Radopholus similis in *Anthurium* Shoot Tissue

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Additional index words. burrowing nematode, plant-parasitic nematode

Abstract. *Radopholus similis* distribution in *Anthurium* plant tissue was determined in a greenhouse experiment. Two thousand mixed life stages of *R. similis* per plant were inoculated onto *Anthurium* cultivars 'Alii' and 'Midori'. Nine months later, nematodes per gram of tissue were determined from stem sections (0–3, 3–6, and above 6 cm from the base), the lowest leaf petiole, and root tissue. *R. similis* occurred in all stem sections, leaf petioles, and roots in both 'Alii' and 'Midori'. Nematode distribution differed between the two cultivars. 'Midori' had higher numbers of nematode in the roots whereas 'Alii' had higher numbers of nematode in the stem sections and first leaf petiole. *Anthurium* apical stem cuttings could be contaminated with *R. similis* and may not be a nematode-free propagation material.

Several genera of plant-parasitic nematodes, such as *Aphelenchoides*, *Anguina*, *Bursaphelenchus*, *Ditylenchus*, and *Rhadinaphelenchus*, infect shoot tissue, whereas other genera infect root tissue (Dropkin, 1989). These different nematode life strategies dictate the use of different assay methods and control tactics. *Radopholus similis* is a migratory endoparasite of roots (Higaki et al., 1994; Holdeman, 1986; Holtzmann et al., 1984). Consequently, assay methods for *R. similis* involve sampling roots, and control recommendations include propagation of plants from cuttings taken at least 30.5 cm above the soil line (Higaki et al., 1994; Holtzmann et al., 1984). However, Goo (1995) observed *R. similis* in stems of anthurium (*Anthurium andraeanum* Linden ex André) 30 cm above the growing medium, even though the subtending aerial roots and cinder medium were free of nematodes. *Radopholus similis* has also been found in stem tissue examined for fungal infection (J. Uchida, personal communication). The objective of this experiment was to examine the frequency and extent of *R. similis* invasion of *A. andraeanum* shoot tissues.

Materials and Methods

Anthurium andraeanum, 'Alii' and 'Midori', *R. similis* intolerant and tolerant

Received for publication 9 June 1998. Accepted for publication 15 Sept. 1998. Supported in part by U.S. Dept. of Agriculture grant no. 93-34199-8355. This paper is a contribution from the Hawaii Institute of Tropical Agriculture and Human Resources, Univ. of Hawaii, Honolulu, Journal Series No. 4368. The cost of publishing this paper was defrayed in part by the payment of page charges. Under postal regulations, this paper therefore must be hereby marked advertisement solely to indicate this fact.

cultivars, respectively (Wang et al., 1997), were transplanted by stem cuttings (30 cm tall for 'Alii', 20 cm for 'Midori') into 3.8-L pots filled with sterile medium. Inoculum of *R. similis* was obtained by placing infected alfalfa root callus cultures in a mist chamber for 48 h. The *R. similis* was placed at the base of each plant in 10-mL aliquots of 2000 nematodes (Ko et al., 1996). Nine months after inoculation, shoot tissue was examined for *R. similis* invasion. The stem of each of four plants of each cultivar was sectioned into three pieces (<3 cm, 3–6 cm, and >6 cm from the base) and the petiole of the first leaf (<3 cm from the base). These sections were individually assayed in a mist chamber (Barker, 1985) for 4 d to extract nematodes. The experiment was repeated three times.

Samples were collected from the mist chamber, the number of *R. similis* counted under the dissecting microscope, and the tissue oven-dried to a constant weight. Number of plants with *R. similis* in a particular section was recorded for incidence of invasion. Incidence data were tested for homogeneity of variance by the Bartlett likelihood ratio test (Steel and Torrie, 1980). As homogeneity was detected throughout the three tests ($P \geq 0.05$), data were pooled and frequency of infection calculated. Data were analyzed for differences in frequency between cultivars for each tissue section using a $2 \times 2 \chi^2$ test, and among tissues within a cultivar using analysis of variance by the general linear model procedure of the Statistical Analysis System (SAS Institute, Cary, N.C.). Means were separated by the Waller–Duncan k ratio t test ($k = 100$).

Results and Discussion

Radopholus similis was found in 83% of the shoot sections. Incidence of plants with

nematodes differed between cultivars. Incidence in stem sections 3–6 cm and >6 cm was greater in 'Midori' than in 'Alii' ($P \leq 0.025$ and 0.005, respectively; Fig. 1). Frequency of nematode invasion of the first leaf petiole was higher in 'Alii' than in 'Midori' ($P \leq 0.05$, Fig. 1). *Radopholus similis* was recovered more frequently in lower stem sections (0–3 and 3–6 cm sections) than in higher sections (>6 cm) in 'Alii' (Fig. 1). In some stems, nematodes were recovered from the 3–6 cm section, but not from the lower section.

Numbers of nematodes per gram of dry tissue did not differ between cultivars or sections ($P \geq 0.05$). Some plants had no nematodes in the roots but nematodes were present in the shoot sections. A higher number of *R. similis* occurred in the root than in the shoot tissue of 'Midori' (Fig. 2). However, nematode numbers tended to be highest in the roots, lower stem sections, and first leaf petiole, with decreasing population densities higher in the shoot (Fig. 2).

Our results confirm anecdotal observations that *R. similis* is not limited to *Anthurium* root tissue, but occurs in stem and petiole tissues. *Pratylenchus* sp., another migratory endoparasitic nematode closely related to *R. similis*, has also been isolated from the stem and leaves of *Aglaonema* (D. Schmitt, personal communication). *Radopholus similis* invades all types of citrus parenchyma tissues, except hardened tissue such as suberized epidermis, exodermis, endodermis, mature xylem, hardened pith, peridermis, and the hardened root cap (DuCharme, 1959). *Radopholus similis* can penetrate the nonsuberized endodermis of young citrus roots and invade the stelar region.

Fig. 1. Incidence of *Radopholus similis* invasion in three stem sections from 0 to 6 cm from the base, and from the first leaf petiole of *Anthurium andraeanum* 'Alii' and 'Midori' 9 months after nematode inoculation. Data are percentages based on 12 replications; * and ** indicate significant differences between cultivars based on χ^2 test at $P \leq 0.05$ and 0.01, respectively.

Fig. 2. Numbers of *Radopholus similis* recovered per gram of dry tissue from *Anthurium andraeanum* sections. Stem sections are designated as the distance from the base of the stem. Means separation in 'Midori' by Waller-Duncan k ratio *t* test ($k = 100$), $P \leq 0.05$.

Members of the aroid family (e.g., *Aglaonema* and *Anthurium*) may have relatively less suberized or hardened tissue (Higaki et al., 1984), thus allowing penetration of these migratory endoparasitic nematodes into the stele. Restriction of nematode invasion to the first leaf petiole in 'Midori' might be related to its higher tolerance to *R. similis*.

Propagation of *Anthurium* by stem cuttings cannot guarantee nematode-free planting material. Furthermore, phytosanitary certification based solely upon the absence of nema-

todes in the root tissue may be problematic, since some plants may harbor nematodes in shoot tissue. A more thorough examination of shoot tissue may be required for certification. Further investigations are planned to determine the frequency of infected cuttings used for propagation.

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