Some Biotic and Climatic Factors Influencing Sporadic Occurrence of Sunflower Downy Mildew

D. E. Zimmer

Research Plant Pathologist, Agricultural Research Service, U.S. Department of Agriculture, Fargo, North Dakota 58102.

Contribution from Agricultural Research Service, U.S. Department of Agriculture, and the North Dakota Agricultural Experiment Station as Journal Series Paper No. 561.

Accepted for publication 27 January 1975.

ABSTRACT

The sporadic occurrence of systemic downy mildew of sunflower, *Helianthus annuus,* incited by *Plasmopara halstedii,* was closely associated with seedling age and rain. Seedlings become increasingly resistant with age, and the rapidity of symptom development was a function of seedling age at inoculation. The period of maximum susceptibility to systemic infection was as short as 5 days in a 22-25 C greenhouse. In the field, where mean air temperatures during seed germination and emergence were as low as 13.2 C, plants remained susceptible to systemic infection for at least 15 days. With enough rain during this period to provide free-soil water for only a few hours, systemic infection was likely on infested land. Any managerial practice that supports rapid seedling emergence, and reduces the chance of free-soil water during the period of susceptibility, should reduce losses from systemic downy mildew.

When seed harvested from plants with systemic symptoms was planted, systemically infected plants were not produced. However, more of these plants had root and basal-stem infection than did plants from seed harvested from healthy plants.

Phytopathology 65:751-754

Additional key words: epidemiology, *Helianthus annuus, Plasmopara halstedii.*

Downy mildew of sunflower (*Helianthus annuus* L.), incited by *Plasmopara halstedii* (Farl.) Berl. & De T., is a potentially destructive disease. Downy mildew manifests itself as damping off (4, 14), local foliar lesions (2, 8, 13), root and basal-stem infections (1, 7, 8, 12), and systemic symptoms (1, 4, 8, 10). Systemic infection is the most destructive, occasionally causing 50% yield reduction in some fields on the heavy clay soils of the Red River Valley of North Dakota and Minnesota (10). The sporadic occurrence of systemic downy mildew from field-to-field, from year-to-year, and from planting-date to planting-date suggests that climate strongly influences its occurrence (10).

Mean temperatures during the normal planting period are well within the range that supports *P. halstedii* activity (4, 8). Rain pattern and intensity vary among years, locations, and dates of planting and may influence the sporadic occurrence of systemic downy mildew. Such a relationship was suggested by Zimmer (10).

Plants with pronounced systemic symptoms occasionally produce abundant seed (10, 15). Seed from such plants harbor the fungus, but seldom give rise to plants with systemic symptoms (1, 3, 7). Plants growing from infected seed, although showing no visible systemic symptoms, have been reported to produce infected seed and disseminate the pathogen (1).

The biology of *P. halstedii* on annual sunflowers has been studied by several workers (3, 4, 6, 8, 9). Infection resulting in systemic symptoms occurs mainly through roots (3, 6) and hypocotyls (1); infection through leaves normally produces local lesions (1, 8, 11). Infection through apical buds has been reported to give a high incidence of systemically infected plants (1). Although zoosporangia are produced in great numbers on systemically infected plants, their role is of doubtful importance in epidemiology (4, 8).

The following study was undertaken to obtain biotic and climatic information relative to the sporadic occurrence of systemic downy mildew in the major sunflower-producing area of the USA.

MATERIALS AND METHODS.—*Seed transmission and dissemination (field trials).*—Fully formed achenes harvested from 20 field-grown 'Peredovik' sunflower plants with pronounced symptoms of systemic downy mildew, and achenes from 20 healthy plants were planted in the spring of 1971 and 1973 on land with no previous history of sunflower cropping. The percentage of emergence of 100 planted seed from each plant, and the relative vigor of the emerged seedlings, were determined both years. The plants were observed weekly for systemic downy mildew symptoms. In 1973, about 10% of the plants from each row were dug at flowering, and examined for basal-stem and root infections.

Date of planting (field trials).—About 1,000 achenes of downy-mildew-susceptible cultivar Peredovik were planted in replicated trials at each of four dates in 1971, 1972, and 1973 on land known to be uniformly and heavily infested with *P. halstedii.* The percentage of seedlings with systemic symptoms was determined 6 and 12 weeks after planting. Daily rain and mean temperature were recorded and correlated with the counts of systemically infected seedlings.

Seedling age and susceptibility to systemic infection (greenhouse trials).—Zoosporangia for greenhouse trails traced from a collection made from cultivar Peredovik near Fargo, North Dakota, in the summer of 1971 and maintained on susceptible cultivars in the greenhouse. Unless specified, inoculum consisted of 10,000 zoosporangia/ml of distilled water, determined with a hemocytometer.

Four inoculation methods were used in each of the three trials. In the first method, seedlings of Peredovik were grown in flats of sterile vermiculite, removed at various ages, and inoculated by the whole-seedling method of Zimmer and Kinman (14) and Cohen and Sackston (1). After inoculation, the seedlings were gently (but thoroughly) washed in running tap water,

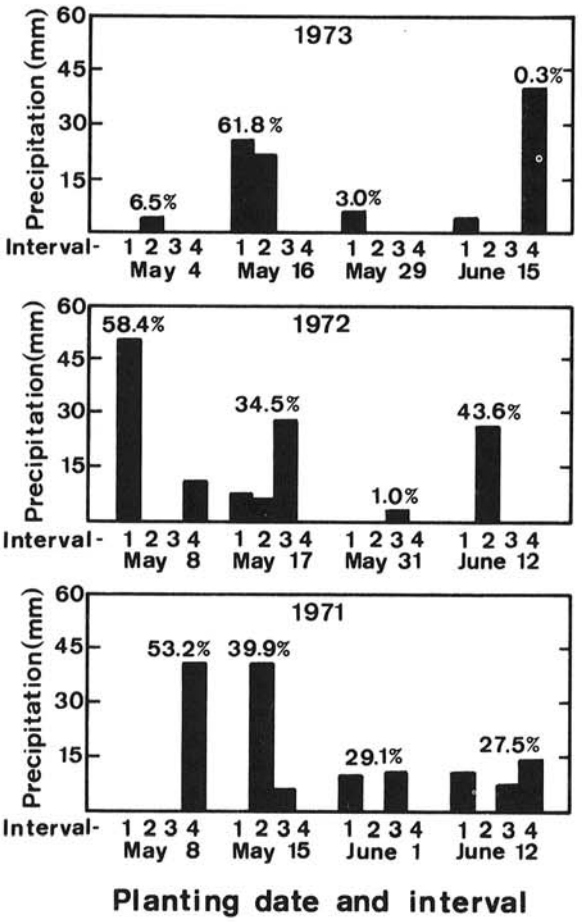

Fig. 1. The percentage of sunflower, *Helianthus annuus*, plants with systemic symptoms of downy mildew and its relationship to the occurrence and distribution of rainfall during four intervals after planting in 1971, 1972, and 1973. Intervals 1, 2, 3, and 4 are 3-5, 6-8, 9-11, and 12-14 days after planting, respectively.

TABLE 1. Percentage of sunflower *(Helianthus annuus)* plants with systemic downy mildew following inoculation of seedings with *Plasmopara halstedii*

Seedling age (days after seeding)	Inoculation method[a]		
	Soil drench	Foliage spray[b]	Whole seedling
3	100	...	100
4	100	...	100
5	87	100	98
6	12	98	85
7	3	91	51
8	0	14	3
9	0	8	1
10	0	0	0
11	0	0	0
12	0	0	0
13	0	0	0

[a]Data were averaged over three experiments. Each experiment contained 100 seedlings.

[b]Seedlings did not emerge until 5 days after seeding; thus foliage inoculation was not possible until after that time.

transplanted to a sterile potting mixture in 15.3-cm diameter clay pots, and observed daily for systemic symptoms.

In the second and third methods, Peredovik seedlings of various ages were grown in a mixture of vermiculite and sterile soil (1:1, v/v) in flats or pots. They were inoculated in place either by drenching the soil around the seedlings with inoculum, or by spraying the entire foliage to wetness with inoculum. Care was exercised during drenching to avoid inoculum contact with the foliage, unfolded cotyledons, or emerged portion of the hypocotyl. After inoculation, the flats and pots were placed overnight in a 20-C mist chamber, returned to greenhouse benches (22-25 C), and observed daily for systemic symptoms.

In the fourth method, seedlings were inoculated at various ages by placing on their cotyledons and leaves one or more 5-mm filter paper disks saturated with 50,000 zoosporangia/ml, as described by Cohen and Sackston (1) or by placing a drop of inoculum directly on the apical meristem. After inoculation, the plants were placed in a 20 C humidity chamber for 24 hours, returned to greenhouse benches, and observed daily for systemic symptoms.

In all greenhouse trials, systemic infections were verified by placing suspect plants overnight in a 100% relative humidity cabinet at 20 C. The fungus sporulated on the under-surface of the true leaves, and on some cotyledons of systemically infected plants.

RESULTS AND DISCUSSION.—*Seed transmission and dissemination (field trials).*—Achenes from plants with pronounced systemic mildew symptoms were much smaller than achenes from healthy plants (5). Thus, it was not unexpected when they gave 20% less emergence in 1971 and 16% in 1973, and produced less-vigorous seedlings. The early differences in vigor did not persist past 5 weeks. The percentage of plants with systemic symptoms did not differ significantly, whether the plots were seeded with achenes from systemically infected plants or with achenes from healthy plants. The low level of systemically infected plants (2.0% in 1971 and 1.2% in 1973) was attributed either to a resident population of *P. halstedii* in the fields before planting or to wind-borne zoosporangia. Other workers (2, 3, 7) have found little evidence that plants with systemic symptoms arise from planting infected seed. Thus, it is highly unlikely that seed transmission can account for the dramatic outbreaks of systemic downy mildew in fields not previously planted to sunflowers and often separated by several miles from fields previously cropped to sunflowers. Wind-borne zoosporangia from mildew-infected volunteer plants in spring-grain fields, summer-fallowed fields, and wayside areas probably were more responsible for such outbreaks.

Surveys in the spring of 1971 and 1972 revealed that more than 20% of the volunteer seedlings in some fields were systemically mildew-infected and produced abundant zoosporangia, coinciding with the emergence of seeded sunflowers in surrounding fields. Zoosporangia are relatively thin-walled and short-lived (3, 4, 8). However, high humidity and wind often occur before rain squalls. Therefore, it seems likely that zoosporangia could travel several miles, be deposited around emerging seedlings, germinate, and produce zoospores that, in turn, could cause systemic infection under favorable

conditions. Delanoe (3) reported that zoosporangia can travel at least 1,500 m and remain viable. Under ideal conditions this distance may be even greater.

Root and basal-stem infection described by Cohen and Sackston (1) and Zimmer (12) was observed on 25.5% of the plants removed from the 1973 plots planted with seed from systemically infected plants, but on only 3.0% of the plants in plots whose seed was from healthy plants. Much infection centered around the location of the pericarp of the achene that had been sloughed in the germination and emergence process. As suggested by Novotelnova (7) and others (2, 4, 5), *P. halstedii* appears able to disseminate and establish in new areas of sunflower production without the occurrence of systemically infected plants. Thus, as proposed by others (2, 4), adequate quarantine is needed when seed are imported from downy-mildew infested areas into noninfested areas. Quarantine would be particularly important when seed are imported from areas of the world where highly virulent strains of *P. halstedii* exist (13).

Date of planting (field trials).—Early, mid-season, and late planting did not consistently affect the relative incidence of systemically infected plants (Fig. 1). The data failed to substantiate that early planting has a much lower incidence of systemic mildew than later planting. The major influence in the occurrence of systemic infection appears to be the relative amount of rain near seeding time. Rain before seeding does not appear to affect the incidence of systemic mildew directly. The amount of rain 3-14 days after seeding was important in the relative occurrence of infection giving rise to systemic symptoms. If this period was relatively free of rain, little or no systemic symptoms developed. However, if this period received enough rain to establish free-soil water, the incidence of systemic infection was high. In general, the heavier the rainfall during the period, the greater the incidence of systemic infection. There is some evidence, however, that this generality may be modified by temperatures within the interval, especially if rain falls mainly on the fringe of the period. For instance, 35 mm of rain fell 12-14 days after seeding on both 8 May 1971 and 15 June 1973 (Fig. 1). The incidence of systemic downy mildew in the plots planted 8 May 1971 was 53.2%, compared with only 0.3% in plots planted on 15 June 1973. The mean air temperature during the 14 days after 8 May 1971 was 13.2 C, which was 6.1 C cooler than the mean air temperature for the same period after 15 June 1973. The higher mean air temperatures after the 15 June 1973 planting date is assumed to have shortened the period of maximum susceptibility to systemic infection, so that the seedlings were no longer susceptible to systemic infection when it rained.

Seedling age and susceptibility to systemic infection (greenhouse trials).—Seedlings become increasingly resistant with age to infection giving rise to systemic symptoms, regardless of the method of inoculation (Table 1). Seedlings more than 10 days old at inoculation were highly resistant to infection resulting in systemic symptoms, even though they were grown until flowering. However, a high percentage of these seedlings showed hypocotyl lesions and root infection. Seedlings inoculated by the whole-seedling method and by complete-foliage spraying were susceptible to systemic infection for 2-3 days longer than seedlings inoculated by soil drenching. Maximum susceptibility seemed to extend from the time of radicle emergence until cessation of hypocotyl elongation.

Resistance to systemic infection probably is associated with the differentiation and elongation processes within the hypocotyl. This is an attractive hypothesis, because Cohen and Sackston (1) reported that most infections that result in systemic symptoms in young plants occur through the hypocotyl. Such a hypothesis would perhaps explain why seedlings inoculated by soil drenching have a shorter interval of susceptibility than seedlings inoculated by foliage spraying or by the whole-seedling method, especially if differentiation and elongation of the hypocotyl occur more rapidly at the base of the hypocotyl than at progressively higher levels.

The downy-mildew-induced damping-off described by Goossen and Sackston (4) and observed by Zimmer and Kinman (14), much like systemic infection, is closely associated with seedling age at inoculation. Very young seedlings with radicles shorter than 5 mm long 3-4 days after planting are more susceptible to mildew-induced damping-off than older seedlings. Poor stands often found in sunflower fields heavily affected by downy mildew probably result from pre-emergence damping-off induced by early infection. Consequently, the importance of downy mildew is probably underestimated when losses are based only on counts of emerged seedlings showing systemic symptoms.

The rapidity of systemic symptom development also seems associated with seedling age at inoculation. The older the seedling, the longer the interval before symptoms are expressed. Seedlings inoculated 3 days after planting under greenhouse conditions showed systemic symptoms on an average of 18 days earlier than seedlings inoculated at 7 days after planting. Seedlings inoculated after 8 days seldom produced systemic symptoms. However, when they did, symptoms were not expressed until flowering. This suggests that mid- and late-season expression of systemic infection of field-grown plants may be manifestations of infection late in the period of seedling susceptibility, and not necessarily the result of infection through the apical bud, as reported by Cohen and Sackston (1).

Foliage and apical bud infection.—Foliage inoculation by the disk method and apical bud inoculation of more than 280 plants at different ages failed to show that such infection contributes significantly to the incidence of mid- or late-season systemic symptoms. Although chlorotic and necrotic lesions reported by Cohen and Sackston (1) always resulted from disk inoculation, only rarely could sporulation be induced from these lesions when plants were placed in humidity chambers. In this study, no evidence that apical inoculation resulted in systemic symptoms was obtained. It has been reported (2) that infection through the leaves, and possibly through the apical meristem, produce systemically infected symptomless plants. Seed was not checked microscopically in this study, therefore no direct evidence either supporting or refuting the results of Cohen and Sackston (2) were obtained.

If sunflower plants were as susceptible to systemic invasion via the apical meristem as reported (1), and if this infection gave rise to systemic symptoms (1), an extremely high percentage of plants in some fields would show late-

season systemic symptoms. This would be true particularly when plants were exposed under favorable conditions to abundant wind-borne zoosporangia arising from systemically infected seedlings in the field. Surveys during 1971 and 1972 revealed that rarely more than 5% of the adult plants showed systemic symptoms even though more than 50% of the seedlings in such fields had been observed at earlier dates to be systemically infected and to produce abundant wind-borne zoosporangia. The adult plants with systemic symptoms probably resulted mainly from seedlings infected so late in the period of susceptibility that symptom expression was delayed until near flowering.

Data obtained from this study suggest that any managerial practice that would reduce the interval of susceptibility to systemic infection, such as shallow seeding and delaying planting until the soil temperature supports rapid germination and emergence, should minimize the probability that seedlings would be exposed during the stage of susceptibility to sufficient rainfall to support systemic infection. Sporadic and unpredictable heavy rain showers during the seeding period in the major sunflower producing area of the USA, however, suggests that such managerial practices alone cannot eliminate serious losses from systemic downy mildew in areas of intensive sunflower production. Other practices, such as long-term crop rotation and early season control of systemically infected volunteer sunflowers shedding air-borne zoosporangia, would probably be more effective. Hybrid oilseed varieties have been developed that possess the resistance gene Pl_2 (14). So far, this resistance has been universially effective.

LITERATURE CITED

1. COHEN, Y., and W. E. SACKSTON. 1973. Factors affecting infection of sunflower by Plasmopara halstedii. Can. J. Bot. 51:15-22.
2. COHEN, Y., and W. E. SACKSTON. 1974. Seed infection and latent infection of sunflower by Plasmopara halstedii. Can. J. Bot. 52:231-236.
3. DELANOE, D. 1972. Biologie et epidemilogie du mildiou du tournesol (Plasmopara helianthi Novot.). Inform. Technol. 29:1-49.
4. GOOSSEN, P. G., and W. E. SACKSTON. 1968. Transmission and biology of sunflower downy mildew. Can. J. Bot. 46:5-10.
5. LEPPIK, E. E. 1962. Distribution of downy mildew and some other seed-borne pathogens on sunflowers. FAO (Food Agric. Organ., UN) Plant Prot. Bull. 10:126-129.
6. NISHIMURA, M. 1922. Studies in Plasmopara halstedii. I. The infection of Helianthus annuus L. by zoospores. II. The method of fertilization and zoospore-formation. J. Coll. Agric., Hokkaido Imp. Univ. 11:185-210.
7. NOVOTELNOVA, N. S. 1963. Nature of parasitism of the pathogen of downy mildew of sunflower at seed infection. [In Russian]. Bot. Zh. 48:845-860.
8. NOVOTELNOVA, N. S. 1966. Downy mildew of sunflowers. [In Russian]. Nauka, Acad. Sci. USSR, Moscow. 150 p.
9. TIKHONOV, O. I. 1973. Peculiarities of infection and forms of sunflower downy mildew. Pages 156-158 *in* Proc. Fifth Int. Sunflower Conf., Clermont-Ferrand, France, 1972.
10. ZIMMER, D. E. 1971. A serious outbreak of downy mildew in the principal sunflower production area of the United States. Plant Dis. Rep. 55:11-12.
11. ZIMMER, D. E. 1972. Field evaluation of sunflower for downy mildew resistance. Plant Dis. Rep. 56:428-431.
12. ZIMMER, D. E. 1973. Basal gall of sunflower incited by Plasmopara halstedii. Plant Dis. Rep. 57:647-649.
13. ZIMMER, D. E. 1974. Physiological specialization between races of Plasmopara halstedii in America and Europe. Phytopathology 64:1465-1467.
14. ZIMMER, D. E., and M. L. KINMAN. 1972. Downy mildew resistance in cultivated sunflower and its inheritance. Crop Sci. 12:749-751.
15. ZIMMER, D. E., and D. C. ZIMMERMAN. 1972. Influence of some diseases on achene and oil quality of sunflower. Crop Sci. 12:849-861.