

PETE VERMEULEN: At what point did you differentiate between basal and second cuttings?

G. TEHRANI: The basal cutting was taken directly from the main stem and the second was taken above this.

RALPH SHUGERT: Have you tried taking cuttings and sticking them directly in the field row?

G. TEHRANI: No, I haven't because of the wet soil conditions at the time we were taking the cuttings but I personally don't think they would root.

CHARLEY HESS: Thank you, Dr. Tehrani. I'd like now to introduce the moderator for this morning's session, Mr. Bill Flemer III, President of the American Nurserymen's Association.

MODERATOR FLEMER: Our next speaker of the morning is Dr. J. N. Cummins of Cornell University who will speak on, "Increased production of rooted *Prunus* cuttings with a pre-planting soak of Benomyl."

INCREASED PRODUCTION OF ROOTED PRUNUS BESSEYI BAILEY SOFTWOOD CUTTINGS WITH PREPLANTING SOAK IN BENOMYL^{1,2}

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INTRODUCTION

Decreasing damage caused by fungi and other microorganisms is among the principal means of improving methods of propagating woody plants by cuttings. Softwood cuttings under mist or in propagation boxes provide particularly favorable conditions for the growth and spread of fungus diseases. To produce important economic benefits, a fungicidal treatment should meet three conditions: (1) the treatment must appreciably reduce the incidence of disease; (2) the treatment must not be harmful to the plant material being propagated; and (3) the treatment must not interfere with the rooting/establishment process.

Under intermittent mist, cuttings of many species of *Prunus* are susceptible to *Botrytis cinerea* and a number of other pathogens. Selection and utilization of asexually propagated rootstocks for peach, plum, apricot and sweet and sour cherries depends in part on development of disease control systems.

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²Benomyl is the coined common name for 1-(butylcarbamoyl)-2-benzimidazole carbamic acid, methyl ester (duPont fungicide 1991 or Benlate). Appreciation is accorded the E.I. duPont de Nemours & Co. for providing the benomyl used in this work. The technical assistance of Miss B. Oakes is gratefully acknowledged.

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Three types of damage by fungi may be distinguished on many lots of *Prunus* cuttings under mist:

(1) Necrosis of the bark below the surface of the rooting medium, typically observed by the 3rd or 4th day under mist; this inevitably culminates in death of the cutting;

(2) Yellowing and/or browning of the leaves, with or without lesions, becomes discernible by the 3rd to 7th day under mist and is followed by leaf abscission during the 2nd or 3rd week.

(3) Necrotic patches on the lower stem are associated with root emergence, especially if a vertical "comb" of roots erupts.

A number of reports (cited below) suggested that benomyl might control a considerable range of fungi in the mist bed. The work reported here was intended to examine the effects of benomyl on rooting and establishment of *Prunus* cuttings under mist, using *P. besseyi* as the test subject.

REVIEW OF LITERATURE

Comparing intermittent mist with closed propagation boxes for rooting apple softwood cuttings, Singh *et al.* (8) concluded that rooting results were primarily dependent on survival, with either dessication or fungus infection capable of eliminating a planting. Many fungicides have yielded promising results when used on cuttings, and some commercial root-inducing preparations contain fungicides as well as regulators.

Doran (1) obtained promising results when he treated cuttings of *Magnolia virginiana* and other woody species with Phygon-XL. Klaus (5) found treatment with zineb dust to be very helpful with cuttings of begonia, chrysanthemum, pelargonium, and salvia. Tinley (10) working with several fungicides, obtained good establishment of *Hevea brasiliensis* using ferbam, Manzate, and Parzate. Tinley, as well as van der Kerk *et al.* (11) suggested the possibility that these related dithiocarbamate compounds might be growth-regulating substances as well as fungicides. Van Doesburg (12) found beneficial effects from captan dust on cuttings of several species of ornamentals. Smith and Evans (9) reported dichlofluanid the most promising of several chemicals for controlling grey mold on chrysanthemum cuttings.

Benomyl has been shown to be a most effective fungicide, controlling a wide range of fungi on many kinds of host plants. Erwin, *et al.* (3) demonstrated that benomyl applied as a soil drench controlled *Verticillium* wilt of cotton by systemic action. Engelhard (2) found benomyl sprays to have unusually long residual effectiveness against black spot of rose; he considered that this long residual activity might be indicative of systemic activity. Schroeder and Providenti (7) found that both soil drenches and pre-planting seed treatment with benomyl gave systemic control of powdery mildew of squash and cucumber.

Gilpatrick (4) reported that soil drenches effectively controlled powdery mildew of both apple and cherry but not apple scab and that the systemic activity is greater by root absorption than by foliar. He found no phytotoxicity at rates up to 100 mg/kg of dry soil. Benomyl was shown by Manning and Glickman (6) to give commercial control of *Botrytis cinerea* on geranium, whether applied as whole cutting dips, as sprays, or as drenches of the medium.

MATERIALS AND METHODS

We selected *Prunus besseyi* as a test subject because our past experience had indicated that the species had both a high capacity for rooting and an extreme susceptibility to damage by disease under mist.

Uniform, subterminal 3-node, 2-leaf softwood cuttings of several unnamed clones were taken from vigorous 10-year-old mother plants. After fungicide treatment, the fresh cuttings were air-dried for about 10 minutes; just before planting, the basal centimeter of each cutting was dipped for about 5 seconds into 1000 ppm IBA (3-indolebutyric acid) in 40% ethanol. Each flat was divided into four rectangular sections, each section receiving a different randomly distributed treatment (Fig. 1). In a shaded greenhouse bed, mist was applied intermittently for 2 seconds every minute, 14 hours a day, throughout the June 16 — August 4, 1969 experimental period.

Sterile media were obtained by autoclaving flats containing the chosen substrate. "Contaminated media" had been used in 1968 for mist propagation of apple, cherry, and geranium; disease had been a serious problem, and many particles of decayed plant tissue were dispersed throughout the substrate.

Evaluations were made 22 days after cuttings were treated, unless otherwise noted. Records taken included:

Survival (a cutting was considered as "surviving" only if the stem, callus tissue, and roots were free of superficially discernible infection),

Rooting (a cutting with any emergent root was considered "rooted". without regard to root size, number of roots, or survival status of the cutting),

Leaf retention (a leaf adhering to the stem was considered to be "retained" if its color was approximately normal and no decay was visible on either blade or petiole).

Trial 1: Benomyl and contaminated medium. — Fresh cuttings of the clones 'PB-10', '-17', and '-19' were soaked for 5 minutes in benomyl at 0, 330, 1000, or 3000 ppm concentration¹. After IBA treatment, cuttings were stuck into flats containing a contaminated perlite/sand medium; each of the 12 plots contained 23 cuttings. Cuttings of 'PB-36' were similarly treated but were planted in sterile perlite-vermiculite substrate, 14 cuttings per plot.

¹All concentrations are expressed in terms of active chemical.

Trial 2: Whole cutting soaks in captan or benomyl. Cuttings of the clone 'PB-17' were soaked for 5 minutes in 1000, 3000, or 6000 ppm benomyl or captan, or in water. Cuttings were then treated with IBA and stuck in a heavily contaminated peat-perlite mixture. The experiment included two replicates, with 25 cuttings per treatment.

Trial 3: Interaction of whole cutting soaks and substrate drenches Fifteen cuttings of each of the clones 'PB-8', '-15', and '-17' were soaked for 5 minutes in 1000, 3000, or 6000 ppm benomyl or in water. After drying, the cuttings were stuck into a heavily contaminated peat-perlite medium which had been drenched with 5 grams per flat of either captan or benomyl.

Trial 4: Effect of whole cutting soaking time. 'PB-1', which had earlier showed an unusual susceptibility to fungus attack, was used to examine the effect of the period of soaking time on protection. Cuttings were dipped momentarily, soaked for 5 minutes, or soaked for 1 hour in 1000 ppm benomyl and were then immediately rinsed in tap water. After IBA treatment, they were planted in a contaminated peat/perlite medium. Four replications, 25 cuttings per plot were used.

Trial 5: Duration of residual effectiveness The previous cuttage trials suggested that effectiveness was of short duration. Accordingly, we planted in contaminated media 100 cuttings soaked for 5 minutes in 1000 ppm benomyl and the same number of untreated cuttings. Weekly thereafter we evaluated these cuttings for leaf loss, petiole infection, death of cuttings, and root establishment.

Statistical treatments. Because so few proportionate data were at percentage extremes, we did not resort to arcsin transformation but rather performed analyses of variance directly on the data. Duncan's multiple range test was applied when significant differences were indicated.

RESULTS AND DISCUSSION

Striking effects of benomyl soaks on leaf retention, cutting survival, and rooting are shown in Tables 1 and 2 and Figs. 1 and 2. With cuttings in contaminated media (Table 1), the

Table 1. Effects of different concentrations of benomyl on cuttings of *Prunus besseyi* planted in contaminated rooting media.

Benomyl concentration (ppm)	No of healthy leaves per cutting (originally 2) ¹	Percentage of cuttings surviving after 22 days ¹	Percentage of cuttings rooted ¹
0	1.1a	50.9a	66.4a
330	1.4b	55.2a	74.2ab
1000	1.7c	80.2b	85.3bc
3000	1.8d	91.4b	98.1c

¹In a given column, means followed by different letters are significantly different at the 5% level, as indicated by Duncan's multiple range test.

Table 2. Effects of benomyl concentration on cuttings of *P. besseyi* 'PB-10' planted in sterile perlite-vermiculite substrate.

Benomyl concentration (ppm)	No. of healthy leaves per cutting (originally 2) ¹	Percentage of cuttings surviving after 22 days ¹	Percentage of cuttings rooted ¹
0	0.93a	71.4a	49.9a
330	1.79b	85.7ab	57.0ab
1000	1.71b	92.9b	71.5c
3000	1.79b	92.9b	64.4bc

¹In a given column, means followed by different letters are significantly different at the 5% level.

activity of the fungicide increased with concentration throughout the range used in this trial. In a sterile medium (Table 2) no concentration effect was observed, but the differences between treated and untreated cuttings was quite marked. There was no indication of phytotoxicity on any plot. Among the cuttings treated with 3000 ppm benomyl, almost all leaves appeared healthy and functional after 3 weeks under intermittent mist; more than 90% were free of visible symptoms of disease.

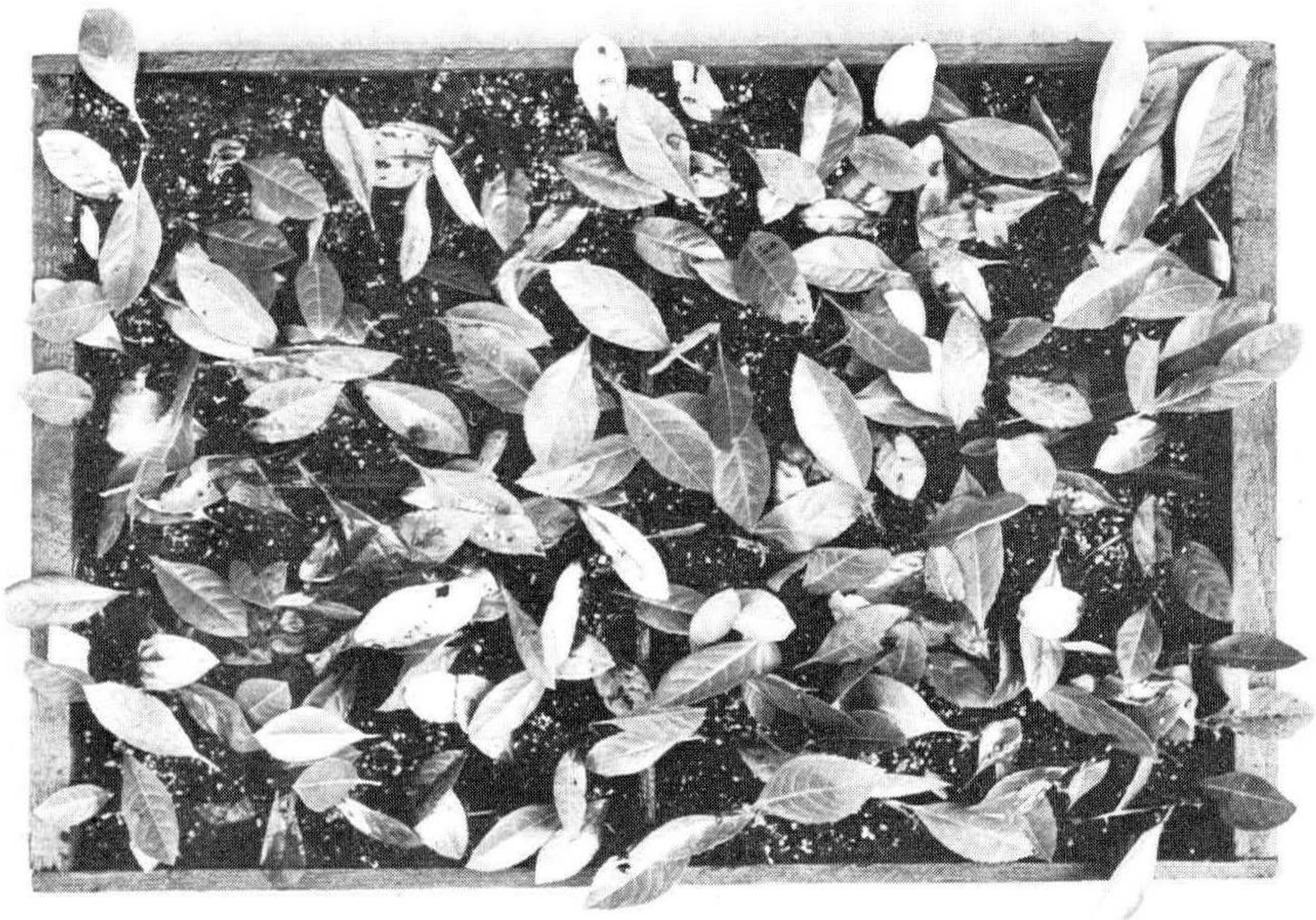


Figure 1. Flat of softwood cuttings of *Prunus besseyi* cv. 'PB-1' after 22 days under intermittent mist. The flat is divided into 4 rectangular quarters, each a treatment plot. Heavy leaf loss and large numbers of lesions on the leaves are shown on the check (lower left) and 330 ppm benomyl (upper left). Foliage in the 3000 ppm benomyl plot (lower right) is conspicuously healthy. 'PB-1' is highly susceptible to *Botrytis*.

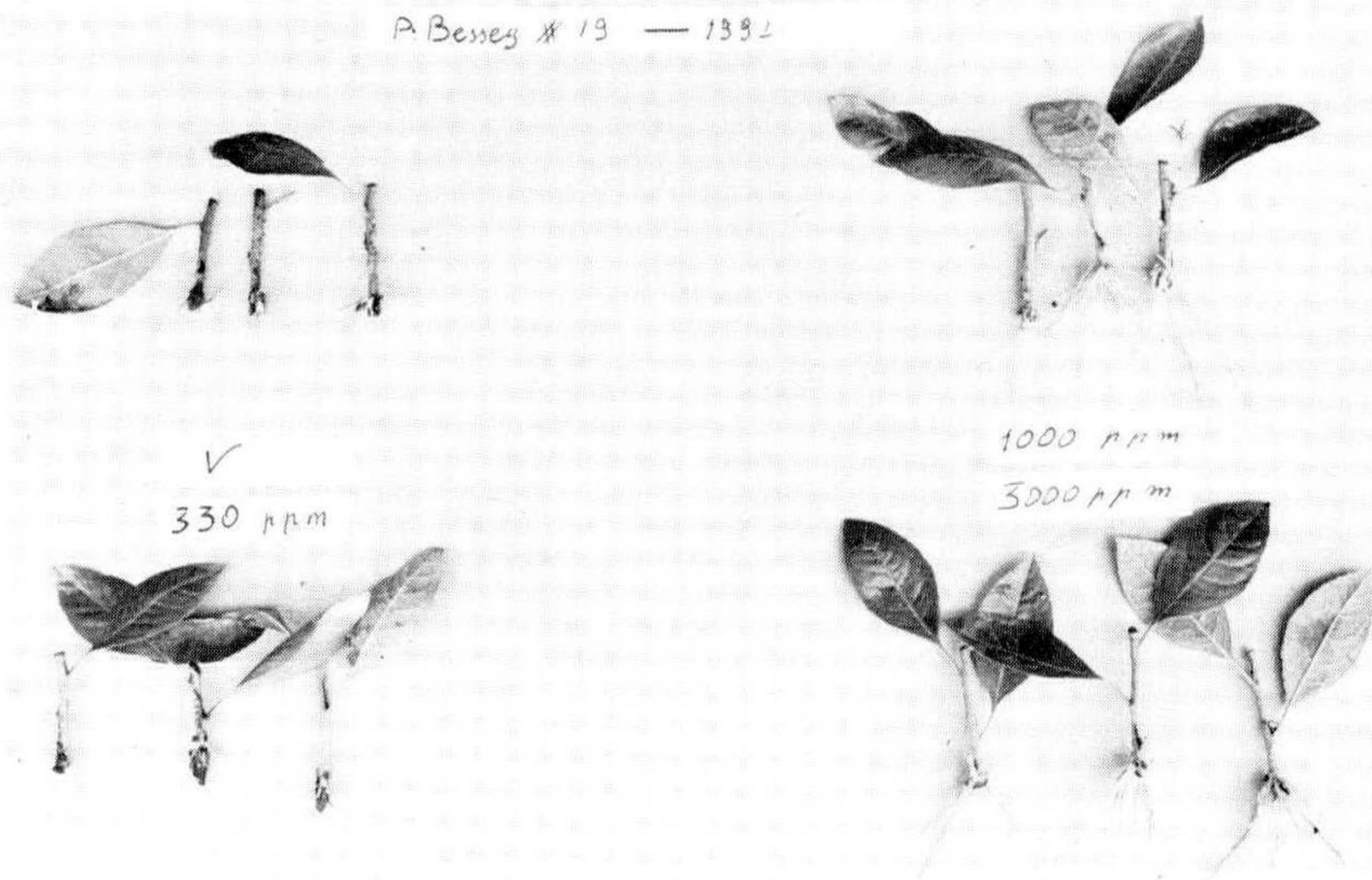


Figure 2. Typical cuttings of 'PB-19' rooted after 22 days under mist. Vigorous root growth, good leaf retention, and healthy stems of the 3000 ppm benomyl group (*lower right*) contrast with the less healthy appearance of the check (*upper left*) and 330 ppm benomyl samples (*lower left*).

Initial infection of cuttings was usually observed as a dark, slimy region at the extreme base of the cutting. With contaminated media, such infections were often observed as early as 3 days after planting. This initial infection usually took place at the basal excision wound. Later infections originated at the base or through leaf scars, through infected buds, or at wounds caused by eruption of roots.

Our observations indicated that the infection came from two sources: the medium itself, and the stock plants. In sterile media (Table 2), even the lowest concentration of benomyl appeared to be effective in disease control; this was probably due primarily to reduction of the inoculum originally on the cuttings. In contaminated media (Table 1), the concentration effect is a response to continuing attack by pathogens from the medium. Supposedly this concentration effect is related to variations in residual surface protection and/or to different amounts of benomyl initially absorbed by the cuttings.

Rooting was increased by fungicide treatments, but no conclusive evidence was obtained to distinguish whether this was due to a direct stimulation of root formation, to a higher rate of cutting survival during the rooting period, or to the number and quality of leaves retained. The data in Table 2 do not support the last hypothesis, although the data of Table 1 indicate that leaf retention is an important factor.

Captan and benomyl soaks compared. (Trial 2). On contaminated media, a five-minute soak in either captan or benomyl materially improved leaf retention, cutting survival, and rooting, as shown by the data of Table 3. There was no concentration effect with captan; even the lowest (1000 ppm) captan soak compared well with the two lower benomyl concentrations. The 6000 ppm benomyl soak was superior to any other treatment in all three criteria; no phytotoxicity was observed even at this extremely high concentration¹. We infer that at this highest concentration enough benomyl was absorbed and/or adsorbed by the cutting to provide long residual protection. It should be noted that the differences in fungicide costs are negligible, since 5 gallons of suspension are adequate for treating many thousands of cuttings.

Interactions between drenches and whole cutting soaks (Trial 3). When used as substrate drenches at the 5 g per flat rate, captan appeared to be slightly superior to benomyl, as shown in Table 4. That both drenches were residually effec-

¹6000 ppm benomyl is equivalent to 10 pounds of 50% wettable powder per 100 gallons of water.

Table 3. Comparison among 5 minute soakings in captan and benomyl (Trial 2). Under mist 21 days. Two replications using clone 'PB-17' only; 25 cuttings.

Fungicide and concentration (ppm)	No. of leaves retained per cutting (originally 2) ¹	Percentage of cuttings surviving ¹	Percentage of cuttings rooted ¹
Control	0.16a	28a	30a
Captan 1000	0.46b	72cde	50ab
Captan 3000	0.48b	60cd	36a
Captan 6000	0.42b	58bcd	48ab
Benomyl 1000	0.38b	36ab	44ab
Benomyl 3000	0.44b	52bc	60ab
Benomyl 6000	0.62c	84e	76b

¹In a given column, means followed by different letters are significantly different at the 5% level.

Table 4. Interactions between substrate drenches using 5 g of benomyl or captan per flat and soaking cuttings in benomyl. Three clones ('PB-8', - '15', and -'17'); 45 cuttings total. Contaminated media.

Concentration of benomyl used for 5 minute soak (ppm)	Number of functional leaves retained per cutting (of 2)		Percentage of cuttings surviving ¹		Percentage of cuttings rooted ¹	
	Captan ¹	Benomyl ¹	Captan ¹	Benomyl ¹	Captan	Benomyl
Control	0.72ab	0.63a	82.2ab	64.6a	75.6	68.9
1000	1.44c	1.11bc	88.9b	91.1b	77.8	68.9
3000	1.54c	1.32c	100.0b	91.1b	77.8	80.0
6000	1.28bc	1.50c	87.8b	91.1b	75.6	77.8

¹In a given column, means followed by different letters are significantly different at the 5% level.

Table 5. Effects of soaking period in 1000 ppm benomyl on leaf retention, cutting survival, and rooting of 'PB-1' in contaminated media.

Period of soaking in 1000 ppm benomyl	No. of leaves retained per cutting (originally 2) ¹	Percentage of cuttings surviving ¹	Percentage of cuttings rooted ¹
0	0.22a	38a	50a
Quick dip	1.10b	78b	78b
5 minutes	1.24bc	66b	76b
1 hour	1.44c	80b	84b

¹In a given column, means followed by different letters are significantly different at the 5% level.

Table 6. Comparison of residual effects 1, 2, and 3 weeks after 5 minute pre-planting soak in 1000 ppm benomyl. Cuttings set into contaminated media.

Week	Leaves retained per cutting		Petioles infected		Cuttings surviving (percentage)		Cuttings rooted (percentage)	
	Check ¹	Benomyl ¹	Check	Benomyl	Check ¹	Benomyl ¹	Check ¹	Benomyl ¹
1	1.56cd	1.88d	0.20	0.00	58b	90c	6a	10a
2	1.04bc	1.52cd	0.26	0.10	46b	84c	30b	38b
3	0.14a	0.46ab	0.14	0.44	20a	52b	40b	56c

¹In a given column, means followed by different letters are significantly different at the 5% level.

tive on the media, rather than within the cuttings, is shown by the difference in leaf retention of cuttings soaked in benomyl and those not soaked in the fungicide.

With quick-rooting species such as *P. besseyi*, loss of leaves may have little direct influence on rooting. For slow-rooting plants, such as *P. domesica* cv. Brompton, retention of leaves appears almost essential for rooting.

Period of soak (Trial 4). A momentary dip in 1000 ppm benomyl gave surprisingly effective disease control (Table 5). Since these cuttings were rinsed immediately after treatment, a very rapid initial uptake and/or strong adsorption is indicated.

Residual effectiveness (Trial 5). The dramatic effect of treatment with benomyl in keeping the cutting healthy through the first critical period of rooting is shown in Table 6. In effect, the data indicate two waves of infection: the first occurs immediately after planting, the second some 10 to 14 days later. The 5 minute soak in 1000 ppm benomyl is effective for the first infection period, but its efficiency has been lost before the onset of the second wave of infection. Our observations suggested that the first infection probably came from inoculum on the mother trees and from the contaminated substrate, with entry being through excision wounds. For the second infection wave, 3 possibilities are suggested: (1) new production of inoculum, with subsequently high infection potential; (2) different parasites involved, one active immediately, the

second, later, which may be resistant to benomyl; (3) development of new avenues of infection into the stem tissue, principally by eruption of roots and production of unuberized callus at the basal cut, on leaf scars, and at lenticels.

CONCLUSIONS AND SUMMARY

The efficacy of benomyl soaks of *Prunus besseyi* softwood cuttings before setting under intermittent mist was clearly demonstrated. No phytotoxicity was observed at concentrations of 1000, 3000, or 6000 ppm actual benomyl. The improved quality of rooted cuttings which had been pre-soaked in benomyl (Fig. 2) should result in improved performance of the plants when transplanted to the nursery. The pre-planting soak is suggested for commercial trial for propagation of *Prunus* and other genera as soon as label clearance is obtained. If the propagator is using contaminated media, drenching this substrate with captan before planting cuttings may be expected to improve disease control.

None of our observations suggest that benomyl in significant quantities is absorbed by the unrooted cutting from the substrate. When benomyl is applied in a whole cutting soak, its initial action may be to destroy the pathogen on or within the host plant. Factors influencing the residual effectiveness of benomyl include initial concentration, use of wetting agent, period of soaking, amount and nature of pathogen in the rooting substrate, inherent susceptibility of the cutting, and possibly extent of leaching of benomyl by the mist. Undoubtedly there are differences in rates of uptake of benomyl among various *Prunus* species and probably among clones within the same species. Although the effect may be expressed over a long period of time, the major action of benomyl appears to be in maintaining good plant health during the first 10 days in the propagation bench.

LITERATURE CITED

1. Doran, W. L. 1952. Effects of treating cuttings of woody plants with both a root inducing substance and a fungicide. *Proc. Amer. Soc. Hort. Sci.* 60: 487-491.
2. Engelhard, A. W. 1969. Preventive and residual fungicidal activity of three benzimidazole compounds and zinc ion + maneb against *Diplocarpon rosae* on two rose cultivars. *Pl. Dis. Rep.* 53 (7) :537-540.
3. Erwin, D. C., H. Mee, and J. J. Sims 1968. The systemic effect of 1-butylcarbamoyl)-2-benzimidazole carbaamic acid, methyl ester, on *Verticillium* wilt of cotton. *Phytopathology* 58:528-529.
4. Gilpatrick, J. D. 1969. Systemic activity of benzimidazoles as soil drenches against powdery mildew of apple and cherry. *Pl. Dis. Rep.* 53 (9) :721-725.
5. Klaus, H. 1957. Zineb zur Saatgutbeizung und Stecklingsvermehrung. *Gartenwelt* 57:110-111.
6. Manning, W. J., and M. Glickman. 1969. Effectiveness of several systemic and non-systemic fungicides in the prevention of *Botrytis* blight of geranium cuttings and stock plants. *Pl. Dis. Rep.* 53 (6) :412-415.
7. Schroeder, W. T., and R. Provvidenti. 1968. Systemic control of powdery mildew on cucurbits with Fungicide 1991 applied as soil drenches and seed treatments. *Pl. Dis. Rep.* 52 (8) :630-632.
8. Singh, S. M., R. J. Garner, and E. S. J. Hatcher. 1957. The influence of source and environment on the performance of apple leaf-bud cuttings. *J. Hort. Sci.* 32 (4) :284-293.

9. Smith, P., and S. G. Evans. 1967. The chemical control of grey mould, *Botrytis cinerea* Pers. ex Fr., in mist-propagated chrysanthemum cuttings. *Pl. Path.* 16:157-159.
10. Tinley, G. H. 1961. Effect of ferric dimethyldithiocarbamate on the rooting of cuttings of *Hevea brasiliensis*. *Nature* 191 (4794): 1217-1218.
11. Van der Kerk, G. J. M., M. H. van Raalte, A. Kaars Sijpesteijn, and R. van der Veen. 1955. A new type of plant growth-regulating substances. *Nature* 176:308.
12. Van Doesburg, J. 1962. Use of fungicides with vegetative propagation. *Proc. XVIth Int. Hort. Congr., Brussels.* I:365-371.

FRED SERBIN: Do you know if the solvent for the benomyl is DMSO (*dimethylsulfoxide*)? If it is, the human toxic factor becomes important.

JIM CUMMINS: I can't speak with authority on this but I don't believe it is.

FRED SERBIN: Did you take any special precautions in handling this material?

JIM CUMMINS: No, we did not. This material has been worked with for 4 years now and there is no report of the kind of difficulty you suggest. I might add that we used no wetting agent with this material but there are reports of wetting agents being used with considerable increase in effectiveness of penetration. This could be used to cut costs in using this material.

CARMINE RAGONESI: Does this material have any inhibiting growth on the cuttings?

JIM CUMMINS: The only observation I can give is on *Prunus besseyi* and just the converse happened; treated plants developed rapidly and made better plants than untreated ones. We've also tried this material as a transplant solution and though it is expensive to use this way, we obtained excellent results with plants subjected to field fungi.

ANDY LEISER: Do you know the trade name under which this will be marketed and who is the manufacturer?

JIM CUMMINS: duPont manufactures it. It has been tested as Fungicide 1991 and will probably be marketed as Benomyl which is the official, coined, common name; it has also been called Benlate and it may come out under this name.

MODERATOR FLEMMER: We will have to cut off the questions now; any additional questions can go into the Question Box. Our next speaker is Dr. Robert Farmer who will speak about mist propagation of black cherry.