Surveying for Virus-vectoring Nematodes in Container-grown Blueberry Plants in Oregon

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Accepted for publication 25 July 2016. Published 8 August 2016.

Sharma-Poudyal, D., Fraley, C., and Osterbauer, N. K. 2016. Surveying for virus-vectoring nematodes in container-grown blueberry plants in Oregon. *Plant Health Prog.* 17:175-176.

The National Clean Plant Network (NCPN) is a joint effort by United States Department of Agriculture's Agricultural Research Service (USDA-APHIS), Animal and Plant Health Inspection Service (USDA-APHIS), and National Institute of Food and Agriculture (USDA-NIFA) to provide healthy clonally propagated plants to nurseries and growers. The NCPN is also establishing national standards for certification of such plants as virus-tested. A draft national certification standard has been developed for the production of certified blueberry (*Vaccinium* spp.) nursery plants (Anonymous 2016). Nematode-vectored viruses are identified as pathogens of concern in the draft standard, with specific mitigation measures required that address the nematode vectors. Oregon is one of the largest producers of blueberry nursery plants in the United States; the production of blueberry nursery plants takes place in containers, usually placed on gravel beds. The goal of this study was to determine the risk of finding virus-vectoring nematodes in containerized plants placed on gravel.

Two nematode-borne viruses that infect blueberry, *Tobacco ringspot virus* and *Tomato ringspot virus*, have been reported in Oregon (Converse and Ramsdell 1982; Martin et al. 2012) and in Arkansas, Connecticut, Illinois, Michigan, New Jersey, and New York (Martin et al. 2012). Dagger nematode (*Xiphinema americanum*), which is widespread in North America, is known to transmit these viruses (Martin et al. 2012; Robbins 1993). The incidence of dagger nematode varies from 21 to 40% in Oregon blueberry fields (Converse and Ramsdell 1982; Zasada et al. 2010); however, its incidence in Oregon blueberry nurseries has not been reported. To detect dagger nematode, soil and potting media samples were collected from 15 and 11 blueberry nurseries in 2014 and in 2015, respectively. All of the nurseries grew plants in containers using soilless potting media; containers were placed on a gravel bed (>5 cm thick) or, for one nursery in 2014, on a plastic sheet placed on the soil surface.

A total of 57 samples were collected in 2014, and 22 samples in 2015 (Table 1). Each sample consisted of a composite of four randomly taken sub-samples up to a total volume of 500 cc. Two types of samples were collected; potting media samples were collected from within pots containing plants, and soil samples were collected from beneath the gravel layer or the plastic sheet. The potting media samples consisted primarily of bark (typically Douglas-fir), peat moss, and either perlite or pumice. Nematodes were extracted from all of the samples using the sucrose

centrifugation technique (Zukerman et al. 1985). In brief, each sample was hydrated, mixed, and filtered three times through 20-mesh and then 500-mesh sieves. The remaining soil on the 500-mesh sieve was collected in a 50-ml tube, centrifuged (1,750 rpm for 5 min), and the supernatant discarded. Sucrose solution was then added to re-suspend the pellet and the sample centrifuged again (1,000 rpm for 30 sec). The resulting supernatant was poured through a 500-mesh sieve and the remaining content on the sieve collected into a beaker for examination under a dissecting stereoscope. Nematode identification was verified morphologically using a compound microscope.

Both years, no dagger or other plant parasitic nematodes were detected in the samples tested (Table 1). These results show soilless potting media is free of dagger and other plant parasitic nematodes. This suggests no treatment of soilless potting media is necessary before planting blueberries into containers. The results also indicate the gravel layer may help suppress the incidence of dagger and plant parasitic nematodes as the soil beneath was consistently free of these parasites (Table 1). The gravel layer may be providing a physical barrier to nematode movement as well as suppressing the growth of broadleaf weeds, which are alternate hosts for nematodes. Only a single soil sample from beneath a plastic sheet was tested. Although this sample tested free of dagger and plant parasitic nematodes, additional information must be collected to verify this is an effective barrier.

The draft national standard requires soil to be tested for virus-vectoring nematodes within 1 year before planting. If the soil is

TABLE 1
Dagger and plant parasite nematodes in potting media and soil collected from container-grown blueberry plants in Oregon nurseries.

Year	Sample type ^x	No. of samples	No. of nematodes ^y	
			Dagger	Plant-parasitic
2014	Potting media	29	0	0
2014	Soil	28	0	0
2015	Potting media	11	0	0
2015	Soil	11	0	0

^x Potting media was collected from containers with blueberry plants and soil was collected from beneath a gravel layer.

^y Total number of nematodes extracted from 500 cc of potting media or soil using the sucrose centrifugation technique.

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doi:10.1094/PHP-BR-16-0029
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infested with nematodes, soil needs to be treated with an approved method to manage these vectors. In addition, the planting site and a surrounding 10-m buffer zone must be free of hosts of soil-borne viruses and virus-vectoring nematodes (Anonymous 2016). Based on these survey results, growing blueberries planted in soilless potting media in containers that have then been placed onto a gravel layer effectively suppresses virus-vectoring nematodes; this best management practice should be taken into consideration in the draft national standard as a mitigation measure for soil-borne viruses that could infect certified blueberry plants.

ACKNOWLEDGMENTS

The authors appreciate the help provided by Jeffery Grant, Shannon Lane, and Sherree Lewis for sample collection. Funding for the project was provided by Farm Bill cooperative agreements 14-8130-0388-CA and 15-8130-0388-CA.

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