

Factors associated with ergot resistance in pearl millet

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Abstract : The host and host-pathogen interaction factors associated with resistance to ergot (*Claviceps fusiformis*) was studied in resistant, intermediate and susceptible cultivars of pearl millet. Protogyny, time between full protogyny and anthesis initiation, and stigma length were significantly shorter and seed set was significantly higher in the resistant and intermediate cultivars than in the susceptible cultivars. Latent periods were comparable in the three groups of cultivars, but the sporulation period, ergot severity, and numbers of conidia and sclerotia produced were significantly lower in the resistant and intermediate than in the susceptible cultivars. Implications of these findings in ergot epidemiology and management are discussed.

Keywords : *Pennisetum glaucum*, *Claviceps fusiformis*, resistance, epidemiology

Infection of pearl millet [*Pennisetum glaucum* (L.) R. Br.] florets by *Claviceps fusiformis* Loveless occurs through fresh stigmas, and withering of stigmas either due to ageing or pollination prevents infection (Thakur and Williams, 1980). Development of constriction in the style, which leads to stigma withering, occurs within 6 h after pollination and more than 36 h after ergot inoculation (Willingale and Mantle, 1985, 1987; Willingale *et al.*, 1986). Protogyny (the time between stigma initiation and anthesis initiation), and the time between full protogyny (the ergot inoculation stage) and anthesis initiation (FP-AI period) are considered as major factors related to self-pollination and ergot resistance in pearl millet. Host-pathogen interaction factors, such as latent period (time between inoculation and honeydew appearance), ergot severity, sporulation period,

and the quantity of conidia and sclerotia produced are likely to be associated with ergot resistance levels of pearl millet lines. This study was conducted to understand the relationships among various factors associated with ergot resistance in some selected cultivars of pearl millet.

MATERIALS AND METHODS

In a greenhouse experiment, six pearl millet cultivars, two resistant (< 10% mean severity) ICMPEs 1, ICMPEs 2 to ergot, two intermediate (10-30% mean severity) ICMPEs 8, ICMPEs 9, and two susceptible (> 30% mean severity) 842A, BK 560 were grown in plastic pots (23-cm dia) filled with a mixture of field soil and farmyard manure 1:1 (v/v), fertilized, and watered regularly. Each cultivar was grown in 5 pots with 3-4 plants per pot. The inoculum used in the experiment was an aqueous conidial suspension (1×10^6 conidia/ml), obtained from panicles of a susceptible pearl millet cultivar BK 560 inoculated with a monoconidial isolate of

C. fusiformis grown on Kirchoff's medium for a month at 25°C.

In each cultivar, five panicles (on the main shoots) were covered at the boot-leaf stage with parchment selfing bags to record the protogyny, FP-AI period and seed set. Bags were briefly opened daily at 0900 and 1600 h to record various observations. At full protogyny, 10 florets from the middle portion of a panicle were removed with a pair of forceps and measured for stigma length (portion protruded from the glume) with a scale and a magnifying lens (15x).

Ten panicles were inoculated (Thakur *et al.*, 1982) in each cultivar; five for recording latent period, ergot severity and sclerotial formation, and the other five for measuring sporulation period (duration of honeydew production) and estimating total conidial production. Ergot severity and sclerotial formation (all infected florets may not support sclerotial development) were recorded using an ergot severity assessment key (Thakur and Williams, 1980).

In each of five inoculated panicles of susceptible and intermediate cultivars, a cluster of five infected florets in the middle of the panicle, and isolated infected florets in resistant cultivars (clusters of five infected florets not found in resistant cultivars) were isolated by carefully clipping the surrounding florets with a pair of fine scissors at the very first appearance of honeydew. Honeydew exudates were collected daily at 0900 h from each of the five florets with a fine camel-hair brush into 10 ml sterile distilled water in screw-capped glass vials throughout the duration of honeydew production. The vials were stored at 5°C in a refrigerator until macroconidia (henceforth referred as conidia) were counted using a haemocytometer. The total number of conidia produced per floret per day, and per floret for a 7-day period (the maximum number of days conidia were produced by any panicle of any cultivar) were calculated.

Data were subjected to proper statistical analyses to determine significant treatment differences and relationship among various factors.

RESULTS AND DISCUSSION

Effect of host factors

The protogyny period varied from 42 h in resistant ICMPEs 1 to 142 h in susceptible 842A. The susceptible cultivars had significantly ($P < 0.05$) longer protogyny than the intermediate and resistant cultivars (Table 1). Between susceptible cultivars, 842A had significantly longer protogyny than BK 560. The FP-AI time period varied from 26 h in ICMPEs 1 to 113 h in 842A. The susceptible cultivars had again significantly ($P < 0.05$) longer FP-AI period than the intermediate and resistant cultivars. Stigmas were shortest in ICMPEs 1 (0.9 mm) and longest in 842A (4.1 mm). The susceptible cultivars had significantly longer stigmas than the intermediate and resistant cultivars. Even within each of the three groups, 842A, ICMPEs 9 and ICMPEs 2 had longer stigmas than BK 560, ICMPEs 8 and ICMPEs 1, respectively (Table 1). The susceptible BK 560 had no seed set under selfing, while the resistant and intermediate cultivars had 84-98% seed set. The results indicated that higher susceptibility to ergot in pearl millet is associated with the longer protogyny, the FP-AI period and stigma length. In susceptible cultivars, because of long protogyny, stigma withering occurs much before anthesis commences and this results in reduced self-pollination, and thus poor seed set and increased ergot infection. In resistant cultivars, because of shorter protogyny, self-pollination is quicker, and thus increased seed set and less ergot infection (Willingale *et al.*, 1986). The FP-AI period could be crucial for the resistance that is based on rapid self-pollination. Cultivars with longer FP-AI periods like those with longer protogyny are generally more susceptible than those with shorter FP-AI periods. The longer stigma of susceptible cultivar exposes larger surface area for infection than

Table 1: Measurements of various host factors in pearl millet cultivars, susceptible (S), intermediate (I) and resistant (R) to ergot in greenhouse at ICRISAT Centre

Cultivars	Ergot reaction ^a	Protogyny period (h) ^b	Time period FP-AI (h) ^b	Stigma length (mm) ^c	Seed set (%) ^b
842A	S	142	113	4.1	0
BK 560	S	105	76	3.0	0
ICMPES 8	I	57	29	1.9	84
ICMPES 9	I	66	48	2.3	97
ICMPES 1	R	42	26	0.9	98
ICMPES 2	R	53	33	1.9	97
CD ($P = 0.05$)		25.5	19.0	0.20	7.3

- a. Based on several years of testing in ICRISAT pearl millet ergot nursery, S = > 30% mean ergot severity; I = 10-30% mean ergot severity; and R = < 10% mean ergot severity.
- b. Mean of 5 panicles under bagged conditions;
FP = Full protogyny when > 75% stigmas have emerged,
AI = Anther initiation.
- c. Mean of 10 fully exerted stigmas in the middle portion of the panicles.

the shorter stigmas of intermediate and resistant cultivars.

Under weather conditions favourable for ergot infection (drizzling rains and cool temperature), pollination is greatly reduced due to pollen wash and the susceptible cultivars, which have longer protogyny and FP-AI period, and larger stigmas become more vulnerable to ergot infection than the intermediate and resistant cultivars.

Host-pathogen interaction factors

Although there was no significant difference in latent period, it was longest (157 h) in resistant ICMPES 1, shortest (108 h) in intermediate ICMPES 9, and in susceptibles 842A and BK 560, it was 113-136 h (Table 2). The sporulation period was shortest (48 h) in resistant ICMPES 2 and longest (125 h) in susceptible BK 560. The resistant cultivars had significantly shorter sporulation period than the intermediate and the susceptible cultivars. Both ergot severity and sclerotial formation in resistant and intermediate cultivars were significantly

lower than in susceptible cultivars. Resistant cultivars showed up to 2% ergot severity and no sclerotial formation compared with 23-29% ergot severity and 3-12% sclerotial formation in intermediate cultivars, and 87-89% ergot severity and 37-39% sclerotial formation in susceptible cultivars (Table 2).

Correlations of protogyny and stigma length were positive and significant ($P < 0.01$) with ergot severity and sclerotial formation, and were negative ($P < 0.01$) with seed set. Similarly, there were positive and significant correlations between duration of sporulation and ergot severity, the FP-AI period and ergot severity, and stigma length and ergot severity.

The daily mean conidial production in resistant cultivars was generally lower than in intermediate and susceptible cultivars, except on day 1 (Fig. 1). The peaks for conidial production occurred on day 1 for resistant, day 2 for intermediate and day 3 for susceptible cultivars. After a sharp decline from the peaks, conidial

Table 2 : Measurements of various host-pathogen interaction factors in pearl millet cultivars, susceptible (S), intermediate (I), and resistant (R) to ergot in greenhouse at ICRISAT Centre

Cultivars	Ergot reaction ^a	Latent period (h) ^b	Sporulation period (h) ^b	Ergot severity (mm) ^b	Sclerotia formation (%) ^c
842A	S	113	82	89	39
BK 560	S	136	125	87	37
ICMPES 8	I	116	77	23	3
ICMPES 9	I	108	82	29	12
ICMPES 1	R	157	58	2	0
ICMPES 2	R	133	48	1	0
CD ($P = 0.05$)		16.1	17.9	9.4	16.7

- a. Based on several years of testing in ICRISAT pearl millet ergot nursery; S = > 30% mean ergot severity; I = 10-30% mean ergot severity; and R = < 10% mean ergot severity.
 b. Based on mean of five artificially uninoculated panicles.
 c. Percentage of florets producing sclerotia.

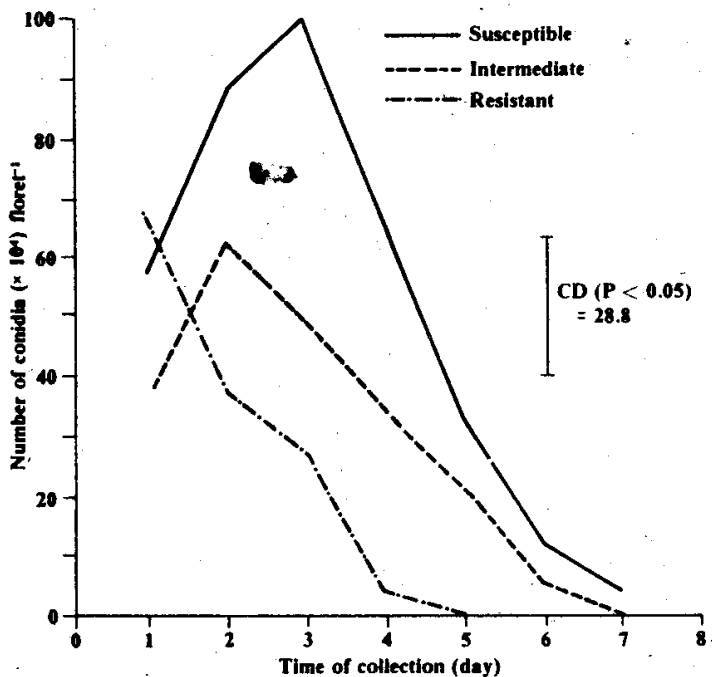


Fig. 1 : Production of conidia in honeydew from a single floret of pearl millet panicles artificially inoculated with *Claviceps fusiformis* in a greenhouse experiment. Individual data point is the mean number of conidia produced from 5 florets of each of 5 panicles of two susceptible (842A, BK 560), two intermediate (ICMPES 8, ICMPES 9) and two resistant (ICMPES 1, ICMPES 2) cultivars. Day 1 indicates the first appearance of honeydew symptoms and day 7 the last appearance of honeydew on any cultivars.

production continued up to day 7 for susceptible, day 6 for intermediate and day 5 for resistant cultivars. The total conidial production over a 7-day period and the rates of conidial production per floret per day were significantly higher in susceptible than resistant cultivars (Fig. 2).

A significant positive correlation ($r = 0.81$) between sporulation period and ergot severity indicates the degree and duration of susceptibility of cultivars. A susceptible cultivar would thus produce conidia for a longer time, would remain susceptible for a longer time and catch more inoculum than a resistant cultivar. The results, thus; suggest that a resistant cultivar would not only be less infected and contribute much less inoculum, but it would also support much less sclerotial formation than a susceptible cultivar.

These findings lead us to conclude that the best means of managing ergot in pearl millet is through the use of ergot-resistant cultivars. Several ergot-resistant lines developed by ICRISAT that are also resistant to smut and downy mildew, and have high grain yield potential (Thakur and King, 1988; Thakur *et al.*,

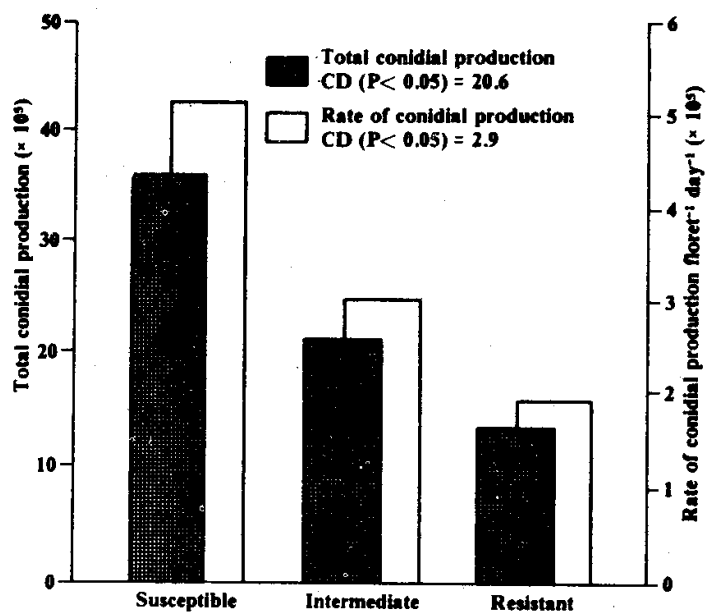


Fig. 2 : Production of total conidia and the rate of conidial production per floret per day by susceptible (842A, BK 560), intermediate (ICMPES 8, ICMPES 9), and resistant (ICMPES 1, ICMPES 2) cultivars of pearl millet following artificial inoculation with *Claviceps fusiformis* in a greenhouse experiment. Each bar represents the mean conidial numbers from two cultivars in each group.

1988), provide the best sources of resistance available for utilization in resistance breeding programmes and for growing in the areas where ergot is a problem.

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