FUNGI ASSOCIATED WITH THE RUSTY ROOT DISORDER OF MUCK-GROWN CARROTS IN ONTARIO

J.C. Sutton[1]

Abstract

Filamentous fungi recovered from carrot roots do not appear to initiate the rusty root disorder of carrots produced in soils at the Bradford and Keswick marshes of Ontario. Species of Alternaria, Cylindrocarnon, Gliocladium, and Fusarium were recovered frequently from carrots grown in eight fields in the marshes, but in pathogenicity tests failed to produce symptoms of rusty root. No filamentous fungus was recovered consistently from carrot roots with symptoms of rusty root. Groups of fungi found in carrots with and without rusty root were similar, but were recovered less frequently from the nonaffected carrots. Although other factors appear to initiate rusty root, the filamentous fungi found in carrot roots are probably important in the development of the disorder.

Introduction

A disorder of carrots (Daucus carota L. var. sativa DC.), referred to as "rusty root" or "early wilt" (1), has damaged a substantial proportion of the carrots grown in the Bradford and Keswick Marshes of Ontario in recent years. Affected carrots characteristically show numerous rusty-brown lateral roots, profuse development of lateral roots, misshapen or stunted tap roots, and stunting and wilting of the foliage. In the field, carrots with rusty root are often distributed in patches that appear larger in successive growing seasons. parsnip (Pastinaca sativa L.) and dill (Anethum graveolens L.) may also develop symptoms of rusty root. In carrots, similar disorders have- been observed in British Columbia (2), Wisconsin (3), Florida (5), and the Netherlands (8).

The cause of rusty root in carrots grown in Ontario is not known, but some observations have indicated that microorganisms may be important in the initiation or development of the disorder. Steam sterilization of affected muck soil prevented rusty root in carrots subsequently grown in the treated soil (unpublished data). Rusty root did not develop on carrots grown in affected soil exposed to about 5 million rads gamma radiation from a cobalt 60 source, but seriously damaged carrots in control soil (Dr. S.G. Fushtey, personal communication). Elsewhere, carrot disorders similar to rusty root have been attributed to species of Pythium (2, 3, 5).

To examine the possible role of fungi in the initiation and development of rusty root, a diagnostic survey of the fungi associated with roots of carrots grown in Ontario muck soils was carried out.

Materials and methods

carrots were sampled in one field on each of eight farms distributed widely in the Bradford and Keswick marshes on June 16, July 14 and August 14, 1972 (Table 1). Rusty root had appeared during the past 3 years on carrots grown in seven of the eight sampled fields. In field 7 (Table 1) the disease had not been observed in the previous carrot crop grown in 1968. Groups of carrots were lifted with some surrounding soil from 8 to 12 random locations in an area about 40 m diam in each field and bulked. The same areas were sampled at each sampling time. Harvested carrots were stored in plastic bags at about 4 C for 3 to 5 days before plating on agar media. The pH of soil samples was determined by the method of Schofield and Taylor (6). Samples of parsnip and dill with symptoms of rusty root were also collected on July 14 from the Bradford Marsh.

To isolate fungi, pieces of lateral and tap roots of carrot, parsnip, and dill showing various degrees of rusty-brown discoloration were washed in tap water, surface-sterilized, and plated on agar media. About 40 root pieces, 5 mm in length, were cut from each root sample, immersed for 5–10 seconds in 70% alcohol and in 0.5% NaOCl ("Javex"), washed in sterilized distilled water, and plated on water agar, potato dextrose agar (PDA), corn meal agar (Difco), carrot agar, and Martin medium RB-MZ (7). All media except Martin RB-MZ were supplemented with chloretetracycline at 0.1

[1] Assistant Professor, Department of Environmental Biology, Ontario Agricultural College, University of Guelph, Guelph, Ontario.

Table 1. Location, soil pH, and cropping history of carrot fields, and rusty root severity and sowing times for carrot varieties grown in 1972

Field no.	Location of marsh	Soil pH	Cropping history 1969	1970	1971	Carrot var. grown in 1972	Time of sowing in 1972	Rusty root rating[1] August 14, 1972
1	Keswick	5.9	onions	carrots	onions	Hipack	Early May	3
2	Bradford	5.2	lettuce	carrots	onions	Pioneer 318	Mid May	3
3	Bradford	6.0	onions	carrots	onions	Hipack	Mid May	1
4	Bradford	5.7	carrots	onions	carrots	Hipack	Early May	2
5	Bradford	6.2	carrots	carrots	onions	Hipack	Mid May	3
6	Bradford	5.2	onions	carrots	onions	Hipack	Early May	2
7	Bradford	6.3	onions	potatoes	lettuce	Carousel	Late April	0
8	Bradford	6.0	onions	carrots	onions	Hipack	Mid May	2

[1] In area of field sampled; o = none, 1 = light, 2 = moderate, 3 = severe.

mg/ml. Plates were incubated at 22 C, and examined frequently for fungus growth. Additional fungus isolations were made from the samples collected in August by grinding surface-sterilized root segments in a tissue grinder and preparing dilution plates with the above media. The recovery of Pythium from these samples was attempted by using the method described by Mildenhall et al. (3).

For detection of chytridiaceous and mycorrhizal fungi about 0.5 g of lateral roots from samples collected in July were fixed in formalin-acetic-acid-alcohol, cleared in potassium hydroxide, and stained in trypan blue by the method of Phillips and Hayman (4).

The pathogenicity to carrot roots of fungi recovered frequently from the root segments was examined by growing carrots in muck soil that was sterilized and infested artificially with fungus propagules. Muck soil in which carrots did not develop rusty root in 1972 was collected at the Muck Research Station, Bradford, Ontario, in September, 1972. The soil was autoclaved at 121 C for 30 min, held at room temperature for 2 days, then reautoclaved for an additional 30 min. Spores of three isolates of Cylindrocarpon destructans (Zins.) Scholten, three isolates of Fusarium solani (Mart.) Sacc., two isolates of Gliocladium sp. and two isolates of Alternaria alternata (Fr.) Keissler were recovered in water from cultures grown on malt-extract agar. For each isolate, 10^4 and 10^5 spores were added per g of separate samples of the autoclaved muck soil. In spore suspensions of Cylindrocarpon and Fusarium both macroconidia and microconidia were present. Noninfested and infested soil samples were placed in 16 oz plastic cylinders, and seeds of carrot 'Gold Pak' were sown. The carrots were grown at 8–12 C for 4 weeks and at 16–20 C for a further 4 weeks; the photoperiod was 16 h and light intensity 200–240 $uE/m^{-2}/sec^{-1}$. The carrot roots were then washed and examined on a dissecting microscope for disease symptoms.

Results

Rusty root symptoms were present on carrot roots collected at each time of sampling from each field except field 7 where no rusty root was found (Table 1).

The fungus genera recovered frequently from the root segments of carrots with symptoms of rusty root were Alternaria, Cylindrocarpon, Fusarium, Gliocladium, Mucor, and Penicillium (Table 2). Species of Penicillium were abundant at each time of sampling. Fusarium and Mucor were recovered commonly only in June and Gliocladium only in July. Cylindrocarpon was the predominant fungus found in July and August, and Alternaria appeared frequently in roots collected in August.

Genera of fungi found in roots collected from the various fields where rusty root developed were similar, but there were quantitative differences, especially in the August samples. In these samples, Cylindrocarpon was the dominant funqus recovered in roots from four fields, Alternaria in roots from two fields and Penicillium in roots from one field.

The fungi recovered from comminuted root segments were generally similar to those found in intact segments, but yeasts and species of Penicillium were notably abundant.

Table 2. Frequency of recovery on agar media of fungi from segments of rusty and non-rusty carrot roots collected at different times in the growing season

Fungus genus	Percent root segments[1]					
	June		July		August	
	Rusty[2]	Non-rusty[3]	Rusty	Non-rusty	Rusty	Non-rusty
Alternaria	0	0	3	0	16	83
Chaetomium	2	0	6	0	2	0
Cladosporium	0	0	6	0	4	5
Cylindrocarpon	2	0	30	12	42	8
Emericellopsis	2	0	4	5	0	0
Fusarium	34	25	10	0	8	8
Gliocladium	0	0	14	0	0	0
Mucor	24	12	10	0	0	0
Papulaspora	0	0	4	0	0	0
Penicillium	27	5	18	0	16	8
Pythium	6	8	4	0	2	0
Rhizoctonia	9	0	5	0	9	4
Rhizopus	6	0	0	0	0	0
Stemphylium	0	0	0	0	2	0
Trichocladium	4	5	0	0	0	0
Trichoderma	8	0	4	0	0	0
Verticillium	0	0	6	0	0	0
Not identified	0	0	4	0	2	0

[1] Percent root segments placed on five selective agar media yielding the fungi indicated.

[2] Samples collected in seven fields where rusty root appeared.

[3] samples collected in one field where no rusty root appeared.

There were no consistent qualitative differences in the fungi found in segments from rusty- and healthy-appearing portions of the roots from affected carrots.

Fungi found in carrots without rusty root were similar to those with rusty root, but were fewer in number and were usually recovered less frequently (Table 2).

Isolates of Fusarium, Cylindrocarpon, and Alternaria were identified, respectively, as F. solani (Mart.) Sacc. (18 isolates), C. destructans (Zins.) Scholten (15 isolates) and A. alternata (Fr.) Keissler (14 isolates). The isolates were from carrots collected in most of the eight fields sampled.

In pathogenicity tests, neither Fusarium, Cylindrocarpon, nor Gliocladium produced visible symptoms of disease in carrot roots. However, all were reisolated on PDA from 30-60% of surface-sterilised segments of the carrot roots grown in soil infested with 10^4 or 10^5 propagules/g.

Chytrids were numerous in roots collected in July from most fields, but were relatively few in roots from fields 3 and 5 (Table 1). In contrast, 50-100% of the lateral roots of carrots from fields 3 and 4 were mycorrhizal,

whereas carrots from the remaining fields were only 1-12% mycorrhizal.

Fungi recovered from affected parsnip roots were Alternaria, Fusarium, Rhizoctonia, Mucor, and Pythium, and from the roots of dill, Cylindrocarpon, Alternaria, and Penicillium.

Discussion

Filamentous fungi recovered from carrot roots do not appear to initiate the rusty root disorder of carrots produced in the Bradford and Keswick Marsh soils. No fungus species was recovered consistently from carrots that developed rusty root. There were marked differences in the kinds and frequency of fungi recovered from affected carrots harvested at various stages of development and from different fields. Several of the fungi found frequently in carrot roots with the rusty appearance were also found in healthy-appearing roots and in the roots of carrots grown in a field where rusty root did not develop. None of the fungi commonly found in affected roots produced symptoms in carrots grown in sterilized soil infested with large numbers of propagules of these fungi, and under environmental conditions conducive to rusty root development in carrots grown in affected soil.

Pythium was not found to be important in the [illegible]ion and development of rusty root which may therefore differ from similar disorders of carrots described previously (2, 3, 5). The fungus was recovered infrequently even when specialized techniques for Pythium isolation were used.

Although other factors appear to initiate rusty root, the filamentous fungi found in carrot roots are probably important in the development of the disorder. There was an abundance of F. solani, C. destructans, and other fungi in carrot roots showing many stages of rusty root development. The ability of several of these fungi to colonize carrot roots which subsequently remain symptomless indicates a possible involvement early in rusty root development.

Crop sequences may influence the prevalence and severity of rusty root. In the seven fields where rusty root appeared in 1972, carrots and onions had each been grown during 1 or 2 of the previous 3 years (Table 1). In field 7, where no rusty root was found, carrots had not been grown during the previous 3 years, and onions were last grown in 1969. Carrot var. Carousel grown in field 7 is known to be susceptible to rusty root (the late C.C. Filman, personal communication).

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