PLANT-PARASITIC NEMATODES IN GRAPE AND RASPBERRY SOILS OF ONTARIO AND A COMPARISON OF EXTRACTION TECHNIQUES[1]

J. L. Townshend [2]

Abstract

Eight plant-parasiticnematodes were associated with grape and raspberry in Ontario. Pratylenchus penetrans (Cobb) Filip. & Stekh. was harmful only to raspberry on sandy soils. Xiphinema americanum Cobb, the only species of dagger nematode found in grape and raspberry soils, was not considered to be important in itself, although it could be a vector of several soil-borne viruses. A greater number of Xiphinema americanum was extracted from soil with a combination of the Cobb and Oostenbrink methods than with the latter method alone. Extraction of Xiphinema from soil by the combined method was affected by the size and the amount of mixing of a sample,

Introduction

The development of virus-free raspberry stock and the introduction of American and European grape cultivars in Ontario necessitated the detection of plant-parasitic nematodes that might be harmful to these crops. Of particular interest were species of Xiphinemaknown to transrnit soil-borne plant viruses **(3).** Grape soils were examinedin 1964 and raspberry soils in 1965 by means of the Oostenbrink cottonwool filtermethod (9), as well as by a modificationof this method to extract Xiphinema from these soils. This paper presents details of the modification and its assessment and the results of the two surveys.

Materials and methods

In the Niagara Peninsula, 90 vineyards located on 19 farms in the counties of Lincoln and Welland were surveyed in 1964. The 90 soil samples were taken from about the roots of 18cultivars: 'Agawam', 'Canada Muscat', 'Catawba', 'Concord', 'Delaware', 'Duchess', 'Elvira', 'Foch', 'Fredonia', 'Niagara', 'Pinot Blanc', 'President', 'Seibel 1000', 'Seibel 9110', 'Seibel 108781, 'Van Buren', 'V-Port' and 'Westfield'. Three or more cultivars were grown on each farm. The soils were predominantly clay loams.

In southern and central Ontario 75 raspberry plantings located on 42 farms in the counties of Norfolk, Wentworth, Lincoln, Welland, Peel, Ontario, Durham and Prince Edward were surveyed in 1965. The 75 soil samples were taken from about the roots of 9 cultivars: 'Columbian', 'Crescent', 'Creston', 'Latham', 'Madawaska', 'Newburgh', 'September', 'Taylor' and 'Willamette'. Two or more cultivars were grown on each farm. The soils were predominately sandy loams.

Nematodes were extracted from two subsamples of 50g each taken from each field sample. One subsample was processed by the Oostenbrinkmethod and the other by a combined Cobb-Oostenbrinkmethod as described below. Nematode counts were recorded as the number per 50g of soil. The field samples were never stored longer than 2 weeks at 40" F before processing.

The Cobb (8) and Oostenbrink (9) methods were combined to extract Xiphinema from soil. The former method was the first phase: a soil sample was mixed in water in a pan and the coarse soil particles were allowed to settle out for a few seconds. The suspension was then poured through a 10-mesh sieve into a second pan, The original sample was resuspended and the preceeding steps repeated. The suspension accumulated in the second pan was stirred, debris allowed to settle momentarily, and then the suspension was poured through a 150-mesh sieve into a sink. Nematodes and fine organic debris trappedon the 150-mesh sieve were suspended in 500 ml of water in a third pan. This suspension was stirred and more debris allowed to settle out briefly before decanting into a beaker. The Oostenbrink method was incorporated at this stage. The suspension of Xiphinema and fine organic debris was poured in a serpentine manner onto #90 cheesecloth clamped in plastic hoops. The hoop with Xiphinema on the cheesecloth was placed in a pan containing a shallow layer of water. For convenience, an extraction period of 7 days was used for the combined method and for the Oostenbrink method alone.

The combined method was assessed by extracting & americanum Cobb from a Fonthill loam. Fresh soil was collected for each of 3 experiments performed. In the first experiment 50g samples of field soil were subjected to the following treatment: (1) the Cobb-Oostenbrink method with only cheesecloth in the hoops; (2) the Cobb-Oostenbrink method with cheesecloth and Kleenex in the hoops; **(3)** as in the first treatment except that the soil was first thoroughly mixed by hand; (4) the unmodified Oostenbrink method as a control.

In the remaining experiments with the combined methodonly cheesecloth was used in the hoops. In

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1 Contribution No. 137, Research Station, Research Branch, Canada Department of Agriculture, Vineland Station, Ontario.

2 Nernatologist

the second experiment the effect of sample size was determined by processing 25, 50, 100, 200 and 400g samples. In the third experiment, the survival of X. americanum in Fonthillloam stored at 40°F for a prolonged period was studied. Samples of 50g were processed weekly for 4 weeks. In each experiment the extraction period for the Cobb-Oostenbrink method was 2 days and for the Oostenbrink method 7 days at room temperature. Treatments in all experiments were replicated 8 times.

Results

In grape soils, eight types of nematodes occurred in small numbers only and were not related to cultivar or soil type (Table 1). Xiphinema was found in 47 of the 90 field samples from 17 of the 19 farms surveyed.

In raspberry soils seven types of nematodes were noted (Table 2). The root-lesion nematode averaged 102 per 50g in sandy soils and only 10 in clay soils. The condition of most plantings due to bad cultural practices prevented a critical assessment of the damage caused by the root-lesion nematode. However, in a single planting, areas of stunted canes were definitely associated with large populations of root-lesion nematodes. Xiphinema was found in only 21 of the 75 field samples from 37 of the 42 farms surveyed.

The identity of the eight nematode types are noted in Table 3 along with the crops with which they were associated.

Almost twice as many X. americanum were extracted from 50g of soil by Cobb-Oostenbrink method as by the Oostenbrink method alone (Fig. 1A). Fungi were seen in many of the dagger nematodes extracted by the latter method. Suspensions of dagger nematodes obtained by the combined method were almost

Table 1. Plant-parasitic nematodes associated with grape in Ontario

	Nematode types							
	Root-lesion	Pin	Dagger	Ring	Spiral	Stunt	Cyst	Root-knot
Average number of nematodes per 50g of soil	31	29	7	4	18	4	2	7
Percentage of samples examined in which each nematode type occurred	83	76	52	16	18	7	4	2
Percentage of farms containing each type of nematode	100	95	89	53	58	26	21	5

Table 2. Plant-parasitic nematodes associated with raspberry in Ontario

	Nematode types						
	Root-lesion	Pin	Dagger	Ring	Spiral	Stunt	Lance
Average number of nematodes per 50g of soil	102	65	2	6	36	2	8
Percentage of samples examined in which each nematode type occurred	68	72	28	3	39	7	3
Percentage of farms containing each type of nematode	81	83	38	2	45	7	2

Table 3. Identity of the nematodes associated with grape and raspberry

Common name	Genus and Species	Crop
Root-lesion	Pratylenchus penetrans (Cobb) Filip. & Stekh.	grape and raspberry
	P. neglectus (Rensch) Filip. & Stekh.	raspberry
Pin	Paratylenchus projectus Jenkins	grape and raspberry
Dagger	Xiphinema americanum Cobb	grape and raspberry
Ring	Criconemoides curvatum Raski	grape and raspberry
Spiral	Helicotylenchus canadensis Waseem	raspberry
	H. digonicus Perry	grape and raspberry
	H. platyurus Perry	raspberry
Stunt	Tylenchorhynchus claytoni Steiner	grape and raspberry
cyst	Heterodera trifolii Goffart	grape
Root-knot	Meloidogyne hapla Chitwood	grape
Lance	Hoplolaimus galeatus (Cobb) Thorne	raspberry

free of debris that is present in suspensions obtained by the Cobb method alone. Both cheesecloth alone, and the Kleenex and cheesecloth trapped this debris but permitted the dagger nematode to move through. Excessive handlingof soil was very destructive to X. americanum (Fig. 1A).

The number of X. americanum extracted from soil by the combined method increased'linearly with the weight of the sample in the range of 25 to 200g (Fig. 1B). In large soil samples extraction was less efficient; only 840 dagger nematodes were extracted from soil samples weighing 400g when approximately 1600 were expected (Fig. 1B). The number of X. americanum in Fonthill loam stored at 40°F did not decrease significantly in 4 weeks.

Discussion

In the grape industry in Ontario, plant-parasitic nematodes, in themselves, are not important. Recently, it was proved that X. americanum could transmit grape yellow veinvirus from one herbaceous host to another (7) but it'was not proved that the nematode could transmit the virus from grape to grape. Moreover, grape yellow vein virus does not seem to spread in vineyards (5) nor has it been found in Ontario vineyards (H. F. Dias - personal communication). Xiphinema index Thorne & Allen, which transmits other soil-borne grape viruses, has not been found in Ontario field soils. Apparently X. index cannot survive the winters. At present, dagger nematodes as vectors of soil-borne viruses in Ontario vineyards are not important.

In raspberry production in Ontario, the root-lesion nematode Pratylenchus penetrans (Cobb) Filip. & Stekh. was the most important nematode because it damaged the crop on light sandy soils where nematode populations were frequently large. This species occurred almost as frequently in raspberry in Ontario as in the eastern United States (4). In British Columbia P. penetrans predominated in raspberry plantings on Vancouver Island whereas P. crenatus Loof predominated in the Fraser valley on the mainland (2).

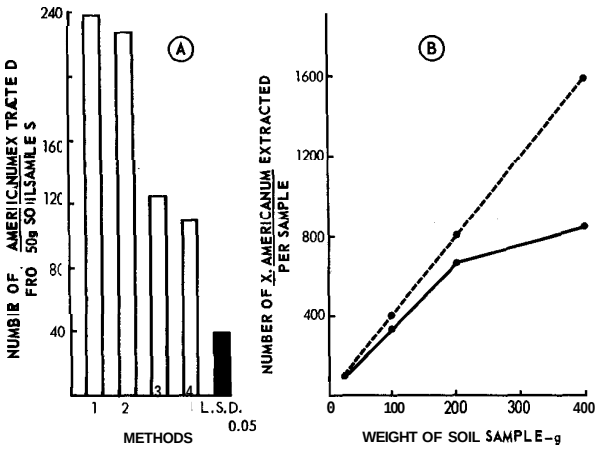

Figure 1 A. Extraction of X. americanum from 50g sail samples; (1) by the Cobb-Oqstenbrinkmethod with only cheesecloth in the hoops; (2) by the Cobb-Oostenbrink method with cheesecloth and Kleenex in the hoops; (3) with method (1) after the soil sample was handmixed; (4) by the unmodified Oostenbrink method. B. Effect of sample size on the extraction of X. americanum from soil by the Cobb-Oostenbrink method. The solid line represents the actual number of X. americanum extracted from the 5 samples of different weights and the broken line represents the number of nematodes expected from these samples.

Lesions were also noted on the roots (2). Pratylenchus neglectus (Rensch) Filip. & Stekh. was noted three times in raspberry in Ontario and infrequently in the eastern United States (4) as well.

Xiphinema americanum was found in many farms where raspberries were grown. This nematode can transmit tomato ringspot virus from cucumber to cucumber (7) but it has not been proven that the nematode can transmit the virus from raspberry to raspberry. Tomato ringspot virus was found in British Columbia (6) but the virus has not been reported in Ontario in recent years. So, at present, X. americanum seems to be of little importance in Ontario raspberry plantings,

In the United States& americanum was found in 60 percent of the raspberry soil samples collected in eight eastern states (4) whereas in Ontario the nematode was found in 28 percent of the raspberry soil samples. Improved cultural practices are essential in the raspberry industry in Ontario.

Greater numbers of X. americanum were extracted from soil by the Cobb-Oostenbrink method when the samples were of 25 to 200g of soil. With heavier samples, a large proportion of dagger nematodes were probably carried to the bottom of the pan before the water was decanted (8) and were therefore lost. In the original Oostenbrink method the soil and endozoic fungi might be responsible for the large losses of X. americanum. The soil on the Kleenex probably obstructed the movement of the dagger nematode and also permitted endozoic fungi to more readily parasitize and kill the nematode. Ectozoic fungi may have been more active as well in such an environment.

Excessive hand mixing of soil destroyed many Xiphinema and this would cause serious reductions in the rate of recovery, regardless of the method of extraction,

The capacity of X. americanum to survive prolonged periods of storage in soil is not unusual as Bergeson and Athow found that the nematode could survive 49 weeks in soil stored at 5°C (2).

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