

Relationship of age of sunflower seedlings to infection by *Plasmopara halstedii*

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Key words: *Helianthus annuus* L., *Plasmopara halstedii*, seedling age, percentage of plant infection

Abstrak

Kaitan di antara umur anak benih pokok bunga matahari dengan jangkitan kulapuk downy, *Plasmopara halstedii*, telah dikaji dengan menginokulasi anak-anak benih yang berumur di antara 3–15 hari. Korelasi yang negatif dan bererti ($r = -0.91^{**}$) didapati di antara umur anak benih dan peratus jangkitan kulapuk downy.

Lebih daripada 83% anak benih yang berumur 8 hari rentan terhadap patogen ini. Walau bagaimanapun, anak benih yang berumur 15 hari menunjukkan kerintangan. Peratus jangkitan menurun dengan bertambahnya umur anak benih.

Abstract

The relationship of age of seedlings to infection by *Plasmopara halstedii* was studied by inoculating 3-day-old to 15-day-old sunflower seedlings. There appears to be a negative and significant correlation ($r = -0.91^{**}$) between sunflower seedling age and percentage of seedling infection of downy mildew.

Over 83% of the seedlings up to 8 days of age were susceptible to this pathogen. However, the 15-day-old plants showed some resistance. The percentage infection decreased with increase in age of plants.

Introduction

Downy mildew [*Plasmopara halstedii* (Farlow) Berl. et de Toni] is a widespread and destructive seed-, soil-, and air-borne disease of sunflower (*Helianthus annuus* L.). The pathogen is an obligate parasite which apparently originated in North America but has spread in recent years with sunflower seed to most areas where sunflower is cultivated (Sackston 1974). Typical symptoms of downy mildew infection appear in the form of chlorosis of the leaves often associated

with severe stunting of systemically infected plants, and various types and degrees of leaf distortion (Goossen and Sackston 1968). Downy mildew was the second predominant disease in the Dakotas and Minnesota in 1984 with prevalence of 59% (Gulya and MacArthur 1984).

This disease is characterized by distinct systemic and localized phases (Zimmer 1975). However, systemic infection is the most destructive, occasionally causing 50% yield reduction

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in some fields of Red River Valley of North Dakota and Minnesota (Zimmer 1971). Immediately after seed germination, the sunflower seedlings are susceptible to systemic infection for only a short time and later the chances of infection are reduced. The aims of this study were to investigate if the age of the plants, when inoculated with downy mildew, determines the severity of infection and also the age of sunflower seedlings most susceptible to *P. halstedii* infection.

Materials and methods

This experiment was conducted in the greenhouse of the Department of Plant Science, South Dakota State University, Brookings, South Dakota, USA, during winter 1982. The sunflower seed of USDA hybrid 894, susceptible to downy mildew, was planted into a potting mixture of soil, sand and peat moss (2:1:1, steamed 2 h) in 54 cm x 27 cm plastic flats of 5.5 cm depth, accommodating 90 plants in each flat. Five dates of planting were made to obtain seedlings 3, 5, 8, 11 and 15 days of age. The experiment was designed in a randomized complete block design with three replications and each experimental unit consisted of one plastic flat.

Inoculum of downy mildew was prepared by washing the sporulating leaves obtained from the infected host sunflower plants kept overnight in the mist chamber to obtain sporangial suspension in sterile distilled water. The inoculum consisted of 10 000 zoosporangia per mL of distilled water, determined with a hemocytometer. The sunflower seedlings were inoculated by drenching the soil around each seedling with 25 mL inoculum. Care was exercised during drenching to avoid inoculum contact with foliage, unfolded cotyledons, or emerged portion of the hypocotyl. After inoculation, the plastic flats were placed overnight in a 16 °C dew chamber.

The plastic flats were then returned to greenhouse benches and observed daily for systemic symptoms. One week later, the systemic infections were verified by placing the plastic flats in a mist chamber overnight in a greenhouse (20–22 °C) to observe for sporulation of *P. halstedii*. Plants showing sporulation and non-sporulation of *P. halstedii* were counted to determine the percentage of infection. The analysis of variance of the data obtained was carried out. The simple correlation coefficient between sunflower seedling age and percentage of seedling infection was also calculated.

Results and discussion

Mean squares of sunflower seedlings inoculated with downy mildew at different ages show that significant differences exist for seedling age (*Table 1*). By using the least significant differences test, the sunflower seedlings inoculated at 15 days of age showed significant differences from the others. Evidently, the sunflower seedlings become less susceptible to downy mildew with increasing seedling age (*Table 2*). It is probable that much of the infection occurred through the hypocotyl, when inoculum was poured onto the soil surface without touching the aerial parts. More than 83% of the 3 to 8-day-old seedlings sporulated when inoculated by this method and placed overnight in mist chamber. The younger seedlings among the 3-day-old showed the maximum susceptibility (94.7%) to downy mildew. However, 15-day-old seedlings that were inoculated showed a lower infection of 24.9%.

The age of sunflower seedlings was significantly and negatively correlated with percentage of seedling infection ($r = -0.91^{**}$). This significant negative correlation may have been because resistance to systemic infection is probably associated with the differentiation and elongation processes within the hypocotyl. Moreover, it has

Table 1. Mean squares of sunflower seedlings with downy mildew infection following inoculation with *Plasmopara halstedii* at different seedling ages in days

Source	df	Mean squares
Replication	2	353.33
Seedling age	4	2 264.78**
Error	8	70.52

**Significant at the 0.01 level of probability

Table 2. The percentage of sunflower seedlings with symptoms of downy mildew and its relationship to seedling age

Sunflower seedling age (days)	Infected plants (%)
3	94.7
5	84.1
8	83.9
11	73.2
15	24.9
LSD (0.05)	15.81

been shown by Cohen and Sackston (1973) that the hypocotyl is the primary site of entry which gives rise to systemic infection in young plants. Such a hypothesis would perhaps explain why sunflower seedlings inoculated by soil drenching have a shorter interval of susceptibility.

The downy mildew-induced damping-off described by Goossen and Sackston (1968) and observed by Zimmer and Kinman (1972) is very much like systemic infection and is closely associated with seedling age at inoculation. Very young seedlings with radicles shorter than

5 mm when inoculated 3–4 days after planting were more susceptible to mildew-induced damping-off than older seedlings. Seedlings inoculated after 8 days seldom produced systemic symptoms (Zimmer 1975). The present results are in conformity with the results of previous workers and suggest that sunflower seedlings up to 8 days of age are best to evaluate for susceptibility to downy mildew in promising lines or cultivars.

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