

## Temperature and Humidity Associated with Sporulation of *Helminthosporium maydis* Race T

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### ABSTRACT

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The relationships of time, temperature, and relative humidity to sporulation of *Helminthosporium maydis* race T were documented by the results of experiments conducted in the dew chamber, in the greenhouse, and in the field. In the dew chamber, precise meteorological criteria were identified for sporulation on plants with Texas male-sterile cytoplasm (Tcms) and on a hybrid of normal (N) cytoplasm; i.e., longer dew period up to 48 hr at higher temperatures to 28 C resulted in greatest spore production. Initial sporulation occurred on

Tcms corn in 5 hr at 22 C and on a hybrid of N-cytoplasm corn after a 12-hr dew period at 25 C. In the field, the relationships of time, temperature, and humidity to initiation and quantity of sporulation were not clear; the results were erratic. None or many spores were produced by the same set of weather variables. The accuracy of forecasts of southern corn leaf blight epidemics in the field on the basis of weather criteria identified in the dew chamber remains in doubt.

*Additional key words:* dew, epidemics, forecasting, N-cytoplasm, T-cytoplasm, sporulation, weather.

Many investigators have reported on the sporulation of the fungus, *Helminthosporium maydis* Nisikado and Miyake race T (1, 2, 3, 4, 5, 6, 7, 8, 9). In 1971, we initiated laboratory experiments to determine a temperature-humidity timetable for the sporulation of the pathogen. We wanted to document the time required for spore production and the quantity produced with time. Sporulation on corn plants in the field was compared with dew chamber results. From these data, we hoped to establish the requirements of temperature, humidity, and time for sporulation for use in predicting epidemics of the corn leaf blight.

### MATERIALS AND METHODS

In the dew chamber experiments, corn seedlings in the three-leaf stage in 10-cm-diameter pots were inoculated with isolates of *H. maydis* race T and placed in the dew chamber overnight (about 16 hr) for infection to occur. The plants were removed and placed on the greenhouse bench for 4 days. After this period, lesions averaging about  $2.5 \times 10$  mm had developed and the plants were placed in the dew chamber for periods of 5, 8, 12, 18, 24, 30, 36, 42, and 48 hr at temperatures of 10, 13, 16, 19, 22, 25, 28, and 31 C. Quantitative estimates of sporulation were obtained by brushing and washing spores off individual lesions and counting the number of spores with

the aid of a hemocytometer.

In one field experiment, three Tcms- and two N-cytoplasm tasseling corn cultivars were sprayed with a spore suspension of *H. maydis* race T, and 100 individual lesions were tagged and observed for sporulation each day over a 24-day period. Lesions were brushed with a camel's-hair brush each morning after readings were made.

In other field trials, 15 and 20 individual lesions in inoculated plots were marked, the lesions were examined with a hand lens for spores, and spores were removed with double-stick tape at 0800 hours daily for a series of 34 and 55 days, respectively. Lesions were brushed after each reading.

Hygrothermograph, dew, and rainfall data were recorded in the plots.

### RESULTS

**Dew chamber experiments.**—Spores from 60 lesions were counted and averaged for each exposure. The average number of spores/lesion after dew periods of 5, 8, 12, 18, 24, 30, 36, 42, and 48-hr at seven different temperatures was counted. Only those counts at 12, 24, 36, and 48 hr are depicted (Fig. 1) because those at 5 and 8 hr were too low to graph.

The following results were obtained:

- (i) At 10 C, in a preliminary experiment, a few spores were produced after 27-hr dew period.
- (ii) At dew period temperatures of 22, 25, 28, and 31 C, 1, 15, 13, and 2 spores per lesion, respectively,

- were produced after 5 hr.
- (iii) At 16, 19, 22, 25, 28, and 31 C, 11, 20, 38, 31, 22, and 4 spores per lesion, respectively, were produced after 8 hr.
  - (iv) The greatest increase in sporulation occurred above 16 C, and, again, above 19 to 28 C at the 12-, 24-, 36-, and 48-hr dew periods.
  - (v) At 25 C, spore production nearly doubled from a 12- to 24-hr dew period, doubled after 36 hr and further increased after 48 hr.
  - (vi) At 28 C, spore production increased 3.6 times from a 12- to 24-hr dew period, then nearly doubled from 24- to 48-hr.
  - (vii) At 19 C, spore numbers increased from 312 at 12 hr to 8,437 per lesion after a 48-hr dew period.
  - (viii) The greatest number of spores per lesion, 27,768 or 731/mm<sup>2</sup>, occurred after a 48-hr dew period at 28 C.
  - (ix) Temperatures  $\geq 19$  C during the dew period  $\geq 24$  hr would be essential for numbers of 2,800 or more per lesion.
  - (x) Generally, the longer dew periods at temperatures  $\geq 16$  C resulted in greater spore production.

**Field experiments.**—During 1973, in a preliminary trial with three Tcms and two N-cytoplasm corns, all the individual lesions that were tagged produced spores intermittently over the 24-day period. Spores were produced on lesions after periods of RH  $\geq 90\%$  for 0, 4, 8, and 12 hr. Although the spore count exceeded 50/lesion on only 3 days, these lesions produced some spores over a long period. A few spores were produced on some lesions of the N-cytoplasm corn.

In 1974, 15 lesions were tagged and observed 34 days during the period 6 August to 25 September. The greatest total number of spores on the 15 lesions was 10,300, or about 700 spores per lesion. Spores appeared on the lesions after periods of RH  $\geq 90\%$  of 0 to 17 hr. The two highest counts, 700 and 665 spores per lesion, occurred after dew periods of 12 and 13 hr with average temperature of 21 C. In the laboratory, these same conditions produced about 3,000 spores per lesion. In the

field, another 17-hr period of RH  $\geq 90\%$  at 17 C produced three spores per lesion on the 15 lesions; yet, after another dew period of 11 hr at 17 C, 213 spores per lesion were produced. On another day, a dew period of 16 hr at 14 C produced 0 spores, and another day, a 12-hr dew period at 14 C produced 85 spores per lesion. In another instance, a 12-hr dew period at 14 C produced only 12 spores per lesion on the 15 lesions.

In this experiment, individual lesions produced spores for a period of 25 days.

In 1975, spore counts were made on 55 days. Sporulation occurred on 20 lesions at periods of RH  $\geq 90\%$  ranging from 0 to 30 hr, with the greatest spore production after 39 hr.

After two 8-hr periods, spore counts were 0 and 213 spores per lesion. The average temperatures during the dew periods were 20 and 16 C, respectively, with no spores produced during the more favorable temperature. Spores produced at four 11-hr periods in the same temperature range varied from three to 470 spores per lesion, with the fewest spores produced on the largest lesions, 87 mm<sup>2</sup> average. Four, 15-hr periods varied from two to 132 spores per lesion on the 20 lesions. The temperature averages of the dew periods were all above 15 C, and the average temperature of a period that produced 80 spores per lesion was lower than that of the period that produced two spores per lesion and the average lesion size was the same, 31 mm<sup>2</sup>. Lesions averaging 90 mm<sup>2</sup> produced fewer spores than those averaging 52 mm<sup>2</sup> at the 15-hr periods. In another instance, an 8-hr period produced two to three times more spores than some 11-, 12-, 13-, 14-, and 15-hr periods; and the longer humidity periods had temperature averages in the mid- and high-teens and low 20's. The average lesion sizes (52-88 mm<sup>2</sup>), of the longer high-humidity periods were equal to or larger than the average lesion size of 52 mm<sup>2</sup> at the 8-hr period so lesion size was not the reason for greater spore production. Several nights during the season, a few spores were produced when no dew or hrs of RH  $\geq 90\%$  or only 2 to 6 hr of RH  $\geq 90\%$ , was recorded. Successive days of precipitation resulted in consecutive days with 8 hr or more of RH  $\geq 90\%$ .

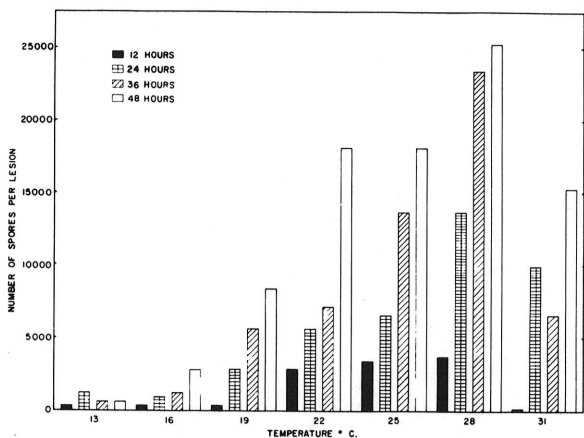


Fig. 1. Number of spores per lesion of *Helminthosporium maydis* race T produced on corn leaves exposed to seven temperatures at four different dew periods in the dew chamber.

## DISCUSSION

Generally, the results of our sporulation experiments at different time, temperature, and humidity regimes in the dew chamber agreed with those of other investigators (1, 2, 4, 5, 6), although we obtained sporulation at lower temperatures, and shorter and longer dew periods than employed by previous investigators. In extending the dew period to 48 hr at 28 C, our spore count, 731/mm<sup>2</sup>, was greater than recorded in the literature. The dew chamber experiments were conducted to demonstrate that such data could be correlated with sporulation on corn plants in the field and thus be employed to predict the behavior of the fungus in the field.

To our knowledge, the present study is the first time that sporulation data from the dew chamber have been related to sporulation in the field, and the results were surprising. The results demonstrate that the behavior of the fungus in the controlled environment does not depict the behavior of the fungus in the field. For example, in the

field, we expected longer periods of  $RH \geq 90\%$  at favorable temperatures to result in greater spore production as occurred in the dew chamber, but this did not always occur. Furthermore, spores were produced in the absence of dew periods or periods of  $RH \geq 90\%$ . During the 34 days samples were taken in 1974, some spores were produced on the 15 lesions every night but one during periods of  $RH \geq 90\%$  ranging from 0 to 15 hr. On the night that no spores were produced, there were 15 hr of  $RH \geq 90\%$  and the average temperature of the period was 14.5 C, favorable for sporulation.

Another unexpected, unexplainable phenomenon observed was the production on three days of very few spores on some lesions in spite of consecutive nocturnal temperature minima of 1.11, 2.22, and 3.89 C at periods of  $RH \geq 90\%$  of 11, 13, and 9 hr, respectively.

We are aware of the limitations in instrumentation, sampling methods, documentation of specific weather parameters, integration of temperature-humidity differentials, and the preconditioning of lesions and conidiophores in the field; however, such methods, instruments, etc., have been used successfully in predicting potato-tomato late blight and *Cercospora* leaf spot of sugar beets. Our findings demonstrate that equating the field- and controlled-environment results is extremely difficult at the present time. Modeling and computerizing corn blight forecasts will evade us until weather variables can be related more reliably to spore production in the field.

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