similarly Jones et al (1973) have obtained very effective to satisfactory control of the oak wilt following pressure injection of solubilized benomyl into the trunks. The Dutch elm disease has also been successfully suppressed and cured in many trees in several parts of the United States with a pressurized trunk injection technique involving the use of solubilized benomyl i.e. MBC-Chloride (Gregory et al 1973, Van Alfen and Walton, 1974).

How long the prophylactic or the curative effects last is not yet fully ascertained but according to Kondo (1973) treated elms with MBC-Phosphate can remain uninfected for at least 2 years. This is because solubilized benomyl (MBC-Chloride or Phosphate) persists in the tree system for about 2 years (Kondo 1973, Prasad & Travnick 1973). If the protection could be prolonged beyond 2 years it would allow municipalities, governments and private owners to afford treatment of elms at a reasonable cost. According to our data, the cost of treatment for a small tree (d.b.h.-7") is $23.75. Assuming the street tree is four times larger, the cost price ($95.00) would still be within the bounds of the owners of the tree and thus the treatment can be justified. In some trees of landmark and historical value (Prasad 1974) even higher costs of treatment can be justified because there is no alternate effective way of protecting these old and highly prized trees. Clearly more research is needed to ensure a guarantee of the treatment as well as improvement

in the technique. Perhaps a mass treatment involving hundreds of trees should be carried out as the first step to improve techniques and reduce cost per treatment.

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SUMMARY AND CONCLUSION

A high degree of remission of the Dutch elm disease was obtained in mature elm trees (d.b.h.-7") following trunk-injection with solubilized benomyl (MBC-Chloride @ 1000 ppm). Whereas the untreated (check) trees showed 85% disease development following inoculation with the DED spores, the treated trees showed 100% suppression of the disease symptoms. The cost of treatment was about $25.00 per tree and it is suggested that more research should be carried out to improve the potentials of this technique for the use of other pesticides, nutrients and hormones in large trees.

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