Digital Video Technology as an Aid for Quantifying Root Rot

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Abstract. Evaluation of disease severity in root systems is usually subjective, based on observation and categorization into an arbitrary scale of several categories. Results obtained using this approach can vary according to the observer's experience. A new, simple method for evaluating and quantifying the root severity index (RSI) was developed. This method consists of surrounding the root pan with a transparent film and tracing all roots with a marker. On a second transparency, only healthy roots are traced. Lengths of both healthy and diseased roots are measured with a root length/area meter (Dias II). The method of peripheral root-ball root tracings was evaluated with poinsettias infected with *Pythium ultimum* (Trow.). Results indicated that it was as effective as the traditional method of determining RSI for total and peripheral roots.

One method of evaluating the effectiveness of fungicides in the prevention or control of soilborne plant diseases in floriculture involves growing crops in containers where the pathogen is added to the medium before or after the fungicide. After infection and disease development, plants are removed from the containers and roots are evaluated for disease severity (DS) either before or after removal of the growing medium. An arbitrary scale of DS is used to evaluate the degree of disease on the affected roots (root disease severity index or DSI); this scale generally has five or six categories. This qualitative method is simple and requires no sophisticated equipment, but is subjective and requires some observational skills by the evaluator, and results can vary among observers. Equipment to measure root length/area has been available for quite some time. A video camera creating an image, and computer software using mathematical algorithms, are used to measure pixels on the screen, and to estimate the area and length of such an image. This computer technology allows for easy quantification of some root characteristics and helps remove some of the subjectivity of human analysis. Although it can in theory distinguish between healthy and damaged roots based on differences in shad-

ing, its use requires the proper balance of lighting to maximize contrast. This balance is not easily achieved by a researcher with limited experience or someone who uses the equipment sporadically. Our objective was to de-

Fig. 1. Tracing technique. The transparency tightly surrounds the root ball, allowing for the roots to be traced with a fiber pen. After root tracing, the transparency is removed, photocopied, and root length is measured with a digitized video image instrument.

velop a simple procedure utilizing digital video technology for the quantification of damaged and healthy roots that could be used by a researcher having limited experience with such a technique.

Materials and Methods

Poinsettia (*Euphorbia pulcherrima* Wild.) plants, cv. Freedom Red, were grown in containers whose interior surface had been coated with a flat latex paint impregnated with metalaxyl { (R)-2[(2,6-dimethylphenyl)-methoxyacetyl amino]-propionic acid methyl ester; Subdue 2E, Ciba-Geigy Corp., Greensboro, N.C. } : 78 $\mu\text{L}\cdot\text{L}^{-1}$, as described by Pasian and Struve (1997). Additional plants were grown in containers that had not been coated (controls). Four weeks after planting, the soil was inoculated with the fungus *Pythium ultimum* as described by Varela et al. (1985). Sixty-two days after inoculation, plants were removed from their pots and root balls were evaluated by visual observation using a root severity index scale (RSI) from 1 through 6 (1 = symptomless, 2 = mild, 3 = intermediate, 4 = severe, 5 = severe and crown infection, and 6 = dead plant) (Boehm and Hoitink, 1992). After evaluating the severity of root damage in the periphery of the root ball, the length of healthy and diseased roots was measured. This last variable was obtained by removing the

Fig. 2. Representative example of poinsettia root tracings in contact with or near the internal surface of the container. Traces indicate the roots that were visible when the container was removed from the root ball: (A) all roots; (B) diseased roots. Actual length of tracings was 24 cm long.

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Table 1. Effect of metalaxyl-coating of pots on total root length, length of root-ball peripheral roots, and root-rot severity indices of poinsettia plants before and after washing the root medium from the root ball. Soil inoculated with *Pythium ultimum* 4 weeks after planting.

Treatment	Total root length ^y (cm)	Length of healthy roots (cm and %) ^y	Root-rot severity index ^z	
			Before washing	After washing
No paint, no metalaxyl	163.4	68.5 (42.0)	3.4	3.6
Paint plus metalaxyl (78 μL^{-1})	231.8	179.4 (77.4)	2.6	2.2
LSD _{0.01}	85.1	90.0	1.2	1.0
Significance	NS	**	NS	**

^zRoot rot rated 0 (none) to 6 (plant dead) before and after washing medium from root ball (root-ball peripheral roots).

^yOn the periphery of the root ball.

NS, **Nonsignificant or highly significant ($P = 0.01$), respectively.

root ball from the pot, placing a transparent film (for plain paper copiers, #901; 3M, Austin, Tex.) over the root ball and tracing all exposed roots with a marker (Fig. 1). The original transparency was removed and a second transparency was used to trace only diseased roots. Photocopies of the transparencies with the root traces were used to measure root length using a DIAS II (Decagon Devices, Pullman, Wash.) root length/area meter. Length of healthy roots was obtained by subtracting the length of diseased roots from total root length (Fig. 2). After root tracing, root balls were washed with water to separate the roots from the growing medium and root-rot severity was again evaluated using the qualitative RSI described above.

Results and Discussion

The method of peripheral root-ball root tracings was as effective as the traditional method of RSIs for total and peripheral roots. Total root length did not differ significantly among treatments (Table 1). Seventy-seven percent of the root length of plants exposed to metalaxyl showed no root-rot symptoms vs. 42% for the controls. Before washing off the medium, RSIs for both treatments did not differ significantly, but after washing off the medium, RSIs indicated that roots treated with metalaxyl were healthier than those not exposed to the fungicide. The root tracing method has the advantages of being less subjective and less dependent on evaluator experience, and

allows a longer period for evaluation because of its nondestructive nature. On the other hand, it requires an easy-to-use, yet expensive, video camera that may not be accessible to everyone. The method takes into account only roots at the periphery of the root ball, ignoring roots located at the center. This should not be a concern because most roots of plants grown in containers tend to be at the periphery of the root ball. Furthermore, in order to reach roots at the center, the media has to be washed away; damaged roots are easily lost, causing an increase in the evaluation error. This problem may be aggravated with herbaceous plants, such as the poinsettia used in this study. In the case of root balls having numerous fine roots too close to each other to trace individually, the root tracing method allows for area measurement as opposed to measurement of only total root length of healthy and diseased roots.

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