

# Effect of Temperature and Relative Humidity on Development of *Cercosporidium personatum* on Peanut in Georgia

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## ABSTRACT

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In controlled environment experiments, a minimum of 4 hr of relative humidity  $\geq 95\%$  per day was required for conidial production by *Cercosporidium personatum*, and the highest numbers of conidia were produced when lesions were subjected to daily periods of 16 or more hours of relative humidity  $\geq 95\%$ . The optimum temperature for spore production was near 20 C. In field studies, numbers of airborne conidia of *C. personatum*, the causal agent of peanut late leaf spot, were monitored with Burkard 7-day recording volumetric spore traps at Athens, Plains, and Tifton, Georgia, in 1986-1988. Trapping of conidia was initiated in mid-July to early August, when late leaf spot lesions were first detected. A diurnal periodicity in spore release was observed, with peak spore catches occurring between 10 A.M. and 6 P.M. Conidia were detected on most days during the trapping period. For the years and locations examined, duration of relative humidity  $\geq 95\%$  generally exceeded 5-hr duration each day and temperatures seldom dropped below 20 C, indicating that conditions in Georgia were generally favorable for spore production.

Additional keywords: aeromycology, *Arachis hypogaea*, spore trapping

Late leaf spot of peanut, caused by *Cercosporidium personatum* (Berk. & M.A. Curtis) Deighton, is an important disease of peanut (*Arachis hypogaea* L.) in the southeastern United States (14). To prevent reductions in pod yield caused by late leaf spot, seven to eight calendar-based fungicide applications are recommended each season, at a cost to producers of approximately \$60 per acre in fungicide alone (11). When susceptible peanut cultivars are grown without the use of fungicides, pod losses due to late leaf spot can exceed 50% (14).

Conidia are produced directly from mycelium in crop debris in the soil (12). Rain splash of soilborne inoculum is probably the predominant mechanism for dissemination of conidia when plants are young. After leaves become infected, conidia from sporulating lesions are readily disseminated by wind during the growing season (8,16).

Temperature and humidity are important environmental conditions which regulate the production and release of conidia. These variables have served as the basis for disease prediction models for similar leaf spot fungi of peanut, such as *Cercospora arachidicola* S. Hori (4,5). Under controlled environmental condi-

tions, infection of peanut by *C. personatum* was optimal at 20 C and at least 12 hr/day of relative humidity (RH) greater than 93% (13). Conidial germination was greatest at 16-20 C (15). The quantitative effects of temperature and high relative humidity on sporulation of *C. personatum* and subsequent infection of peanut under field conditions have not been described. The objectives of this study were to determine the effect of temperature and relative humidity on sporulation of *C. personatum* and on development of late leaf spot on peanut under both controlled environments and field conditions in Georgia.

## MATERIALS AND METHODS

**Controlled environment experiments.** Six-week-old peanut plants cv. Florunner in 250-cm<sup>3</sup> containers were inoculated with an aqueous suspension of *C. personatum* containing  $2.5 \times 10^4$  conidia per milliliter. Ninety-eight plants were placed in an area 60  $\times$  60 cm and uniformly inoculated with 40 ml of the suspension by using an atomizer. Following inoculation, plants were incubated in a dew chamber for 6 days at 25 C with an 8-hr/day fluorescent light photoperiod. After 6 days, plants were moved to a growth chamber at 25 C and 50-60% RH with a 16-hr/day fluorescent light photoperiod. Six days in the dew chamber were required to obtain a cohort population of lesions of the same size and maturity. Although nearly 100% of the spore population germinates within 36 hr after inoculation, germ tube penetration of stomates occurs over a period of 3-17 days after inoculation (F. W. Nutter and S. C. Alderman, unpublished).

Twenty-four days after inoculation, lesions were harvested by excising leaf disks containing single lesions with a 5-mm-diameter cork borer. The effect of temperature on sporulation over time was determined by placing lesion disks, abaxial side up, in plastic chambers lined with moistened filter paper and incubating them at temperatures ranging from 15 to 35 C. No sporulation was observed on any lesion disk prior to incubation. Each temperature treatment included 30 lesions in each of six replications.

After 2, 4, 6, 8, and 10 days incubation, lesion disks were placed in vials (30 lesions per vial) containing 15 ml of de-ionized, distilled water with Tween 20 (1 drop/100 ml). A micro stirring bar was placed in each vial, and lesion disks were agitated for 10 min on a magnetic stir plate to dislodge conidia. Six 2-ml samples were drawn from the resulting suspension, and 2 ml of 4% copper sulfate was added to each sample to prevent conidial germination prior to enumeration. The number of spores per milliliter was determined using a hemacytometer and expressed as the number of spores per lesion. Lesion disks were rinsed with de-ionized, distilled water and returned to the appropriate moisture chamber and temperature until the next spore harvest. The cumulative number of spores per lesion as affected by temperature was then determined, and the area under the curve (AUC) was calculated as previously described (7). All experiments were repeated at least once.

The effect of daily periods of RH  $\geq 95\%$  on sporulation was investigated using the same procedures described above, except that daily RH periods  $\geq 95\%$  were used instead of different temperatures. The daily RH periods  $\geq 95\%$  were 4, 8, 12, 16, 20, and 24 hr/day for a total of 10 days. When not exposed to RH periods  $\geq 95\%$ , lesion disks were placed in an incubator at 20 C and 60-70% RH.

**Field studies.** Field plots of peanut cv. Florunner were established at Athens, Plains, and Tifton in 1986-1988. Plots measured 50-75 m long and 24-48 rows wide (0.97 m apart). Fertilization and weed control practices were in accordance with extension recommendations (6). No fungicides were applied.

At weekly intervals beginning in mid-July to mid-August (depending on when lesions were first observed), four 30-cm segments of row were selected at random, and leaflet number was determined. Le-

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sions were counted on 20 samples (two main stems per sample), and lesions per leaflet were determined by dividing the total number of lesions in each sample by the corresponding number of leaflets. Lesion number per 30 cm of row was estimated by multiplying lesions per leaflet by leaflet number in 30 cm of row. Total leaflets (intact + defoliated) were determined by multiplying total stem nodes by four leaflets per node. Percent defoliation was determined by dividing the number of defoliated leaflets by total leaflets. The change in defoliated leaflets per 30 cm of row between each sampling date ( $\Delta d$ ) was determined. Each  $\Delta d$  was multiplied by lesions per leaflet (average of value for a given date and previous date). The sum of  $\Delta d$  values was added to intact leaflets on the last sample date to estimate cumulative lesions per 30 cm of row.

Conidial populations were monitored with Burkard 7-day recording volumetric spore traps (Burkard Mfg., Ltd., England). A trap was placed at each location in the center of each plot with the orifice positioned 42 cm above ground level. Spore slides were prepared and examined as previously reported (2). The traps were set to continuously sample 10 L ( $1 \text{ m}^3$ ) of air per hour. A correction factor of 0.6, as suggested by the manufacturer, was multiplied by air-intake rate, resulting in a sampling volume of  $14.4 \text{ m}^3$  of air per day. Hourly and daily conidial counts were used to determine diurnal and seasonal periodicities of *C. personatum*. Reference slides of conidia of *C. personatum* were prepared from late leaf spot lesions collected within the field plots and also from infected plants in the greenhouse.

Hygrothermographs were positioned in standard white weather shelters at midcanopy and within 3 m of each spore trap. Prior to placement in the field, hygrothermographs were calibrated according to manufacturer instructions. Each week, RH and temperature readings by the hygrothermographs were compared to readings from an aspirated

wet-dry bulb thermometer, and readings were  $\pm 2\%$  of the temperature readings.

**Statistical analyses.** The number of conidia trapped/lesion density (conidia/lesions in 30-cm row) in relation to hours of  $\text{RH} \geq 95\%$  and temperature during high-humidity periods was determined with regression analysis. The significance of each regression equation was determined based on the *F* test, coefficient of determination ( $r^2$ ), and examination of residuals. The relationship between environmental conditions and numbers of conidia was evaluated 1–10 days prior to spore release. Environmental conditions averaged over 1- to 5-day periods prior to conidial release were also examined to determine the relationship between average conditions prior to spore release and number of conidia trapped.

## RESULTS

**Controlled-environment studies.** The most favorable temperature for spore production during periods of  $\text{RH} \geq 95\%$  was 20 C (Fig. 1). Area under the spore production curve was significantly higher at 20 C compared to all other temperatures tested (Table 1). Areas under the spore production curve for 15 C were significantly higher than values for 30 and 35 C. Cumulative conidia per leaflet increased with increasing hours of daily periods of  $\text{RH} \geq 95\%$ , with the greatest number of spores being produced when high-RH periods exceeded 12 hr per day (Fig. 2).

**Field studies.** Conidia of *C. personatum* were trapped throughout day-night periods, with the greatest number trapped between 10 A.M. and 6 P.M. (Fig. 3). The spore-trapping periods at Athens extended from day 242 to 282, 234 to 260, and 202 to 264 in 1986, 1987, and 1988, respectively. In 1986 and 1987, lesions were observed late in the season (day 255) (traps were not placed in the field until 2 wk prior to harvest) (Fig. 4). At Plains, spore trapping extended from day 226 to 243, 208 to 242, and 215 to 254 in 1986, 1987, and 1988, respectively (Fig. 5). Lesions appeared 10–20 days later in 1986 than in 1987 or 1988. At Tifton, the spore-trapping period extended from day 197 to 232,

210 to 262, and 214 to 256 in 1986, 1987, and 1988, respectively. In each year, lesion number increased after day 220 (Fig. 6).

Lesions appeared earlier at Plains and Tifton than at Athens. The greatest number of lesions per leaflet was observed at Plains, and the smallest at Athens (Fig. 7). Defoliation was observed earlier at Tifton than at Plains or Athens, although the rate of defoliation was similar at each location (Fig. 8, Table 1). Defoliation progressed 10–20 days later at Athens than at Plains or Tifton.

Disease progress, expressed as logit cumulative lesions and the logit of cumulative conidia/ $14.4 \text{ m}^3/\text{day}$  increased linearly with time (Tables 2 and 3). The ranges in slope parameters for cumulative lesions and cumulative conidia were 0.16–0.31 and 0.18–0.33, respectively.

At each location, temperature conditions during periods of high humidity were generally very favorable for sporulation throughout all spore-trapping periods, as indicated by the very narrow temperature range found during periods of high RH. Regressions of conidia (number/ $14.4 \text{ m}^3$  air/day or number/ $14.4 \text{ m}^3$  air/day/lesion density) and temperature were not significant.

Temperature conditions during periods of high RH were similar in Plains and Tifton, and 1–5 C cooler in Athens (Fig. 9). Hours of  $\text{RH} \geq 95\%$  per day (Fig. 10) were more variable than temperature, but regressions of conidial numbers and RH periods  $\geq 95\%$  were not significant. Spore counts were not correlated with cumulative hours of  $\text{RH} \geq 95\%$  in the period 5 or 10 days prior to trapping or in any period between 1 and 10 days prior to trapping.

## DISCUSSION

*C. personatum* and *C. arachidicola* are common leaf spot fungi on peanut in the United States (15). Although late leaf spot predominates in Georgia and Florida, and early leaf spot predominates in North Carolina and Virginia (10), these

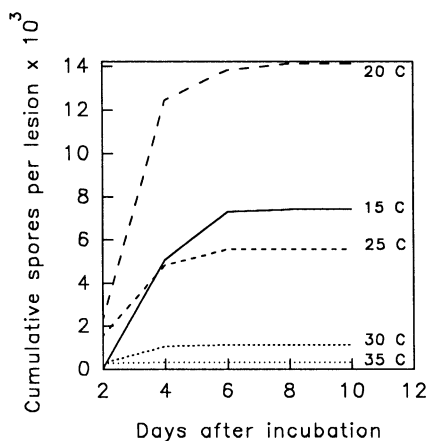


Fig. 1. Cumulative conidia of *Cercosporidium personatum* per lesion after various days incubation in a dew chamber at 15–35 C.

Table 1. Effect of temperature during periods of relative humidity ( $\text{RH} \geq 95\%$ ) under controlled conditions on area under the spore production curve of *Cercosporidium personatum*

Temperature during $\text{RH} \geq 95\%$ (C)	Area under the spore production curve <sup>2</sup>
15	86.1 b
20	185.4 a
25	64.1 bc
30	15.0 cd
35	4.8 d

<sup>2</sup> Means with the same letter in the same column are not significantly different according to the Waller-Duncan *k*-ratio test.

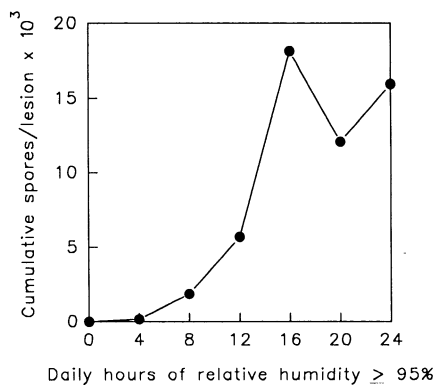


Fig. 2. Cumulative conidia of *Cercosporidium personatum* per lesion at various hours of daily leaf wetness.

two fungi were believed to respond similarly to environmental conditions. Because of consistently warm conditions during the spore-trapping periods in Georgia, it was not determined how *C. personatum* and epidemics of late leaf spot would develop under cooler conditions. However, we observed that epidemics of late leaf spot began later in the season in the cooler, northernmost site at Athens.

Although temperatures were cooler at Athens, disease progress curves, represented by lesions per leaflet or defoliation, paralleled those from Tifton and

Plains. Delays in leaf spot development at Athens may have resulted from cooler temperatures and/or a lower level of overwintering inoculum due to the heavier Piedmont soils, which may be less conducive to pathogen survival. Inoculum for the 1986 experiment at Athens was introduced shortly after plant emergence by spreading late leaf spot infested debris within the plot. Thus, earlier disease development and defoliation in subsequent years may have resulted from an increasing level of overwintering inoculum. Athens is outside the range of peanut production in Georgia, and inoculum

from neighboring fields is not as readily available as it is in central and south coastal plain areas of Georgia.

Based on the results of controlled environment and field experiments, humidity conditions during the spore-trapping periods in Georgia were often favorable for late leaf spot development at all locations and years studied. In the controlled environment studies, temperatures  $\geq 30^{\circ}\text{C}$  during periods of high RH had a greater effect on reducing spore production than did the recommended rate of chlorothalonil applied at 7- to 10-day intervals (7). However, high tem-

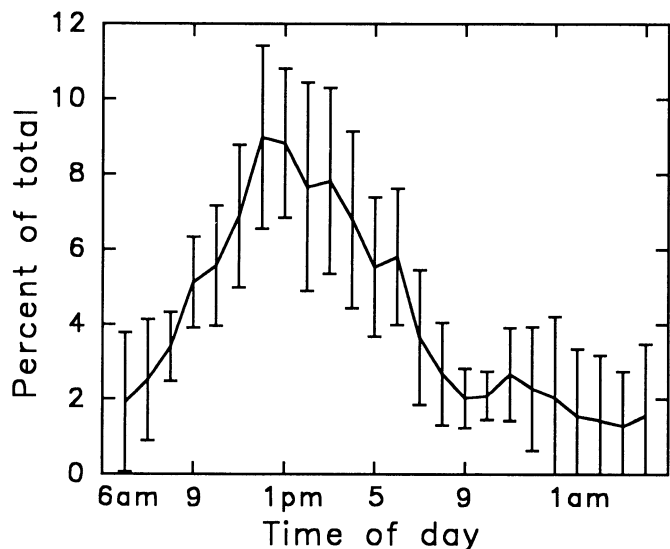


Fig. 3. Percentage of total conidia of *Cercosporidium personatum* trapped/14.4 m<sup>3</sup> air/day/lesion density. Means and standard deviations were based on three sites in each of 3 yr (1986-1988).

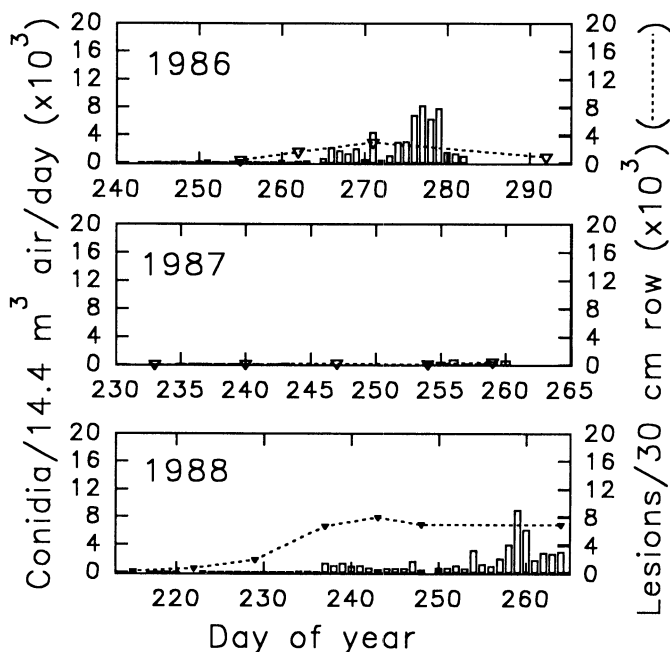


Fig. 4. Number of conidia of *Cercosporidium personatum* trapped/14.4 m<sup>3</sup> air/day and lesions/30-cm row observed at Athens in 1986, 1987, and 1988.

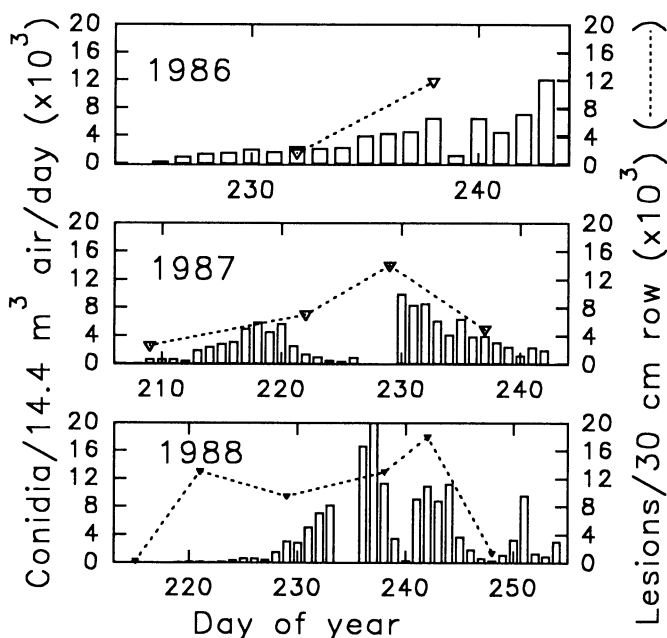


Fig. 5. Number of conidia of *Cercosporidium personatum* trapped/14.4 m<sup>3</sup> air/day and lesion/30-cm row observed at Plains in 1986, 1987, and 1988.

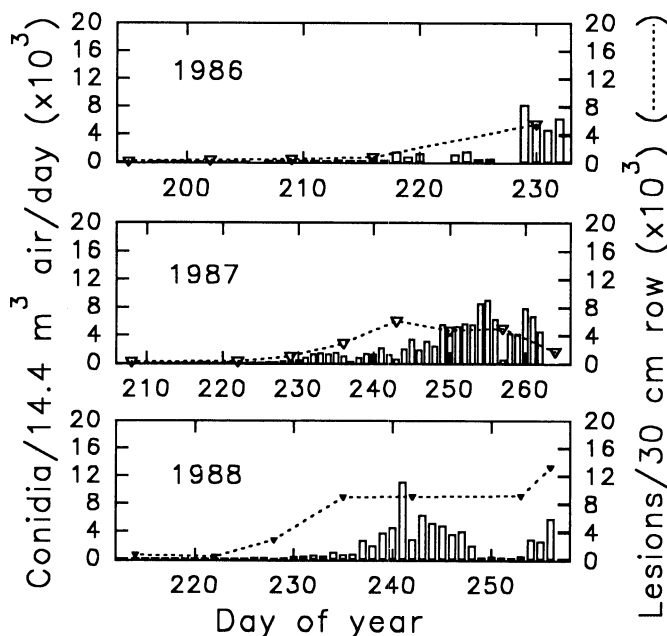


Fig. 6. Number of conidia of *Cercosporidium personatum* trapped/14.4 m<sup>3</sup> air/day and lesions/30-cm row observed at Tifton in 1986, 1987, and 1988.

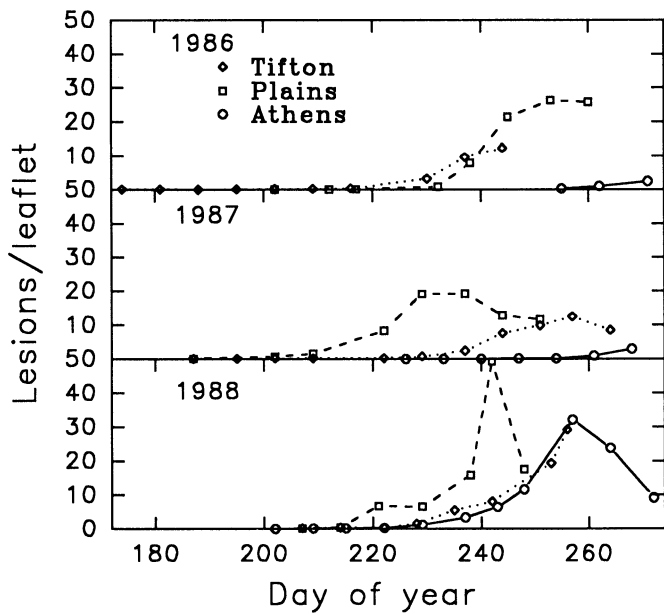


Fig. 7. Number of late leaf spot lesions/leaflet observed in peanut at Tifton, Plains, and Athens in 1986, 1987, and 1988.

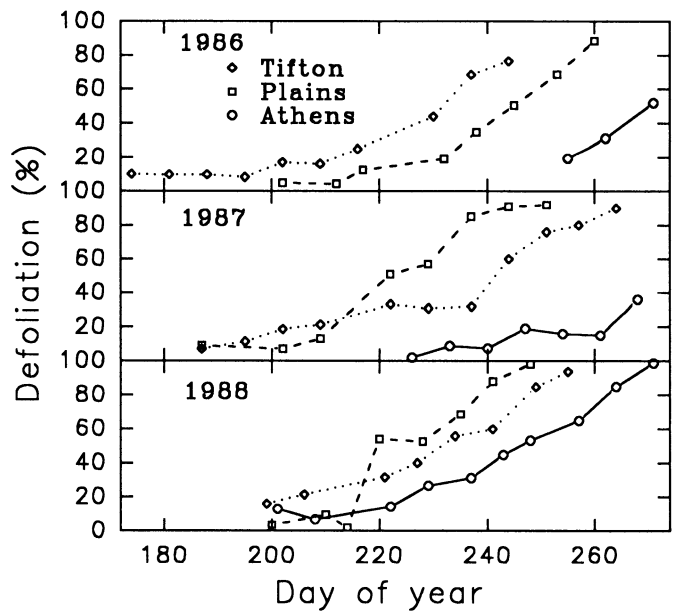


Fig. 8. Defoliation (%) in peanut observed at Tifton, Plains, and Athens in 1986, 1987, and 1988.

Table 2. Regression equations of logit cumulative late leaf spot lesions with respect to time. Logits were calculated based on highest value obtained in each data set

Location	Year	Equation	$r^2$
Athens	1986	$y = 0.31x - 83.92$	0.91
	1987	$y = 0.31x - 34.24$	0.93
	1988	$y = 0.16x - 39.50$	0.99
Plains	1986	$y = 0.23x - 55.65$	0.98
	1987	$y = 0.18x - 38.55$	0.97
	1988	$y = 0.24x - 55.87$	0.93
Tifton	1986	$y = 0.13x - 31.22$	0.94
	1987	$y = 0.15x - 34.94$	0.87
	1988	$y = 0.16x - 39.16$	0.99

Table 3. Regression equations of logit cumulative conidia of *Cercosporidium personatum*/14.4 m<sup>3</sup> air/day with respect to time. Logits were calculated based on highest value obtained in each data set

Location	Year	Equation	$r^2$
Athens	1986	$y = 0.25x - 70.53$	0.90
	1987	$y = 0.19x - 47.94$	0.86
	1988	$y = 0.20x - 50.61$	0.97
Plains	1986	$y = 0.26x - 63.30$	0.95
	1987	$y = 0.28x - 63.35$	0.93
	1988	$y = 0.33x - 78.34$	0.99
Tifton	1986	$y = 0.21x - 48.87$	0.97
	1987	$y = 0.18x - 44.75$	0.97
	1988	$y = 0.32x - 78.09$	0.93

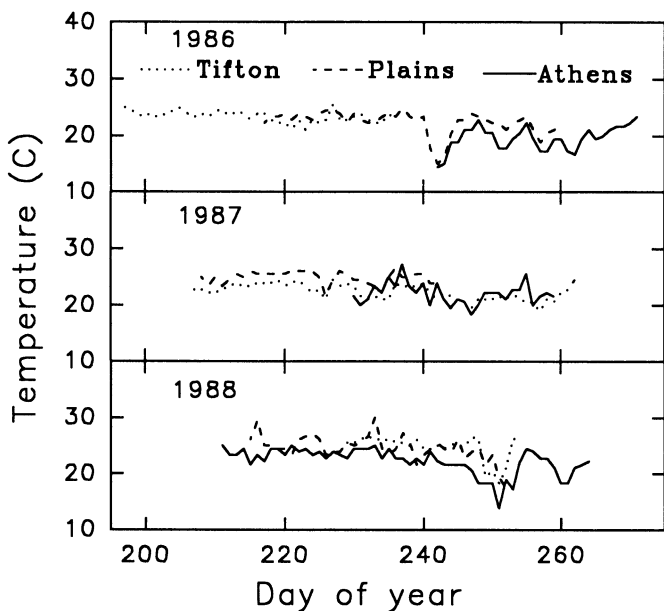


Fig. 9. Temperature conditions during daily periods of relative humidity  $\geq 95\%$  during the period of spore trapping for *Cercosporidium personatum* at Athens, Plains, and Tifton in 1986, 1987, and 1988.

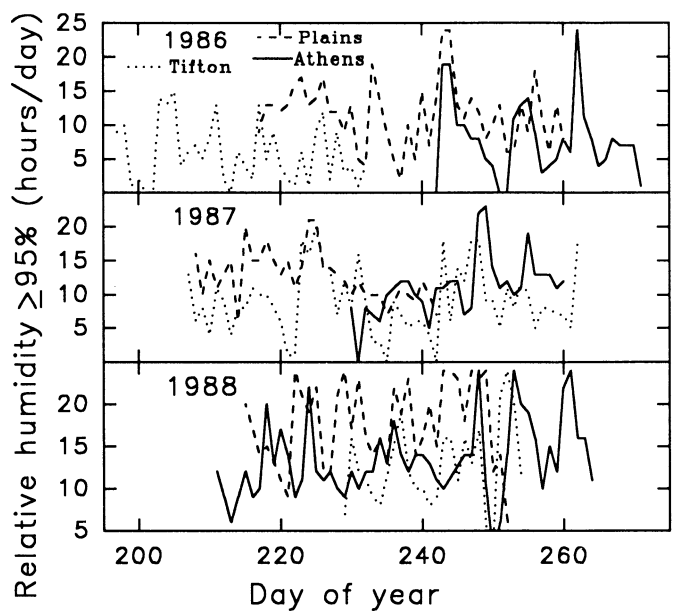


Fig. 10. Daily hours of relative humidity  $\geq 95\%$  during the period of spore trapping for *Cercosporidium personatum* at Athens, Plains, and Tifton in 1986, 1987, and 1988.

peratures did not occur during periods of high RH in the field studies. Based on area under the sporulation curve, temperatures between 15 and 20 C during periods of high RH would be very favorable for spore production in the field.

The lack of any significant regression models relating discrete periods of high RH vs. conidia trapped and disease development may indicate that *C. personatum* is not as sensitive to duration of high RH as is *C. arachidicola*. Histological studies describing the development of haustoria in *C. personatum* (9) suggest this fungus may be more dependent on the water status of the host. Conidia of *C. personatum* arise on distinct stomata which develop within leaf tissue with minimal disruption to the epidermis. This would provide more favorable moisture conditions within lesions, and therefore *C. personatum* would be relatively less dependent on external moisture conditions for inoculum production compared to *C. arachidicola*. Sporulation of *C. arachidicola* results in the rupturing of the epidermis and subsequent drying of lesions, which makes this fungus relatively more dependent on external sources of moisture (1). Thus, RH periods may be more limiting for this pathogen in relation to disease development because of the effect of RH periods on sporulation. Alderman and Nutter (3) earlier reported that there was a strong relationship between RH periods  $\geq 95\%$ , conidia trapped, and disease progress of early leaf spot caused by *C. arachidicola*.

A large decrease in lesions per 30-cm row at Plains in 1987 resulted from a substantial increase in defoliation. An

associated decline in conidia trapped suggests that lesions on defoliated leaflets may not produce as many conidia as lesions on intact plants.

Most conidia of *C. personatum* were trapped between 10 A.M. and 6 P.M., with peak spores trapped about 12 P.M. A similar periodicity was observed by Sreeramulu (16) and Mallaiah and Rao (8) in India.

Additional studies are needed to understand the dynamics of late leaf spot development. More research is needed to assess the importance of primary inoculum and the effects of environment on development of late leaf spot in the field. In addition, studies are needed to determine the early-season effects of temperature and moisture on initiation of late leaf spot epidemics and the mid-season environmental effects on rate of disease development.

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#### LITERATURE CITED

- Alderman, S. C., and Beute, M. K. 1987. Influence of temperature, lesion water potential, and cyclic wet-dry periods on sporulation of *Cercospora arachidicola* on peanut. *Phytopathology* 77:960-963.
- Alderman, S. C., Matyac, C. A., Bailey, J. E., and Beute, M. K. 1987. Aeromycology of *Cercospora arachidicola* on peanut. *Trans. Br. Mycol. Soc.* 89:97-103.
- Alderman, S. C., and Nutter, F. W., Jr. 1987. Sporulation of *Cercospora arachidicola* — a major component of early leafspot forecasting. *Proc. Am. Peanut Res. Ed. Soc.* 19:25.
- Cu, R. M., and Phipps, P. M. 1993. Development of a pathogen growth response model for the Virginia peanut leaf spot advisory program. *Phytopathology* 83:195-201.
- Jensen, R. E., and Boyle, L. W. 1965. The effect of temperature, relative humidity, and precipitation on peanut leafspot. *Plant Dis. Rep.* 49:975-978.
- Johnson, W. C., III, Beasley, J. P., Thompson, S. S., Womack, H., Swann, C. W., and Samples, L. E. 1987. Georgia Peanut Production Guide. Ga. Coop. Ext. Serv. Publ. SB23.
- Labrinos, J. L., and Nutter, F. W., Jr. 1993. Effects of a protectant versus a systemic fungicide on disease components of peanut late leaf spot. *Plant Dis.* 77:837-845.
- Mallaiah, K. V., and Rao, A. A. 1980. Aerobiology of two species of *Cercospora* pathogenic to groundnuts. *Proc. Nat. Acad. Sci. India, Sect. B* 46:215-222.
- Mims, C. W., Luttrell, E. S., and Alderman, S. C. 1989. Ultrastructure of the peanut late leaf spot fungus *Cercosporidium personatum*. *Can. J. Bot.* 67:1198-1202.
- Nutter, F. W., Jr., and Shokes, F. M. Management of foliar fungal pathogens. In: *Peanut Health Management*. H. A. Melouk and F. M. Shokes, eds. American Phytopathological Society, St. Paul, MN. In press.
- Phipps, P. M. 1993. IPM in peanuts: Developing and delivering working IPM systems. *Plant Dis.* 77:307-309.
- Ramapandu, S., and Appa Rao, A. 1982. Survival of *Cercospora arachidicola* and *Cercosporidium personatum* on plant debris of groundnut. *Madras Agric. J.* 69:767-768.
- Shew, B. B., Beute, M. K., and Wynne, J. C. 1988. Effects of temperature and relative humidity on expression of resistance to *Cercosporidium personatum* in peanut. *Phytopathology* 78:493-498.
- Smith, D. H., and Littrell, R. H. 1980. Management of peanut foliar diseases with fungicides. *Plant Dis.* 64:356-361.
- Sommartya, T., and Beute, M. K. 1986. Temperature effects on germination and comparative morphology of conidia for Thai and USA isolates of *Cercosporidium personatum*. *Peanut Sci.* 13:67-70.
- Sreeramulu, T. 1970. Conidial dispersal in two species of *Cercospora* causing tikka leaf-spot on groundnut (*Arachis hypogaea* L.) Indian J. *Agric. Sci.* 40:173-178.