Factors Influencing Uptake, Concentration, and Persistence of Benomyl in American Elm Seedlings

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ABSTRACT

Benomyl fungicide, applied to 4- to 5-month-old container-grown American elm seedlings, was absorbed by the roots and translocated throughout the plants. Zones of inhibition, around tissue sections placed into petri plates containing potato-dextrose agar seeded with *Ceratocystis ulmi* conidia, were considered indicative of the relative concentration of fungitoxicant in the plants. The concentration of fungitoxicant in seedlings that received three or more applications of benomyl increased only slightly beyond the third application. Seedlings contained a high concentration of fungitoxicant 110 days after the last of three applications of benomyl. The amount of fungitoxicant in seedlings, similarly treated and then transplanted to untreated sand, declined between 10 and 40 days after treatment. Composition of the planting medium influenced uptake of benomyl. The sizes of the diameter zone of inhibition around tissue sections from plants grown in sand were 1.5 to 2.5 times larger than from plants grown in a silt loam soil, and 2 to 6 times larger than from plants grown in a mixture of soil, peat, and perlite. Phytopathology 60:1619-1622.

Additional key words: systemic fungicide, methyl 1-(butylcarbamoyl)-2-benzimidazolecarbamate.

Evidence is accumulating that methyl 1-(butylcarbamoyl)-2-benzimidazolecarbamate (benomyl) is effective in suppressing or reducing symptoms associated with certain vascular wilt diseases (1, 2, 5, 7, 11). There is, however, a lack of information on the effect of such factors as soil type, composition, porosity, pH, and microbial populations upon the uptake of benomyl by plants. We conducted this investigation to determine (i) the number of applications of benomyl required to saturate tissues of American elm, *Ulmus americana* L., seedlings; (ii) the residual longevity of benomyl or a fungitoxic benomyl derivative in elm tissues; and (iii) the effect that different planting media have upon the uptake of benomyl by seedlings.

MATERIALS AND METHODS.—We transplanted 1-month-old greehouse-grown American elm seedlings into 7-inch plastic containers in either white silica sand (flint shot, grain size 34), soil (Morley silt loam), or potting mixture (soil 1:peat 2:perlite 2). Supplementary incandescent light (approx 600 ft-c) provided a 16-hr photoperiod. Weekly applications of 200 ml of modified Hoagland's solution (8) were added to each container. Benomyl was applied to the plants either as a drench or incorporated directly into the planting medium when the seedlings were 4 to 5 months old. A min of 5 days elapsed between the final benomyl treatment and a bioassay of the plants for the presence of a fungitoxicant in the tissues. Fifteen treated and three untreated seedlings were used for each bioassay. The plants were severed at the base of the stem and divided into three regions: (i) top, characterized by fully expanded terminal leaves; (ii) center; and (iii) bottom, located about 50 mm above the severed base of the stem. Leaf discs (13 mm diam) and wood and bark sections (25 mm long) from each region were placed into petri dishes which contained 12-14 ml potato-dextrose agar (PDA) seeded with 1 ml of a standardized conidial suspension of *Ceratocystis ulmi* (Buism.) C. Moreau (OD 0.1 at 450 mμ) per 100 ml PDA (6, 7). After the plates were incubated at room temp for 48 hr, the diam of the zones of inhibition around each tissue section was measured to determine the relative concn of fungitoxicant present in the seedlings.

Seedlings grown in sand were drenched 1, 3, 5, 7, or 10 times with 200 ml of a 500 ppm active aq suspension of benomyl/treatment. Plants were treated twice a week. The plants were bioassayed 5 days after the last of each designated number of treatments according to the procedures described above.

Another experiment was conducted to obtain information on the persistence of benomyl or a fungitoxic derivative in the plant tissues and in the planting medium. Plants grown in sand and drenched 3 times with benomyl (same amt and concn as previously used) were bioassayed 5, 10, 20, 40, 80, and 110 days after the last application. A second group of seedlings was treated similarly and then transplanted to untreated sand 5 days following the final application of benomyl. This was necessary in order to determine the longevity of the fungitoxicant in the plants without influence from the residual benomyl present in the planting containers. Prior to replanting, the roots were washed thoroughly with tap water to remove all treated sand and as much adsorbed fungicide as possible. A group of seedlings was bioassayed to determine initial uptake of benomyl the same day the remainder of the seedlings were transplanted. These seedlings were subsequently bioassayed 5, 10, 20, 40, and 60 days after transplanting.

Studies were undertaken to determine the effect of different planting media on the uptake of benomyl by 4-month-old elm seedlings. Plants grown in soil, sand, or

potting mixture were drenched 3 times with 200 ml of a 500 ppm active aq suspension of benomyl and then bioassayed 5 days after the final treatment. A second group of seedlings was transplanted into sand, soil, or potting mixture after benomyl had been incorporated at the rate of 300 mg active material/container of planting medium. The benomyl and planting media were mixed in a cement mixer to assure uniform distribution of benomyl throughout the media. Seedlings were also transplanted into untreated sand, soil, or potting mixture and drenched with one application of 200 ml of a 1,500 ppm active aq suspension of benomyl. The transplanted seedlings were bioassayed 30 days following treatment.

RESULTS AND DISCUSSION.—Only minor differences occurred in the sizes of the zones of inhibition around tissue sections from plants which had been treated 3, 5, 7, or 10 times with benomyl (Fig. 1). The amt of fungitoxicant increased slightly in the leaves and bark between the third and tenth treatment, but the amt in the wood remained essentially unchanged. Apparently there is a saturation point in the elm tissues beyond which additional treatments contribute little or nothing to the level of fungitoxicant in the plants, but merely increase the amt of chemical in the planting medium.

Sections of seedlings which had been treated 3 times still contained an inhibitory concn of fungitoxicant 110 days following the final application of benomyl (Fig. 2). The top and center leaves even contained a slightly greater amount of fungitoxicant after 110 days than after 5 days. Data beyond 40 days were not obtainable for the bottom leaves because of natural leaf drop.

In seedlings which were treated similarly and then transplanted into untreated sand, the level of toxicant dropped sharply in the wood after 10 days and more gradually in the bark and leaves after 20 days (Fig. 3). By 40 days, the fungitoxicant could not be detected in the wood, and by 60 days only trace amt remained in the center bark sections. Biehn & Dimond (1) demonstrated a similar situation in benomyl-treated tomato plants which had been transplanted to fresh sand. In their study, a residual fungitoxicant was detectable only up to 5 days in stem sections and about 2 weeks in leaves. The small amt of active chemical applied to each tomato may in part account for the short residual toxicity.

The level of toxicant remained quite high in the leaves in both experiments designed to determine the residual longevity of benomyl. Benomyl or a fungitoxic derivative (4, 9, 10) is being apparently concd in the leaves at the expense of the wood and bark. Furthermore, a reservoir of fungicide must be maintained in

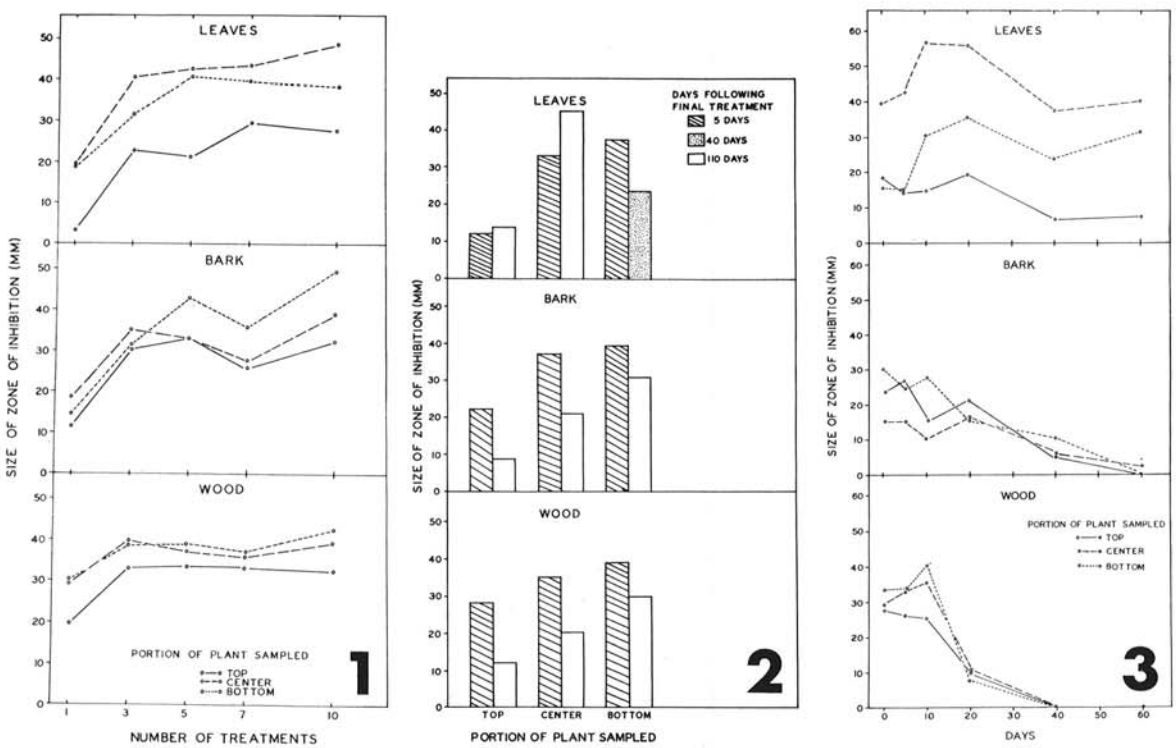

Fig. 1-3. Residual fungitoxicant present in top (characterized by fully expanded leaves), center, and bottom (50 mm above severed base of stem) leaf, wood, and bark sections of American elm seedlings following treatment with benomyl fungicide. **1)** After 1, 3, 5, 7, and 10 treatments. **2)** After 5 and 110 days following three treatments. **3)** In transplanted seedlings which had been treated 3 times prior to being transplanted to untreated sand. Benomyl was applied as a drench at the rate of 200 ml of a 500 ppm active ingredient aqueous suspension per treatment. Leaf discs are 13 mm in diam; bark and wood sections are 25 mm long.

TABLE 1. Effect of different planting media on the uptake of benomyl by American elm seedlings

Treatment and medium	Portion of plant and tissues sampled[a,b]								
	Top			Center			Bottom		
	Leaf	Bark	Wood	Leaf	Bark	Wood	Leaf	Bark	Wood
	Size of zone of inhibition (mm)								
Drench[c]									
Sand	9	25	25	42	28	35	18	32	38
Soil[f]	1	11	10	25	15	19	15	18	20
Mix[g]	0	4	5	10	8	9	16	15	12
Drench[d]									
Sand	33	33	41	55	30	48	56	45	51
Soil	18	13	18	40	7	19	38	28	25
Mix	7	1	4	24	2	8	28	21	10
Incorporation[e]									
Sand	29	25	28	44	18	30	44	31	36
Soil	24	14	17	27	11	21	33	18	24
Mix	11	2	7	23	2	10	10	9	10

[a] Top (characterized by fully expanded terminal leaves), center, and bottom (50 mm above potting medium).
[b] Leaf discs (13 mm diam), bark and wood sections (25 mm long).
[c] Three applications of 200 ml of a 500 ppm active ingredient aq suspension; plants bioassayed 5 days after final application.
[d] One application of 200 ml of a 1,500 ppm active ingredient aq suspension; plants bioassayed 30 days after treatment.
[e] 300 mg active ingredient mixed dry/container planting medium; plants bioassayed 30 days after treatment.
[f] Morley silt loam.
[g] Soil:peat:perlite, 1:2:2.

the planting medium in order to retain a high level of toxicant in the wood and bark and afford protection to newly developing tissues. This was substantiated by the experiment in which seedlings were transplanted to untreated sand.

All treated plants grown in sand took up more benomyl than did those grown in soil or in potting mixture (Table 1). The sizes of the diam zone of inhibition around tissue sections from plants grown in sand were 1.5 to 2.5 times larger than from plants grown in soil and, in most cases, 2 to 6 times larger than from plants grown in the potting mixture. These differences were consistent regardless of the type and the location of the sampled tissues. The amt of fungitoxicant increased in most plants when the period for chemical uptake was extended from 5 to 30 days, but the relative differences in the amt of toxicant present in plants grown in sand, soil, and potting mixture remained unchanged.

Maximum uptake of benomyl by elm seedlings occurred in sand, a porous medium; whereas, uptake was reduced when plants were grown in the less porous soil medium. On the other hand, the potting mixture which is quite porous restricted uptake of benomyl by the seedlings.

Cimanowski et al. (3) found that systemic control of powdery mildew of apple by benomyl was reduced when plants were grown in perlite and attributed this reduction to the increased porosity of the planting medium. Perlite constitutes 40% of the potting mixture used in our studies, and may be the ingredient responsible for restricting the uptake of benomyl; however, the porosity of the planting medium was probably not the major factor influencing uptake, in light of our results when benomyl was incorporated directly with the three different planting media (Table 1). The sizes of the diam zone of inhibition around tissue sections from seedlings transplanted to treated sand were about 1.5 times larger than from seedlings transplanted to treated soil, and 2 to 4 times larger than from seedlings transplanted to the treated potting mixture.

Benomyl may be inhibited by certain chemical, physical, or biological constituents of the soil and potting mixture. The effect of different media on the uptake of benomyl by plants will be investigated further.

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