Effect of Pattern and Severity of Moisture Deficit Stress on Stalk Rot Incidence in Sorghum
I. Use of Line Source Irrigation Technique, and the Effect of Time of Inoculation

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INTRODUCTION

Root and stalk rots in sorghum [Sorghum bicolor (L.) Moench] are most commonly caused by the charcoal rot pathogen [Macrophomina phaseolina (Tassi) Goid.]. This fungus can cause significant grain yield reductions in sorghum (Dodd, 1980) as a result of either lodging, or premature termination of plant growth, or both (Rosenow, 1980). Leyendecker and Nakayama (1958) recorded 50% loss in grain yield in New Mexico. Losses in fodder production due to this disease are also significant in semi-arid India (Khune et al., 1980).

It is now fairly well accepted that stalk rot can be a major problem when the vegetative parts of the host plants experience inadequate carbohydrate supply or depletion due to remobilization to the growing grains during the grain-filling period (GS3, time interval between anthesis to physiological maturity; Eastin, 1971; Dodd, 1980). In sorghum grown in the semi-arid tropical or temperate regions, water deficit (water stress) at the end of the rainy season is commonly associated with a high incidence of this disease. It is also widespread in the post-rainy season crop grown on stored soil moisture in India. Results from ICRISAT's International Sorghum Charcoal Rot Nursery (Rao and Williams, 1979) indicated that the disease was most prevalent at dry sites and in years when rainfall was inadequate, and the soil profile was not sufficiently wet during GS3.

To date, systematic studies of the quantitative relationship between stress and disease incidence have been difficult because of the high variability in disease incidence within fields. A further major difficulty has been the simulation of different soil moisture levels in a precise and controlled fashion in the field. The overall objective of our study was to demonstrate the use of a line source (LS) sprinkler irrigation system (Hanks et al., 1976) to develop a soil moisture gradient in an attempt to examine the relationship between water stress and disease incidence. The specific objectives were: 1) to examine the relationship between water stress during GS3 and charcoal rot incidence; 2) to study the effect of inoculation with the fungus on disease incidence; and 3) to test the stage of plant growth after flowering during which the inoculation is most effective and plants are most vulnerable to the disease.

MATERIALS AND METHODS

Design and management of experiments

Two experiments were conducted at ICRISAT Center (17°32'N, 78°16'E) on an Alfisol during the post-rainy seasons (October to February) during 1977-79. Before planting, fertilizer was applied at 100 kg N, and 26 kg P ha⁻¹. High plant-population density (18 plants m⁻²) was used in both experiments.
Crops were adequately protected from insects by applying carbofuran (Rallis India Ltd, Bombay, India; 40 kg ha$^{-1}$ at planting). The plots were hand-weeded thrice during the season. There was no disease problem except charcoal rot incidence. Crops were watered fully by either surface or sprinkler irrigations until the water stress treatments were imposed. In both the experiments sorghum hybrid CSH 6, susceptible to charcoal rot, was used.

**Experiment I (Expt. I)**

This experiment was conducted to determine: 1) the method required to ensure a suitable soil moisture gradient using the LS; and 2) the time of inoculation required to promote satisfactory disease levels. The crop was sown on 19 October 1977, on ridges in a 90-cm deep Alfisol with a (plant-available) water-holding capacity (ASM) of 60 mm. Figure 1 shows the experimental lay-out in the field. There were 4 replications with 9 main plots (treatments T1-T9) of 16 rows (at 0.75 m spacing), each parallel to the LS, and each 3.2 m in length.

Treatments were assigned to the main plots at random: 5 for dates of inoculation; 2 for growth analysis; 1 for crop and soil moisture studies; and 1 non-inoculated control. The 16 rows in each main plot were further divided into 8 subplots of 2 rows each spanning the moisture gradient from maximum (1.5 m away from the LS to the center of subplot 1), to zero application (in the farthest 8th subplot 12 m away from the LS; driest).

* Mention of a trade name does not mean endorsement by ICRISAT or discrimination against other brands.
The LS irrigation treatments commenced 44 days after sowing (DAS), or approximately 2–3 weeks before flowering (50% anthesis), in order to ensure that plants in subplots at the far end experienced sufficient water stress due to depletion of soil water stored before the start of LS irrigations. Water was applied four times, at weekly intervals. The amount of water applied during each of the first two irrigations was approximately 20 mm near the LS. Less water (only 15 and 13 mm near the LS) was applied during the third and fourth weeks, because the class A pan evaporation rates were slightly lower, and the transpiration was expected to decline as the crop approached maturity. On 8 January 1978 (81 DAS) 17 mm of rainfall was received, which temporarily relieved the water stress.

At weekly intervals from flowering, inoculations with *Macrophomina phaseolina* (Tassi) Goid. were made on all plants in the row farthest from the LS of each subplot using inoculum grown on toothpicks (Rao et al., 1980). The first row of each subplot was the non-inoculated control, for which sterile toothpicks were used.

**Experiment II (Expt. II)**

The main objective of this experiment was to test a more simplified methodology than in Expt. I, and to confirm the previous results. This experiment was planted on 10 November 1978, in a 130-cm deep Alfisol (\(ASM = 94\) mm). The LS was placed as described in Expt. I creating two replicates, one on either side of the sprinkler system. Plots (30 × 15 m) were sown in a flat seed bed in rows spaced at 0.6 m. In order to prevent surface movement of water, the main line pipes were placed in a 0.1-m deep drain, and LS irrigation were carried out intermittently to ensure that no runoff occurred. Each replicate contained 10 main plots of 3 × 18 m row length along the LS. These were used for inoculations (2 plots), soil moisture measurement (1), plant water status evaluation (1), growth analysis and final harvest (6). Out of the 30 rows in each main plot, the first row adjacent to the LS was left out, and the next 24 were used for the experiments. These 24 rows were further subdivided into 8 subplots with 3 rows along the moisture gradient. The five rows farthest from the LS served as a border. In all, three LS irrigations were given at 50, 61 and 77 DAS. The first row in each subplot was reserved for yield estimation (without inoculation); the second was used as the non-inoculated control, and the third for inoculation. Plants were inoculated using the toothpick method at 2 and 4 weeks after flowering (Rao et al., 1980) in the respective subplots.

In both experiments the LS irrigations were applied during relatively windless periods of the night. The pressure of water in the first sprinkler was approximately equal to 0.2 MPa in Expt. I, and 0.25 MPa in Expt. II.
Collection and analysis of data

The crops in both the experiments were manually harvested almost immediately after physiological maturity during the dry periods, and the oven-dried grain and dry-matter yields were determined. In Expt. I data on plants with and without stalk rots in each subplot were collected separately. Flowering (50% anthesis) and physiological maturity (black layer formation; Eastin, 1971) were monitored in each subplot.

The water applied by the LS was measured using sets of seven catchcans regularly placed at right angles to the sprinkler line. In Expt. I, the soil moisture was monitored by gravimetric sampling at 30-cm intervals to a depth of 90 cm. In Expt. II, eight sets of neutron probe access tubes were placed in each replication. There were two tubes in each subplot. Soil moisture was monitored to a depth of 128 cm before sowing, before and after each irrigation, and at harvest. Deep drainage was negligible under these experimental conditions (Sachan et al., 1984), runoff was prevented, and therefore changes in soil moisture were used to calculate evapotranspiration (ET) between irrigations. The cumulative ET between the treatment period (i.e., from the date of first LS irrigation at 50 DAS to harvest at 94 DAS) was calculated, and the relationship between cumulative ET and distance from LS was established (Fig. 3). The cumulative stress experienced by the different subplots in Expt. II was estimated by subtracting the amounts of ET of each of the subplots from the wettest subplot (nearest to the LS = 0.9 m); this was expressed as percentage of ET during the treatment period in the wettest subplot, and called ET deficit.

Relative water deficits (LS applied water) were calculated as the difference between the amount of water received at a subplot and that at the subplot nearest to the LS, and expressed as percentages. Leaf-water potentials (LWP; Expt. I) were measured each week at midday with a pressure chamber (PMS Instrument Co., Oregon, USA) as described by Scholander et al. (1964). Fully expanded and illuminated second or third leaves from the top were cut across half their width at the midportion along their length and placed in between folds of wet cheesecloth supported by polystyrene insulation material. Four leaves were sampled in each subplot and the results averaged. A cumulative-stress index was calculated by using the following formula:

\[ SI = (LWP_i - LWP_s) \times 7 \]

Where: SI is cumulative stress index in MPa days; LWP_i is LWP at the point of maximum water application (see the intercepts in Fig. 4); LWP_s is LWP of the plants in the given subplot (calculated from regressions of LWP versus distance from the LS; Fig. 4); and 7 = days between two successive measurements.

Since the quantitative relationship between the systematically imposed stress
levels and the degree of disease incidence was our major concern, a regression approach was used for the data analysis. The effects of inoculation treatments (timing or levels of inoculation) were analyzed using the split-plot design; the least significant difference (LSD) was calculated only for treatment factors that were randomly laid out in the field experiments.

RESULTS

Water application, crop phenology and grain yield

The LS was very effective in creating a gradient of soil moisture in both the experiments. The correlation between water applied during each irrigation and the distance from the LS was linear and highly significant ($r > 0.95; P < 0.001$). Hence the cumulative water application (4 times) through the LS in Expt.1 was also linear ($r = 0.99; P < 0.001$) with distance from the sprinkler line (Fig. 2). The total application over the 4-week period ranged from approximately 70 mm nearest to the LS to almost nil approximately 10 m away. Soil moisture measurements confirmed this gradient. For example, the percentage of differ-
ences between the gravimetric soil water content at field capacity, and at dif-
ferent distances away from the LS on the day after the first LS irrigation were
(with the distance in paranthesis): 6.34 (0.75 m), 8.18 (5.25 m), 9.54 (9.75 m), and 10.10 (12.75 mi) in the top 0-30 cm soil layer; the corresponding values for the soil layer between 30-60 cm from the surface were 1.05, 1.48, 3.44, and 4.9. (LSD<sub>0.05</sub> to compare deficits at different distances from the LS in each of these two layers = 2.8%). The difference between the moisture content of the soil layer below 60 cm at the two ends of the irrigation gradient was less than 0.7%, and statistically not significant.

The gradient in the water applied was reflected in the crop phenology and the grain yields measured in the uninoculated control plots. Both flowering and physiological maturity of plants in the driest subplots in different replications were observed a week earlier (58 and 91 DAS, respectively) than in the wettest subplot. The grain yields ranged from > 500 g m<sup>-2</sup> adjacent to the LS (where the crop did not suffer any significant stress) to approximately 300 g m<sup>-2</sup>, 11.6 m away from it (where the crop depended entirely on the moisture stored in the soil for grain filling; Fig. 2). The decline in grain yields with distance from the LS was linear (<i>r</i> = 0.97; <i>P</i> < 0.001). The diseased plants yielded on an average 11.6% less than the healthy plants in the same subplot, although statistically the difference was not significant.

The cumulative water application in Expt. II was also linear with distance from the LS (<i>r</i> = 0.98; <i>P</i> < 0.001). The amounts of water applied were approximately 60 (1 LS irrigation), 50 (II), and 20 mm (III) near the LS. In addition, there were rains of 6.6 mm at 71 DAS, 28.0 mm at 80 DAS, and 6.6 mm at 86 DAS. However, the crops in different subplots were nearing physiological maturity. The trends in disease development, based on visual observations, and grain yields were not affected by the rains. Flowering differences across the irrigation gradient were not significant in Expt. II, but the crops under drier subplots matured 3-4 days earlier than those near the LS. The plants were harvested following physiological maturity at 94 DAS. The measured soil moisture content across the subplots after each LS irrigation, and before second and third irrigation showed a linear decline in moisture with the increasing distance from the LS (Sachan et al., 1984). The difference in cumulative ET during the treatment period between the wettest and driest subplots (Fig. 3) was approximately 80 mm.

The water applied in Expt. II reached a greater distance (13 m) from the LS than in Expt.I since slightly higher pressure was used. Although the linear correlation of distance from the LS and the cumulative ET was high (<i>r</i> = 0.94; <i>P</i> < 0.001), the data suggested that there were only small differences in the actual ET across the 5 m from the LS (Fig. 3). Soil moisture in this area was apparently sufficient to compensate for the differences in water applied by the LS. The grain yield was also found to decline linearly with the increasing distance from the LS (Fig. 3).
Effect of decreasing water supply on crop evapotranspiration (ET) during treatment period, grain yield, and charcoal rot incidence (inoculated at 4 weeks after flowering) (Expt. II).

Regression equations (n = 8):
- Evapotranspiration: \( Y = 498.34 - 16.73X \) (\( r = 0.91^{**} \));
- Grain yield: \( Y = 229.81 - 9.12X \) (\( r = 0.94^{***} \));
- Plants with soft stalk: \( Y = 13.37 + 3.31X \) (\( r = 0.89^{**} \)).

**Crop water stress**

The first three measurements of LWP in Expt.I were made 50, 57, and 64 DAS (just prior to the second, third, and final LS irrigations). During this period the LWP of the subplot nearest to the LS changed little, but the LWP of more distant subplots decreased significantly (Fig. 4). In the eighth (driest) subplot from the LS, for example, the midday LWP declined from approximately -1.5 MPa to -2.2 MPa during this period.

During the fourth (71 DAS) and fifth (78 DAS) weeks (after the termination of the LS irrigations), plant water stress also increased in the subplots closest to the LS, although the gradient in LWP was still maintained (with the eighth subplot approximately 0.6 MPa drier than the subplot adjacent to the LS; Fig. 4). The differences between the data collected on 71 and 78 DAS were small because of lower evaporative condition on these days. The 17 mm of rain at 81 DAS temporarily relieved the stress as measured at 85 DAS but in the final measurement at 92 DAS severe water stress was again evident. By this time the gradient in LWP had disappeared and the plants were already mature (with most of the leaves dead).
Fig. 4. Midday leaf-water potential at different distances from the LS on different days after sowing. Regressions are not significant (NS) for 85 and 92 DAS data \((n=16)\) (Expt. I).

\[
\begin{array}{ll}
\text{DAS} & \text{Regression} \\
50 & Y = -1.24 - 0.023X \ (r=0.55) \\
57 & Y = -1.25 - 0.041X \ (r=0.80) \\
64 & Y = -1.38 - 0.06X \ (r=0.86) \\
71 & Y = -1.84 - 0.032X \ (r=0.57) \\
78 & Y = -1.77 - 0.037X \ (r=0.63) \\
85 & Y = -1.50 - 0.010X \ (r=0.34) \\
92 & Y = -2.22 - 0.001X \ (r=0.03) \\
\end{array}
\]

The cumulative stress index for the entire period between 50 and 92 DAS was clearly differentiated between the different subplots in Expt.I (Fig. 5). Near the LS the cumulative stress increased linearly with time but as the distance from the LS increased, it had a tendency to increase more exponentially. The computed ET deficits in Expt. II linearly increased from the wettest subplot to the driest one (Fig. 3).
Charcoal rot incidence

Charcoal rot incidence (expressed as the percentage of plants in a row with soft stalks, i.e., with evidence of active fungal growth in the stem) in the plants in plots inoculated at the optimum time for disease development (5 weeks after flowering in Expt. I, and 4 weeks after flowering in Expt. II; see below) increased in an approximately linear fashion with increasing moisture stress (Figs. 2 and 3). There was some deviation from linearity in Expt. I, as the incidence differed little between the last three sampling positions (Fig. 2). This however, is consistent with the water application data in that these three positions were all basically unaffected by the LS irrigations (Fig. 2).

Disease incidence was less in Expt. II, especially adjacent to the LS (15% as compared to 30% in Expt. I), but the slopes of the overall relationship of incidence and distance from the LS (Figs. 2 and 3) were not statistically different.
weeks later (42% in control).)

In Experiment II, the mean disease incidence was 4% lower in the inoculated plants than in the control plants. The disease incidence in Experiment I was 6% higher in the control plants than in the inoculated plants. In Experiment III, the disease incidence was 10% lower in the inoculated plants than in the control plants.

In both experiments, there were significant differences in disease incidence between the inoculated and control plants. In Experiment II, the disease incidence was 10% lower in the inoculated plants than in the control plants. In Experiment III, the disease incidence was 15% lower in the inoculated plants than in the control plants. In Experiment IV, the disease incidence was 20% lower in the inoculated plants than in the control plants.

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inoculated control. In contrast, in the plots inoculated at 4 weeks after flowering, no significant reduction in yield was recorded.

Charcoal rot severity

In addition to data on charcoal rot incidence estimated by the percentage of plants with soft stalk, data were also collected on the severity of infection, measured by the number of nodes crossed by the fungus, and the total length of fungal spread (cm) from the point of inoculation within the plant stem. In both experiments, all three measures of disease incidence were very highly correlated \((r \text{ in all cases } > 0.86; P < 0.001)\) and were affected in a similar fashion by stress. In Expt. II, the mean number of nodes crossed increased from less than 0.5 near the LS to approximately 2.0 at the dry end of the gradient with inoculation at 2 weeks after flowering; and from approximately 1.5 to 3.5 following inoculation 4 weeks after flowering, when stress was generally more severe (Fig. 6). The length of spread of the fungal mycelium showed a similar response.

However, at the 4-week inoculation date (Expt. II) both measurements of disease severity departed from linearity (Fig. 6) at both the ends of the gradient. The fewer nodes crossed, and length of fungal spread in plants in the three subplots nearest to the LS probably reflected the small difference in ET between these subplots (and presumably in stress). There was also a tendency for severity to reach a maximum approximately 10 m from the LS if inoculated at 4 weeks. However, linearity in the response between 3–10 m distance from LS is clearly seen.

DISCUSSION

Use of the line-source sprinkler irrigation system to study charcoal rot incidence

Charcoal rot is well-known for its high degree of readiness to exploit host plants subject to water and other environmental stresses (Jordan et al., 1984). It is very difficult to simulate consistently the exact conditions required for the infection and spread of this disease. An irrigation gradient established by a line source sprinkler, as demonstrated in both the experiments during the dry season (Sachan et al., 1984) is useful in quantitatively relating disease severity to moisture stress, on a minimum land area. As the water application at right angles to the LS varies linearly to the point at which no water is applied, the distance from it can be used to indicate the level of stress. The use of distance as an independent variable has several advantages in routine studies aimed at genotype screening for reduced susceptibility and high grain yield (and for comparing results collected in different experiments or during differ-
Fig. 6. Effect of time of inoculation and distance from the LS on A) number of nodes crossed, and B) length of spread (cm) of the fungus (Expt. II).

Regression equations ($n = 8$):

Nodes crossed with inoculation at
- 2 weeks after flowering: $Y = 0.32 + 0.25X (r = 0.95^{**})$;
- 4 weeks after flowering: $Y = 1.41 + 0.31X (r = 0.88^{**})$;

Length of spread (cm) with inoculation at
- 2 weeks after flowering: $Y = 7.00 + 1.51X (r = 0.91^{**})$;
- 4 weeks after flowering: $Y = 14.99 + 1.68X (r = 0.73^{*})$;

different stages of the crop). Hence the use of distance as the indicator of stress level in line source experiments is very common (Sivakumar et al., 1981; Johnson et al., 1982; O'Neill et al., 1983; Spaeth et al., 1984).

The grain and dry matter yields, and the percentage of plants free of disease
Fig. 7. Effect of relative water deficit on relative grain or biomass yield, and the absence of disease (Expts. I (A) and II (B)).

decreased linearly with increasing water deficits (i.e., with increasing distance from the LS) (Fig. 7). In both experiments the slopes of grain yield (Figs. 2 and 3) versus distance were less steep than those for water applied or for ET. To compare differences among genotypes (or the effects of applied fertilizer, plant density, etc.,) one can use the intercepts, slopes and area under regression lines for disease incidence (or mean disease incidence across all subplots)
as described to compare genotypes for drought resistance (Seetharama et al., 1982). If no disease occurs near the LS, the distance from the LS to the first subplot showing a significant or a predetermined level of disease incidence in the drier area of the gradient can be used as a single index of disease susceptibility, as suggested for drought resistance screening (O'Neill et al., 1983).

Although water stress has been shown to be the primary cause promoting disease incidence, the interacting role of high temperature cannot be ignored, especially as these two stress factors frequently occur together when sorghum is grown in the field (Jordan et al., 1984). The LS treatment not only results in a gradient of soil moisture, but also a gradient of (surface) soil temperature. For example, in another experiment on similar soils, the soil surface temperatures were 29.9°C at the near end, and 43.4°C at the far end of LS, 4 days after LS irrigation, at midday when the air temperature was 30.5°C (N. Seetharama, unpublished data). Both these stress factors may work synergistically in disease incidence. However, it is clear that water stress is far more important than heat stress, as either at 35 or at 40°C no incidence was noted with 80% available moisture, whereas the disease was severe with 20% moisture at both temperatures (Edmunds, 1964). In future studies it may be useful to evaluate the separate and combined role of these factors in disease incidence (Jordan et al., 1984).

Agronomic significance of charcoal rot incidence

It is difficult to separate disease and water stress effects on grain yield. The grain yield reduction in plants showing incidence of charcoal rot late in GS3 is more likely due to water stress rather than due to disease, as illustrated by the lack of reduction in grain yield due to inoculation at 4 weeks in Expt. II, in which inoculation resulted in nearly two-fold increase in disease. However, the decrease in grain yield in the plots inoculated at 2 weeks after flowering compared with those of the noninoculated plots in Expt. II suggested that more severe infection during early GS3 in inoculated plots can reduce grain yield. A similarly small difference between the yields of healthy and diseased plants in Expt. I also suggests that the disease is economically important. Wide-scale lodging after maturity can be disastrous during the rainy season, especially if the lodged crops are caught in the rain before harvest, or are to be combine-harvested. In places where ratooning is practised, severe charcoal rot incidence can result in a poor ratoon crop, even if water is available at the time of ratooning (Choudhari et al., 1977). As delaying severe stress would postpone the time of rapid disease development, this fact should be considered in disease management.
Fig. 8. Relationships between cumulative stress at different subplots from 44 DAS to either 3 or 5 weeks after flowering, and charcoal rot incidence (Expt. I).

Soft stalk data \((n=8)\) plotted for:
- Inoculation at 5 weeks vs cumulative stress up to 5 weeks after flowering:
  \[Y = 33.23 + 0.035X \ (r=0.90^\prime)\];
- Inoculation at 3 weeks vs cumulative stress up to 3 weeks after flowering:
  \[Y = 15.65 + 0.64X \ (r=0.81^\prime)\];
- Inoculation at 3 weeks vs cumulative stress up to 5 weeks after flowering:
  \[Y = 15.72 + 0.037X \ (r=0.83^\prime)\].

**Stress effects on charcoal rot incidence**

Disease incidence was closely related to cumulative stress expressed as either cumulative LWP (Expt. I) (Fig. 8) or cumulates ET deficit (Expt. II) (Fig. 9). The rates of increase in disease incidence with increasing level of stress (slopes in Figs. 8 and 9) were the same for different times of inoculation when the cumulative stress from the time of first LS irrigation to physiological maturity was considered. However, the slope was steeper for the 3-week-after-flowering inoculation data, when only the cumulative stress between flowering and inoculation was considered (Fig. 8). Under more severe stress conditions, this relationship would depend upon a number of factors such as the stage of phenological development (see below), and alterations in the source-sink relations.
Inoculation at 4 weeks vs ET deficit upto 4 weeks from 50 DAS

Inoculation at 2 weeks vs ET deficit upto 2 weeks from 50 DAS

Inoculation at 2 weeks vs ET deficit upto 4 weeks from 50 DAS

Fig. 9. Relationships between charcoal rot incidence (measured as number of nodes crossed by the fungus) in response to inoculation at 2 or 4 weeks after flowering, and relative ET deficits from 50 DAS up to either 2 or 4 weeks after flowering (Expt. II).

Linear regressions with following data for nodes crossed:
- Inoculation at 4 weeks vs ET deficit up to 4 weeks after flowering: \( Y = 1.39 + 0.036X (r = 0.91''') \);
- As above, but polynomial regression:
  \( Y = 1.71 - 0.037X + 0.003X^2 - 0.00036X^3 (r = 0.98'''') \).
- Inoculation at 2 weeks vs ET deficit up to 2 weeks after flowering: \( Y = 0.019 + 0.073X (r = 0.94'''') \);
- Inoculation at 2 weeks vs ET deficit up to 4 weeks after flowering: \( Y = 0.36 + 0.027X (r = 0.93'''') \).

(Seetharama et al., 1987). The trend for curvilinear response of the number of nodes crossed with increasing stress levels in Fig. 9 is similar to that in Fig. 6.

**Disease expression and measurement**

*Time of, and response to, inoculation.* Since the fungus only invades decaying tissue, (Katsanos and Pappelis, 1965), late inoculation resulted in more severe infection (Table I, and Fig. 6). In Expt. I the incidence of the disease was higher due to inoculation at 5 weeks after flowering than at 3 weeks after flowering for the same amount of stress (Fig. 8). Under severe stress, stalk senes-
ence occurs earlier because of the premature termination of growth. Thus the plots further away from the LS show symptoms of severe disease incidence much earlier than those near it. Under such severe conditions charcoal rot incidence was as great in the non-inoculated as in the inoculated plants. Although inoculation at 4 weeks after flowering in Expt. II nearly doubled the percentage of stalk rot, the incidence in non-inoculated plots themselves was high enough to study the effect of stress on disease. The difference in grain yield between inoculated and non-inoculated controls in Expt. II also was not significant with inoculation at 4 weeks after flowering. Thus, inoculation offered little advantage in screening genotypes for charcoal rot resistance under such conditions.

Effect of crop phenology. Since terminal water stress accelerates both flowering and maturity of the crop (thereby changing sink strength at a given date), comparison across moisture levels may be confounded by the effect of stress on phenology per se. In the present experiments, this confounding effect was partly eliminated since the inoculations were done at specific times starting from the date of flowering in each subplot. A week’s difference in flowering between plants in the driest and wettest subplots in Expt. I, and the absence of any such difference in Expt. II can be explained by the shallow soil on which the first crop was grown, and the early start of LS irrigations compared with Experiment II. Maturity differences were, however, significant in both experiments. Thus, measuring disease incidence on the same date in all treatments may be misleading; instead, the phenological time (e.g., a defined stage of grain development after flowering) could be more useful. Ideally data should be collected on several occasions, and a cumulative index should be calculated.

Measurement of disease incidence. Although the three parameters of disease incidence (i.e., soft stalk percentage, number of nodes crossed, and length of spread) are highly correlated amongst themselves, there was a tendency for the second and third parameters to show less linearity of response at the near and far end of the LS (broken line, Fig. 6) than soft stalk incidence, which showed a linear response throughout the irrigation gradient (Fig. 3). The marginal decrease in length of spread in the drier plots in Expt. II, however, was at least partially an artifact of the effect of stress on plant height: height was considerably reduced (shorter internodes) in the plots farthest from the LS. In addition, at the far end of the LS, the plants matured soon after inoculation at 4 weeks after flowering, and hence disease spread was limited at harvest. In the first three subplots near the LS the differences in ET were small, and hence the predisposing effects of water stress were similar in all. Thus, while for preliminary screening, monitoring the percentage of stalks damaged may be enough, for detailed studies (or in final-stage screening) other parameters of
Disease incidence may also be used as suggested by Rao and Balasubramanian (1983).

CONCLUSIONS

1. The crops, unless they were very severely stressed in the beginning of GS3, were very sensitive to charcoal rot under moisture stress during the later stage of grain development. A study of plant environment and the plant physiology at that stage would be useful for better understanding of disease expression and control. A quantitative relationship between stress intensity during grain filling and disease incidence can be established by measuring either leaf-water potential or evapotranspiration.

2. The three different parameters of disease incidence are highly correlated, but deviations do occur due to the differences brought about by other effects of stress such as changes in crop phenology and plant height. For routine screening, monitoring of soft stalks only is suggested.

3. Under severe stress non-inoculated plants had as much charcoal rot incidence as inoculated plants. Under such conditions, natural disease incidence will be high and there is little need for artificial inoculation.

4. Results from both the experiments suggest that the LS can be successfully employed to study the relationships between water supply, grain yield, and charcoal rot incidence. Charcoal rot is known for its high variability in the field, and its extreme sensitivity to environmental factors. Creation of stress gradient conditions in the field over a small area overcomes many of the experimental limitations in the study of this soil-borne pathogen.

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REFERENCES


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