Screening of Sugarcane Varieties/Lines against Whip Smut Disease in Relation to Epidemiological Factors

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Abstract
Whip smut caused by (*Ustilago scitaminea*) is an important fungal disease, which is widely distributed all over the world causing huge losses in sugarcane crop. Sugarcane crop basically required humid and hot climate for its development which is also favorable for the different diseases in sugarcane. Epidemiological factors play an important role for the development and the management of different diseases. They are also used as disease prediction models. Epidemiological factors are very important for the development and spread of pathogen causing smut of sugarcane. Out of fifteen promising varieties/lines, eight were found resistant (S2006-US-469, S2006-US-272, S2005-US-54, S2008-US-130, S2008-AUS-190, S2008-AUS-107, S2009-SA-169), six were moderately susceptible(S2008-M-34, S2008-AUS-133, S2003-US-127, S2003-US-704, S2008-Fd-19, S2008-AUS-87) and one (S2003-US-618) had susceptible reaction against the disease. There was positive correlation of relative humidity with disease incidence and negative correlation of maximum and minimum temperature with disease incidence.

Keywords: Sugarcane; Smut disease; Epidemiological factors

Introduction
Sugarcane (*Saccharum officinarum* L.) (Punjabi: Ganna, Urdu: Naishkar, Kamad) belongs to family *Poaceae* and crop is grown under 30° north to 30° north latitude with climatic conditions ranging from sub-tropical to tropical regions [1]. In Pakistan, Sugarcane is cultivated on a range of one million hectare. Sugarcane growing zones of Pakistan fall between 24° N latitude in Sindh to 34° N latitude in KPK. Pakistan is at fifth position with respect to Sugarcane production in the world. The most important thing is that the sugar industry shares of Pakistan economy about 1.9% of GDP [2]. There are numerous restrictions, including diseases such as Whip Smut, Red Rot, Pokkah Boeng, Red Stripe, Rust and Sugarcane Mosaic and Brown stripe [3,4]. In 1877 the smut of sugarcane was first time reported in (Natal) South Africa. Whip Smut is extremely critical disease of Sugarcane wherever the crop is grown. Whip smut is caused by *Ustilago scitaminea*, which belongs to the phylum Basidiomycota [5] occurs in a few physiological races [6,7]. The temperature ranges between (25-30°C) supports the disease development. The smut of sugarcane is prevalent in all the world countries where the sugarcane crop is cultivated. Use of susceptible varieties show more losses because of intensive cultivation, secondary infection, and poor management practices [8]. 52-73% yield losses occur in ratoon crops [9]. Sandhu, [10] specified yield losses of 70.7% to 75.3%. Total crop failure is possible if susceptible varieties are used and conditions are favorable for disease development [11]. It can cause major losses as well as juice quality losses. 3-7% sucrose content of infected variety is reduced [12]. Disease incidence increase was found to be linked with increasing age of the crop and varietal susceptibility. After 120 days of planting the appearance of the apical whips was found. When the second flush of whip was produced, it produces very large quantity of teliospores and these spores effect the lateral and terminal buds of rapidly growing crop. The emergence of the third level of whips and the infection caused by this level is supposed to be very serious in the epidemiology of whip smut disease [13].

Smut inoculation techniques in sugarcane plantlets and examined the chance of screening for smut resistance at the plantlet period. Injury paste technique was found the extreme event of whip smut production, followed by paste; on the other hand, soaking method had the minimum occurrence of smut [14]. The susceptible varieties show significant losses due to poor management practices, secondary infection and intensive cultivation. The most suitable and economical process to control the disease is the use of resistant varieties. The resistant germplasm of sugarcane plays a leading role for assessment of resistant varieties through breeding program [15]. Disease development is dependent on the environmental conditions and the resistance of the sugarcane varieties grown. The most recognizable diagnostic feature of a smut infected plant is the emergence of a "smut whip" [16]. According to Sreeramulu [17], the day time dispersal of spores is maximum. The maximum dispersal of spores takes place at 24 to 27°C and 50 to 60% R.H. Crop age and cycle at the time of infection appear to be important [18]. Resistance of a variety retain only for a few years. A variety resistant previously pertaining race may become susceptible to the new physiological race with change in climatic conditions. Pre-release evaluation of varieties/lines, is therefore, important in relation to epidemiological factors. The objective of my present research to screen the sugarcane clones for the smut tolerance and to study the influence of epidemiological factors on the occurrence of smut disease in sugarcane. This research work was based on the hypothesis, through evaluation of sugarcane varieties in relation to epidemiological factors may be helpful for management of whip smut disease. Use of resistant lines/varieties along with proper management practices and study of epidemiological factors will be helpful to reduce the losses caused by whip smut.

Materials and Methods
Fifteen (15) varieties/lines were grown in field area of Ayub Agriculture Research Institute (AARI), Sugarcane Research Institute
(SRI), Faisalabad during 2015-2016. The varieties were planted under RCBD design with three replication. The plot size was kept as 2.4 m width and 3 m length [19]. The varieties sown were cut into small pieces (setts). The length of one sett was about 45 cm with 3 buds present on it. Forty eight (48) sets of each variety were taken for three row plantation (with 16 sets in one row). These setts were dipped in spore suspension for 30 min prior to plantation. The fungal spores entered into the cane setts which were used to evaluate the disease incidence. Plantation of sugarcane inoculated setts was done in February 2015 in three meter long plot under RCBD design with three replications / repeats at sugarcane experimental area, Sugarcane Research Institute (SRI), Faisalabad in clay loam soil. Thus each treatment was comprised of 48 smut-inoculated setts per variety [20]. Data for the number of smutted tillers was collected with a regular interval of 30 days. The data was collected by counting the number of smutted tillers in each variety. The layout plan for the sugarcane varieties was made in such a way that there were 15 varieties, 3 replications and 3 factors (maximum temperature, minimum temperature and relative humidity) were studied with respect to disease incidence in each variety each month. Data was collected monthly from June to December 2015. To collect the data, number of smutted tillers and total number of tillers were counted.

**Meteorological data**

Meteorological data for temperature and humidity were collected from the meteorological department, AARI, Faisalabad. Meteorological data was in the form of computerized spread sheet on which day to day information for maximum temperature, minimum temperature and relative humidity was listed. Meteorological data was calculated to conclude mean values of maximum temperature, minimum temperature and relative humidity for the whole month.

**Statistical analysis**

The analysis of the information was done based on the percentage of infected strains of the last observation and these were processed using the statistical parametric analysis for randomized blocks.

Correlation and regression analyses with epidemiological factors to determine the relationship between epidemiological factors and disease incidence. The prediction equation used was

\[
Y = a + b1x1 + b2x2 + b3x3
\]

Where, \(Y\) = Predicted disease incidence; \(a\) = Intercept; \(b1 – b3\) = Regression coefficients; \(X1\) = Average maximum temperature (°C), \(X2\) = Average minimum temperature (°C), \(X3\) = Average relative humidity (%), \(R2\) = coefficient of determination.

**Recording smut incidence**

The trial was closely monitored for appearance of first smut whips and recorded monthly intervals until the trial was completed (Table 1).

**Smut reaction**

Due to its vegetative mode of propagation sugarcane is prone to infect by systemic pathogens. Among, smut disease caused by *Ustilago scienmae* is a dreadful disease of sugarcane and is endemic in most of the tropical regions. The most eco-friendly means to contain the pathogen through the use of resistant varieties/lines. In the present investigation, fifteen sugarcane promising clones were evaluated for their resistance against whip smut pathogen under field conditions. It was concluded that, out of fifteen promising lines/varieties eight were found resistant, six moderately susceptible and one had susceptible reaction against the disease (Table 2). The resistance/susceptibility of the variety were determined by bud morphological characters. In the most resistant varieties the germplasm adapted sub apical position in the bud whereas the susceptible varieties the position was apical. The position was considered to be associated with the tendency of the bud to sprout which makes it more vulnerable to the entry of promycelium and hence more prone to infection. Hence, bud scales acted as morphological barrier and restricted smut pathogens. Source of resistant against whip smut available in sugarcane clones and it can be further manipulated through breeding program for evolution of new high yielding sugarcane varieties [15,21].


**Epidemiological factors on smut incidence**

Temperature (max. temperature and min. temperature), relative humidity are important factors in smut epidemiology.

**Characterization of environmental conditions conductive for whip smut of sugarcane disease development on seven varieties**

Seven varieties of sugarcane (S2003-US-618, S2008-M-34, S2008-AUS-133, S2003-US-127, S2003-US-704, S2008-Fd-19, S2008-AUS-87) showed significant correlation with temperature (maximum and minimum) and relative humidity. These varieties were employed to characterize the critical ranges of environmental conditions (maximum and minimum temperature and relative humidity) conductive for the

<table>
<thead>
<tr>
<th>Response</th>
<th>Disease incidence (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Resistant</td>
<td>R</td>
</tr>
<tr>
<td>Moderately Resistant</td>
<td>MR</td>
</tr>
<tr>
<td>Moderately Susceptible</td>
<td>MS</td>
</tr>
<tr>
<td>Susceptible</td>
<td>S</td>
</tr>
</tbody>
</table>

Table 1: Smut description, rating and infection were done as explained by Rao et al.

<table>
<thead>
<tr>
<th>Varieties</th>
<th>D.I. (%)</th>
<th>Response</th>
</tr>
</thead>
<tbody>
<tr>
<td>S-2003-US-618</td>
<td>46.43</td>
<td>S</td>
</tr>
<tr>
<td>S-2008-M-34</td>
<td>25.02</td>
<td>MS</td>
</tr>
<tr>
<td>S-2006-US-469</td>
<td>0</td>
<td>R</td>
</tr>
<tr>
<td>S-2006-US-272</td>
<td>0</td>
<td>R</td>
</tr>
<tr>
<td>S-2005-US-54</td>
<td>0</td>
<td>R</td>
</tr>
<tr>
<td>S-2008-AUS-133</td>
<td>18.45</td>
<td>MS</td>
</tr>
<tr>
<td>S-2008-AUS-130</td>
<td>0</td>
<td>R</td>
</tr>
<tr>
<td>S2003-US-127</td>
<td>21.80</td>
<td>MS</td>
</tr>
<tr>
<td>S-2006-US-658</td>
<td>0</td>
<td>R</td>
</tr>
<tr>
<td>S-2008-AUS-190</td>
<td>0</td>
<td>R</td>
</tr>
<tr>
<td>S-2008-Fd-19</td>
<td>24.42</td>
<td>MS</td>
</tr>
<tr>
<td>S-2008-AUS-107</td>
<td>0</td>
<td>R</td>
</tr>
<tr>
<td>S-2008-AUS-87</td>
<td>19.34</td>
<td>MS</td>
</tr>
<tr>
<td>S-2009-SA-169</td>
<td>0</td>
<td>R</td>
</tr>
</tbody>
</table>

Table 2: Evaluation of sugarcane clones to smut (%) incidence.
whip smut of sugarcane disease development. The results demonstrate that at maximum temperature of 38.75°C the variety S2008-AUS-133 showed the minimum disease incidence of 3.28% (Figure 1). While at the minimum temperature of 25.5°C the variety S2003-US-618 showed the maximum disease incidence of 46.52% (Figure 2). In case of relative humidity at 70.5% relative humidity the minimum disease incidence of 3.28% was recorded in the variety S2008-AUS-133 (Figure 3).

These results clearly demonstrated that the maximum temperature and minimum temperature were negatively correlated with the whip smut disease incidence, while relative humidity was positively correlated with the whip smut disease incidence as shown in Table 3.

**Discussion**

Whip smut of sugarcane (U. scitaminea) is very destructive disease in all sugarcane grown areas of the world. It usually causes losses from germination to maturity of the crop. There was a need to highlight resistant lines among different clones of sugarcane. To fulfill this need, research on screening of different varieties was done on the basis of disease rating scale [22]. Conditions are critically important in the development and spread of the pathogen causing smut of sugarcane. Some of these can be utilized to form the basis of disease prediction model. They may vary in their combinations in different agro climatic zones and influence not only the pathogen but also the host. The present findings are in accordance with Sreeramulu et al. [17] reporting that there is definite diurnal and seasonal rhythms in the spore incidence, the day time dispersal of spores is maximum. The maximum dispersal of spores takes place at 24 to 27°C and 60 to 70% R.H. The difference in diseases severity may be attributed to the environmental conditions. Factors such as maximum temperature, minimum temperature and relative humidity were studied with special reference to the varietal reactions of different varieties. It was observed that all the factors maximum temperature, minimum temperature and relative humidity had statistically significant correlation with varieties. Disease severity was maximum at temperature range 25-27°C. With the decrease in temperature 38-25°C from June 2015 to December

![Figure 1: Correlation of maximum temperature with whip smut disease incidence on different varieties.](image1)

![Figure 2: Correlation of minimum temperature with whip smut disease incidence on different varieties.](image2)
2015, disease incidence or severity was increased. On the other hand, it had been observed that disease severity was maximum at relative humidity range 65-70%. With increase in relative humidity 65-70%, overall disease incidence was increased whereas Singh and Budhraja, [23] reported that disease incidence was maximum at optimum temperature of 28°C as this temperature favours the maximum growth of smut pathogen (Ustilago scitaminea). The smut spores are killed instantaneously at 62°C but can survive more than three day in ice [24,25]). The smut spores are killed

\[ Y_i = 57.76 - 1.48x \quad r = 0.87 \]

\[ Y_4 = 30.94 - 0.66x \quad r = 0.83 \]

\[ Y_5 = 48.23 - 1.01x \quad r = 0.80 \]

\[ Y_6 = 38.31 - 0.81x \quad r = 0.83 \]

\[ Y_7 = 57.76 - 1.48x \quad r = 0.87 \]

**Table 3:** Correlation of environmental factors with whip smut of sugarcane disease on different varieties.

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Varieties</th>
<th>Max Temp. (°C)</th>
<th>Min Temp. (°C)</th>
<th>RH (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>S2008-AUS-133</td>
<td>-0.89**</td>
<td>-0.96**</td>
<td>0.79</td>
</tr>
<tr>
<td>2</td>
<td>S2008-M-34</td>
<td>-0.97**</td>
<td>-0.85**</td>
<td>0.91**</td>
</tr>
<tr>
<td>3</td>
<td>S2003-US-127</td>
<td>-0.99**</td>
<td>-0.80**</td>
<td>0.94**</td>
</tr>
<tr>
<td>4</td>
<td>S2003-US-704</td>
<td>-0.98**</td>
<td>-0.92**</td>
<td>0.92**</td>
</tr>
<tr>
<td>5</td>
<td>S2008-Fd-19</td>
<td>-0.98**</td>
<td>-0.93**</td>
<td>0.90**</td>
</tr>
<tr>
<td>6</td>
<td>S2008-AUS-87</td>
<td>-0.98**</td>
<td>-0.89**</td>
<td>0.91**</td>
</tr>
<tr>
<td>7</td>
<td>S2003-US-618</td>
<td>-0.98**</td>
<td>-0.88**</td>
<td>0.94**</td>
</tr>
</tbody>
</table>

Upper value indicate Pearson’s correlation coefficient, while the lower value indicate level of significance at 1% (0.00-0.01) and at 5% (0.02-0.03) probability.

**Conclusion**

It can be concluded that the intensity of sugarcane smut incidence highly influenced by the epidemiological factors. The prevalence of optimum temperature during the crop stage of germination to tillering, increased the setts and soil borne teliospores germination subsequently it may give rise to infection hyphae which are capable of infecting sugarcane bud. In addition, the temperature has an enhanced effect on the release and dispersal of smut spores in the air. Fifteen sugarcane varieties / lines were screened out to find the resistant lines. When these fifteen varieties /lines were compared on the basis of recommended scale under natural conditions, Eight (8) varieties or lines (S2006-US-469, S2006-US-272, S2005-US-54, S2008-AUS-130, S2006-US-658, S2008-AUS-190, S2008-AUS-107, S2009-SA-169) were graded as resistant, (S2008-M-34, S2008-AUS-133, S2003-US-127, S2003-US-704, S2008-Fd-19, S2008-AUS-87) were found as moderately susceptible and (S2003-US-618) was found susceptible. Maximum Disease Incidence was observed at (25-27°C) and at R.H (65-70%) and minimum disease incidence was observed at (38.75°C) and at R.H (48.5%). Environmental conditions especially maximum temperature, minimum temperature and relative humidity, which showed that maximum and minimum temperature and relative humidity had great influence on the incidence of whip smut disease of sugarcane. There is a negative correlation between maximum and minimum temperature and disease incidence whereas the correlation between relative humidity and disease incidence was recorded as positive.

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