

*Alternaria helianthi*: A Pathogen of Sunflower New to Minnesota

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**ABSTRACT**

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*Alternaria helianthi* on oilseed sunflowers is reported for the first time in Minnesota. The fungus was first found associated with a severe seedling blight affecting more than 280 ha of sunflowers in June 1980. The fungus was later found in lesions on stems and leaves of plants in all 17 sunflower fields in nine Minnesota counties surveyed in September 1980. Greenhouse studies showed that stem, leaf, and petiole tissues are susceptible to infection by conidia of *A. helianthi*. All 27 entries of the 1980 National Oilseed Nursery were susceptible to *A. helianthi*.

Additional key words: *Helianthus annuus*

In 1979 and 1980 a seedling blight of oilseed sunflowers was noted in western Minnesota. After heavy rains in the first two weeks of June 1980, symptoms appeared on young sunflowers. The plants had dark brown, oval to diamond-shaped, slightly sunken lesions on the stems at or slightly below the soil line. Some of the plants with stems completely girdled by lesions were killed. Foliage on affected plants usually was stunted and often completely blighted, but no lesions were observed on the leaves.

Laboratory and greenhouse tests were done to establish the identity of the causal organism as *Alternaria helianthi* (Hansf.) Tubaki and Nishihara and to investigate the effects of inoculum density and plant age on disease development. A survey was made in September 1980 to determine its prevalence in commercial sunflower fields in Minnesota.

**MATERIALS AND METHODS**

Sunflower stem tissue with lesions was dipped in 1% NaOCl, rinsed in sterile water, and transferred to water agar in petri plates. Isolates were cultured on Difco (Detroit, MI 48232) potato-

dextrose agar (PDA), Difco corn meal agar, Difco Czapek's agar, and Difco lima bean agar at 24 and 27 C in the dark, or on PDA and lima bean agar at 24 C under 12 hr of fluorescent light per day. Size and septation of conidia were determined for three monoconidial isolates obtained from different fields. Spores for measurement were taken from lesions on leaves of sunflowers that had been spray-inoculated in the greenhouse or from colonies grown on PDA for 11 days at 24–27 C with 12 hr of fluorescent light per day.

Inoculum for greenhouse studies was obtained from cultures growing on PDA at room temperature or at 24 C under 12 hr of fluorescent light per day. Conidia were suspended in sterile distilled water, and the suspension was strained through two layers of cheesecloth. Sunflower cultivar DO844 was used in all tests unless otherwise indicated. A steam-pasteurized greenhouse soil and sand (2:1) mix and 10.2- or 12.6-cm diameter clay pots were used in all experiments.

Relationships between spore concentration, plant age, and symptom expression on all plant parts were examined in a greenhouse at 22–28 C. The effects of soilborne inoculum were investigated by infesting the soil with  $10^2$ ,  $10^3$ ,  $10^4$ ,  $2 \times 10^4$ , or  $10^5$  conidia per pot, 11

days after sunflowers were planted, three per pot. Spore suspensions (30 ml) were poured over the surface of the soil in each pot and covered with 5–10 mm of soil. A completely random design was used. The effects of plant age and inoculum density were investigated by infesting soil as described with  $0.5 \times 10^3$ ,  $10^4$ , or  $4 \times 10^4$  conidia per pot at 0, 2, 4, 6, or 8 days after planting. A randomized complete block design was used, with blocks separated in space on a greenhouse bench. At the time of inoculation, seedlings planted 4 days earlier were emerging from the soil.

The effects of plant age and inoculum concentration on disease development on foliage were investigated by spray-inoculating sunflower plants to runoff with  $0$ ,  $10^2$ ,  $10^3$ , or  $10^4$  conidia per milliliter 10, 12, 14, 16, or 32 days after planting. Each pot was covered with a plastic bag for 24 hr. In addition, all 27 cultivars of the 1980 National Oilseed Nursery (which included some confectionary types) were grown, five plant per pot, and spray-inoculated 14 days after planting with a suspension of  $5 \times 10^3$  conidia per milliliter.

One control plant in each pot was sprayed with water and covered with a smaller bag before the rest of the plants were inoculated with the spore suspension. The larger bags were removed after 24-hr incubation. The smaller bags were removed from control plants when the foliage of the spore-inoculated plants had dried.

A survey of 17 Minnesota sunflower fields for *A. helianthi* was conducted during September 1980. Isolations were made from surface-disinfected suspect leaves and stems that showed symptoms.

**RESULTS**

Most of the isolations from blighted seedlings yielded the same fungus, which was subsequently identified as *A.*

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**Table 1.** Dimensions of conidia of three isolates of *Alternaria helianthi* from Minnesota

Isolate	Source of spores	Spore dimensions ( $\mu\text{m}$ ) <sup>1</sup>
BS186-80	Leaves	37–97 (67.8 $\pm$ 1.7) $\times$ 12–23 (16.3 $\pm$ 0.4)
BS253-80	Leaves	20–92 (67.8 $\pm$ 1.2) $\times$ 5–18 (14.8 $\pm$ 0.3)
BS184-80	Potato-dextrose agar	26–128 (85.3 $\pm$ 1.1) $\times$ 12–23 (21.4 $\pm$ 0.2)

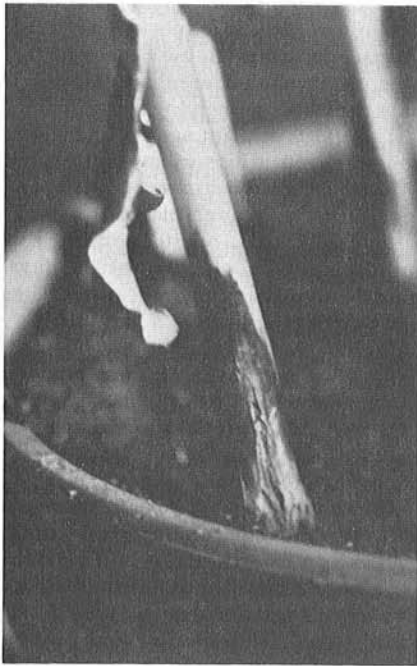
<sup>1</sup> Ranges of length  $\times$  width of 100 randomly selected conidia. Numbers in parentheses indicate mean values  $\pm$  standard deviation.

*helianthi* (8). Conidial dimensions (Table 1) ranged slightly larger and smaller than those reported by Tubaki and Nishihara (8) and Islam and Marić (4), which were  $40\text{--}110 \times 13\text{--}28 \mu\text{m}$  and  $25\text{--}125 \times 12.5\text{--}35 \mu\text{m}$ , respectively. The number of

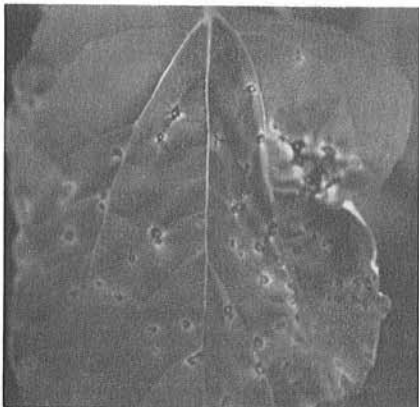
**Table 2.** Effects of initial numbers of *Alternaria helianthi* conidia added to soil on severity of sunflower basal stem necrosis

Days after soil infestation	Stem necrosis (%)				
	Conidia per pot				
	$10^2$	$10^3$	$10^4$	$2 \times 10^4$	$10^5$
20	0.8 <sup>a</sup>	5.5	22.0	51.6	89.4
32	0.5	10.6	45.3	78.1	95.0

<sup>a</sup>Values represent the visual estimate of the percentage of the circumference of the plant stem at the soil line that was necrotic and/or cracked. Each number is the mean of five pots, each pot containing three plants.



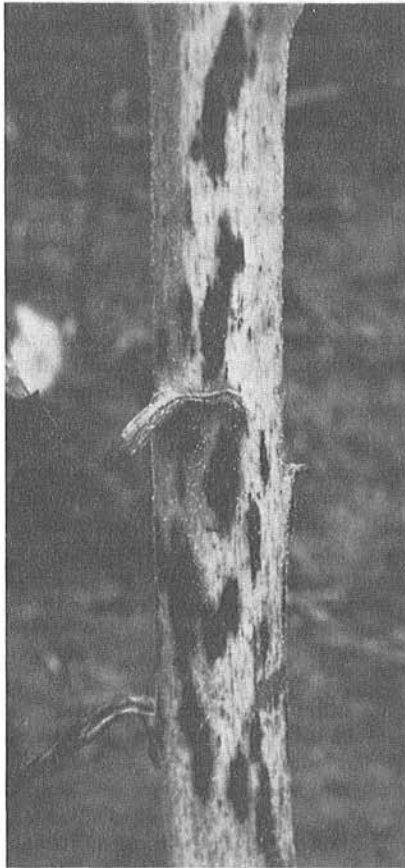
**Fig. 1.** Basal stem symptoms on sunflower seedling grown in soil infested with conidia of *Alternaria helianthi*.



**Fig. 2.** Leaf symptoms on sunflower that was spray-inoculated with conidia of *Alternaria helianthi*.

transverse septa in conidia ranged from 0 to 11 in our isolates, which essentially agrees with other published values (4,6,8). Longitudinal septa were rare in conidia from plant material but were more common in old PDA cultures. Constrictions at transverse septa were also more common and pronounced in conidia from old cultures.

Radial growth was greatest with Czapek's and lima bean agar and least with corn meal agar. Sporulation (conidia per colony) was greatest on PDA. Colonies of *A. helianthi* grown



**Fig. 3.** Symptoms on a sunflower stalk naturally infected in the field by *Alternaria helianthi*.

under fluorescent lights sporulated more profusely, and after 15 days the mean diameter of the colonies was 1.6 times greater than that of colonies grown in the dark.

Sunflowers were susceptible to infection by spores of *A. helianthi* in soil. Small rust-colored spots appeared on infected plants at the soil line within 3 days of soil infestation. Splitting of the basal stem was evident after 4 days and often extended up from the soil line. The basal stem of severely infected plants often became completely necrotic (Fig. 1). Basal stem symptoms observed in greenhouse studies were similar to those occurring naturally on seedlings in the field. *A. helianthi* was reisolated consistently from surface-disinfected tissue with these symptoms. Severity of the basal stem necrosis ranged from less than 1% to more than 90% with a 1,000-fold difference in inoculum concentration (Table 2).

Plant age at time of inoculation influenced infection by *A. helianthi* in infested soil (Table 3). Disease severity was greatest in plants inoculated 6 and 8 days and least in plants inoculated 0 and 2 days after planting (Table 3). Disease severity on the youngest two groups was less, perhaps because they were not in direct contact with inoculum at the time of inoculation. Necrotic lesions appeared on cotyledons of sunflowers that were 4 days old at inoculation because these tissues were at the soil surface at the time of inoculation.

Stems and leaves of all sunflower plants tested at different ages were susceptible to infection by *A. helianthi* conidia. Many small brown spots appeared on leaves and petioles within 24 hr of spray-inoculation. Many spots developed into irregular dark lesions, often with grey centers and often surrounded by a chlorotic halo (Fig. 2). Severe infections resulted in leaf distortion and blight, the latter being most common when young petioles were infected. Angular brown lesions were common on stems and petioles, and large, circular, sunken necrotic lesions appeared

**Table 3.** Relationship between sunflower seedling age, *Alternaria helianthi* inoculum density, and sunflower basal stem necrosis

Plant age at time of infestation (days)	Days after soil infestation	Stem necrosis (%) in pots			
		Conidia per pot			
		0	$5 \times 10^3$	$10^4$	$4 \times 10^4$
0	18	0.4 <sup>a</sup>	5.0	3.3	7.5
	24	0.4	5.0	6.5	21.8
2	18	0.0	0.0	0.4	4.2
	24	0.0	1.4	1.0	16.6
4	18	0.4	1.6	11.7	29.1
	24	0.9	3.0	22.1	55.0
6	18	0.0	10.4	23.2	82.4
	24	0.0	11.6	33.7	92.5
8	18	0.0	6.9	22.1	50.1
	24	0.0	10.0	34.1	65.4

<sup>a</sup>Values represent the visual estimate of percentage of the circumference of plant stem at the soil line that was necrotic and/or cracked. Each number is the mean of four pots; each pot contained three plants.

on cotyledons.

All 27 cultivars of the 1980 National Oilseed Nursery were susceptible to infection by foliar inoculations of *A. helianthi* conidia. No obvious response differences were seen among the cultivars.

*A. helianthi* was isolated from sunflower stems and leaves from all 17 fields surveyed in nine Minnesota counties. *A. helianthi* was consistently isolated from leaves with round to irregular black lesions, some with chlorotic halos. The fungus was isolated from black, elliptical, longitudinal stem lesions ranging from 2 to more than 70 mm long (Fig. 3). In some fields most stems had more than 30 lesions.

## DISCUSSION

*A. helianthi* has been known to be a pathogen of sunflowers since 1943 (3). In many countries it causes severe stem and foliar disease problems (2,4,6,8), but it was not known to be present in North America before 1978 (9). Recently the organism was seen in Mississippi (7), Wisconsin (C. R. Grau, *personal communication*), and North Dakota (V. L. Jons and T. Gulya, *personal*

*communication*). The symptoms in these locations and in Minnesota were leaf, petiole, and stem lesions that threatened full development of sunflower heads.

Our paper is the first known report of a seedling blight caused by *A. helianthi*. The seedling blight was a major concern in Minnesota in 1980, since 280 or more ha of sunflowers had to be replanted.

The seedling blight was found on both volunteer and cultivated sunflowers, but only in fields where sunflowers had been planted in the previous season. Our studies indicate that *A. helianthi* has a major role in the seedling blight problem. It is not known if the fungus was the sole pathogen causing blight in all fields where blight was observed because *Alternaria zinniae* Pape can cause a similar disease (5).

Little is known about the sources of primary inoculum, but infested plant debris may be an important overwintering medium. However, no correlation was noted between previous cropping history and the presence of foliar, stem, or head infections on older plants. *A. helianthi* has been detected on seed in our laboratory and by other researchers (1,2) but the relationship between seed contamination and the occurrence of all forms of the disease in the field has yet to be determined.

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