Pak. J. Bot., 51(5): 1865-1875, 2019. DOI: http://dx.doi.org/10.30848/PJB2019-5(10)

SCREENING OF MAIZE GENOTYPES UNDER NORTHERN CORN LEAF BLIGHT (NCLB) EPIPHTOTIC AT RAWLAKOT AZAD KASHMIR

TAHSIN RAZZAQ[1*], MUHAMMAD FAREED KHAN[2,] SHAHID IQBAL AWAN[2], HUMA TARIQ[3] AND MUHAMMAD ILYAS[4]

Department of Plant Breeding and Molecular Genetics, Faculty of Agriculture, The University of Poonch Rawalakot, Azad Kashmir, Pakistan
[*]Corresponding author's email: Tahsinkhan750@yahoo.com

Abstract

Northern corn leaf blight (NCLB) is most devastating maize foliar disease in Himalayan region of the world, caused by fungus *Exerohillum turcicum*. The present investigations were carried out to screen the 30 maize genotypes against NCLB under agro-climatic field conditions of Rawalakot, Azad Kashmir-Pakistan. Artificial inoculation was done at four to six leaf stages during spring 2017. The disease severity of maize genotypes was assessed using 0-5 points scale. Data were recorded on number of lesions on leaf, lesion length, number of infected leaf, number of infected plants, percentage leaf infection, percentage plant infection, disease severity, disease index, disease incidence and area under disease progressive curve (AUDPC). Genotypes had significant differences for NCLB severity and reactions and were classified into resistant, highly resistant, moderately resistant, susceptible and highly susceptible categories. The percent disease incidence ranged from 20-60%, disease index (20-66%), disease intensity (16-34%) and area under disease progressive curve was 22-362%-dsu (percentage development stage unit). Based on disease rating scale and pathological traits, it was reported that three genotypes i.e., Karamat-Bar-25, SZP-13200 and NCEV-1530-11 had lowest values for all pathological traits and severity rating of 5R, 5R and 10R respectively and marked as highly resistant (R) genotypes, whereas three genotypes viz., Soan-3, Ghuari-122 and Kissan-60 were evaluated as highly susceptible (S) within germplasm and had highest values for all pathological traits and severity rating of 90S, 90S and 80S respectively. Fourteen genotypes were moderately resistant (MR), 10 were moderately susceptible (MS) under field conditions. Results suggested that the genotypes found resistant to leaf blight might be utilized in future breeding program. Alternatively, the said promising genotypes might also be used as parents in hybridization in order to transfer the gene for resistance to early blight to existing adapted high yielding cultivars.

Key words: Northern leaf blight, *Exerohillum turcicum*, Genotypes, Severity, Resistance.

Introduction

Northern corn leaf blight (NCLB).also known.as *Turcicum* leaf blight is major foliar diseases of maize, remains problematic in Himalayan region of the world, caused by the fungus *Exserohilum turcicum* (Ribeiro *et al*., 2016). It is endemic in the areas of the North Western Himalayan regions and is considered to be very important in terms of its geographical distribution and potential, to cause yield losses (Chandrashekara *et al*., 2012). It might be severe in medium-altitude humid. regions, where cloudy weather, high humidity and relatively low temperature thrive, during the maize growing season. Yield losses can easily exceed 50% if the disease appears before flowering. However the losses get reduced if the infection takes place at a later stage (Singh, 2004).

Northern corn leaf blight is known to infect maize plants from seedling stage towards maturity. The symptoms first appear.as small oval spots on the leaves as grayish-green lesions with water soaked lesion going parallel to the leaf margins, the spots increase in size with age and turn gray greenish, finally getting a spindle form with long elliptical tan or grayish lesions (Vieira *et al*., 2014).

The ideal conditions for NCLB incidence is relatively cool seasons with long period of wetness (more than five hours) at temperatures between 18 to 27°C (64 and 81°F) are most conducive to disease.development. Leaving huge amounts of infected residue in the maize field and ongoing to maize crop in these fields will promote disease progress by providing sufficient inoculum in the early season (Wise, 2011).

If the disease starts at an initial stage, it causes early or premature death of infected leaves result in the loss of nutritive value of crop and reduced germination capability, grain yield and vigor. The blight start from the lower leaves and spreads towards the developing ear and then flag leaf of the plant (CIMMYT, 2004). Most of the maize breeding programs initiated with the aim to improve disease resistant and increase grain yield (GY), also consider other related traits i.e., plant height, days to 50% silking, days to 50% tasseling, ear height, and days to maturity, since these traits are also used in diseases management. The best and long term, environmentally and economically safe method for control of NCLB is planting of resistant varieties. Slow development of disease is characterized by several important components like less lesion number, small lesion size and low area under disease progress curve (Bashyal *et al*., 2011).

Northern corn leaf blight is major foliar disease of maize in the Himalayan region particularly in Rawalakot, Azad Kashmir, Pakistan. The disease became epidemic due to environmental factors i.e. high humidity, frequent rain fall, and low temperature. The climate of this area favors the epidemic? of NCLB and cause the serious economic loss.

The present study was initiated with the objectives to explore the resistant and susceptible genotypes within the germplasm, may help to evaluate the valuable gene pools and select improved genotypes to meet the current demands.

Materials and Methods

Field screening against Northern Corn Leaf Blight (NCLB) was carried out at the Plant Breeding and Molecular Genetics, Faculty of Agriculture, University of Poonch, Rawalakot (Latitude 33°51'32.18"N, Longitude 73°45'34.93"E, Elevation 5500 ft). A total of 30 maize genotypes were planted during June 2017 in a randomized complete block design (RCBD) with three replications (Table 1). The seeds were sown in well and fine prepared soil with plants and rows spacing of 25 and 75 cm, respectively. Recommended dose of fertilizers "Urea" (180kh/ha) and "NPK" (360kg/ha) were applied during seed bed preparation.

Preparation of inoculum: Collection of infected plant's leaves (sample) was done during fall 2016 from natural maize field at harvest stage. The samples were sun dried and put in oven for 48 hrs at 40°C. After oven drying process, samples were grind by using grinder in order to attain inoculum in fine powder form (Ali *et al*., 2014). This powder was well- sealed for artificial inoculation for the proceeding season.

Procedure of inoculation: The artificial inoculation was done at four to six leaf stages. The inoculum was manually dropped in leaves whorls at the seedling stage (Khalil *et al*., 2010; Ali & Yan, 2012).

Table 1. Maize genotypes used in studies.

Entry no.	Genotypes	Origin	Source
1.	Azam-140	Pakistan	"CCRI, Pirsabak, Nowshera, KPK"
2.	Ghauri-122	Pakistan	"CCRI, Pirsabak, Nowshera, KPK"
3.	Jalal-29	Pakistan	"CCRI, Pirsabak, Nowshera, KPK"
4.	Kissan-60	Pakistan	"CCRI, Pirsabak, Nowshera, KPK"
5.	Sadaf-141	Pakistan	"CCRI, Pirsabak, Nowshera, KPK"
6.	Sarhed White-27	Pakistan	"CCRI, Pirsabak, Nowshera, KPK"
7.	Golden-199	Pakistan	"Agricultural Research Institute, D.I. Khan, KPK"
8.	Iqbal-78	Pakistan	"Agricultural Research Institute, D.I. Khan, KPK"
9.	Karamat Bar-25	Pakistan	"Agricultural Research Institute, D.I. Khan, KPK"
10.	Pahari-68	Pakistan	"Agricultural Research Institute, D.I. Khan, KPK"
11.	Aziz-2003	Pakistan	"Agricultural Research Institute, D.I. Khan, KPK"
12.	NCEV-1530-1	Pakistan	NARC, "Islamabad, Pak."
13.	NCEV-1530-2	Pakistan	NARC,"Islamabad, Pak."
14.	NCEV-1530-3	Pakistan	NARC,"Islamabad, Pak."
15.	NCEV-1530-4	Pakistan	NARC,"Islamabad, Pak."
16.	NCEV-1530-5	Pakistan	NARC,"Islamabad, Pak."
17.	NCEV-1530-6	Pakistan	NARC,"Islamabad, Pak."
18.	NCEV-1530-7	Pakistan	NARC,"Islamabad, Pak."
19.	NCEV-1530-9	Pakistan	NARC,"Islamabad, Pak."
20.	NCEV-1530-10	Pakistan	NARC,"Islamabad, Pak."
21.	NCEV-1530-11	Pakistan	NARC,"Islamabad, Pak."
22.	NCEV-1530-12	Pakistan	NARC,"Islamabad, Pak."
23.	NARC-W	Pakistan	NARC,"Islamabad, Pak."
24.	NP-1	Pakistan	NARC,"Islamabad, Pak."
25.	NP-2	Pakistan	NARC,"Islamabad, Pak."
26.	NP-3	Pakistan	NARC,ISLAMABAD
27.	TP-1217	Pakistan	NARC,"Islamabad, Pak."
28.	SZP-13200	Pakistan	NARC,"Islamabad, Pak."
29.	Rakaposhi	Pakistan	NARC,"Islamabad, Pak."
30.	Soan-3	Pakistan	NARC,"Islamabad, Pak."

Disease assessment: Maize field trail was visually observed at two–three weeks after inoculation. Randomly ten plants were selected and tagged for disease assessments. Plants were rated and monitored with the interval of 10 days. Monitored plants were marked with the marker so that plants could be traced and monitored again. Data on number of lesions on leaf, lesion length, number of infected leaves, number of infected plants were visually observed and recorded while leaf disease incidence was calculated by formula LDI= n / N × 100 as given by Ishfaq *et al.,* (2014). Where n is number of infected leaves, N is total number of leaves assessed while percentage disease index was calculated by formula as defined by Pandey *et al*., (2003).

$$\text{DI (\%)} = \frac{\text{n1x1+n2x2+n3x3}}{\text{Total number of leaves observed x Maximum rating value}} \text{ x 100}$$

where, N- Number of days after lesion observes (interval), 5, 0, 5 etc., X-Number of lesions after interval. The disease intensity was calculated as per formula given by Harlapur, (2005), D In. (%) = {(nv/NG)} x 100. Where, (nxv) - Sum of score, N - Total number of leaves counted, G - Highest score. Percentage of leaf infection was estimated as per observation by the formula PLI = number of leaves infected/total number of leaves × 100 and percentage plant infection was calculated as PPI= number of plants infected / total number of plants × 100 and area under disease progress curve was calculated according to the formula given by Campbell & Madden (1990).

$$AUDPC = \sum_{i=1}^{n-1} \frac{[(y_i + y_{i+1})\ (t_{i+1} - t_i])}{2}$$

where, n is the number of observations, ti is the days after planting for the Ist disease assessment and yi is the disease severity.

Reaction and scoring of disease

The response of maize genotypes was recorded by the following capital letters.
O- Highly resistant - No visible infection.
R- Resistant - Necrotic areas with or without minute uredia.
MR- Moderately resistant - Small uredia present surrounded by necrotic areas
MS - Moderately susceptible - Medium uredia with no necrosis or distinct chlorosis
S – Susceptible - large uredia and little or no chlorosis present severity

All the leaves on infected plants were scored using 0-5 scale adopted by maize pathology unit CIMMYT (2004) as
0 = no visible lesion
1 = one to few scattered lesion on leaves covering up to 10% of leaf area
2 = lesions on leaf covering 11- 25% leaf area
3 = lesions on leaf covering 26-50% leaf area
4 = lesions abundant on leaf covering 51-75% leaf area
5 = lesions abundant on almost all leaf, plant prematurely dried with 76-100% leaf area covered.

According to scale 0 means complete resistant and 5 being complete susceptible. Based on this rating scale, maize genotypes were classified into four groups namely, resistant (R) genotypes with a score < 2.0; moderately resistant (MR) 2.1-3.0; moderately susceptible (MS) 3.1-3.5 and highly susceptible (S) > 3.5.

Statistical analysis: Data on Northern Corn leaf blight assessment were subjected to analysis of variance (ANOVA). Mean separation was done based on the LSD at the 5% probability level. Simple statistics and numerical taxonomic techniques were utilized for cluster analysis with the help of computer software 'Statistica' (www.statsoft.com), PAST (Hammer *et al*., 2001).

Results

Number of lesions on leaf: Mean values for number of lesions on leaf ranged from 1-16 (Table 2). Maximum mean value was shown by genotype Soan-3 (16) followed by Ghuari-122 (15) and Kissan-60 (14). Whereas minimum mean values for number of lesion on leaf was shown by genotype Karamat-Bar-25 (01) followed by SZP-13200, NCEV-1530-11 (02) and Aziz-2003 (03) (Fig. 1).

Lesions length (cm): Mean values for lesions length from 1-17 cm is given in Table 2. Maximum mean value for lesion length was shown by genotype Soan-3 (17cm) followed by Ghuari-122 (15cm) and Kissan-60 (13cm). Whereas minimum mean values for lesion length was observed in genotype Karamat-Bar-25 (3 cm) followed by SZP-13200 and NCEV-1530-11 (2cm), respectively (Fig. 2).

Number of infected leaves: Mean values for number of infected leaves were displayed in the range of 2-14 (Table 2). Maximum mean value for number of infected leaves was shown by genotype Soan-3 (14) followed by Ghuari-122 (12) and Kissan-60 (11). However, minimum mean values were found in genotype Karamat-Bar-25 (2) followed by SZP-13200 and NCEV-1530-11 (3), respectively (Fig. 3).

Number of infected plants: Mean values for number of infected plants were displayed in the range of 2-20 (Table 2). However, maximum mean value for number of infected plants was shown by genotype Soan-3 (20) followed by Ghuari-122 (18) and Kissan-60 (17). Whereas minimum mean values for number of infected plants were observed in genotype SZP-13200 (2), NCEV-1530-11 (2) and Karamat-Bar-25 (3), respectively (Fig. 4).

Leaf disease incidence (LDI): Mean values for leaf disease incidence ranging from 20-66% are given in Table 2. Maximum mean value for leaf disease incidence was shown by genotype Soan-3 (66%) followed by Kissan-60 (60%) and Ghuari-122 (53%). Whereas minimum mean values were shown by genotype SZP-13200 (20%), NCEV-1530-11 (20%) and Karamat-Bar-25 (20%) followed by TP-1217 (21%) and NCEV-1530-12 (23%), respectively (Fig. 5).

Table 2. Mean ± Standard deviation for pathological traits of 30 maize genotypes.

Genotypes	NLL	LL	NIL	NIP	LDI%	DI%	Din. %	PLI	PPI	AUDPC
Azam-140	9 ± 1.000	12 ± 2.416	4 ± 2.081	6 ± 0.532	26 ± 2.645	60 ± 0.000	26 ± 1.000	33 ± 0.000	54 ± 0.225	200 ± 2.000
Ghauri-122	15±2.081	15±1.000	12±2.516	18 ± 0.100	53 ± 4.041	29 ± 0.534	31 ± 2.345	77 ± 0.112	82 ± 1.345	362 ±1.543
Jalal-29	8±1.527	8±0.576	5±2.516	10 ± 0.000	33 ± 0.000	36 ± 0.526	27 ± 1.009	45 ± 1.987	67 ± 0.000	322 ±0.112
Kissan-60	13±1.527	13±1.527	11±1.000	17 ± 0.456	60 ± 3.511	35 ± 1.511	33 ± 1.234	72 ± 2.965	70 ± 1.045	322 ±1.000
Sadaf-141	10±1.000	8±0.577	4±.577	6 ± 1.056	26 ± 1.567	47 ± 1.548	23 ± 2.005	38 ± 1.035	46 ± 1.547	242 ±2.345
Sarhed white-27	12±2.516	13±1.000	5±1.000	7 ± 0.675	38 ± 2.516	40 ± 2.468	26 ± 2.003	50 ± 2.563	67 ± 0.456	262 ±2.346
Golden-199	12±1.000	8±0.000	5±0.000	7 ± 0.463	33 ± 2.516	67 ± 3.546	28 ± 1.005	62 ± 0.000	60 ± 2.567	200 ±1.345
Iqbal-78	11±1.000	8±0.577	7±1.534	5 ± 2.098	46 ± 2.081	49 ± 0.534	25 ± 1.000	70 ± 2.764	28 ± 1.567	82± 2.685
Karamat Bar-25	5±2.516	4±2.081	2±2.000	3 ± 3.545	20 ± 1.078	90 ± 0.467	19 ± 1.234	18 ± 0.567	8 ± 1.087	22± 0.038
Pahari-68	11±1.000	5±2.516	7±1.000	5 ± 2.523	46 ± 1.567	34 ± 1.356	29 ± 1.054	63 ± 0.546	45 ± 1.056	200± 1.849
Aziz-2003	3±1.527	2±0.577	4±0.000	8 ± 0.586	26 ± 0.000	8 ± 2.356	27 ± 2.453	28 ± 0.537	14 ± 0.065	42± 0.2000
NCEV-1530-1	8±0.577	10±1.527	8±0.577	5 ± 0.538	26 ± 1.056	14 ± 2.534	29 ± 2.000	33 ± 0.578	23 ± 0.567	282 ±1.349
NCEV-1530-2	9±1.000	9±1.527	5±1.527	8 ± 0.000	33 ± 1.110	22 ± 1.500	26 ± 1.054	42 ± 1.056	23 ± 0.543	242 ±0.000
NCEV-1530-3	5±2.516	2±2.516	6±0.476	7 ± 0.523	24 ± 1.511	46 ± 1.000	29 ± 2.546	26 ± 0.000	14 ± 0.913	162 ±1.220
NCEV-1530-4	10±1.000	9±1.000	4±0.000	6 ± 0.567	25 ± 1.567	45 ± 1.367	20 ± 2.000	25 ± 0.245	22 ± 1.111	42 ± 0.1567
NCEV-1530-5	8±1.527	7±1.000	4±1.809	10 ± 1.543	26 ± 1.675	33 ± 0.000	23 ± 0.345	31 ± 0.235	40 ± 1.000	162 ± 0.345
NCEV-1530-6	11±2.000	8±1.527	3±1.527	5 ± 1.034	25 ± 1.534	31 ± 2.685	23 ± 1.567	23 ± 1.546	26 ± 0.566	242 ± 0.567
NCEV-1530-7	5±2.518	2±2.516	5±2.534	8 ± 0.532	33 ± 1.587	42 ± 2.546	30 ± 1.546	41 ± 2.000	38 ± 0.656	242 ± 1.000
NCEV-1530-9	11±1.237	4±2.081	3±0.000	3 ± 1.532	25 ± 1.534	35 ± 1.566	22 ± 0.555	25 ± 1.534	23 ± 1.356	82 ± 1.567
NCEV-1530-10	4±2.081	2±1.000	5±0.223	5 ± 0.000	33 ± 2.543	37 ± 1.456	23 ± 0.567	38 ± 0.000	20 ± 4.322	162 ± 0.345
NCEV-1530-11	2±0.577	1±0.000	3±0.112	2 ± 1.058	20 ± 2.000	76 ± 1.567	16 ± 0.534	21 ± 0.345	10 ± 3.267	42 ± 1.034
NCEV-1530-12	6±0.577	2±1.732	4±1.908	6 ± 1.052	23 ± 2.534	50 ± 1.000	26 ± 0.543	25 ± 0.536	37 ± 0.543	62 ± 1.367
NARC-W	10±.577	8±1.527	5±2.523	7 ± 1.543	33 ± 2.511	42 ± 1.543	28 ± 0.567	42 ± 0.563	36 ± 0.000	122 ± 1.553
NP-1	11±1.237	8±0.000	7±2.511	10 ± 1.052	53 ± 2.578	34 ± 0.534	25 ± 0.543	58 ± 0.578	40 ± 1.456	322 ± 2.000
NP-2	9±1.000	3±1.000	5±2.518	6 ± 2.000	25 ± 1.500	34 ± 0.534	25 ± 0.576	20 ± 1.500	24 ± 0.567	162 ± 1.546
NP-3	9±0.000	8±1.527	6±0.577	10 ± 1.154	40 ± 1.456	41 ± 0.115	28 ± 0.556	46 ± 2.005	40 ± 0.453	102 ± 2.076
TP-1217	6±1.527	6±0.577	5±1.000	11 ± 1.237	21 ± 0.000	31 ± 1.983	20 ± 0.543	23 ± 2.047	23 ± 0.521	242 ± 1.034
SZP-13200	2±0.577	2±0.000	3±0.000	2 ± 1.000	20 ± 1.579	86 ± 0.000	17 ± 0.123	20 ± 1.034	12 ± 0.567	22 ± 1.000
Rakaposhi	5±2.518	5±1.732	4 ± 2.081	6 ± 2.000	26 ± 0.547	34 ± 0.345	31 ± 0.533	36 ± 0.354	28 ± 1.354	82 ± 1.034
Soan-3	16±2.785	17±2.309	14 ± 2.081	20 ± 2.081	66 ± 0.000	62 ± 0.298	34 ± 0.546	90 ± 0.372	80 ± 0.000	362 ± 1.045
Statistics										
Grand Mean	8.53	6.86	5.66	7.73	32.2	41.30	25.0	39.9	35.1	179.80
SE	0.660	0.753	0.50	0.787	2.37	3.54	0.98	3.65	3.8	19.32

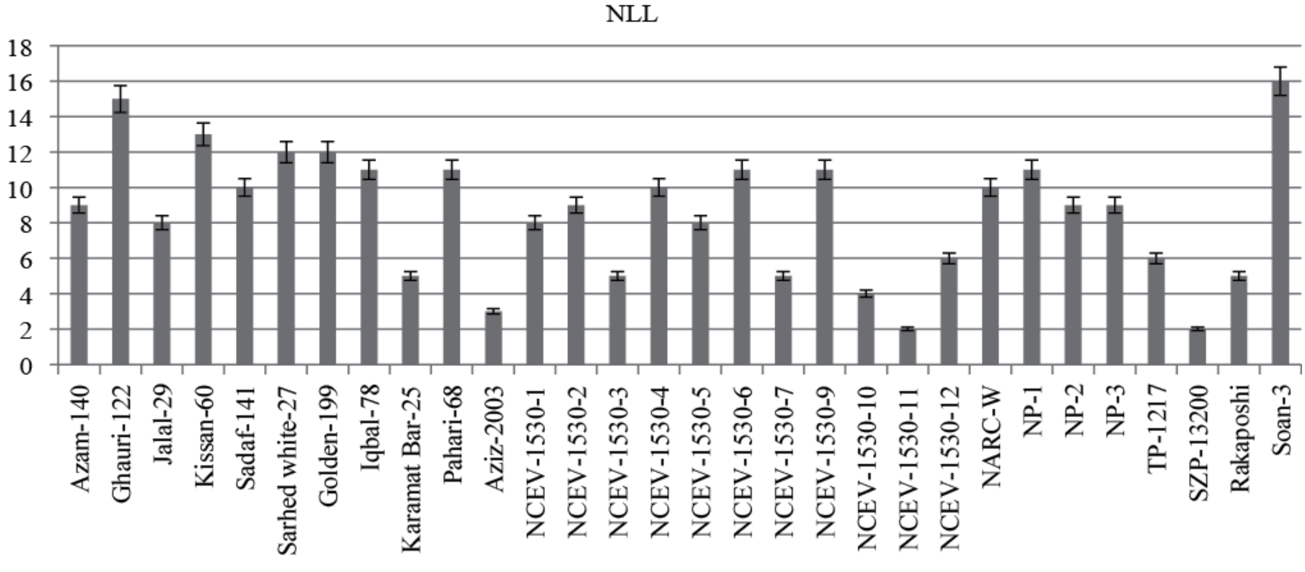

Fig. 1. Number of lesion on leaf of 30 maize genotypes.

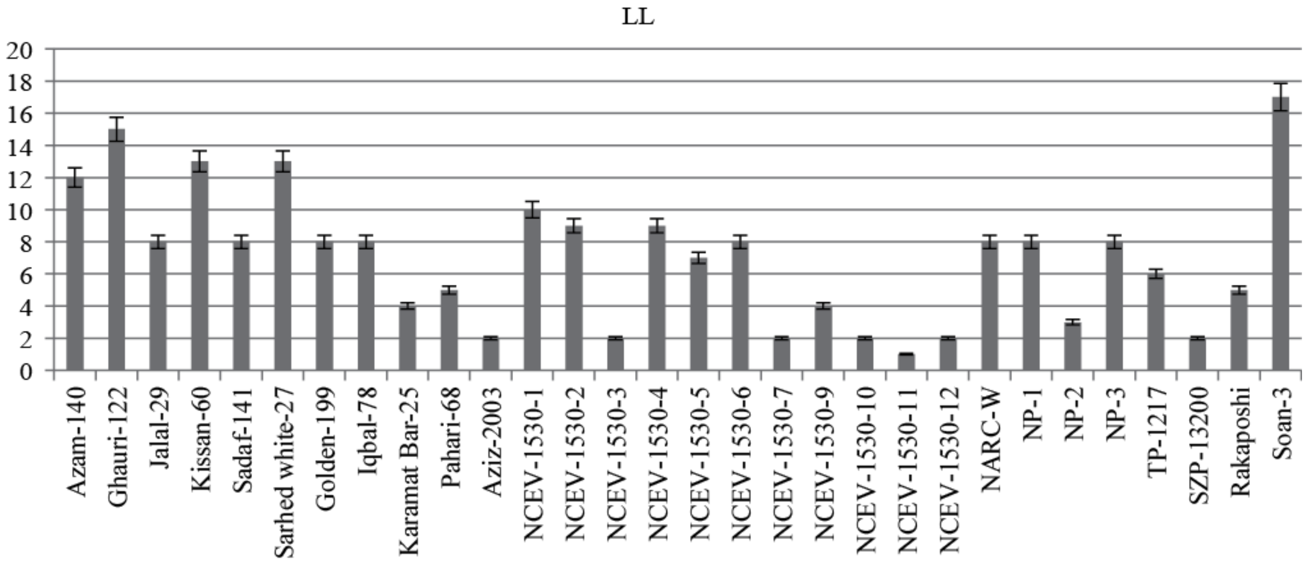

Fig. 2. Lesion length (cm) on leaf of 30 maize genotypes.

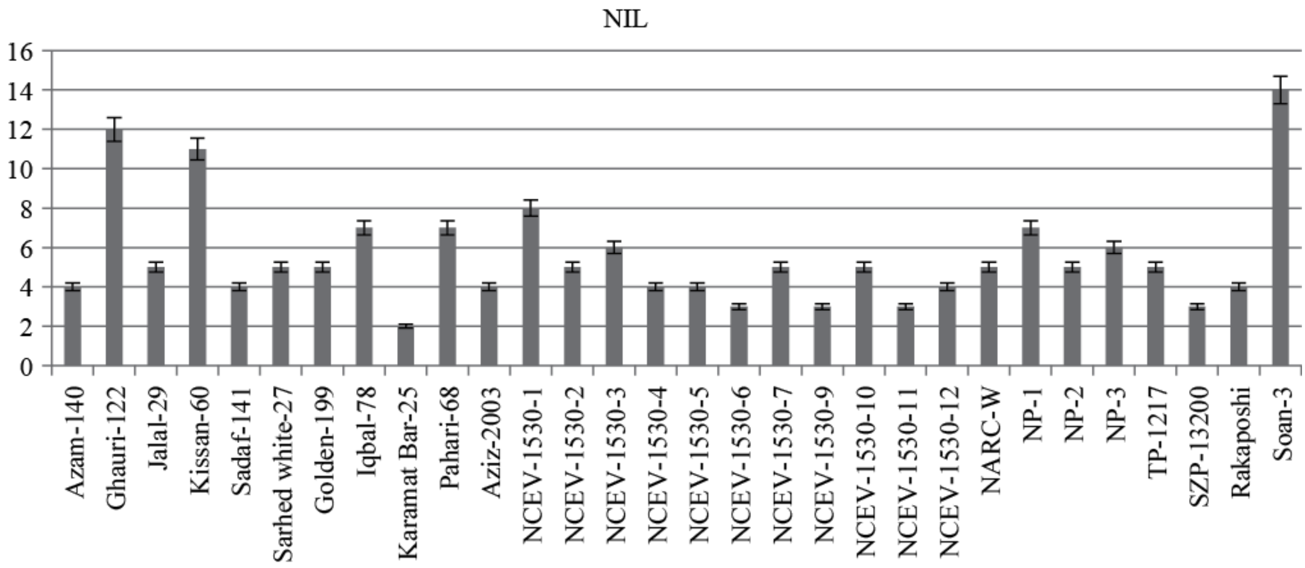

Fig. 3. Number of infected leaf of 30 maize genotypes.

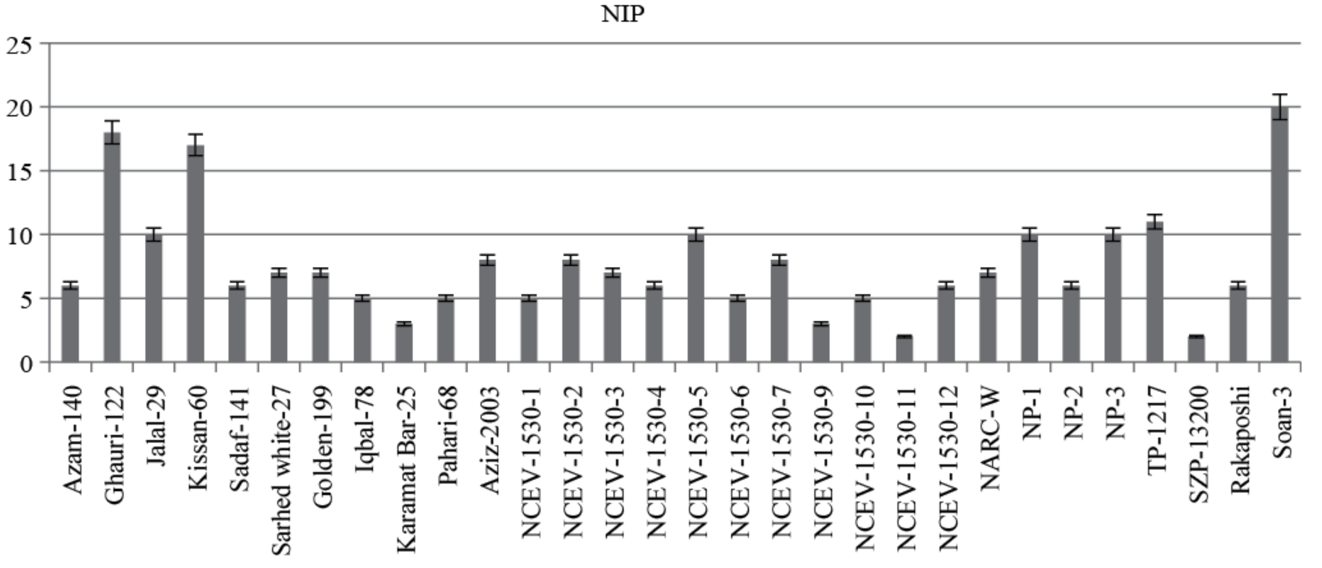

Fig. 4. Number of infected plants of 30 maize genotypes.

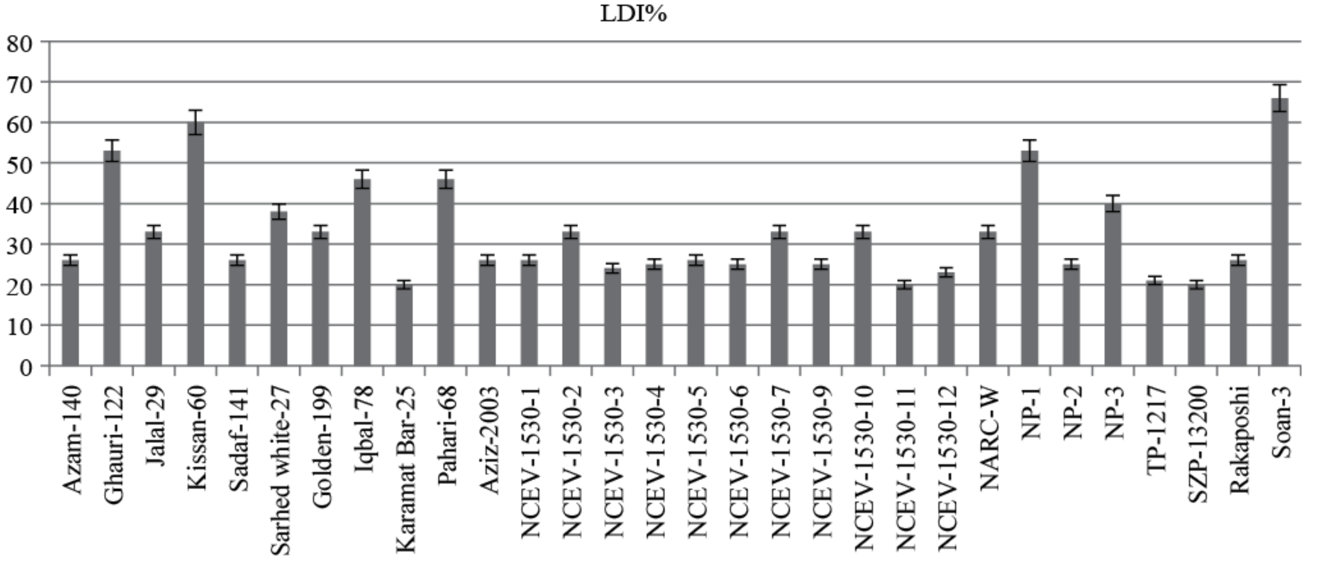

Fig. 5. Leaf disease incidence of 30 maize genotypes.

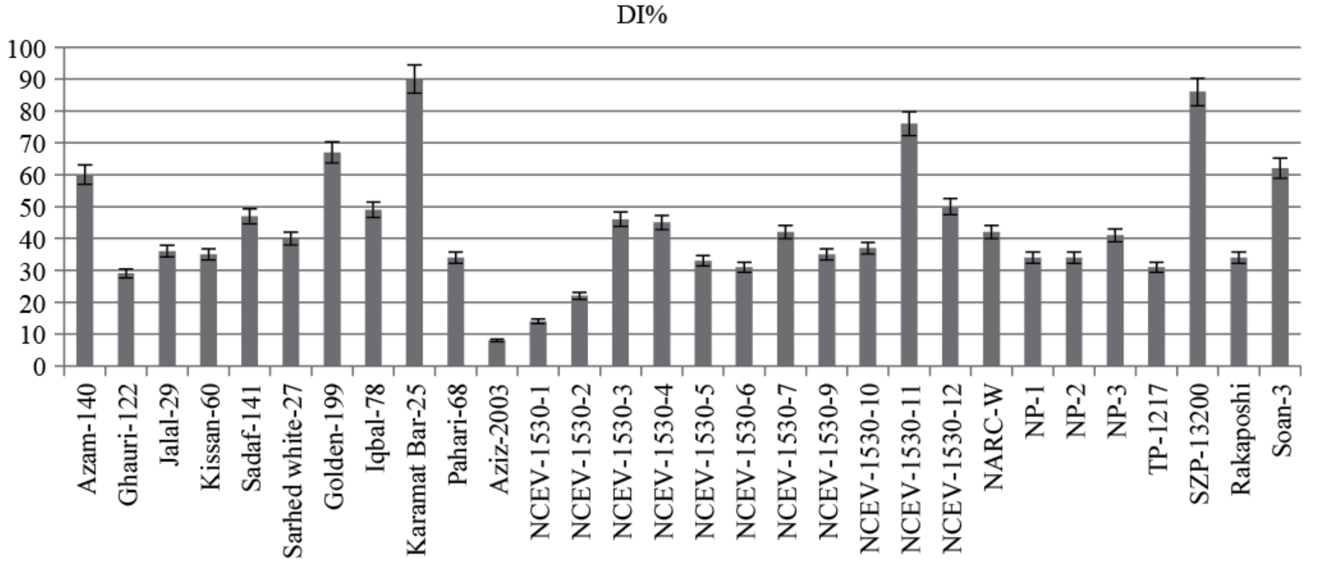

Fig. 6.Disease index of 30 maize genotypes.

Disease index: Mean values for leaf disease index ranged from 20-66% are given in Table 2. Maximum mean value for disease index was shown by genotype Soan-3 (66%) followed by Kissan-60 (60%) and Ghuari-122 (53%). Whereas minimum mean values were shown by genotype SZP-13200 (20%), NCEV-1530-11 (20%) and Karamat-Bar-25 (20%) followed by TP-1217 (21%) and NCEV-1530-12 (23%), respectively (Fig. 6).

Disease intensity: Mean values for disease intensity ranging from 16-34% are presented in Table 2. Maximum mean value for disease intensity was shown by genotype Soan-3 (34%) followed by Kissan-60 (33%), Ghuari-122 and Rakaposhi (31%). Whereas minimum mean values were shown by genotype NCEV-1530-11 (16%), SZP-13200 (17%) and Karamat-Bar-25 (19%) followed by TP-1217 (20%), respectively (Fig. 7).

Percentage leaf infection: Mean values for percentage leaf infection were displayed in the range of 20-90% (Table 2). Maximum mean value for percentage leaf infection was shown by genotype Soan-3 (90%) followed by Ghuari-122 (77%), Kissan-60 (72%) and Iqbal-78 (70%). Whereas minimum mean values were shown by genotype Karamat-Bar-25 (18%) followed by SZP-13200 (20%) and NCEV-1530-11 (21%), respectively (Fig. 8).

Percentage plant infection: Mean values for percentage plant infection ranging from 8-80% are given in Table 2. Maximum mean value for percentage plant infection was shown by genotype Soan-3 (80%), Ghuari-122 (80%) followed by Kissan-60 (70%). Whereas minimum mean values was shown by genotype Karamat-Bar-25 (8%) followed by NCEV-1530-11 (10%) and SZP-13200 (12%), respectively (Fig. 9).

Area under disease progressive curve (AUDPC): Mean value for area under disease progressive curve ranging from 22-362 is given in Table 2. Maximum mean value for area under disease progressive curve was shown by genotype Soan-3 (362), Ghuari-122 (362) followed by Kissan-60 (322). Whereas minimum mean values were shown by genotype Karamat-Bar-25 (22) followed by SZP-13200 (22) and NCEV-1530-11 (42), respectively (Fig. 10).

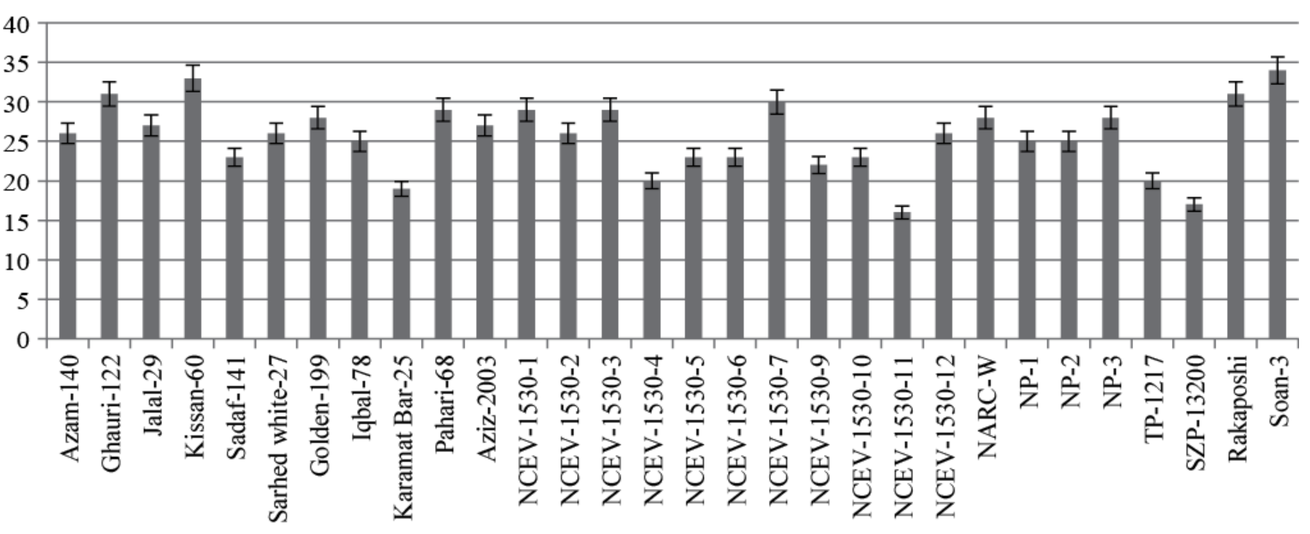

Fig. 7. Disease intensity (%) of 30 maize genotypes.

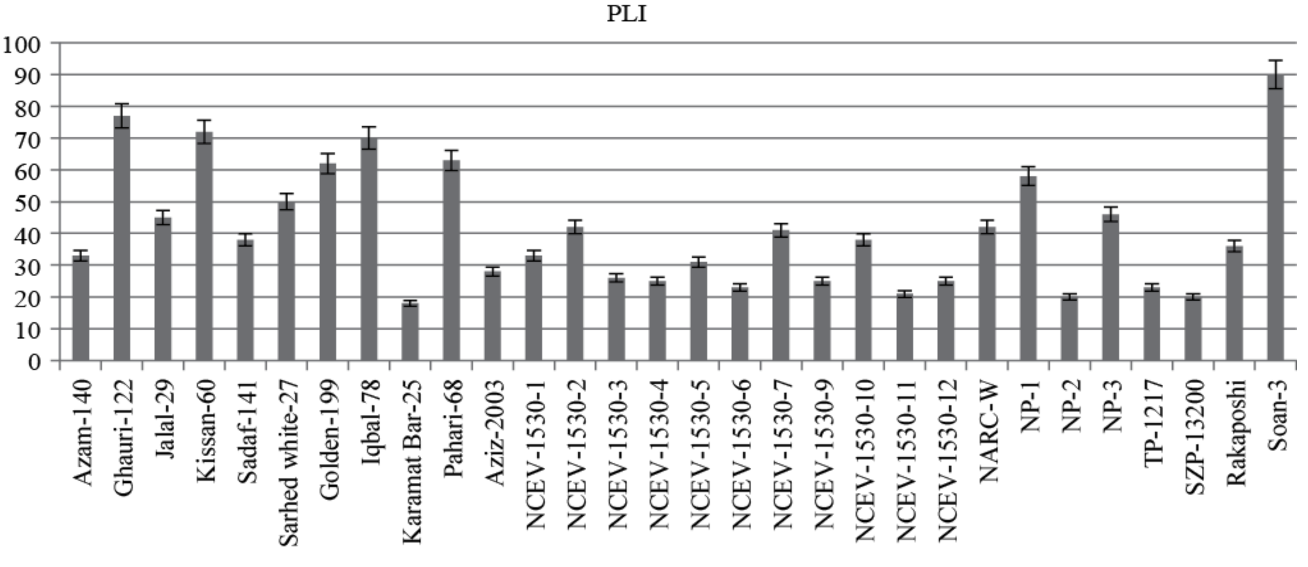

Fig. 8. Percentage leaf infection (%) of 30 maize genotypes.

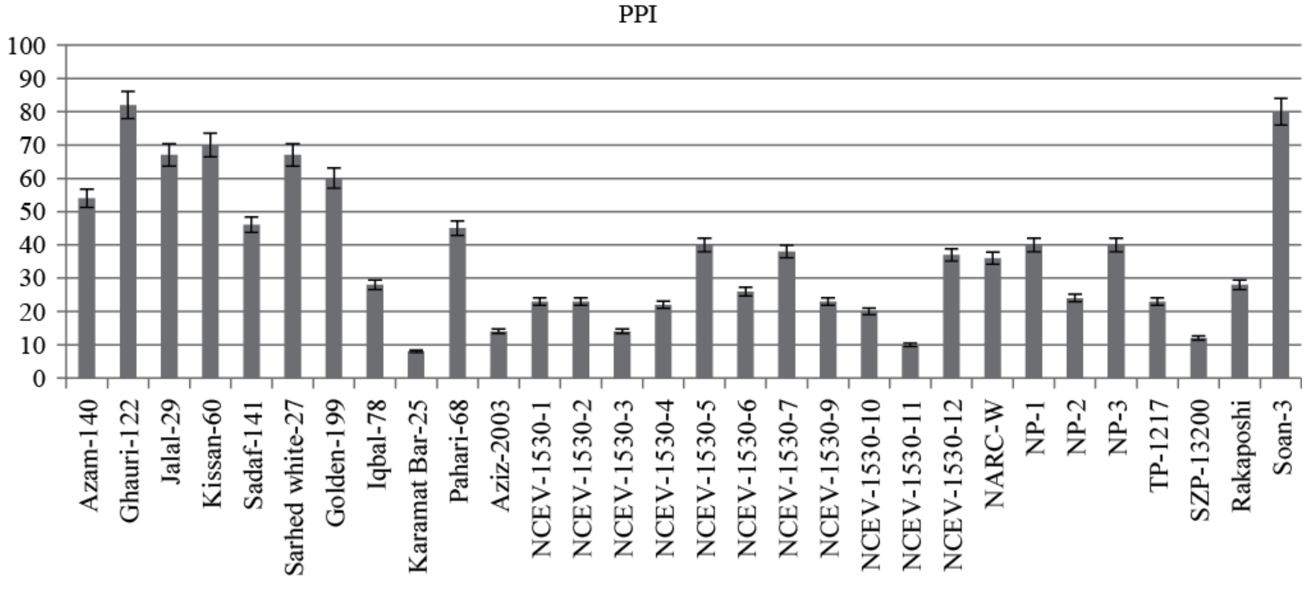

Fig. 9. Percentage plant infection of 30 maize genotypes.

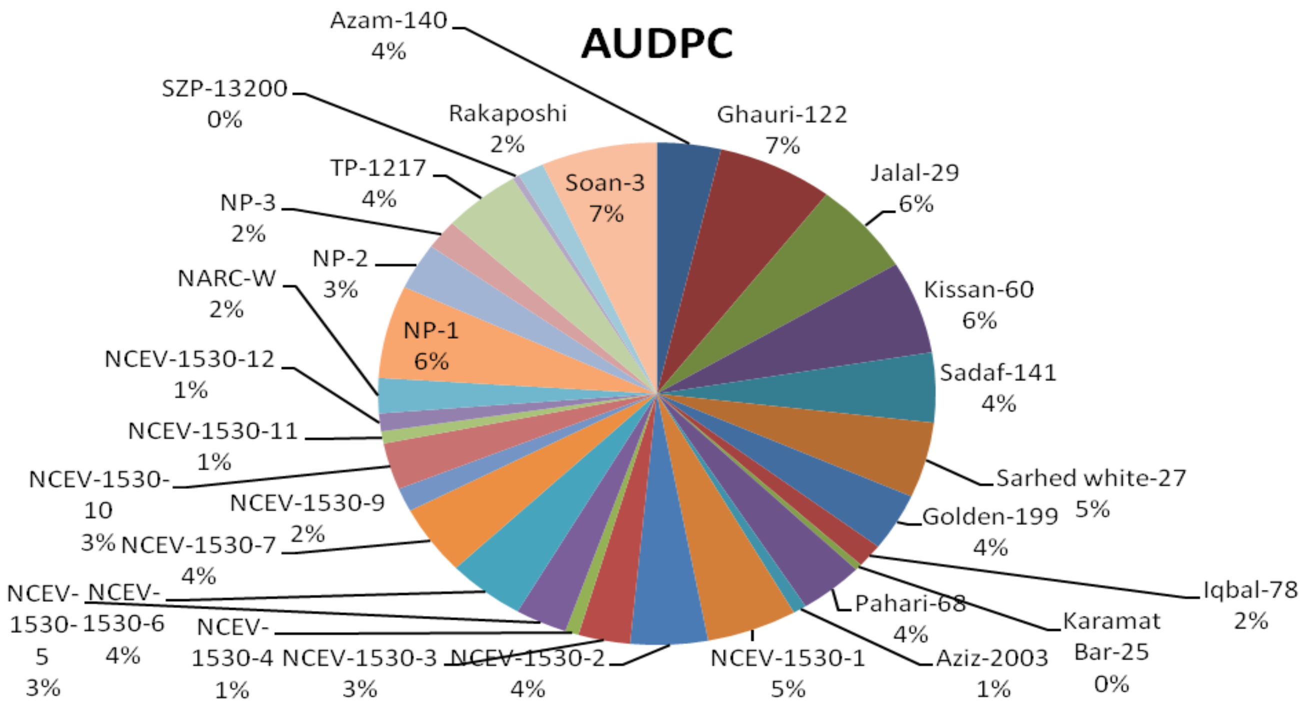

Fig. 10. Area under disease progressive curve (AUDPC) of 30 maize genotype.

Reaction and scoring of disease: Under field conditions the genotypes Kramat-Bar-25, SZP-13200 and NCEV-1530-11 displayed the disease severity of 5R, 5R, 10R, respectively and showed resistance type of reaction against Northern Corn Leaf Blight (Table 3). While genotypes Soan-3, Ghuari-122, Kissan-60 and Iqbal-68 were found susceptible and their reaction rating values were 90S, 90S and 80S, respectively.

Genotypes Jalal-29, Sarhad White-27, NCEV-1530-1 NCEV-1530-3 NCEV-1530-6 NCEV-1530-7, NCEV-1530-10, NCEV-1530-12, NP-1 and NP-2 showed moderate susceptible type reaction. Whereas genotypes Azam-140, Sadaf-141, Golden-199, Pahari-68, Aziz-2003, NCEV-1530-2, NCEV-1530-4, NCEV-1530-5, NCEV-1530-9, NCEV-1530-12 NARC-W, NP-3, TP-1217 and Rakaposhi showed moderate resistant reaction under field screening conditions.

Cluster analysis

Members of clusters based on average linkage distance in maize genotypes: The pathological traits of 30 maize genotypes were classified in two main clusters, cluster I and cluster II (Fig. 11). Cluster I was further divided into two sub-clusters Ia and Ib and cluster II was further divided into two sub-clusters IIa and IIb. Subcluster IIb was further divided into IIc and IId. Sub-cluster Ia was divided into sub-sub clusters which contained genotypes NCEV-1530-11, Karamat-Bar-25 and SZP-13200. Sub-cluster Ib contained genotypes Iqbal-78, NARC-W, NP-3, Aziz-

2003, NCEV-1530-4, NCEV-1530-12, NCEV-1530-9 and Rakaposhi. Sub-cluster IIa contained genotypes Phari-68, Azam-140, Golden-199, NCEV-11530-5, NCEV-1530-10, NCEV-1530-3 and NP-2. Sub-cluster IIc contained genotypes Sadaf-141, Sarhed-White, NCEV-1530-1, NCEV-1530-2, NCEV-1530-6, NCEV-1530-7 and TP-1217 whereas sub-sub-cluster IId contained genotypes Jalal-29, Kissan-60, NP-1, Ghuari-122 and Soan-3.

The highly resistant genotypes i.e., Karamat-Bar-25, NCEV-1350-11 and SZP-13200 fall into cluster I and showing close genetic relation and genotypes which are highly susceptible i.e., Kissan-60, Ghuari-122 and Soan-3 fall cluster II which also showed close genetic relation with respect to NCLB. The genotypes which were moderately resistant fall into same and nearly close sub clusters and showing their close genetic distance whereas moderately susceptible genotypes also revealed close genetic relationship by falling into same sub-sub-cluster.

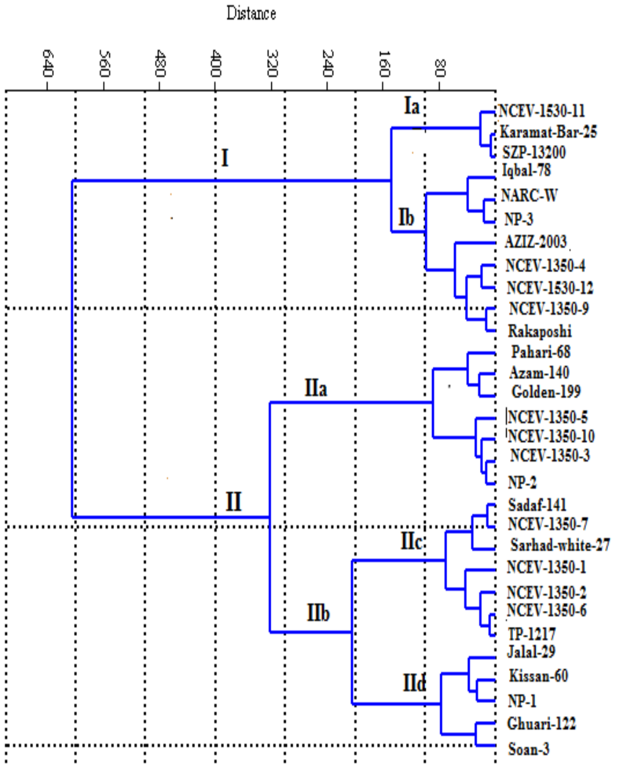

Fig. 11. Dendogram based on average linkage distance in 30 Maize genotypes.

Discussion

The present study resulted in identification of three highly resistant and three highly susceptible genotypes from germplasm on the basis of rating, incidence scoring and other related pathological traits. Karamat-Bar-25, NCEV-1530-11 and SZP-13200 showing highly resistance response and genotypes i.e., Kissan-60, Ghuari-122 and Soan-3 showing highly susceptible reaction toward the Northern Corn Leaf Blight under field screening condition. Resistant genotypes showed least lesion length, disease incidence, disease severity, disease intensity and area under disease progressive curve. The differences amongst the genotypes for resistance to NCLB diseases indicated the potential inherent genetic diversity in the genotypes. Genotypes which were tolerant to NCLB had few lesions on their leaf despite being subjected to the same disease pressure as the susceptible genotypes. The genotypes that were susceptible to NCLB suffered from blighting and necrosis of photosynthetic tissues and showed blighting. About 13 genotypes were observed to be moderately resistant to NCLB. In general, resistant and moderately resistant test materials were greater in number in germplasm than susceptible and highly susceptible types.

The Hierarchal Clustering (HC) based on average linkage distance was helpful for the selection of disease resistant and high yielding genotypes for future breeding program. Similar results were reported by Biabani *et al.,* (2005) while working on nuclear polyhedrosis virus against silk worm (*Bombyx mori*). They reported that Agglomerative hierarchical clustering (AHC) analysis and classified various hybrids into two classes on the basis of their degree of resistance.

In maize NCLB resistance was reported in previous studies by (Muriithi & Mutinda, 2001; Harlapur, 2005). Kumar *et al.,* (2011) evaluated twenty inbred genotypes as sources of resistance against NCLB and described that genotype NAI-147 and composite Girija showed resistance to the NCLB; Adipala *et al.,* (1993) notified that genotypes Babungo 3, population 42 and KWCA significantly showed high resistance to NCLB as compared to susceptible check B73. Likewise, Rai *et al.,* (2009) while studying the screening of maize genotypes from maize germplasm and reported that out of the fifty one genotypes, 26 were noted as resistant (R), 8 as moderately resistant (MR), 13 as moderately susceptible (MS), 2 susceptible (S) and 2 showed highly susceptible reactions towards NCLB.

Resistance to NCLB in maize germplasm is reported by qualitative and quantitative mechanisms (Hooker, 1981; Ogliari *et al*., 2007). Quantitative resistance is described by less number of lesions, smaller lesion size with necrotic area, as well as minimum severity and low area under disease progress curve (AUDPC) values while qualitative resistance is characterized by small lesions bounded by chlorotic halo. In current study, resistant plants had typically quantitative type of resistance to NCLB. Quantitative resistance with slight epidemics and variation allow the evolution of pathogens (McDonald & Linde, 2002).

The significant Maize genotypes x disease type interaction suggest that some genotypes perform better across the environment. This indicates that the genotypes were able to tolerate high disease pressure. The results are similar to Ramdutta *et al*., (2005) they screened maize genotypes under glasshouse environment and perceived that the genotypes showed resistance to NCLB. These results were in agreement with the finding of Chandrashekara *et al*., (2012) while working with Northern Corn Leaf Blight in maize. The new sources of NCLB resistance identified in the present study will be helpful for their utilization in breeding program and as donor parents for various basic and applied researches. In areas where NCLB is prevalent, the resistant genotypes can be grown and improve breeding programs.

Table 3. Disease scoring and disease reaction.

Genotypes	Disease scoring	Disease reaction	Genotypes	Disease scoring	Disease reaction
Azam-140	50	MR	NCEV-1530-5	40	MR
Ghauri-122	90	S	NCEV-1530-6	60	MS
Jalal-29	80	MS	NCEV-1530-7	60	MS
Kissan-60	80	S	NCEV-1530-9	20	MR
Sadaf-141	60	MR	NCEV-1530-10	40	MS
Sarhed white-27	90	MS	NCEV-1530-11	10	R
Golden-199	50	MR	NCEV-1530-12	15	MR
Iqbal-78	20	MR	NARC-W	30	MR
Karamat Bar-25	5	R	NP-1	80	MS
Pahari-68	50	MR	NP-2	40	MS
Aziz-2003	10	MR	NP-3	25	MR
NCEV-1530-1	70	MS	TP-1217	60	MR
NCEV-1530-2	60	MR	SZP-13200	5	R
NCEV-1530-3	40	MS	Rakaposhi	20	MR
NCEV-1530-4	20	R	Soan-3	90	S

Conclusion

The current research resulted in identification of 3 resistant (Kramat-Bar-25, SZP-13200 and NCEV-1530-11) and 3 susceptible genotypes (Soan-3, Ghuari-122, Kissan-60) genotypes. Among them Azam-140, Sadaf-141, Golden-199, Pahari-68, Aziz-2003, NCEV-1530-2, NCEV-1530-4, NCEV-1530-5, NCEV-1530-9, NCEV-1530-12, NARC-W, NP-3, TP-1217, Rakaposhi were moderately resistant while Jalal-29, Sarhad White-27, NCEV-1530-1, NCEV-1530-3, NCEV-1530-6, NCEV-1530-7, NCEV-1530-10, NCEV-1530-12, NP-1, NP-2 were moderately susceptible genotypes. In general, resistant and moderately resistant test materials were greater in number than susceptible and highly susceptible types. The new sources of NCLB resistance identified in the present study will be helpful for their deployment in breeding program.

Acknowledgement

The authors would like to thank the National Agriculture Research Center (NARC), Islamabad and Cereal Crop Research Institute, Pirsabak, Nowshehra, Khyber Pakhtunkhwa for providing the plant material.

References

Adipala, E., P.E. Lipps and L.V. Madden. 1993. Occurrence of *Exserohilum turcicum* on maize in Uganda. *Plant Disease*, 77: 202-205.

Ali, F. and J.B. Yan. 2012. The phenomenon of disease resistance in maize and the role of molecular bc34reeding in defending against global threat. *J. Integ. Plant Biol.,* 55: 134-151.

Ali, G., A.Z. Ishfaq and A. Dar. 2014. Disease reaction studies of maize (*Zea mays* L.) against *Turcicum* leaf blight involve indigenously identified cytosterile source. *Afr. J. Microb. Res.,* 8: 2592-2597.

Bashyal, B.M., R. Chand, L. Prasad and A.K. Joshi. 2011. Partial resistance components for the management of spot blotch pathogen *Bipolaris sorokiniana* of barley (*Hordeum vulgare* L.). *Act. Phytopathol. Entom. Hung,* 46: 49-57.

Biabani, M.R., A.R. Seidavi, M.R. Gholami, K. Etebari and L. Matindoost. 2005. Evaluation of resistance to nuclear polyhedrosis virus in 20 commercial hybrids of silkworm (*Bombyx mori*). *Form. Ento*., 25: 103-112.

Campbell, C.L. and L.V. Madden. 1990. Introduction to Plant Disease Epidemiology. *John Wiley and Sons, New York*.

Chandrashshekara, C.S., K. Jha, P.K. Agrawal, N.K. Singh and J.C. Bhatt. 2012. Screening of extra early maize inbred under artificial epiphytotic condition for North- Western Himalayan region of India. *Maize Genetic Coop. Newsl.,* 86: 1-4.

CIMMYT. 2004. Maize disease, a guide for field identification. 4th Edition. Government of Pakistan. Ministry of food, Agriculture and livestock, economic wing. Islamabad, Pakistan.

Hammer, O., D.A.T. Harper and P.D. Ryan. 2001. PAST: Paleantological statistics software package for education and data analysis. *Palaeon. Elec*., 4: 1-9.

Harlapur, 2005. Epidemiology and management of turcicum Leaf blight of maize caused by *Exserohilum turcicium* (pass.) Leonard and suggs. Ph.D. (Plant Pathology). Thesis, University of Agricultural Sciences, Dharwad.

Hooker, A.L. 1981. Resistance to *Helminthosporium turcicum* from *Tripsacumfloridanum* incorporated into corn. *Maize Genetic Coop. News.,* 55: 87-88.

Ishfaq, A.Z.A., A. Dar, G. Lone, A. Ali, B. Gazal and F.A. Mohiddin. 2014. Disease reaction studies of maize against *turcicum* leaf blight involving indigenously identified cytosterile source. *Afr. J. Micro. Res*., 8: 2592-2597.

Khalil, I.A., H. Rahman, Durrishahwar, I. Nawaz, H. Ullah and F. Ali. 2010. Response to selection for grain yield under maydis leaf blight stress environment in maize (*Zea mays*). *Biodicon*., 3(1): 121-127.

Kumar, S., K.T. Pardurange, S.K. Pant, M. Shekhar, B. Kumar and B.H. Parsanna. 2011. Sources of resistance to *Exserohilum turcicum* (Pass.) and *Puccinia polysora* (Underw.) incitant of *Turcicum* leaf blight and polysora rust of maize. *Arch. Phytopath. Plant Prot.,* 44: 528-536.

McDonald, A.B. and C. Linde. 2002. Pathogen population genetics, evolutionary potential and durable resistance. *Ann. Rev. Phytol.,* 40: 349-379.

Muriithi, L.M. and C.J.M. Mutinda. 2001. Genetic variability of maize genotypes for resistance to *Exerohilum turcicum* in Kenya. In: 7th Eastern and South Africa Regional Maize Conf., pp. 11-15.

Ogliari, J.B., M.A. Guimaraes, I.O. Geraldi and L.E.A. Camargo. 2005. New resistance genes in the *Zea mays* - *Exserohilum turcicum* pathosystem. *Gene. Mol. Bio.,* (28): 435-439.

Pandey, K.K., P.K. Pandey, G. Kalloo and M.K. Banerjee. 2003. Resistance to early blight of tomato with respect to various parameters of disease epidemics. *J. Gen. Plant Pathol.,* 69: 364-371.

Rai, D., A. Kumar, M. Kumar and R. Prasad. 2009. Field Screening of Maize genotypes against Maydis leaf blight caused by *Helminthosporiun maydis* Nisicado and Miyake. *Inter. J. Pant. Protec*., 2: 265-266.

Ramdutta, M.S. and L.A.L. Sangam. 2005. Evaluation of maize genotypes for location source of resistant to *turcicum* incitant of *turcicum* leaf blight of maize. *Ind. Phytopathol*., 58: 67-70.

Ribeiro, R.M., A.T Junior, L.S.A Goncalves, L.S. Candido, T.R.C. Silva and G.F. Pena. 2016. History of Northern Corn leaf blight disease in the seventh cycle of recurrent selection of an UENF-14 popcorn population. *Acta Scientiarum*.

Singh, R. 2004. Identification of additional sources of resistance to *Exserohillum turcicum* in maize (*Zea mays* L.). SABRAO *J. Breeding* and *Gen.,* 36: 45-47.

Vieira, R.A., M. Renata, N. Cleiltan, T. Fernando, J. Dauri and A. Carlos. 2014. A new diagrammatic scale for the assessment of Northern corn leaf blight. *Crop Protec.,* 56: 55-57.

Wise K. 2011. "Diseases of Corn: Northern Corn Leaf Blight". Purdue University Extension Publication. Purdue University.

(Received for publication 28 April 2018)