Effects of Container Bases on the Spread of *Meloidogyne incognita* in a Hawaiian Ornamental Nursery

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ABSTRACT

Ko, M. P., Schmitt, D. P., and Saxby, M. 1997. Effects of container bases on the spread of *Meloidogyne incognita* in a Hawaiian ornamental nursery. Plant Dis. 81:607-613.

The influence of container bases on nematode spread from infested pots or ground in an ornamental foliage nursery was investigated with *Meloidogyne incognita* as the test nematode and cowpea as the indicator plant. The container bases examined were black cloth, concrete masonry blocks, gravel, crushed cinders, and bare ground. Masonry blocks raised 46 cm above ground (raised bench) served as the control treatment. Under the standard nursery practice of irrigation by rainfall and supplemental overhead sprinkler, nematodes spread from contaminated pots or ground to adjacent nematode-free pots on all tested container bases. Incidence of spread, measured as the percentage of nematode-free pots that became contaminated, increased over time and occurred more extensively from ground to pot than from pot to pot. Ground to pot spread increased rapidly following heavy rains, indicating rain was an important contributing factor. On the *M. incognita*-infested nursery ground, the nematode was found more frequently associated with weeds than with bare soil, suggesting that weeds were important reservoirs of nematode inoculum for rain-splash dispersal. Placement of containers on a raised bench was more effective than on any of the unraised container bases in preventing the ground to pot spread.

Additional keywords: concrete masonry units, plant-parasitic nematodes, shade cloth, weed reservoir

Floriculture and nursery products, mostly exported, generated a revenue of \$68 million in Hawaii during 1994 (15). Plant-parasitic nematodes, especially *Radopholus* spp. and *Rotylenchulus reniformis*, can be a problem in Hawaiian nurseries (2,17,34). The nematodes directly damage many of the nursery and ornamental crops (16,27), indirectly increase the costs of production (16) and decrease potential markets for export due to quarantine regulations (16,19). Because of the economic importance of these nematodes, costly local quarantine and growing guidelines are established (16).

The possible sources of nematode infestation or contamination include the following: infected propagation stock (9,37); infested soils or growth media (6,23); contaminated plant debris (6,23); infested soil adhering to vehicles, tools, and boots (7); contaminated irrigation water (2,19); and alternative weed hosts (8,19). However, the spread of the nematodes from these sources to healthy plants is difficult

to demonstrate and has not been done under Hawaiian nursery conditions.

Spread of nematodes depends on the survival of the nematodes and production of infective and dispersal stages. Nursery production practices in Hawaii are amenable to adoption of innovative measures to minimize nematode damage. Typically, ornamental foliage plants are grown in containers placed on a black cloth base or other bedding (hereafter referred to as container base) used for weed control. Container bases, with the appropriate physico-chemical properties (e.g., pH, moisture, or heat-holding capacities), may differ from the normal habitats of nematodes such that the rate of nematode survival is reduced. Container bases have been shown to alter the impact force of incident rain drops, affecting the distance and height of fungal propagule dispersal (38,39). The use of pinewood bark on black cloth in place of black cloth alone as ground cover has reduced the incidence of *Phytophthora* diseases in container-grown azaleas (4). Similarly, the change of bedding in cow hutches from hay-straw to ground corn cobs decreased the survival of maggots and, subsequently, the flies that decreased milk production in dairy pens (36). Therefore, a suitable container base in an ornamental nursery, along with other sanitary measures, such as clean propagation stock and weed eradication, may help eliminate the source of nematode inoculum

and the need for expensive elevated benches that are now the norm in Hawaiian certified nurseries (16).

The objectives of this study were: 1) to demonstrate the occurrence of nematode spread from contaminated pots or ground under the typical nursery conditions of rainfall and irrigation; and 2) to determine the effect of container bases on the spread of the nematode from ground to pot or pot to pot. Cowpea (*Vigna unguiculata* (L.) Walp. cv. California Blackeye) was used as the indicator plant and a local population of *Meloidogyne incognita* (Kofoid & White) Chitwood, originally isolated from ginger at Hilo, Hawaii and maintained on Pixie tomato, was used as the test nematode.

MATERIALS AND METHODS

Nursery conditions and plant growth.

The experiments were conducted from October 1993 to July 1994 at a commercial nursery in Keaau, Hawaii. The experimental area (19 × 24.5 m²) was a section of a nursery where ornamental and foliage plants such as *Rhapis excelsa*, *Cycad revoluta*, *Pritchardia* spp., *Spathiphyllum* spp., and *Aglaonema* spp. were grown. The experimental ground, consisting of cinder, soil, and small rocks, was under the cover of black cloth during the past five years. The ground was found to be initially free of weeds and plant-parasitic nematodes through preliminary sampling and tomato-seedling bioassay (3). The area was divided into two portions: area A for the ground-to-pot spread experiment, and area B for the pot-to-pot spread experiment (Fig. 1). In both experiments, six types of container base (each with 5 replicates) were arranged in 1-m²-microplots in a randomized complete block design (Fig. 1). The container bases evaluated were: 1) black cloth (Pak Ground Cover - Landscape, PAK Unlimited Inc., Norcross, GA); 2) concrete masonry units (CMUs), 20.3 × 20.3 × 40.6 cm, laying flat on the 40.6-cm-side (J. W. Glover, Hilo, HI); 3) a 7.6-cm-layer of ca. 1.9-cm-diameter pieces of gravel; 4) a 7.6-cm-layer of 1- to 2-cm-diameter pieces of crushed cinders; 5) bare ground (mixtures of rock, soil, and cinder); and 6) control, a level bench constructed from CMUs raised to 46 cm above ground (the current practice in Hawaiian certified nurseries). Nematode-free or *M. incognita*-infested cowpea seedlings (5/pot) growing in 3.8-liter plastic pots (15.2 × 17.8 × 12.7

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Accepted for publication 20 February 1997.

Publication no. D-1997-0422-06R
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cm) containing a cinder-peat (3:1 vol/vol, pH 6.4) mix were used to simulate the clean and contaminated foliage plants, respectively. The plants were fertilized with 5 cm³ of Nutricote/pot (18-6-8 N-P-K, 270-days formulation, Chisso-Asahi Fertilizer Co., Ltd., Tokyo) and sprayed with diazinon (48% a.i., Ciba-Geigy, Greensboro, NC) once every two weeks at 1.8 g/liter to control insects. Plants were irrigated by natural rainfall supplemented with overhead sprinklers to attain an average weekly precipitation of at least 7 cm. A 73% shade cloth (Pak Shade, PAK Unlimited Inc.) was placed over the entire

nursery area. Plant growth in a pot container was estimated at mid-course (19 January and 16 March for the pot-to-pot and ground-to-pot spread experiments, respectively) by measuring the weights of two arbitrarily chosen cowpea plants from each pot after the plants had been dried at 70°C for 72 h. Climatological data (Table 1 and Fig. 2) during the course of experiments were obtained from weather stations at Hilo Airport and Keaau, Hawaii, located 10.0 and 0.7 miles from the experimental site, respectively (30). Actual temperatures at the experimental site were also measured with a Taylor maximum/

minimum thermometer (Thermometer Corporation of America, Fletcher, NC).

Nematode spread from ground to pot. In this experiment (area A) (Fig. 1A), six nematode-free pots of cowpea seedlings (indicator pots) were placed at the center of each experimental unit or microplot (Fig. 1C). *M. incognita* was introduced into the ground by planting three 1-month-old nematode-infected cowpea seedlings (inoculated when 5 days old with 7,500 eggs of *M. incognita*) at the interspace between adjacent microplots. A 30-cm-deep fiberglass (Lasco, Florence, KY) barrier was installed around the area to confine the introduced *M. incognita*. These cowpea plants, along with any subsequent volunteer weeds in this area, were trimmed to 5-cm-height at least once a month so they would not hinder rain-splash dispersal. At mid-course of the experiment (16 March 1994), weeds on the experimental ground were identified to species according to Haselwood and Motter (14), Hubbard (18), or Neal (31), and counted.

Assays for nematodes were conducted at 1- to 3-month intervals. Three types of samples were collected: 1) roots and growth media taken from the middle of each indicator pot to evaluate the incidence of nematode spread (pot samples); 2) 30 samples taken from arbitrarily chosen locations between the container bases to determine the quantity of inoculum on the ground (ground samples); and 3) 30 cowpea and 30 weed plants taken arbitrarily from the ground to determine the incidence of *M. incognita* around their roots (rhizosphere/rhizoplane samples). The pot samples, each taken in three 2-cm-diameter cores with a coring device (cork borer no. 12) that was flamed between sampling to prevent cross-pot contamination, consisted of 50 cm³ mixed roots and media. The ground samples, each taken to a depth of 15 cm with a hand trowel from bare areas between the microplots, consisted of 300 cm³ soil. Each rhizosphere/rhizoplane sample consisted of varying amounts of root segments and adhering soil cut into 1- to 2-cm-pieces after the bulk soil around roots was removed.

The soil and root samples were stored in a cooler and processed within 24 h. Fifty cm³ of each sample was placed in a mist chamber on Baermann funnels (3) at 22°C. Second-stage juveniles (J2) of *M. incognita* emigrating from the samples were collected after 3 days and counted. The incidence of nematode spread in an individual microplot was defined as the percentage of indicator pots becoming infested with *M. incognita* in that microplot.

At the termination of the experiment, additional 250-cm³ ground and pot samples were similarly taken and placed individually in 7.6-cm-diameter clay pots for tomato seedling bioassay. One-month-old tomato (*Lycopersicon esculentum* Mill. cv. Pixie) seedlings were transplanted into

Fig. 1. Schematic of experimental layout to determine the effect of container bases on the spread of *Meloidogyne incognita* in a commercial nursery (not to scale). (A) Arrangement of microplots (squares) and their container-base treatments (treatment number placed inside a square) for the ground-to-pot experiment. (B) Arrangement of microplots (squares) and their container-base treatments (treatment numbers placed inside a square) for the pot-to-pot experiment. (C) Close-up view of a microplot in A showing pot arrangement to determine dynamics of spread from the contaminated ground to indicator pots of nematode-free cowpea plants. (D) Close-up view of a microplot in B showing pot arrangement to determine dynamics of spread from the source pot to indicator pots of nematode-free cowpea plants. Pots were placed on the following container base treatments: 1) black cloth on ground; 2) concrete masonry units (CMUs) on ground; 3) a 7.6-cm layer of gravel (approximately 1.9-cm-diameter pieces) on the ground; 4) a 7.6-cm layer of crushed cinder (1- to 2-cm-diameter pieces) on ground; 5) bare ground (mix of cinder, rock, and soil); and 6) control, level benches consisting of CMUs raised 46 cm above ground.

each pot, and the pots were maintained in a greenhouse at $30 \pm 5^\circ\text{C}$. Tomato plants were harvested from the pots eight weeks later. Roots from the plants were individually washed free of media and rated for root-knot galling on a scale of 1 to 100 (3).

Nematode spread from pot to pot. In this experiment (area B) (Fig. 1B), an *M. incognita*-infested pot of cowpea seedlings (source pot) encircled with eight similar nematode-free pots (indicator pots) was placed at the center of each container-base microplot (Fig. 1D). Infested pots had received 7,500 eggs of *M. incognita* when the cowpea seedlings were 5 days old. Pots were similarly placed in the control microplot, with the exception that the 'source pot' was also nematode-free. In contrast to area A, the ground was not infested with *M. incognita*. Any volunteer weeds in this area were rogued or spot treated with Roundup (62% a.i., Monsanto, St. Louis, MO) at 7.9 ml/liter to prevent establishment of the nematode.

Incidence of nematode spread and quantity of inoculum from the source pots in each container-base or control microplot were determined at 1- to 3-month intervals from pot samples taken from indicator or source pots as described in the previous section for nematode spread from ground to pot. Quantity of inoculum on the ground was likewise determined from ground samples taken at 1- to 3-month intervals, and from tomato-seedling bioassays (for root-knot galling) conducted with the additional 250-cm³ ground and pot samples taken at the termination of the experiment as described in the previous section.

Statistical analysis. Data were subjected to analysis of variance and correlation analysis with SAS software (SAS Institute) procedures. Means were separated according to Duncan's New Multiple Range test if a significant treatment effect was detected. In some cases, data were transformed to $\log(x + 1)$ values to normalize variances before the analyses. Un-

less otherwise stated, all differences were significant at $P = 0.05$.

RESULTS

Nursery conditions and plant growth.

The average monthly temperature measured at the nearby Hilo Airport weather station during the experimental period was 23.1°C , with variation from -4 to $+5\%$ (Table 1). Average daily temperatures on the sampling days at the experimental site did not differ by more than 0.7°C from the reported temperatures at Hilo Airport weather station; the coefficient of correlation between the temperatures at both sites was 0.97 ($P = 0.0001$). The temperatures at the weather station, therefore, were taken as the temperature at the experimental site. Accordingly, the accumulated thermal degree-days above 10°C (DD10) for the pot-to-pot spread experiment (mid-October to mid-April) and ground-to-pot spread experiment (mid-January to mid-July) were 2,899 and 2,975 degree-days, respectively (Table 1). Thus, the conditions were conducive to plant growth and *M. incognita*

development. Nematode-free cowpea plants in the indicator pots grew normally, although somewhat etiolated because of partial shade. All nematode-infested cowpea plants in the source pots in area B or on the ground in area A were stunted (average dry weight of a 3-month-old plant was 0.51 g) compared with the nematode-free plants (average dry weight/plant was 0.88 g), especially during the final stage of the experiments. A few plants in the source pots died early and were replaced with new cowpea seedlings. Weeds were found two weeks after initiation of the experiment (Table 2). Densities of cowpea plants and weeds averaged $5/\text{m}^2$ on the bare surface in area A at the mid-course of the experiment.

Nematode spread from pot to pot.

Spread of *M. incognita* from the source pots to the indicator pots occurred with all container bases except the control and bare-ground treatments 3 months after the initiation of the experiment (19 October 1993) (Fig. 3). Spread occurred as soon as 1 month on the CMUs and as late as 3 months on the bare-ground base. After 6

Fig. 2. Climatological data during the 10-month experimental period (1 October 1993 to 31 July 1994) at Keauau weather stations located 0.7 mile from the experimental site. Pot-to-pot and ground-to-pot spread experiments were set up at time *a* (upward arrowhead) and *b* (downward arrowhead), respectively. Sampling dates within the same experiment were denoted by the same directional arrowheads.

Table 1. Summary of climatological data at Hilo and Keauau Weather Stations, October 1993 to July 1994, including deviations from the norms^a

Date	Monthly temperature (°C)			Degree-days ^c	Monthly rainfall (cm)	
	Range	Hilo ^b			Keaua ^b	
		Average	Deviation (%) ^d		Average	Deviation (%) ^d
October 1993	28.2–20.7	24.4	+0.7	607	31.17	+11.9
November 1993	26.7–19.3	23.0	–1.9	509	23.80	–24.4
December 1993	25.9–18.2	22.1	–1.7	474	31.57	–4.8
January 1994	25.3–16.9	21.1	–4.3	423	27.71	–1.1
February 1994	25.4–18.2	21.8	–1.0	415	67.82	+117.4
March 1994	25.9–18.2	22.1	–0.8	471	49.10	+24.6
April 1994	26.7–19.3	23.0	+1.7	510	24.10	–42.1
May 1994	27.8–19.8	23.8	+2.4	569	21.64	–23.2
June 1994	28.2–20.7	24.4	+1.8	587	33.86	+91.0
July 1994	29.7–21.9	25.6	+5.3	669	33.32	+26.4

^a Calculated from data obtained from NOAA (30).

^b Hilo Weather Station located at $19^\circ 43' \text{N}$ $155^\circ 4' \text{W}$, 8.2-m elevation. Keauau Weather Station located at $19^\circ 38' \text{N}$ $155^\circ 2' \text{W}$, 67.1-m elevation.

^c Thermal degree-days = sum of the numbers of days $\times$ daily mean temperature above 10°C .

^d Deviation from the 30-year averages, 1951–1980 for 1993, or 1961–1990 for 1994.

months, contamination of the indicator pots ranged from 0% in the control to 5 to 15% in various container bases (Fig. 3). Incidences of nematodes on all container-base treatments were not different ($P = 0.05$) because of the large variations among replicates of the same treatment. Similar results were obtained with the tomato bioassay. The correlation coefficient between the incidences determined by the two methods was 0.89 ($P = 0.01$). Rainfall was heavy in December, February, and March (Fig. 2), but this did not appear to significantly increase nematode incidences in the months following these rainy periods on any of the container bases (Fig. 3).

J2 were detected in the source pots during the period within limits of 10 to 100/50 cm³ soil (Fig. 4). The population densities of J2 in the source pots of all container bases and in the control were not different from one another at any sampling date throughout the experimental period, although large temporal fluctuations occurred ($P = 0.05$) (Fig. 4). J2 were not detected on any samples taken from the ground in area B, either by the mist chamber or by the tomato bioassay.

Nematode spread from ground to pot.

Contamination of indicator pots on the bare-ground base appeared as soon as 2 months from the inception of the experiment (19 January 1994), and on all other container bases including the control shortly after 3 months (Fig. 5). By 6 months, nematode incidences, as assayed by the mist chamber, ranged from 5% on the control to 45% on the bare-ground or gravel base. Similar values of incidence were obtained with the tomato bioassay. The correlation coefficient between the nematode-incidence values obtained by the mist chamber and tomato bioassay was 0.85 ($P = 0.01$). Contamination of indicator pots placed on the crushed cinder was less than on other container bases ($P = 0.05$) (Fig. 5). Rainfall was extremely heavy in February and March (Fig. 2), about 117 and 25% above normal, respectively (30). In contrast to the results in the

pot-to-pot experiment, increases in nematode incidences on all container bases were notable in April (Fig. 5), about 2 months or 886 degree-days later (Table 1).

J2 were detected throughout the experiment on cowpea plants, volunteer weeds, and exposed ground (Fig. 6). J2 were present in roots of all the cowpea plants grow-

Fig. 3. Effects of container bases on the incidence of pot-to-pot contamination or spread by *Meloidogyne incognita* during the 6-month period between 19 October 1993 and 20 April 1994. Incidence of nematode spread for a container base was defined as percent of indicator pots on the base that were positive for *M. incognita* juveniles. Each data point is a mean of five replicates.

Fig. 4. Population densities of *Meloidogyne incognita* juveniles in the source pot of each container base for pot-to-pot spread during the 6-month period between 19 October 1993 and 20 April 1994.

Table 2. Incidence of weed species and their association with *Meloidogyne incognita* on the ground of the experimental nursery site in March 1994.

Weed species	Common name	Frequency of occurrence (%)	Association with <i>M. incognita</i> ^a
<i>Drymaria cordata</i> (L.) Willd. ex. R. & S.	Drymaria	21	+++
<i>Cyperus brevifolius</i> (Rottb.) Hassk.	Green kyllinga	18	+
<i>Stachy arvensis</i> L.	Staggerweed	15	+++
<i>Begonia foliosa</i> HBK	Begonia	14	+++
Fern & Allies ^b	Ferns	10	+
<i>Cyperus javanicus</i> Houtt.	Marsh cyperus	6	+
<i>Alternanthera sessilis</i> (L.) R. Br. ex. R. & S.	Sessile joyweed	5	++
<i>Chrysaliocarpus lutescens</i> (Bory) Wendl.	Areca palm	5	++
<i>Cyperus esculentus</i> L.	Yellow nutsedge	3	+
<i>Centella asiatica</i> (L.) Urban	Asiatic pennywort	1	+
<i>Cyperus hypochlorus</i> Hbd.	Cyperus	1	+
<i>Digitaria sanguinalis</i> (L.) Heist. in Scop.	Crabgrass	1	+

^a Density of *M. incognita* second-stage juveniles (J2) from 50 cm³ root and associated soil (rhizosphere/rhizosphere) (3): +, ++, and +++ denote the presence of an average of 1–99, 100–999, and ≥1,000 J2, respectively.

^b Including *Asplenium unilaterale* Lamarck (Pamoho fern), *Cyclosorus dentatus* (Forsk.) Ching (Oak fern), *Microlepia setosa* (Smith) Alston (Palapalai fern), and *Sphenomeris chusana* (Linnaeus) Copeland (Palaa fern) (18,31).

ing on the ground at all sampling times. They also were present in 40 to 100% of samples associated with weed species, but in only 10 to 30% of the samples associated with exposed ground during the same period (Fig. 6). Surveys of the ground in area A in March (midway through the experiment) revealed that there were at least 12 weed species present (Table 2). All of the weed species examined were found to harbor J2 in the rhizospheres/rhizoplanes of their roots.

DISCUSSION

We detected nematode spread in our experiment by measuring the number of progenies of the primary J2 inoculum that were successfully dispersed from the contaminated pots or ground. Unlike bacterial and fungal foliar pathogens which impart symptoms in leaves within a few days (11,22), these secondary J2 did not appear until 1 to 2 months later. *M. incognita* requires 534 to 718 DD10 (41 to 55 days at 23°C) to complete a life-cycle (20); therefore, any J2 spread would not be detected visually until approximately 60 days after the initial contamination. Our detection methods, though insensitive (28), did have the advantage of measuring the effective nematode multiplication in a host, a better indication of whether the nematode would persist in a particular nursery plant.

Our data demonstrated that nematodes readily spread from contaminated pots or ground under Hawaiian nursery conditions. The data also showed that the ground-to-pot spread was more extensive than the pot-to-pot spread. There was little difference among the container bases in preventing pot-to-pot spread, even though the container bases tested varied greatly in physico-chemical properties such as water drainage, surface morphology, and physical hardness. In view of the absence of vectors, nematodes probably spread from contaminated pots, ground, or contaminated container base via: 1) active vertical migration through drainage holes of the pots; 2) free running water from rain or irrigation; or 3) rain-splash dispersal. Rapid upward migration of root-knot nematodes (e.g., *M. chitwoodi*) through a soil column was demonstrated, but the extent of movement depends very much on soil texture, population density, and the length of the growing season (29). The influence of cinder/peat mix used in our experiment on *M. incognita* movement is not clear, but the absence of any notable spread in the pot-to-pot experiment at the end of 6 months likely precludes significant contribution of this route to the spread. Also, dispersal via running water or vertical migration would imply the existence of differences among the container bases for allowing nematode spread. For example, black cloth, with relatively poor water drainage and long retention period for free water to pond between the drain-

age holes of source and indicator pots, would be expected to cause higher nematode dispersal to the indicator pots. This was not observed, suggesting that the route of spread was independent of the container base or ground cover. Also, puddles were observed in rainstorms in certain parts of the nursery ground where an experimental block was situated. However, there was no increase in spread in this block relative to the other blocks. Therefore, direct splashing of nematode-contaminated medium from source pots to adjacent indicator pots was the most likely route of nematode spread. This route would be influenced by the intensity and duration of splashing as well as the surface area and topography of the contaminating areas (39). The former aspect was illustrated in our experiment as the ground-to-pot spread was more extensive than pot-to-pot spread. The surface

area of contaminated ground was much greater than the surface area of a contaminated pot, so the probability of rain drops splashing from the contaminated ground to the indicator pots was much greater than that from a source pot. The route of spread by rain splashing was also augmented by the observation that a majority of the nematode contamination was detected in April, following the February rainstorm 886 DD10 earlier. With our detection method, a lag of 534 to 718 DD10 was required to detect in the indicator pots any J2 spread from a source pot. The sampling days in March and April were 415 and 886 DD10 following the rainstorm, respectively. Subsequently, the observation that incidence of spread was low in the March (short of the lag period) but high in the April (exceeding the lag period) sampling was consistent with the argument that,

Fig. 5. Effects of container bases on the incidence of ground-to-pot contamination or spread by *Meloidogyne incognita* during the 6-month period between 19 January 1994 and 13 July 1994. Incidence of nematode spread for a container base was defined as percent of indicator pots on the base that were positive for *M. incognita* juveniles. Each data point is a mean of five replicates.

Fig. 6. Incidence of *Meloidogyne incognita* infestation associated with cowpea plants, weeds, or soil on the nursery ground where the ground-to-pot spread experiment was conducted during the 6-month period between 19 January 1994 and 13 July 1994. Thirty samples each, consisting of cowpea plants, weeds, or soil, were taken separately at a sampling date. Incidence, calculated separately for the plant, weed, or soil samples, was defined as percent of the respective samples that were positive for *M. incognita* juveniles.

whereas daily irrigation and/or vertical nematode migration might contribute to spread, heavy rain was much more important to the nematode splash-dispersal. Although correlation analysis between the amount of rainfall and nematode incidence 534 to 718 DD10 later is not possible because of presumable secondary contamination from the infested indicator pots (which were not removed), there are other indications that rainfall played a vital role in the spread of nematodes. Grenvold (13) showed that incident drops of water 5 mm in diameter falling 2.2 m on a cow pat of 500 g contained 4,560 third-stage larvae of *Cooperia* spp., a trichostrongyle nematode, resulting in splash dispersal of larvae to a maximum distance of 63 cm from the point of impact. Kuske and Benson (22) found rainfall was significantly correlated with incidences of *Phytophthora parasitica* on rhododendron and the incidences were greatest after periods of rain. Gottwald et al. (11) found also the spread of *Xanthomonas campestris* pv. *citri*, a causal agent of citrus canker, was coincident with rainstorms.

The overhanging shade cloth or fabric might have contributed to the observation that rain was the major factor in the ground-to-pot spread, but not in the pot-to-pot spread. In a heavy rain, the fabric, equivalent to an US standard sieve number 25 mesh (0.75-mm-aperture), might have atomized the large raindrops in most spots but accumulated them into pools where the fabric droops. Thus, raindrops filtering through the shade fabric were not uniform and consisted of mostly fine drops (with insignificant effects on nematode splash dispersal) and random large drops (with great impact on splash dispersal). Because the surface area of the contaminated ground was larger than the contaminated pot, the probability of being struck or splashed by forceful random large drops was much higher for the ground than for the pot; thus, the ground-to-pot spread was more successful than the pot-to-pot spread. The observed large variation in nematode incidences among replicates of the same container-base treatment also was consistent with the non-uniformity of downpours of large water drops. It may be worthwhile to conduct simulation rain experiments similar to those conducted by Yang and TeBeest (38) under the microclimate of the shade house (12) to determine exactly how the pore size of a shade cloth in a nursery affects the size of raindrops, the distribution of rainfall, and, thus, the dispersal of plant pathogens beneath the cloth. Such information is needed not only to design better shade cloth for light penetration, air circulation, and rainfall distribution, but also to minimize disease spread.

Our results also give evidence of the importance of weeds as a reservoir for nematode spread. Weed reservoirs have contributed to the spread of nematodes to

other economic crops (8), and are particularly important in the tropics because of the year-long growing season and the wide host range of the quarantined nematodes (5,24,27,32). The absence or low number of *M. incognita* J2 in bare or exposed ground indicates that the nematode did not survive under the wet and moist tropical conditions in the absence of a host. Goodell and Ferris (10) found that more than 90% of *M. incognita* J2, in the absence of a host, became nonmotile at soil moisture near field capacity. This phenomenon may be common among plant-parasitic nematodes in the tropics. For instance, Mateille et al. (25) found that *Radopholus similis* did not persist after fallowing or flooding in banana plantations, even when nematode-free plants were re-planted to the areas. Thus wet fallow, the application of water to cropland in the absence of a host, increases plant-parasitic nematode mortality. Survival of nematodes such as *R. reniformis* in soil under drying conditions was documented (1,19), but it would be equally important to investigate their survival under the tropical wet and moist conditions prevalent in the nursery production areas.

Although the container bases that we tested did not prevent nematode spread from the contaminated pots or ground, an appropriate container base would play an important role in reducing the source of nematode or other pathogen inoculum (4,22,35,38). Container bases could achieve this end by avoiding container-to-soil contact, improving water drainage away from the containers, suppressing weeds, or reducing the height and distance of rain splash-dispersal. For example, less splash dispersal of *Colletotrichum acutatum* occurred from infected strawberry fruit on soil covered with fresh straw than covered with plastic or soil alone (39). Dispersal of *C. gloeosporioides* was higher and farther via contaminated raindrops rebounding from concrete than from soil and water surfaces (38). Container bases may also cause nematode injury directly, as demonstrated by McGrady and Cotter (26) and Kokalis-Burelle et al. (21), who found pine bark in media was nematicidal to root-knot or cyst nematodes.

Cowpea, the indicator plant used in the experiments, is a susceptible nematode host quite distinct from most of the ornamental plants. However, the mechanics of nematode spread is mostly independent of the host status so that the data obtained here with cowpea are applicable to other host plants. Nonetheless, because host status is important to nematode population increase and survival, and thus, the inoculum source, host influence should be included in future studies.

In summary, spread of nematodes was found to be influenced by a range of factors, only some of which are under the control of a nursery grower. Little controls

over rain intensity or rain duration can be expected, but weeds, height of the benches and the nature of container base can be controlled, as black cloth, cinder, and elevated benches are commonly used in foliage production (16). As shown in this study, the difference in nematode spread between the practices of placing pots on raised benches and on ground-level was great. Container plants grown on the latter can result in very high nematode incidences if the ground is contaminated relative to those cultured on the former. Growing plants on raised benches for nematode-free certification still gives the best results, as indicated by our results and those of others (16,33). However, modification of current systems may prove successful, especially when the nursery ground is kept free of weeds and rain splashing is minimized. The current practice of covering the nursery floor with black cloth to prevent weed growth is effective, but weeds that occasionally grow on top of the black cloth must be eradicated. The use of container bases that absorb splash impact, and shade cloth that breaks the size of raindrops while not pooling rain water, are other possibilities that can be developed in the future. In the meanwhile, a pitch in the shade cloth (to prevent puddling) coupled with raised benches should be effective as a control strategy. Most important of all is the elimination of the primary source of nematode contamination from the mother-stock plants, as suggested earlier (9,16,17,37). A rigorous inspection system by primary producers of nursery stocks would aid in the elimination of nematode contamination from their production systems.

ACKNOWLEDGMENTS

This research was funded in part by a grant (Contract No. 92-08) from the State of Hawaii Governor's Agricultural Coordinating Committee. This paper is a contribution from the Hawaii Institute of Tropical Agriculture and Human Resources, University of Hawaii, Honolulu. Journal Series No. 4241. We thank D. Meyer, G. Nagai, K. Okazaki, M. Young, and the staff at Puna Certified Nursery, Inc., Hilo for their technical assistance; and M. Aragaki, B. S. Sipes, and J. Starr for critical reviews of the manuscript.

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