

INFLUENCE OF ENVIRONMENTAL FACTORS ON PRODUCTION AND DISPERSAL OF *TOLYPOSPORIUM PENICILLARIAE* SPORIDIA

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The influence of environmental factors on production and dispersal of airborne sporidia of *Tolyposporium penicillariae*, the causal agent of smut of pearl millet (*Pennisetum glaucum*), was studied for 4 years (1984-1987) at the ICRISAT Centre farm. Sporidia were monitored once every 2 weeks, using rotorod spore samplers in two fields each cropped with pearl millet (June-October) and groundnut (November-April), located 500 m apart; one was a pearl millet smut nursery and the other was not. Most sporidia were trapped during the latter half of the pearl millet growing season, the time when the crop is at flowering and susceptible to infection. Maximum numbers of sporidia were recorded when the maximum temperature was 31-32° C and the minimum temperature was 20-21°C, and maximum relative humidity (RH) was more than 80% and minimum RH was 41-50%, with some rainfall (11-30 mm) and low wind speed (4-6 km/h), calculated on biweekly basis. Temperature, RH and wind speed were the principal environmental factors influencing production and dispersal of *T. penicillariae* sporidia.

Tolyposporium penicillariae Bref., the causal agent of smut of pearl millet (*Pennisetum glaucum* (L. R. Br.)), is widely distributed in the semi-arid tropics^{1,2}. The pathogen perpetuates from one season to another through teliospores deposited on the ground from previously infected crops. Under favourable moisture and temperature conditions, teliospores germinate to release numerous airborne sporidia³. When deposited on pearl millet panicles at flowering, these sporidia infect the florets through emerging stigmas⁴. Sporidia may also play a role in secondary spread of the disease^{5,6}.

Survival of *T. penicillariae* from one crop

season to another in areas traditionally growing pearl millet, where only one crop is taken in a year, depends on weather factors during the noncrop season of about 8 months (November—June). Teliospores, released from the infected crop in the preceding season and deposited on the ground, are exposed to various environmental stresses during the noncrop season. Depending upon prevailing soil moisture and temperature conditions, some teliospores might germinate or die during the noncrop season, whereas others would survive till the next growing season and produce sporidia that infect the crop. Weather factors, such as air temperature, relative humidity (RH), rainfall,

wind speed and wind direction all can influence the survival, production and dispersal of the pathogen, and they are important factors in the disease epidemiology. The purpose of this study was to investigate the effects of environmental factors on production and dispersal of *T. penicillariae* sporidia.

Materials and Methods

Selection of fields

Two fields were selected for this study at the ICRISAT farm near Hyderabad, India. Pearl millet has been grown in both fields each year during the rainy season from mid-June to October. A smut nursery is grown in one field (RP 13), where pearl millet plants are artificially inoculated with *T. penicillariae*⁷. High humidity is provided by sprinkler irrigation for 30 min on each rainfree day for about a month (July—August) during inoculation. Within 20 days after inoculation, smut sori are fully developed and, as the crop matures during September—October, these sori rupture and release masses of teliosporeballs. Most of these settle on the ground in the field itself, but some might be blown outside the field by wind. In the other field (RL 19), pearl millet plants are not artificially inoculated and since smut does not naturally occur in this field, it was used as a control with a low background inoculum level. It is located about 500 m south-east of RP 13. Both fields were cropped with groundnut during the off season (November—April) and were irrigated once a month.

Spore sampling

At fixed locations in each field, airborne spores were sampled every 2 weeks, using battery-operated rotorod spore samplers (Metronics Associates Inc. City, California, USA). Each time samplers were operated for 5 min between

14.00 and 15.00 hours during the rain-free period at the crop canopy level. The sampling heights varied with crop growth from 30 to 150 cm, and during the noncrop period it was 100 cm. For each sampling, two plastic rods coated with a thin film of silicone grease were exposed. Spore samplings were done for two full years (1984 and 1986), which included a pearl millet crop (mid-June–October) followed by a groundnut crop (November–April) each year, and two half years (1985 and 1987), only during the pearl millet crop. *T. penicillariae* sporidia were counted on a 40 mm² surface of each rod under a microscope, by fixing the rod on a grooved plastic slide. The sum of sporidial counts from the two rods were taken to represent one sample.

Weather data

Daily readings of air temperature (max. and min.), RH recorded at 07.00 (max.) and 14.00 (min.), rainfall, and wind speed for the entire period of experimentation were obtained from the ICRISAT Meteorological Observatory located on the farm. Means for each 2-week period for each weather factor in relation to sporidial counts were used for the presentation of results.

Statistical analysis

Correlation and stepwise regression analysis, using logtransformed values for sporidial number and weather factors individually and in combinations, were done, using a SAS program (SAS Institute Inc. SAS Circle, Cary, N.C., USA).

Results

Time of sporidial production in two fields

Sporidial production was consistently higher every year in the field where the smut nursery was located, RP 13 than in the other field, RL 19.

Between January and September 1984 and 1986, sporidial production was low in both the fields. The highest sporidial counts were obtained on 9 October 1984 (Fig. 1a), 24 September 1985 (Fig. 1b), 22 January 1986 (Fig. 1c), and 1 October 1987 (Fig. 1d) in both fields. Virtually no sporidia were trapped between February July. The differences in sporidial numbers in the two fields were most pronounced during peak periods of sporidial production, especially from July to September 1985 and August to October 1987.

Effect of temperature

Sporidial counts were highest when the temperatures were in the range of 31–32°C (max.) to 20–21°C (min.) in both fields. There was a gradual increase in mean sporidial number with increasing temperatures from 27 to 30°C (max.) and 16 to 21°C (min.); the numbers however, declined significantly when temperatures exceeded 32°C (max.) and 22°C (min.) (Fig. 2a, b). In all the 4 years (Figs. 1a–d) the peak of sporidial counts occurred when the maximum temperatures were around 30°C, except in 1986 (Fig. 1c) when the peak was reached at below 30°C in January in both fields. Correlations of sporidial numbers, with maximum and minimum temperatures were negative and significant ($P < 0.05$) in 1984, and that with maximum temperatures combined over 4 years were negative and significant (Table 1).

Effect of RH

There was a gradual increase in the numbers of sporidia trapped with increasing RH, and the maximum number of sporidia occurred when maximum RH was $> 80\%$. This usually occurred from about mid-July to early November (Figs. 1a–d). Peaks in sporidial counts were generally recorded in both fields when the maximum RH was above 90% and the minimum RH 41–50%;

the mean maximum numbers of sporidia trapped per 80 mm² were 1126 in RP 13 and 481 in RL 19 (Fig. 2c, d). The lowest numbers of sporidia were observed when minimum RH was below 40%. Correlations of mean sporidial numbers with maximum RH, and minimum RH, were positive and significant ($P < 0.05$) over 4 years (Table 1).

Effect of rainfall

Maximum numbers of sporidia were observed in both the fields when there was 11–30 mm rainfall (Fig. 20). The highest sporidial counts were obtained during the end of September to mid-October with rainfall ranging from 13 to 136 mm in 1984, 1985 and 1987 (Figs. 1a, b and d), but in 1986 (Fig. 1c) the highest sporidial count was obtained in January; with heavier rains there were reductions in sporidial counts, except in 1987.

Effect of wind speed

Wind speed varied from 4 to 23 km/h during the 4 years (Figs. 1a–d). Maximum sporidia were trapped in both fields when wind speeds were in the range of 4–6 km/h. With increasing wind speed, there was a decline in sporidial number, and at wind speeds > 19 km/h sporidial counts were minimal in both fields (Fig. 2f). Correlations between sporidial numbers (log values) and wind speed were negative for all the years and it was significant ($P < 0.05$) during 1986, as well as for all the years combined (Table 1). Although wind directions were variable throughout the year, the wind direction during September–November was generally south-easterly i.e. from RP 13 towards RL 19.

Effect of weather factor combinations

The stepwise regression analysis of environmental factors against sporidial counts (log values) identified three significant factors (RHmax., wind speed \times RHmax, and RHmin.)

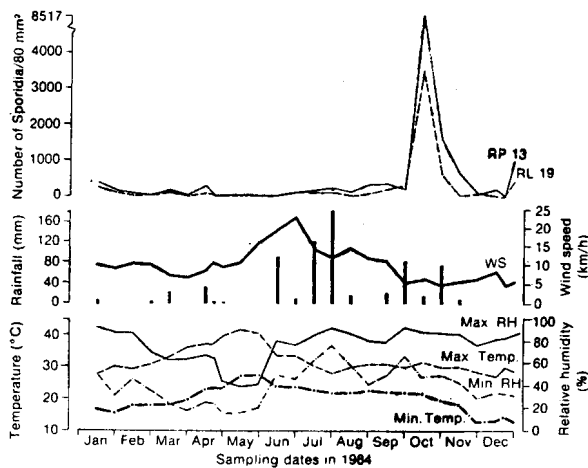


Fig. 1a

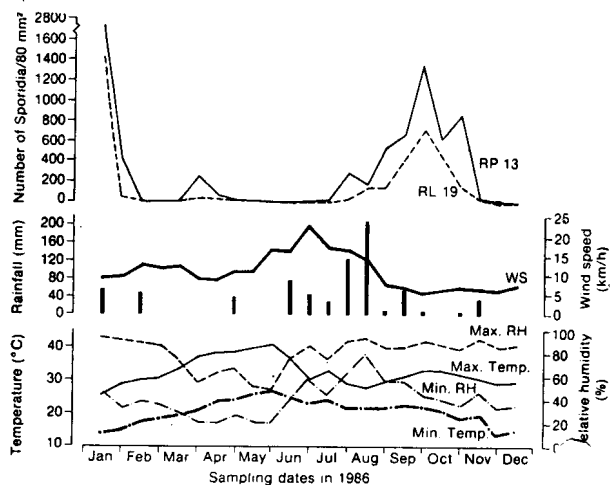


Fig. 1c

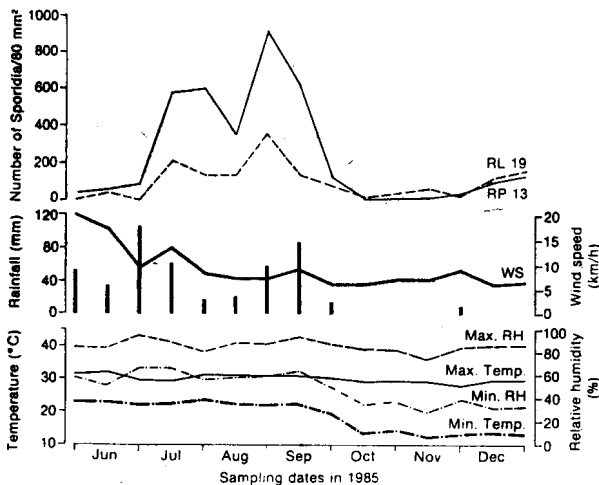


Fig. 1b

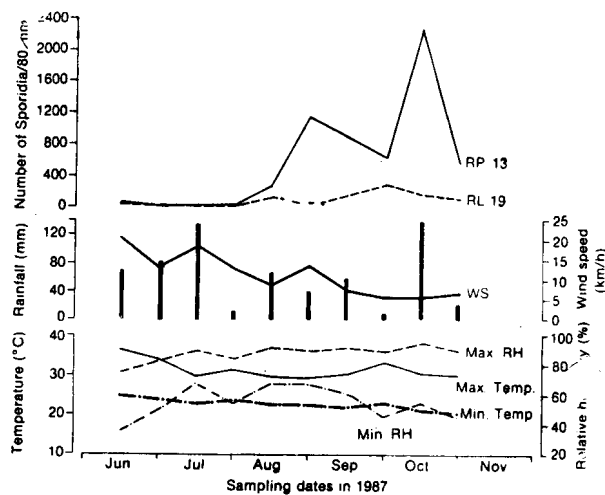


Fig. 1d

Fig. 1. Number of *Tolyposporium penicillariae* sporidia trapped, using rotorod samplers, in relation to temperature (max. and min.), RH (max. and min.), wind speed, and amount of rainfall in two fields (RP 13, a pearl millet smut nursery, and RL 19, not a smut nursery) at ICRISAT Center during a. 1984; b. 1985; c. 1986; and d. 1987. Note that each data point for each environmental factor is the mean of a 2-week period.

Table 1. Correlations between environmental factors and *Tolyposporium penicillariae* sporidial counts (log-transformed values) during four years in two fields, one a pearl millet smut nursery (RP 13) and the other not a smut nursery (RL 19) at ICRISAT Center

Environmental factors	1984		1985		1986		1987		Overall	
	RP 13	RL 19	RP 13	RL 19	RP 13	RL 19	RP 13	RL 19	RP 13	RL 19
Temp. (max.)	-0.67*	-0.40*	0.39	0.11	-0.21	-0.31	-0.39	-0.09	-0.42*	-0.31*
Temp. (min.)	-0.42*	-0.25	0.58*	-0.09	-0.06	-0.10	-0.56	-0.41	-0.14	-0.20
RH (max.)	0.71*	0.46*	0.38	-0.10	0.25	0.38	0.56	0.34	0.49*	0.36*
RH (min.)	0.50*	0.32	0.61	-0.010	0.28	0.39	0.09	-0.26	0.41*	0.24*
Rain fall	0.15	0.25	0.42	0.32	0.09	0.15	-0.14	0.07	0.13	0.10
Wind speed	-0.37	0.32	-0.05	-0.47	-0.48*	-0.49*	-0.62	-0.57	-0.40*	-0.44*

*Significant at $P=0.05$

for RP 13 and two significant factors (wind speed \times Temp. max., and Temp. max. \times RHmin.) for RL 19.

DISCUSSION

The two fields used in this study were similar in every respect except that smut inoculum was more plentiful in RP 13 than in RL 19, and accordingly sporidial counts in RP 13 were consistently higher than in RL 19. Among the environmental factors, temperature and RH seem to be significant in production of sporidia and wind speed for sporidial dispersal. The maximum number of sporidia were trapped at 31-32°C maximum air temperature, with >80% maximum RH. At temperatures above 35°C, and RH<80%, relatively few sporidia were trapped, indicating that high temperatures and low RH (max.) may not be conducive for sporidial production. Low sporidial counts during certain months (May-June) despite good rains could be due to the prevailing high air and soil temperatures (35-40°C), and low maximum RH (<76%) which could affect teliospore germination. During the summer 1987 following

light rains there was an increase in sporidial number to a certain extent, but with excessive rains (>180 mm/2-week period) sporidial counts declined significantly. In December 1984 and 1985 more than 100 sporidia/80 mm² were trapped despite no rains during the period; this may have been because of soil wetting in the RP 13 field caused by irrigation given to the postrainy season groundnut crop.

High RH is a critical factor in smut development⁷, and, in the present study, the maximum sporidia were trapped when the maximum RH was >90%, a condition that generally prevails during the rainy season, the main season of pearl millet production. This explains why smut becomes severe when flowering coincides with wet weather.

The correlations of mean sporidial numbers over 4 years with temperature (max), RH (max. and min.) and wind speed were generally weak but significant for both fields. Sporidial counts were generally higher during September-October, when air temperatures were moderate and RH higher than in other months. In the smut

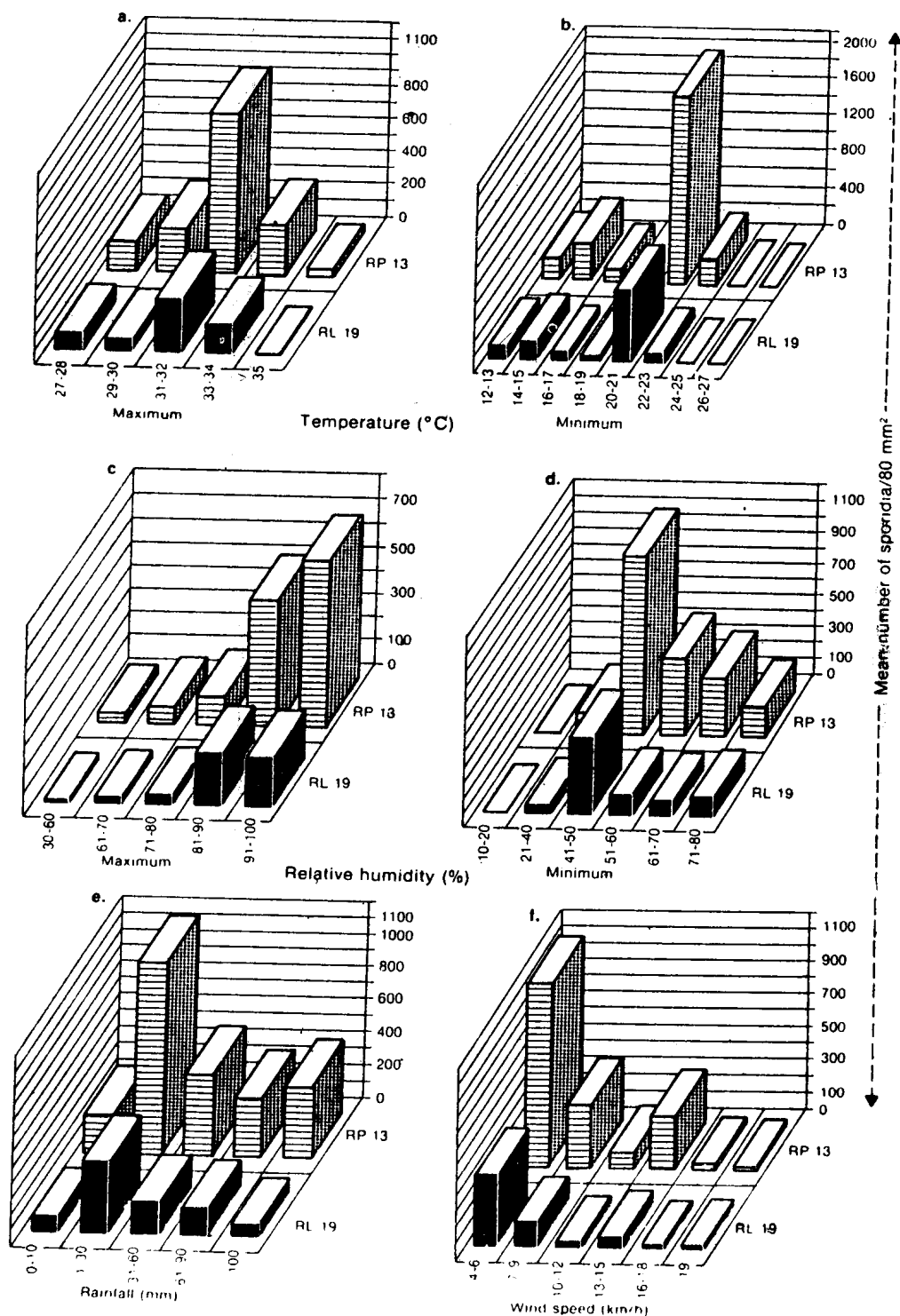


Fig. 2. Mean numbers of *Tolyposporium penicillariae* sporidia, collected, using rotorod samplers in relation to a. maximum temperature; b. minimum temperature; c. maximum RH; d. minimum RH; e. amount of rainfall; and f. wind speed in two fields (RP 13, a pearl millet smut nursery, and RL 19 not a smut nursery) at ICRISAT Center farm over 4 years (1984-1987).

nursery (RP 13) where sprinkler irrigations were provided on rain-free days for about a month during which only two spore samplings were done, there was no significant temperature effect, however, there was a significant effect in RL 19 where sprinkler irrigation was not provided.

The seasonal distribution patterns of airborne sporidia indicate that they are abundant in the air during the period from early July to mid-November, and to a lesser extent during the dry season following light rains or irrigations. At ICRISAT Centre, artificial inoculations for out screening start at the end of July, and teliospores from these early-infected plants could also contribute to the increased airborne sporidial number from mid-August onwards. Teliospores produced from these infected panicles could infect the late-planted (late August) pearl millet at ICRISAT Centre.

Dissemination of sporidia from RP 13 to RL 19 with south easterly wind during the peak period of sporidial production is clearly indicated by relatively higher sporidial counts during the September–November period than in other months, except in 1985. Sporidial trapping was generally more effective at lower wind speeds than at higher wind speeds probably because at higher wind speed sporidia are blown to greater distances and there is less concentration in the location of the rotorods. The relationship between the number of sporidia may also depend on the prevailing wind speed and direction. In studies with barley loose smut, caused by *Ustilago nuda*, under fluctuating wind directions, Mathre and Johnson obtained a linear relationship between infection and the distance from the inoculum source⁸.

The results thus indicate the significance of environmental factors, particularly temperature, RH and wind speed in release and dispersal of *T. penicillariae* sporidia. We, however, realize

that based on the limited weather data set, it will be inappropriate to generalize the findings of this study for a wider application. More elaborate studies, which would include more locations, more frequent spore sampling, using an improved sampling procedure, and precise measurements of the microenvironment in the fields, supported by studies in controlled environment will be helpful to clearly understand the relationship between various weather factors and smut development in pearl millet.

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References

1. Rachie, K.O. & Majumdar, J.V. 1980. Pearl millet. Pennsylvania State University Press, University Park, PA. pp. 307.
2. Thakur, R.P. & King, S.B. 1988. Smut disease of pearl millet. Information Bulletin no. 25. Patancheru, A.P. 502 324, India: International Crops Research Institute for the Semi-Arid Tropics. pp. 17.
3. Subba Rao, K.V. & Thakur, R.P. 1983. *Tolyposporium penicillariae*, the causal agent of pearl millet smut. *Trans Br. mycol. Soc.* **81** : 597-603.
4. Bhatt, R.S. 1946. Studies in Ustilaginales. 1. The mode of infection of the bajra plant by smut *Tolyposporium penicillariae* Bref. *J. Indian Bot Soc* **26** : 163-186.
5. Ajrekar, S.L. & Likhite, V.N. 1933. Observations on *Tolyposporium penicillariae* (The bajra smut fungus). *Curr. Sci.* **1** : 215.
6. Vasudeva, R.S. & Iyengar, M.R.S. 1950. Secondary infection in bajra smut disease caused by *Tolyposporium penicillariae* Bref. *Curr. Sci.* **19** : 123.
7. Thakur, R.P., Subba Rao, K.V. & Williams, R.J. 1983. Evaluation of a new field screening technique for smut resistance in pearl millet. *Phytopathology* **73** : 1255-1258.
8. Mathre, D.E. & Johnston, R.H. 1975. Dispersion of loose smut in foundation barley seed fields in a semi-arid climate. *Plant Dis Rep.* **59** : 979-981.