

CULTIVAR-ISOLATE INTERACTION IN INFECTION OF SUNFLOWER SEED BY VERTICILLIUM DAHLIAE

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INTRODUCTION

Verticillium wilt or leaf mottle of sunflowers induced by *Verticillium dahliae* Kleb. is known to occur in most countries where sunflowers are grown. Resistance to Verticillium wilt has been found in cultivated sunflowers and in wild *Helianthus* spp. Isolates of the pathogen from various hosts and from different areas differ in their virulence on sunflowers. The pathogen may be carried on and in sunflower seed, but seed infection is erratic. Environmental factors such as temperature and photoperiod have been shown to affect the development of Verticillium wilt on sunflowers, but little is known about the factors which influence ability of the pathogen to infect the seed. This paper reports differences in infection of seed of two sunflower lines by two isolates of *V. dahliae*.

MATERIALS AND METHODS

Seed of the Verticillium wilt susceptible line CM 162 and the resistant line CM 144, both developed at the Agriculture Canada Research Station, Morden, Manitoba, was provided by Dr. W. Dedio and Dr. J.A. Hoes of that station. Culture VNur of *V. dahliae* was isolated from sunflowers with typical leaf mottle symptoms in the wilt nursery at Macdonald College. Culture VM was isolated by Dr. Hoes at Morden.

Plants were grown in a pasteurized soil mixture, with complete fertilizer added, and were thinned to 1 per pot after emergence. Effects of pot size, plant age at time of inoculation, and point of

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inoculation along the stem were investigated, but as both sunflower lines and both cultures were used in most experiments, the data were combined where possible. Plants were grown in a greenhouse thermostatically controlled at about 22° C day and 18° C night, with daylight supplemented by cool white fluorescent tubes, or in cabinets maintained at $24 \pm 1^\circ$ C day and $18 \pm 1^\circ$ night temperature, 14 hr day, and measured light intensity of 295 to 297 micro Einsteins provided by cool white VHO fluorescent tubes supplemented with incandescent bulbs.

Plants were inoculated with a suspension of spores washed from the surface of agar cultures, strained through gauze, and injected with a hypodermic syringe fitted with a 27 gauge needle, at predetermined points along the stem of plants of different ages. Sterile water was injected into control plants. At the conclusion of the experiments, sections of stems were cut from the top internode of each plant, surface sterilized, and disks cut from them were plated on potato dextrose agar (pda). Ten seeds were removed from each capitulum, surface sterilized, and plated on pda.

RESULTS

Plants of CM 162 and CM 144 grown in various sized pots were inoculated with VNur or VM injected into the fourth internode 46 days after sowing. One replicate was grown in the controlled greenhouse, 3 replicates were grown in controlled environment cabinets. The experiment was concluded when the seeds appeared ripe, 98 days after sowing.

Verticillium grew from most of the disks plated from inoculated plants of CM 162, and from discs of many inoculated plants of CM 144 (Table 1). *Verticillium* grew from some seeds of about 30% of inoculated CM 162 plants, and from some seeds of about 15% of inoculated CM 144 plants (Table 1). It did not grow from stem disks or seeds of control plants.

In another experiment, plants of CM 162 and CM 144 were inoculated with VM at 46 or 65 days after sowing. Inoculum was injected into the base of the stem, halfway along the stem, or at the base of the capitulum. Plants were grown in a controlled environment cabinet until the seeds appeared ripe, 98 days after sowing.

Verticillium grew from the disks of most inoculated plants of CM 162 except those injected at the base of the capitulum, from some seeds of about 50% of the plants inoculated at 46 days, and about

TABLE 1

Effect of cultivar and of culture on establishment of Verticillium dahliae in sunflower stems and seeds

Cultivar	Verticillium culture	No. of plants			No. of seeds	
		Plated	Yielding Vert.		Plated	Yielding Vert.
			from stem disks	from seeds		
CM 162	VNur	24	18 ²	4 ³	240 ³	13
	VM	24	22	12	224 ⁴	49
CM 144	VNur	24	20	6	240	9
	VM	21 ¹	11	1	210 ¹	1

¹ 3 plants were lost before the experiment was completed.

² 10 disks were plated from the top internode of each plant.

³ 10 seeds were plated from each plant.

⁴ 3 plants had only 6, 6, and 2 seeds respectively per head. All 14 seeds yielded *Verticillium* colonies.

Verticillium did not grow from any disks or seeds of control plants injected with water.

10% of those inoculated at 65 days (Table 2). *Verticillium* gre from disks of some plants of CM 144 inoculated at 46 days, from disks of fewer plants inoculated at 65 days, and from only one seed of each of two plants (Table 2). It did not grow from disks or seeds from control plants.

TABLE 2

Effect of cultivar and plant age at inoculation on establishment of Verticillium dahliae (isolate VM) in sunflower stems and seeds

Cultivar	Age at inoculation (days)	No. of plants			No. of seeds	
		Plated	Yielding Vert.		Plated	Yielding Vert.
			from stem disks	from seeds		
CM 162	46	9	71	5 ²	73 ³	15
	65	9	6	1	90	1
CM 144	46	9	6	2	90	2
	65	9	4	0	90	0

¹ 10 disks were plated from the top internode of each plant.

² 10 seeds were plated from each plant.

³ 2 heads contained only 1 seed and 2 seeds respectively.

Verticillium did not grow from any disks or seeds of control plants injected with water.

DISCUSSION

CM 162 is very susceptible to *Verticillium* wilt. Symptoms induced on it by isolate VM appear slightly sooner and are more severe than those induced by VNur, and VM apparently infects a higher proportion of seeds than VNur does.

CM 144 is resistant to *Verticillium* wilt. Relatively few plants develop symptoms even when inoculated by injection of spores into the stem, indicating that resistance is not confined to the roots. The pathogen can survive in stems of inoculated plants, however, and can infect the seeds in some of them. Seed infection is even more limited than in CM 162; apparently the resistance of CM 144 is operative at this level as well. The fact that seed infection occurs at all is disquieting, however, as it may mean that seed of resistant varieties may also disseminate *Verticillium*. Whether or not seed infection can occur in resistant plants in the field must be investigated.

It is interesting that isolate VNur, although less aggressive than VM on susceptible sunflowers, can be reisolated from a higher proportion of stems of symptomless resistant CM 144 plants than can VM, and from an appreciably higher proportion of seeds. The resistance of CM 144 appears to be isolate specific to some extent, and more effective against *Verticillium* isolated in the area where CM 144 was selected than from other regions. This difference in reaction to the two cultures appears to be more pronounced at the level of the seed than in the stem.

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