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**Journal of the Saudi Society of Agricultural Sciences**

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## FULL LENGTH ARTICLE

# Study of southern corn leaf blight (SCLB) on maize genotypes and its effect on yield

Samavia Mubeen, Mazhar Rafique, Muhammad Farooq Hussain Munis, Hassaan Javed Chaudhary \*

Department of Plant Sciences, Faculty of Biological Science, Quaid-i-Azam University, Islamabad 45320, Pakistan

Received 2 June 2014; revised 30 April 2015; accepted 30 June 2015

### KEYWORDS

*Cochliobolus heterostrophus*;  
 Maize genotype;  
 Southern corn leaf blight;  
 Inbred line;  
 Hybrid

**Abstract** Southern corn leaf blight is considered the most devastating disease of maize crop, which causes noticeable reduction in crop yield. Inbred lines are useful because they are genotyped, multiple time phenotyping is possible, and genetic uniformity, genetic stability and its vigor make inbred lines suitable to study in diversified environment. In present investigation, 12 maize genotypes viz: NC-2703 (hybrid), NC-2003 (hybrid), SP-3 (inbred line), NCML-73 (inbred line), NRL-6 (inbred line), NRL-4 (inbred line), Soan-3 (variety), Rakaposhi (variety), Margala (variety), EV-1097 (variety), Local-Y (variety), Local-W (variety) were tested against southern corn leaf blight under laboratory and field conditions. According to disease severity scale (0–5) inbreds SP-3 and NCML-73 were found highly resistant; Local-W moderately resistance and rest of the genotypes were least resistance in *in vitro* analysis. In field screening, Margala, NRL-4, EV-1097 showed maximum resistance followed by moderately resistant SP-3, NCML-73, NC-2703, NRL-6 and Local-Y maize genotypes. NC-2003, Rakaposhi and Soan-3 showed least resistance during field evaluation. *Cochliobolus heterostrophus* showed considerable effects on yield of crop. Significant difference was found in grain yield, plant height, ear height and ear weight while ear placement, ear per plant and infected ear data were non-significant. The results clearly showed the effect on maize genotypes and its yield.

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## 1. Introduction

Maize (*Zeamays* L.) is third most widely grown cereal crop after rice and wheat in Pakistan (Hussain et al., 2011). It belongs to family Poaceae (grass family) and tribe Maydaea. Maize is a highly cross pollinated crop henceforth it does not survive in its wild form (Ram and Singh, 2003; Bothastr and Schlicher, 2005). Maize crop is infected by approximately 65 pathogens which include fungi, bacteria and viruses (Rahul

\* Corresponding author. Tel.: +92 051 90643004.

E-mail addresses: [samavia.mubeen@gmail.com](mailto:samavia.mubeen@gmail.com) (S. Mubeen), [mazharrafique88@gmail.com](mailto:mazharrafique88@gmail.com) (M. Rafique), [plantpath7@gmail.com](mailto:plantpath7@gmail.com) (M.F.H. Munis), [hassaan\\_javed@yahoo.com](mailto:hassaan_javed@yahoo.com) (H.J. Chaudhary).

Peer review under responsibility of King Saud University.



Production and hosting by Elsevier

<http://dx.doi.org/10.1016/j.jssas.2015.06.006>

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Please cite this article in press as: Mubeen, S. et al., Study of southern corn leaf blight (SCLB) on maize genotypes and its effect on yield. Journal of the Saudi Society of Agricultural Sciences (2015), <http://dx.doi.org/10.1016/j.jssas.2015.06.006>

and Singh, 2002). Leaf blight, stalks rot, seedling blight and smuts are the most important diseases of corn crop (Hafiz, 1986). Southern corn leaf blight (SCLB) is an important foliar disease of maize crop and caused by fungi *Cochliobolus heterostrophus*, and also known as *Bipolaris maydis* (ascomycetes). It is reported from most maize growing regions but most devastating in hot and humid tropical and temperate areas of the world. Almost 70% yield loss is recorded due to SCLB (Wang et al., 2001; Ali et al., 2011a).

Use of inbred lines for research activities is an important factor in genetic uniformity. It is preferable to use inbred lines. They have advantages including prior characterization, stability, vigor and uniformity along with the potential for research information. Most inbred lines shows a high degree of genetic stability and reproducibility, besides that in some cases unstable inbred lines have also been documented (Bogenschutz and Russell, 1986). Three races (C, O and T) of *B. maydis* have been identified in maize crop. Race O and T are most prevalent in Pakistan while race C is only identified in China. Severity and symptoms of the disease depend on the host germplasm and the race of the pathogen (Ali et al., 2011a). Race C shows high pathogenicity to cultivar with Charrua male-sterile cytoplasm (Wei et al., 1988). Race O is most prevalent in tropical and sub-tropical areas. It infects the broad range of maize genotype despite the type of cytoplasm. When susceptible lines were inoculated with Race O, 50% yield loss is recorded (Fisher et al., 1976; Gregory et al., 1978). Initially Race O causes small lesion and becomes diamond shape and rectangular as they mature. These lesions are restricted to leaf veins (Ali et al., 2011b). Race T is considered as the most common and attacks the maize cultivar with Texas male-sterile cytoplasm (cms-T). It is the major cause of epidemic in USA in 1970 and 1971. It attacks on leaves, husks as well as ears and produces small lesion on maize (Ullstrup, 1972). Race O and T can be discriminated by pathogenicity test of cms-T plants and physiological/morphological characters on culture media (Leonard, 1977; Warren et al., 1977). Randomly amplified polymorphic DNA (RAPD) and DNA finger printing are also used for race discrimination (Nicholson et al., 1993).

Current practices for controlling disease and crop improvement involve selection, formation, evaluation, genetic recombination of inbred lines or variable families and use of synthetic pesticide (Emmert and Handelsman, 1999; Pixley et al., 2006). Inbred lines are more useful because of the number of reasons such as they have been entirely genotyped, their multiple time phenotyping is possible and moreover their cost effectiveness makes them more suitable to study various traits in diversified environment (Atwell et al., 2010). Development of resistance varieties has been the primary way for the control of southern corn leaf blight. Maize crop is resistant to pathogen with normal cytoplasm. SCLB disease caused by race T has been controlled by eliminating cms-T cultivars from cultivar of elite germ plasm (Hyre, 1970; Ullstrup, 1972). A broad range of maize genotype is attacked by race O. The *rhm* recessive gene of *C. heterostrophs* confers resistance to race O (Zaitlin et al., 1993). Various screening methods including detached leaf techniques (Lakshmi and Sharma, 1987), tissue culture (Kuehnle and Earle, 1988), and seedling assay (Tajimi et al., 1985) for disease resistance have been investigated. Conventional breeding or recurrent selection is effective method to improve resistance against SCLB in breeding (Shieh and Lu, 1993).

This research was aimed to determine the resistance level of different maize inbred lines genotype against the disease SCLB through the process of screening and evaluation because of its prime importance to address the fungal disease. As a consequence the information acquired from the study could be of vital use for the scientific research of phytopathologists, plant breeders and also for the farmers.

## 2. Materials and methods

### 2.1. Isolation of *C. heterostrophus*

The causal organism of SCLB (*C. heterostrophus*) was isolated by the technique of Ricker and Ricker (1935). The infected leaves of three different isolates viz: V-31, V-32 and B-202 were cut into small pieces and surface sterilized with 1% aqueous solution of chlorox plus and two drops of tween twenty for about one minute. These sterilized pieces were blotted and inoculated on moist filter paper and Potato Dextrose Agar (PDA). The plates were incubated at 29 °C for 24 h and 48 h under light and dark conditions respectively. Pure cultures were prepared by isolating single conidia.

### 2.2. Pathogenicity test

Pathogenicity of the fungus was studied on susceptible varieties of maize in earthen pots (diameter 30 cm) containing autoclaved sterilized soil. The maize genotypes were inoculated at 3–4 leaf stage with the inoculum ( $2 \times 10^4$  spores/ml) of the *C. heterostrophus* (conidial suspension) and the seed sprayed with simple distilled water was used as control. The data of the percent infection were taken after every 7 days.

### 2.3. Virulence test of different isolates

The maize germplasm of representative variety Soan-3 was inoculated with three different isolates of *C. heterostrophus* to check their maximum virulence. The sterilized seeds with inoculum of each isolate were put together in test tubes under sterilized conditions. Data were recorded after 1st, 2nd and 3rd week of the inoculation and observed disease severity by IRR (1996).

### 2.4. Effect of culture media, temperature, and pH on the growth of *C. heterostrophus*

The effect of different culture media on growth of *C. heterostrophus* was studied on PDA medium, Basal medium, Richard's agar, Corn meal agar and Czapek's dox agar medium. To determine the most suitable optimum growth temperature, *C. heterostrophus* was incubated at 20 °C, 25 °C, 30 °C, and 35 °C. Effect of different levels of pH on the growth of *C. heterostrophus* was studied at pH 3, 5, 7, 9 and 11. These experiments were run in three replicates. Data were collected after 7th day of incubation.

### 2.5. In vitro screening

The twelve different maize genotypes viz. NC-2703 (hybrid), NC-2003 (hybrid), SP-3 (inbred line), NCML-73 (inbred line), NRL-6 (inbred line), NRL-4 (inbred line), Soan-3 (variety), Rakaposhi

(variety), Margala (variety), EV-1097 (variety), Local-Y (variety), Local-W (variety) were obtained from Crop sciences institute (CSI), National Agricultural Research Center (NARC) Islamabad, Pakistan. Maize seeds were placed with inoculum in sterilized test tubes containing filter paper plus distilled water. Each genotype was replicated thrice for analysis. The data were recorded after 1st, 2nd and 3rd week of inoculation. Leaf lesions scale of 0–5 (IRRI, 1996) was used as disease severity ratings.

## 2.6. Field experiment

Field experiment was performed at National Agricultural Research Center (NARC), Islamabad. The experiment was carried out in a randomized complete block design (RCBD) with three replicates. Maize seeds were planted in a five row plot of 5 × 3 m size. The seeds were spaced at 20 cm in 75 cm wide rows. Half dose of urea (at the rate of 150 kg ha<sup>-1</sup>) and full dose of phosphatic fertilizer (at the rate of 75 kg ha<sup>-1</sup>) were applied at the time of planting. Herbicides used were Primextra (pre-planting) and Round up (post-planting). An insecticide Furadon was applied at the rate of 15 kg ha<sup>-1</sup> when the crop was at 5–6 leaf stage.

### 2.6.1. Method of inoculation

The genotypes were inoculated with spore suspension of isolate V-31. For the preparation of spore suspension, colonies with spore were harvested and transferred into flask containing sterilized distilled water. The suspension was calibrated as 2 × 10<sup>4</sup> spores ml<sup>-1</sup> by Haemocytometer. Each single squeeze of wash bottle delivered 4 ml of spore suspension. Each plant was inoculated at leaf whorl stage.

### 2.6.2. Disease assessment

Southern corn leaf blight was assayed on the parameters of disease incidence (%), disease severity and disease index (%). Effect of *C. heterostrophus* on disease incidence was determined by the number of disease plants/total number of plants × 100. Disease severity is rated according to 0–9 scale (Lim et al., 1996). Data were recorded on 4 leaves (4th, 5th, 6th and 7th leaf) in randomly selected plants/subplot. The first data were taken 10 days after the inoculation and subsequent data of 15-days interval. The collected pathological data were analyzed by RCBD factorial design.

## 2.7. Measurements of yield and its components

Yield (kg ha<sup>-1</sup>) and its components were recorded at the maturity of the crop and was adjusted to 15% storage moisture content. Other data included plant height (cm), ear height (cm), grain weight, infected ear (%), ears per plant, ear placement, per ear weight (g ha<sup>-1</sup>) were recorded. All the data were analyzed statistically using RCBD factorial design.

## 3. Results

### 3.1. Pathogenicity of *C. heterostrophus* and its virulence effect on maize germplasm

Pathogenicity of the fungus was studied on maize plants. Data on un-inoculated (control) and inoculated plants were taken as

the symptoms of disease appeared on the leaves. Percent plant infection and disease severity on un-inoculated plants were 2.1 and 0.56, while on inoculated plants it were 55.8 and 2.7 respectively. There was 2557.1% increase in percent plant infection and 382.1% increase in disease severity over control.

The virulence of isolate was decided on the basis of disease severity ratings. The isolates showed significantly different results at  $P = 0.05$  in disease severity at different time intervals. In mean values the most virulent isolate at 2.67 was V-31. Isolates V-32 and B-202 were non-significant in their disease severity ratings. So V-31 was mass cultured for further experimentation.

### 3.2. Effect of different culture media, temperature and pH level on *C. heterostrophus*

In all media the radial colony growth increased with an increase in incubation period. The data after 7th day of incubation showed that the Basal (69.22 mm), Richard (69.33 mm) and PDA (69.56 mm) media were non-significant in their difference but significantly different from Czapek's Dox agar (57.67 mm) and Corn meal agar (58.00 mm) media.

The most suitable temperature for mycelial growth of the pathogen was found to be 30 °C as the diameter at this temperature was 69.44 mm. Whereas, other temperature levels (25 and 35 °C) showed reduction in the colony growth (41.56 and 26.22 mm respectively). While, minimum growth was observed at 20 °C showing 19.00 mm colony growth.

Observations revealed that mycelial growth of the pathogen was non-significant at pH levels 7 and 9. The lowest growth was noted at pH 3 (37.78 mm in diameter). Also colony growth was good at pH levels of 5 and 9 (54.00 and 58.44 mm in diameter).

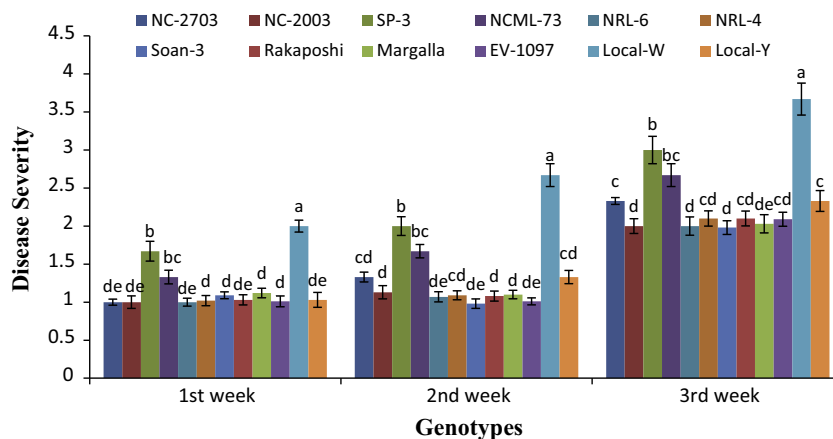
### 3.3. In vitro screening of different maize genotypes against *C. heterostrophus*

There is significant difference in disease severity ( $P = 0.05$ ) after each week of data recording. In mean values Local-W (variety) was observed as moderately susceptible followed by SP-3 (inbred line) and NCML-73 (inbred line) at disease severity 2.78, 2.22 and 1.89 respectively as they were least susceptible. The other genotypes are least resistant as they are non-significant in their disease severity ratings (Fig. 1).

### 3.4. Field experiment

Field experiments include artificial inoculation and subsequent recording of pathological and yield data. Pathological data consist in all three data recordings of disease incidence (%), disease severity and disease index (%) after artificial inoculation with *C. heterostrophus*. Effect of *C. heterostrophus* on these parameters showed significant difference at  $P = 0.05$ . In inoculated plot, genotypes Local-W had maximum disease incidence (%) followed by SP-3, NCML-73, NC-2703, NRL-6 and Local Y while Soan-3 had minimum disease incidence (%). In control plot the difference was non-significant between genotypes (Fig. 2).

1st, 2nd, and 3rd data record of disease severity showed that the inoculated genotypes Local-W showed maximum disease severity followed by SP-3, NCML-73, NC-2703 and



**Figure 1** Analysis of disease severity of different maize genotypes screened *in vitro* against *C. heterostrophus* after 1st, 2nd and 3rd week of inoculation.

NRL-6 while minimum disease severity was shown by Soan-3 and Rakaposhi respectively. In un-inoculated plot the genotypes were non-significant in their difference (Fig. 3).

Disease index of the three data record revealed that maximum disease index values were of Local-W followed by SP-3 and NCML-73 and was shown non-significant difference between them while minimum disease index was of Soan-3 followed by Rakaposhi, NC-2003, EV-1097 and NRL-4 as they were non-significant in their difference. There was non-significant effect of *C. heterostrophus* on disease index against genotypes of control plot (Fig. 4).

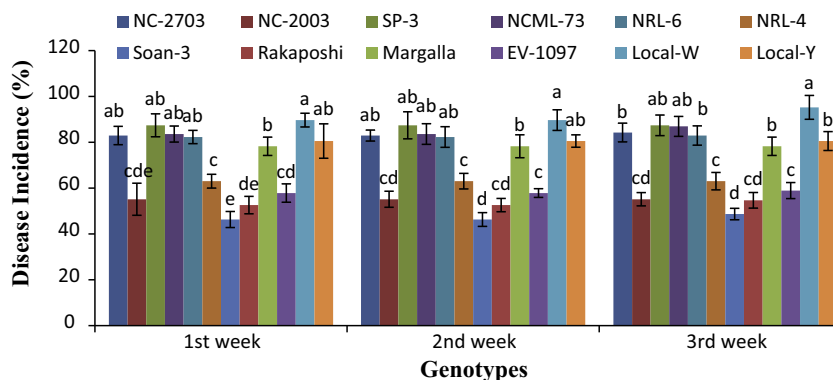
### 3.5. Measurement of yield and its components

The effect of pathogen on grain yield ( $\text{kg ha}^{-1}$ ) of different maize genotypes was significant at  $P = 0.05$ . The treatments were significant in the mean values. In the inoculated plot highest yield was given by Soan-3 at  $5378.92 \text{ kg ha}^{-1}$  followed by Rakaposhi, NC-2003 and EV-1097 at  $4534.70 \text{ kg ha}^{-1}$ ,  $3951.93 \text{ kg ha}^{-1}$  and  $3832.29 \text{ kg ha}^{-1}$  respectively, while poor yield was given by Local-W at  $950.54 \text{ kg ha}^{-1}$  followed by SP-3 at  $1565.11 \text{ kg ha}^{-1}$  (Table 1). In the control (un-inoculated) plot the genotypes Soan-3 also gave maximum grain yield at  $5728.36 \text{ kg ha}^{-1}$  followed by Rakaposhi at  $4710.64 \text{ kg ha}^{-1}$  and minimum yield by Local-W at  $1130.16 \text{ kg ha}^{-1}$ . In the mean values the combined  $\text{kg ha}^{-1}$

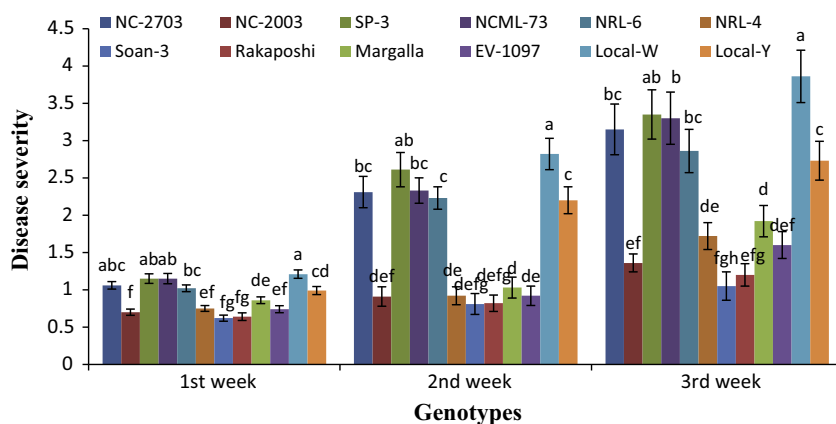
value of inoculated and control plot was significant in yield. In this maximum yield was also given by genotype Soan-3 at combined value of  $5553.64 \text{ kg ha}^{-1}$  while minimum yield was given by Local-W.

Maize varieties are significantly differed in plant height. In the inoculated plot, Rakaposhi was highest at 156.9 cm and NRL-6 was lowest at 107.4 cm of plant heights while in control plot the positions were in similar pattern but heights (cm) were more, as Rakaposhi was at 160.3 cm and NRL-6 was at 111.1 cm (Table 1). The combined value of inoculated and control plots revealed that maximum height was of Rakaposhi and lowest height was of NRL-6 at 158.6 cm and 109.3 cm respectively. The genotype effect on ear height was significant. In the inoculated plot maximum height was attained by Rakaposhi (70.3 cm) while lowest height was of NRL-6 (33.7 cm) (Table 1). In the control plot Rakaposhi was also at maximum height of 72.6 cm while lowest was of NRL-6 (37.1 cm). The combined value of inoculated and un-inoculated plots showed same results with respect to maximum and lowest height.

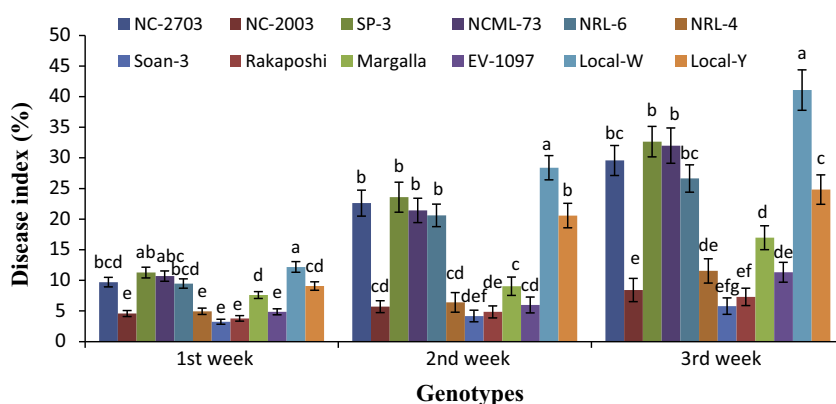
There was significant difference in the genotypes of inoculated plots to infected ears. In the inoculated plot maximum infection was in genotype Local-W and SP-3 at values of 36.13 and 35.57 as they were non-significant in difference. The lowest percent ear infection was of Soan-3 and Rakaposhi also as they had non-significant difference (Table 1), while in control plot the maximum percent infection



**Figure 2** Effect of *C. heterostrophus* on disease incidence (%) against different maize genotypes.



**Figure 3** Effect of *C. heterostrophus* on disease severity against different maize genotypes.



**Figure 4** Effect of *Cochliobolus heterostrophus* on disease index (%) against maize genotype.

**Table 1** Mean values of twelve selected maize genotype yield and its components which include grain yield ( $\text{kg ha}^{-1}$ ), plant height (cm), ear height (cm), infected ear (%), ear weight ( $\text{g ha}^{-1}$ ), ear placement and ears per plant.

Genotypes	Grain yield ( $\text{kg ha}^{-1}$ )	Plant height (cm)	Ear height (cm)	Infected ear (%)	Ear weight ( $\text{g ha}^{-1}$ )	Ear placement	Ears per plant
NC-2703	1942.5	121.1	54.1	31.06	73.2	42.74	1.65
NC-2003	3951.93	149	63.9	14.64	116.8	44.26	1.68
SP-3	1565.11	141.3	57.1	35.57	57.9	43.03	1.25
NCML-73	1787.73	120.4	52.2	31.71	61.8	46.81	1.35
NRL-6	2163.69	107.4	33.7	15.5	85.9	40.92	1.88
NRL-4	3699.44	117.8	37.4	13.36	106.8	41.07	1.53
Soan-3	5378.92	149.1	64.5	9.21	148.3	39.52	2.04
Rakaposhi	4534.7	156.9	70.3	9.46	129.2	41.74	1.83
Margalla	3342.3	144.2	58	15.96	103	42.33	1.7
EV-1097	3832.29	135.5	61.7	18.04	108.9	43.15	1.68
Local-W	950.54	124.9	63.8	36.13	52.3	43.02	0.59
Local-Y	2618.97	126.3	59.1	26.1	96.9	44.08	1.37
Total mean	2988.8	132.8	56.3	21.37	95.129	42.72	1.66

was shown by NRL-6 and Local-W at values of 11.96 and 9.59. Genotype showed significant difference in grain weight ( $\text{g ha}^{-1}$ ). Maximum weight was gained by the genotypes Soan-3 at value of 148.3 g and minimum was by Local-W at 52.3 g (Table 1), while in the control plot the maximum weight was gained also by Soan-3 at 158.4 g and similarly minimum

by Local-W at 59.5 g. Statistical analysis of ear placement of different maize genotype revealed no significant difference and interaction of the genotype. The maximum ears per plant were in Soan-3 followed by Rakaposhi and they showed non-significant difference, while lowest numbers of ears were in Local-W (Table 1).



#### 4. Discussion

In the present research work, different maize genotypes were screened against *C. heterostrophus*, the causal organism of foliar blight disease of maize SCLB. Pathogenicity of the fungus was taken as the symptoms of disease appeared on the leaves. The maize germplasm of representative genotype Soan-3 (variety) was inoculated with three different isolates of *C. heterostrophus*. The most virulent isolate in disease severity was V-31. There was an increase in severity after each interval. This is due to difference in physiological activity and intensity of infection among three different isolates of *C. heterostrophus*.

Growth of fungal colonies needs some nutritional requirements, optimum ambient temperature, optimum pH level and certain favorable environments. The synthetic capabilities of fungi differ in different groups. Some fungi are unable to synthesize certain key compounds that they require and must obtain them from the medium, upon which they culture. All the fungi require almost the same essential elements but differ widely in their ability to obtain them from the growth medium. The most effective mediums for supporting the growth of the fungus were PDA, Richard's medium and Basal medium. On the other hand, somewhat different results were obtained by Orillo (1959) who studied the pathogen, *C. heterostrophus* thoroughly which grew and sporulated well on a variety of natural agar media as well as plant tissues and also found that the pathogen grew well but sporulated sparingly in Richard's and Czapek's media. The most favorable temperature was found to be 30 °C, while minimum radial colony growth (19.00 mm) was recorded at 20 °C. Results similar to our findings were reported in other studies (Orillo, 1959; Singh and Singh, 1966; Misra, 1976). Another important factor in growth of fungal pathogen is its optimum pH level. Growth of the pathogen *C. heterostrophus* was obtained at all pH levels (3, 5, 7, 9 and 11) but it was maximum (59.78 mm in diameter) at pH 7 after 7 days of incubation. Observation revealed that pH 9 was also favorable and showed good colony growth (58.44 mm in diameter). Singh and Singh (1966) studied the physiology of *C. heterostrophus* and observed that the pathogen grew best at pH 5–7. These findings strongly support our results as well as the fact that fungus grows well in environment of neutral pH with respect to particular pathogen *C. heterostrophus*.

The genotypes were evaluated *in vitro* on the basis of disease severity. Maximum value was gained by Local-W (variety) followed by SP-3 (inbred line) and NCML-73 (inbred line). These genotypes were categorized as moderately susceptible. NC-2703 and Local-Y were categorized as least susceptible. The other genotypes are least resistant as they are non-significant in their disease severity ratings. The susceptibility and resistance to particular pathogen were due to difference in their genetic makeup, as some were inbred, varieties and hybrids. Shah et al. (2006) reported highly significant differences ( $p < 0.01$ ) for SCLB severity in two populations of maize while significant differences along with the maize varieties for resistance to SCLB were observed by Rahman et al. (2005).

##### 4.1. Field experiment

Disease incidence (%) gives detail about the calculated prevalence of particular disease, as in the results of the studies by

Reddy and Reddy (1989) Ahmed and Bhutta (1989) and Harlapur et al. (2000). In our study, genotypes Local-W, SP-3, NCML-73, NC-2703, NRL-6 and Local-Y were highly susceptible while Soan-3, Rakaposhi, NC-2003 and EV-1097 were least susceptible. Margalla and NRL-4 were moderately susceptible in disease incidence analysis. Over all inbred lines and varieties showed susceptibility which is not congruent to the results of Choi et al. (1994) and Miura et al. (2004).

Disease severity is the intensity of disease prevalence based on specific scale of rating as described in the results of Lambert and White (1997). After three data recordings Local-W was found moderately susceptible, SP-3, NCML-73, NC-2703, NRL-6 and Local-Y were least susceptible while rest of genotypes were found least resistant. The difference of genotypes in disease severity was due to diversity in their genetic makeup as reported by Williams and Hallauer (2000) and Kraja et al. (2000).

Disease index shows the cumulative value of disease in our target genotypes. Its value is based on disease incidence and disease severity. In all three recordings of disease index (%), the genotype Local-W showed maximum susceptibility. The genotype SP-3, NCML-73, NC-2703, NRL-6 and Local-Y confirmed moderate susceptibility while Margalla, NRL-4 and EV-1097 were found least susceptible. The least resistance was shown by NC-2003, Rakaposhi and Soan-3. Field screening of genotypes against *C. heterostrophus* showed various level of susceptibility and resistance (Ali and Ahmed, 1992). It was not prominent in *in vitro* screening because in laboratory screening all genotypes excluding Local-W, SP-3 and NCML-73 showed partial resistance to *C. heterostrophus* because of inoculum potential or abiotic factors such as temperature, humidity which cannot be controlled in field conditions. The diversity in response of different genotypes to *C. heterostrophus* was also evident in the results of Rathee et al. (2000).

##### 4.2. Yield and its components

Grain yield is the most complex and significant character. The effect of pathogen on grain yield ( $\text{kg ha}^{-1}$ ) of different maize genotypes was significant at  $P = 0.05$ . This difference in yield had also been shown by Ali and Ahmed (1992). The Local-W was lowest in yield as it was highest in susceptibility while Soan-3 was highest in yield as result of its resistance to *C. heterostrophus*. The effect of *C. heterostrophus* on yield was reported by Raziq and Ahmed (1992) and Sain et al. (2000). Maize germplasm was evaluated for resistance to *H. maydis* and MLB had negative impact on grain yield was confirmed in numerous studies (Rahman et al., 2005; Shah et al., 2006).

Plant height is an important trait which affects the overall grain yield of the crop. Extremely dwarf and tall varieties result in crop yield reduction. Maximum height was gained by Rakaposhi and lowest height was of NRL-6. The difference in plant heights was due to effect of disease SCLB and their genetic potential. The resistant genotypes were highest in plant height (cm). Their results indicated that there was significant difference in plant height (cm) of different maize genotypes. The ear height has great impact on the maximum yield production of crop. If the ears are placed extremely high, plants are liable to be damaged by stalk lodging and low ear height effect crop by the attack of wild animal (Ali et al., 2011b). Maximum

height was attained by Rakaposhi while lowest height was of NRL-6. The genetic potential of different genotype showed the difference in ear height (cm) which is congruent to the result of Ali et al. (2011a) due to the SCLB disease effect. The effect of *C. heterostrophus* on per ear weight ( $\text{g ha}^{-1}$ ) of different maize genotypes was significant at  $P = 0.05$ . Local-W showed lowest and Soan-3 highest per ear weight ( $\text{g ha}^{-1}$ ) as a result of intensity of disease which is analogous to the outcome of Raziq and Ahmed (1992).

Ear infection is non-significant in difference but ear placement is significant in only treatments and non-significant difference in mean values of the genotypes. In this case the disease SCLB had no effect on ear placement of genotypes. The effect of the pathogen on ears per plant was only significant in only the mean values of the genotypes. The difference in genotypes was due to its genetic makeup as showed by the results of Josephson et al. (1971).

## 5. Conclusion

Pathological parameters and yield (and its components) results showed significant effect of SCLB on different genotypes of maize. The genotypes of the control also showed disease incidence (%), disease severity and disease index (%) due to the presence of wind inoculum of SCLB that had traveled from inoculated to control or already the presence of soil borne *C. heterostrophus*. However none of the genotypes evaluated under this program showed complete resistance to the disease. So genotypes identified on the level of resistance should be recommended for end users after further trials.

## Conflict of interest

The authors declare that there are no conflicts of interest.

## Acknowledgement

The authors acknowledge the Dean; Faculty of Biological Sciences and Chairperson; Department of Plant Science Dr. Asghari bano, Nadia Batool Zahra (Department of Biotechnology, Quaid-i-Azam University Islamabad) and Dr. Yasmin Ahamd for their assistance in this research project. We also thank National Agricultural Research Centre for providing laboratory and field facilities.

## References

- Ahmed, S.I., Bhutta, A.R., 1989. Seed-borne fungal pathogens of maize in Pakistan. *J. Sci. Ind. Res.* 32, 107–109.
- Ali, F., Muneer, M., Rahman, H., Noor, M., Durrishahwar, Shaukat, S., Yan, J.B., 2011a. Heritability estimates for yield and related traits based on testcross progeny performance of resistant maize inbred lines. *J. Food, Agric. Environ.* 9, 438–443.
- Ali, F., Rahman, H., Durrishahwar, Nawaz, I., Munir, M., Ullah, H., 2011b. Genetic analysis of maturity and morphological traits under maydis leaf blight (MLB) epiphytotic in maize (*Zea mays* L.). *J. Agric. Biol. Sci.* 6, 13–19.
- Ali, K., Ahmed, S., 1992. Genotype assay of maize for resistance to maydis leaf blight under artificial field epiphytotic of Peshawar region [Pakistan]. *Sarhad J. Agric.* 8, 547–549.
- Atwell, S., Huang, Y.S., Vilhjalmsjon, B.J., Willems, G., Horton, M., Meng, D., Platt, A., Tarone, M., Hu, T.T., 2010. Genome-wide association study of 107 phenotypes in *Arabidopsis thaliana* inbred lines. *Nature* 465, 627–631.
- Bogenschutz, T.G., Russell, W.A., 1986. An evaluation for genetic variation within maize inbred lines maintained by sib-mating and self-pollination. *Euphytica* 35, 403–412.
- Bothastr, J., Schlicher, M.A., 2005. Biotechnological processes for conversion of corn into ethanol. *Appl. Microbiol. Biotechnol.* 67, 19–25.
- Choi, K.J., Lee, H.S., Park, S.E., Chin, M.S., Park, K.Y., 1994. Stay-green characteristics and characters related to stay-green in maize inbred lines. *RDA J. Agric. Sci.* 36, 127–134.
- Emmert, A.B.E., Handelsman, J., 1999. Biocontrol of plant disease: a (Gram-) positive perspective. *FEMS Microbiol. Lett.* 171, 1–9.
- Fisher, D.E., Hooker, A.L., Lim, S.M., Smith, D.R., 1976. Leaf infection and yield loss caused by four *Helminthosporium* leaf diseases of corn. *Phytopathology* 66, 942–994.
- Gregory, L.V., Ayers, J.E., Nelson, R.R., 1978. Predicting yield losses in corn from southern corn leaf blight. *Phytopathology* 68, 517–521.
- Hafiz, A., 1986. Plant Diseases. Pakistan Agricultural Research Council, Islamabad, pp. 93–102.
- Harlapur, S.I., Mruthunjaya, C.W., Anahosur, K.H., Muralikrishna, S., 2000. A report survey and surveillance of maize diseases in North Karnataka. *Karnataka J. Agric. Sci.* 13, 750–751.
- Hussain, N., Khan, M.Y., Baloch, M.S., 2011. Screening of maize varieties for grain yield at Dera Ismail Khan. *J. Anim. Plant Sci.* 21, 626–628.
- Hyre, R.A., 1970. Epidemiology of southern corn leaf blight exploratory experiments. *Plant Dis. Rep.* 54, 1131–1133.
- IRRI, 1996. IRRI Discussion Paper Series 18, Field Sampling Methods for Crop Pests and Disease in Kenya and IRRI Evaluation System for Rice.
- Josephson, L.M., Graves, C.R., Kinier, H.L., Hilty, J.W., 1971. Reduction in yield of corn from southern corn leaf blight. *Plant Dis. Rep.* 58, 219–220.
- Kraja, A., Dudley, J.W., White, D.G., 2000. Identification of tropical and temperate maize populations having favorable alleles for disease resistance. *Crop Sci.* 40, 948–954.
- Kuehnle, A.R., Earle, E.D., 1988. Evaluation of tissue culture-derived methomyl resistant cms-T lines. *Maize Genet. Co., Newslett.* 62, 68.
- Lakshmi, P., Sharma, R.C., 1987. Evaluation of maize germplasm to *Helminthosporium maydis* using detached leaf technique. *Ann. Agric. Res.* 8, 34–40.
- Lambert, R.J., White, D.G., 1997. Disease reaction changes from tandem selection for multiple disease resistance in two maize synthetics. *Crop Sci.* 37, 66–69.
- Leonard, K.J., 1977. Races of *Drechslera maydis* in the Southeastern U.S. from 1974–1976. *Plant Dis. Rep.* 61, 914–915.
- Lim, K.G., Gorbacheva, A.G., Ivashchenko, V.G., 1996. Method for evaluating the resistance of maize to southern leaf blight. *Kukuruza I Sorgo* 4, 23–24.
- Misra, A.P., 1976. *Helminthosporium* Species Occurring on Cereals and Other Gramineae. Catholic Press, Ranchi, 834001.
- Miura, Y., Sato, H., Hasegawa, H., Monma, E., Enoki, H., Shigemori, I., Takamiya, Y., Okabe, T., 2004. Breeding of a flint maize inbred line “Ho49” and its characteristics. *Bull. Natl. Agric. Res. Center Hokkaido Region.* 180, 19–31.
- Nicholson, P., Rezanoor, H.N., Su, H., 1993. Use of random amplified polymorphic DNA (RAPD) analysis and genetic fingerprinting to differentiate isolates of race o, C and T of *Drechslera maydis*. *J. Phytopathol.* 139, 261–267.
- Orillo, F.T., 1959. Conditions for the growth of *Helminthosporium maydis*. *Philippines Agric.* 38, 27–32.

- Pixley, K.V., Dhaliwayo, T., Tongoona, P., 2006. Improvement of maize populations by full-sib selection alone versus full-sib selection with selection during inbreeding. *Crop Sci.* 46, 1130–1136.
- Rahman, H., Raziq, F., Ahmad, S., 2005. Screening and evaluation of maize genotypes for southern leaf blight resistance and yield performance. *Sarhad J. Agric.* 21, 231–235.
- Rahul, K., Singh, I.S., 2002. Inheritance of resistance to banded leaf and sheath blight (*Rhizoctonia solanf. sp. Sasakii*) of maize. In: *Proc. 8th Asian Regional Maize Works*. Bangkok, Thailand, vol. 5, pp. 356–365.
- Ram, H.H., Singh, H.G., 2003. Maize. In: *Crop Breed and Genetics*. Kalyani Publishers, India, pp. 105–109.
- Rathee, V.K., Guleria, S.K., Sharma, B.K., 2000. Evaluation of maize germplasm against leaf blights. *Res. Crops* 1, 253–254.
- Raziq, F., Ahmed, S., 1992. Comparison of Maydis leaf blight epidemics at different stages of maize development. *Sarhad J. Agric.* 8, 569–573.
- Reddy, V.K., Reddy, S.M., 1989. Seed mycoflora of maize. *Ind. Bot. Rep.* 8, 76–77.
- Ricker, A.J., Ricker, L.S., 1935. *Introduction to Research on Plant Diseases*. Journal Swift Co., New York.
- Sain, D., Pawan, A., Dhanju, K.S., Surender, K., Padam, S., Dass, S., Arora, P., Kumar, S., Singh, P., 2000. Gene effects for maydis leaf blight (*Drechslera maydis*) resistance and grain yield in white maize (*Zea mays*). *Ind. J. Agric. Sci.* 70, 317–319.
- Shah, S.S., Rahman, H., Khalil, I.H., Rafi, A., 2006. Reaction of two maize synthetics to maydis leaf blight following recurrent selection for grain yield. *Sarhad J. Agric.* 22, 263–269.
- Shieh, G.J., Lu, H.S., 1993. Diallel analysis of mature plant resistance to *Helminthosporium maydis* in maize. *J. Agric. Res. China* 42, 12–18.
- Singh, G.P., Singh, B., 1966. A leaf spot disease of maize caused by *Drechslera maydis* (Nisikado) Shoemaker. *Prod. Nat. Acad. Sci. Ind., Sect. 36*, 303–305.
- Tajimi, A., Ando, N., Teranaka, M., 1985. Seedling test for resistance to southern leaf blight in corn under greenhouse conditions. Virulence of six isolates of *Drechslera maydis* collected at six localities in southwest Japan. *Bull. Natl. Grassland Res. Inst., Jpn.* 31, 68–72.
- Ullstrup, A.J., 1972. Impacts of the southern corn leaf blight epidemic of 1970–71. *Annu. Rev. Phytopathol.* 10, 37–50.
- Wang, X.M., Dai, F.C., liao, Q., Sun, S.X., 2001. *Field Corn Pest Manual*. China Agricultural Science and Technology Publishing House, Beijing, pp. 4–102.
- Warren, H.L., Jones, A., Huber, D.M., 1977. Morphological and physiological differences between *Drechslera maydis* races O and T. *Mycologia* 69, 773–782.
- Wei, J.K., Liu, K.M., Chen, J.P., Luo, P.C., Stadelmann, O.Y.L., 1988. Pathological and physiological identification of race C of *Drechslera maydis* in China. *Phytopathology* 78, 132–157.
- Williams, T.R., Hallauer, A.R., 2000. Genetic diversity among maize hybrids. *Maydica* 45, 163–171.
- Zaitlin, D., DeMars, S., Ma, Y., 1993. Linkage of *rhm*, a recessive gene for resistance to southern corn leaf blight, to RFLP marker loci in maize (*Zea mays*) seedlings. *Genome* 36, 555–564.