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GENETIC ASSESSMENT OF CSISA - SPOT BLOTCH LINES OF WHEAT (*TRITICUM AESTIVUM* L.) FOR SPOT BLOTCH RESISTANCE AND YIELD TRAITS

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Abstract

Fifty two CSISA- spot blotch lines were analysed in randomised block design for various genetic parameters, diversity and principal components. Significant variability was found among the accessions for all the traits. High heritability coupled with high genetic advance was observed for lesion mimic. All the 52 lines were grouped into 8 clusters based on the genetic divergence. Maximum diversity was assessed between Cluster III and VI whereas maximum intra-cluster distance was found in the cluster I and in cluster VIII suggesting that member of these clusters can be utilised in hybridization breeding programmes. The first two principal components (PCI and PCII) cumulatively explained a major portion of total variability (46.52%). Spike length and lesion mimic in PC1 showed highest positive weights. Days to heading, biomass and plant height in PCII were the characters possessing highest positive weights. The findings of this study pave ways for combining breeding for higher yield with enhanced resistance to spot blotch, a major biotic constraint in south-east Asia as the accessions involved in the study have shown wide genetic variance and exhibited genetic divergence that are prerequisites of transgressive and recombination breeding.

Introduction

India, Nepal, Bangladesh and Pakistan are major wheat producing countries in Indian subcontinent. The main aspects of wheat improvement are dependent upon search for lines with promising performance in the changed ambience and on the development of transgressive segregants. Biotic stresses have central role among the many determinants of production and productivity of any crop (Pandey *et al.* 2014). The chief biotic constraint faced by wheat in south Asia is the spot blotch of wheat caused by the fungal pathogen *Bipolaris sorokiniana* (Sacc.) Shoem syn. *Drechslera sorokiniana* (Sacc.) Subrm and Jain (syn. *Helminthosporium sativum*, teleomorph *Cochliobolus sativus*) (Dubin and van Ginkel 1991, Joshi *et al.* 2004a,b, Sharma *et al*. 2004). The disease has shown the potential to cause significant yield loss ranging from overall 22 per cent to complete failure of crop under severe epidemics (Saari 1998 and Chowdhury *et al*. 2013). Although the efforts focussed to develop spot blotch resistant lines combined with appreciable yield and other desired agronomic characters have not yet made substantial progress. Due to continuous breeding efforts the genetic background of most of the available wheat varieties has been narrowed. The cluster and the principal components analysis will be helpful in selection of diverse parents for hybridization programmes to generate variability and also to select for transgressive segregants.

The present investigation was intended to estimate the variability and divergence for yield and spot blotch resistance under artificial epiphytic conditions.

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Materials and Methods

A set of 52 lines included in CSISA- SB (Cereal System Initiative for south Asia-spot Blotch) programme of CIMMYT including the susceptible check Sonalika were grown in RBD with spacing of 23×10 cm replicated twice at Agricultural Research Farm, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi (U.P.), India during the rabi season of 2012-13. The individual pot size was 3 × 1 m. Along with standard agronomic practices to ensure a good crop stand it was taken care that the crop must not face any moisture deficit in order to facilitate the disease development. The artificial epiphytotic was created by spray of the inoculum. Observations were recorded from randomly tagged five plants form each line and averaged value was used in analysis.

The observations on disease severity were recorded at different growth stages **(**Zadoks *et al.* 1974) on double - digit scale as proposed by Saari and Prescott (1975). The area under disease progress curve was calculated according to the following formula given by Shaner and Finney (1977) from the observations on the disease severity.

$$\mathrm{AUDPC} = \sum_{i=1}^{n=1} [\{(Y_i+Y_{(i+1)})/2\}\times(t_{(i+1)} - t_i)]$$

Analysis of variance was made according to procedure given by Panse and Sukhatame (1967). The D^2-statistics of Mahalanobis (1928) was applied to assess the genetic distance among lines as measured by allelic frequencies at a sample of loci. The D^2 values of all combinations of pair of lines were arranged in ascending order of magnitude. The grouping of lines into different clusters was done by Ward's method as described by Rao (1952).

Results and Discussion

All the characters (Table 1) showed significant variation among the genotypes. Here, very high estimate of GCV was observed for the traits like lesion mimic (49.76), glaucouseness (waxiness) (34.22) and area under disease progress curve (AUDPC) (20.57). Traits like peduncle length (17.12), biomass (13.96), 1000-grain weight (10.63) and plot yield (7.54) were among the relatively moderate variable traits. The traits like chlorophyll content, canopy temperature and days to heading were least variable traits. Heritability (broad sense) was calculated for all the characters and the traits like 1000-grain weight (0.95), biomass (0.93), lesion mimic (0.91) and plant height (0.90) were found to be the most heritable traits thus, having higher proportion of genotypic variation. Similar results were reported by Ali *et al.* (2008). Characters like plot yield (0.26), chlorophyll content (0.42) and canopy temperature (0.56) were least heritable traits showing a major proportion of the variability for these traits to be due to environmental factors only.

Very high estimates of genetic advance for the traits like lesion mimic (97.61), glaucouseness (62.57) and AUDPC (39.72) envisages the applicability of these traits for immediate response to selection. Since lesion mimic had shown significant positive correlation with AUDPC (Pandey *et al.* 2014), the very high heritability coupled with high genetic advance for lesion mimic paves the way to improve spot blotch resistance by applying lesion mimic as morphological marker in negative selection programme.

All the 52 line included in this investigation were grouped in to eight clusters (Table 2) based on Euclidean dissimilarity using complete linkage method i.e. based on Ward's method at 40% linkage distance (Fig. 1). The euclidean distance among the clusters i.e. inter and intra cluster distances based on Mahalonbis (1928) D^2 statistics are presented in the Table 3. The maximum extent of within cluster distance was noticed for cluster I followed by cluster VIII and cluster II

Table 1. Analysis of variance and other genetic variability parameters for 12 yield and spot blotch resistance traits.

Source of variation	MSS												
	Degree of freedom	Days to heading	Glauco-useness	Plant height (cm)	Peduncle length (cm)	Spike length (cm)	Chloro-phyll content at 80 DAS	Canopy temp. at 80 DAS	Lesion mimic %	1000-grain weight (g)	Plot yield (g)	Biomass (g)	AUDPC
Replication	1	0.15	0.24	0.09	5.09	0.24	0.28	1.89	10.16	0.01	294.47	216.35	17572.10
Treatment	51	21.41**	1.32**	79.81**	16.64**	1.45**	6.45**	1.85**	85.12**	22.39**	4331.81*	91693.06**	72686.99**
Error	51	1.25	0.16	4.07	2.26	0.40	2.66	0.53	4.15	0.54	2554.81	3476.15	4685.04
GCV		4.13	34.22	6.50	17.12	6.94	2.82	3.98	49.76	10.63	7.54	13.96	20.57
PCV		4.38	38.56	6.84	19.63	9.23	4.37	5.34	52.24	10.89	14.85	14.50	21.94
h^2 (broad sense)		0.89	0.79	0.90	0.76	0.57	0.42	0.56	0.91	0.95	0.26	0.93	0.88
Gen. adv. as % of mean		8.03	62.57	12.72	30.75	10.76	3.74	6.12	97.61	21.37	7.89	27.68	39.72

suggesting that these cluster are composed of highly diverse lines. The lowest distance was found between genotypes CSISA 46720 and CSISA 46722 (Standardized Euclidean distance = 6.786) in the cluster IV suggesting the genotypes included in this cluster are more alike and can be utilised in backcross breeding (Khodadadi *et al.* 2011**)**. The highest distance was estimated to be between genotypes CSISA 46738 from cluster V and CSISA 46747 from cluster VIII (standardized Euclidean distance = 87.333). The maximum inter cluster distance was recorded between cluster III and VI leading to the possibility of recovery of transgressive segregants from the inter crosses

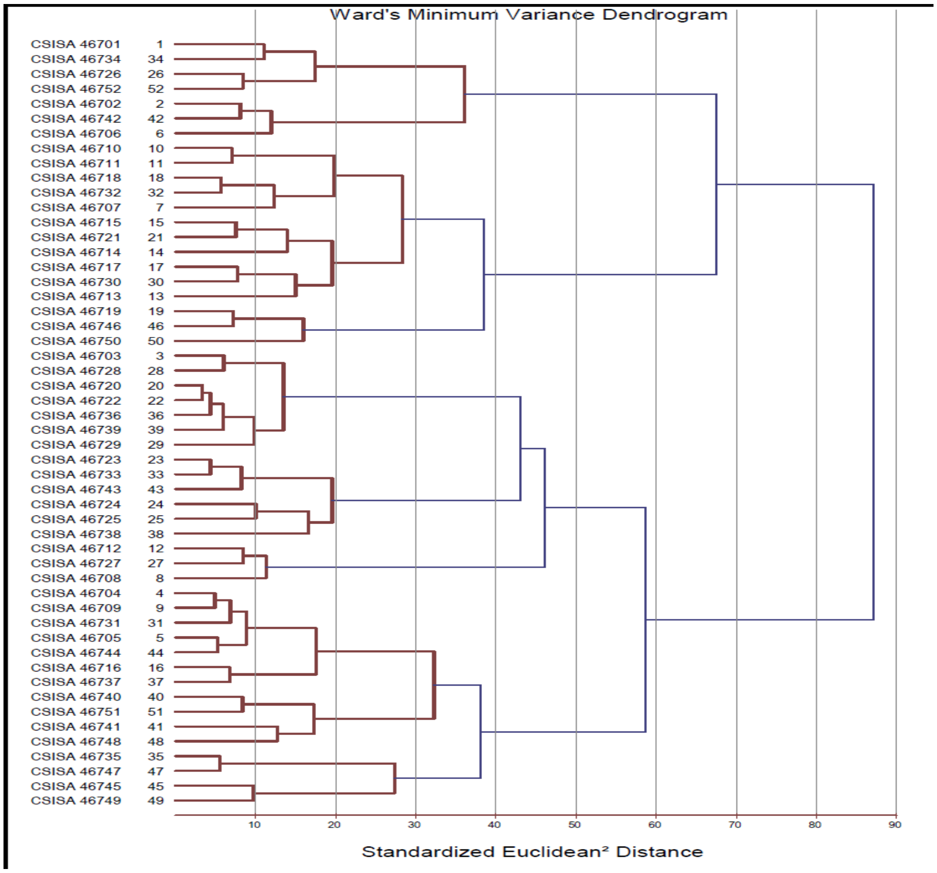

Fig. 1. Dendrogam showing 8 clusters of 52 accessions based on yield and spot blotch resistance traits at 40% linkage distance.

between the members of these two clusters. From the Table 4 showing cluster means for the traits studied it can be speculated that the two clusters (III and VI) are quite contrast for the traits like glaucouseness, plant height, spike length, canopy temperature, lesion mimic, 1000- grain weight, biomass and AUDPC. As most of these traits are highly associated with yield and disease resistance (Pandey *et al*. 2014), there is a scope of getting transgressive segregants combining high yield with appreciable level of spot blotch resistance from inter-crosses between cluster III and IV. In consonance with the findings of Khodadadi *et al.* (2011) and Ali *et al.* (2008) who reported the usefulness of cluster analysis for obtaining high yielding lines in wheat, this study also showed the presence of genetic divergence for yield and spot blotch resistance traits.

Table 2. Grouping of genotypes into 8 different clusters based on Euclidean distance.

Cluster	Total number of lines	Cluster members
I	7	CSISA 46701, CSISA 46734, CSISA 46726, CSISA 46752, CSISA 46702, CSISA 46742, CSISA 46706
II	11	CSISA 46710, CSISA 46711, CSISA 46718, CSISA 46732, CSISA 46707, CSISA 46715, CSISA 46721, CSISA 46714, CSISA 46717, CSISA 46730, CSISA 46713
III	3	CSISA 46719, CSISA 46746, CSISA 46750
IV	8	CSISA 46703, CSISA 46728, CSISA 46720, CSISA 46722, CSISA 46736, CSISA 46739, CSISA 46729 CSISA 46723,
V	5	CSISA 46733, CSISA 46743, CSISA 46724, CSISA 46725, CSISA 46738
VI	3	CSISA 46712, CSISA 46727, CSISA 46708
VII	11	CSISA 46704, CSISA 46709, CSISA 46731, CSISA 46705, CSISA 46744, CSISA 46716, CSISA 46737, CSISA 46740, CSISA 46751, CSISA 46741, CSISA 46748
VIII	4	CSISA 46735, CSISA 46747, CSISA 46745, CSISA 46749

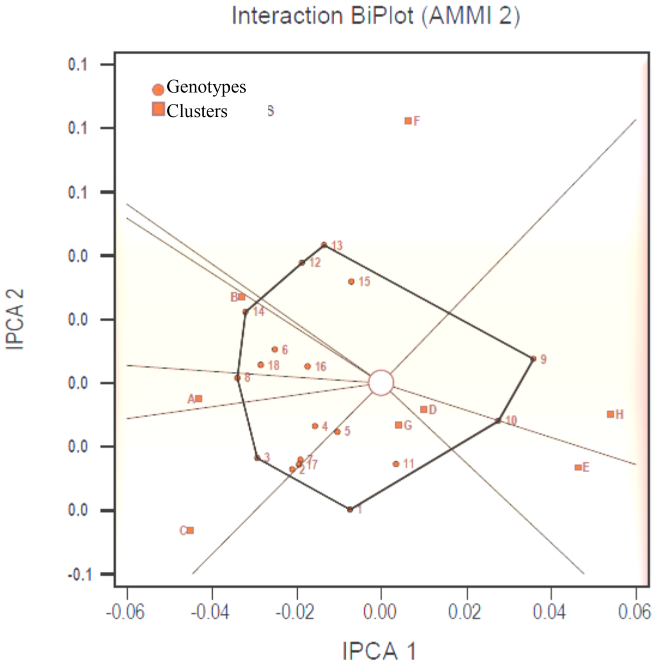

Fig. 2. The biplot of genotypes and clusters based on PC 1 and PC 2.

The mean value of different traits of each cluster has been given in the Table 4. The differences among the clusters are highly significant for almost all the traits studied. Cluster I had exhibited the highest mean value for AUDPC and lowest values for the traits like glaucouseness, plot yield and biomass. Thus, cluster I is supposed to be consisted of most susceptible lines thereby, resulting in lowest mean plot yield and biomass. Cluster II indicated to have highest mean value for traits like lesion mimic, 1000-grain weight and plot yield. The genotypes falling in cluster II are utilisable in yield improvement programmes. Highest mean value for glaucouseness, chlorophyll content and biomass was exhibited by cluster III which also had the lowest canopy temperature mean. Thus, components lines of cluster III are potential of keeping their canopy cooler probably due to high glaucousness which is supposed to be involved in dissipation of incoming radiation. The member of cluster III may therefore find their application in development of promising wheat lines for south Asia where the spot blotch often coupled with terminal heat is major threat to wheat production (Lillemo *et al.* 2013). Cluster IV had lower mean values for many traits and that too lowest for peduncle length and chlorophyll content among the clusters. Cluster V was supposed to be consisted of the lines which showed highest mean days to heading and spike length but the least means for lesion mimic and 1000-grain weight. The mean value for AUDPC was estimated to be the lowest for cluster VI which was accompanied with lowest means for plant height and days to heading. The member genotypes of cluster VI represent their potential for utilization in breeding programmes aimed at development of early and resistant lines with short stature. An average value for most of the traits but highest mean plant height was expressed by cluster VII. Cluster VIII was characterised by highest mean peduncle length but lowest spike length.

Table 3. Average inter and intra- cluster distances among 8 clusters.

	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V	Cluster VI	Cluster VII	Cluster VIII
Cluster I	**31.044**	40.738	46.505	37.808	46.429	43.902	39.672	47.209
Cluster II		**27.415**	36.553	29.717	45.656	39.600	33.833	43.477
Cluster III			**23.201**	36.867	45.702	59.863	35.736	45.080
Cluster IV				**14.4**	29.298	33.389	27.095	35.261
Cluster V					**23.509**	40.516	35.546	39.118
Cluster VI						**19.833**	39.975	44.550
Cluster VII							**24.195**	34.656
Cluster VIII								**28.447**

The variance and covariance matrix was applied in order to reduce the various quantitative parameters into a single index of similarity. The principal component analysis resulted into 12 Eigen values for 12 Eigen vectors out of which first six (PC1, PC2, PC3, PC4, PC5 and PC6; each having Eigen value more than 1) are described here and these six accounted for 77.221 percent of the total variation. The high estimate of the cumulative variation (46.522) explained by the first two principal components i.e. PC1 and PC2 revealed a high degree of correlation among the characters studied for these genotypes (Parihar *et al.* 2014). Eigen vector of the individual components indicates their level of association with the original traits. In case of PC1 characters spike length and lesion mimic were found to possess higher positive weights while canopy temperature, chlorophyll content, plant height, peduncle length and biomass were shown to have high negative weights. Days to heading, biomass and plant height had shown maximum positive weights in the PC2 whereas lesion mimic and AUDPC exhibited high magnitude of negative weights. Canopy temperature, peduncle length and days to heading for PC3; yield per plot, glaucouseness and canopy temperature for PC 4; lesion mimic, glaucouseness and days to heading for PC5 and peduncle length, glaucouseness and AUDPC for PC 6 had exhibited higher positive weights. The biplot of the PC1 and PC2 has been presented as Fig. 2.

Table 4. Mean values of 12 quantitative traits studied for all the 8 clusters identified based on D^2- statistics.

	Days to heading	Glauco-useness	Plant height (cm)	Peduncle length (cm)	Spike length (cm)	Chlorophyll content at 80 DAS	Canopy temp. at 80 DAS	Lesion mimic	1000-grain wt. (g)	Plot yield (g)	Biomass (g)	AUDPC
Cluster I	74.857	1.571	89.643	15.357	10.000	49.564	20.207	14.286	28.760	343.143	1221.429	1138.624
Cluster II	76.545	2.182	94.545	15.091	10.500	48.486	20.368	20.455	33.771	434.864	1484.091	825.421
Cluster III	77.833	3.833	99.833	17.500	10.500	51.833	19.367	15.000	32.553	423.167	1700.000	1049.383
Cluster IV	79.143	2.286	93.429	12.214	10.071	47.571	19.950	10.000	32.190	386.786	1625.000	856.878
Cluster V	79.250	3.083	92.167	13.667	11.583	49.125	20.892	5.000	28.211	374.917	1425.000	782.716
Cluster VI	74.333	1.667	86.000	14.500	9.500	49.050	22.350	15.000	32.907	391.000	1345.833	720.988
Cluster VII	75.500	1.682	100.500	18.091	10.864	48.841	20.250	10.455	30.917	399.773	1621.591	908.586
Cluster VIII	78.625	2.875	96.750	19.625	9.375	48.037	20.950	8.750	28.340	390.750	1618.750	892.593
Mean	76.865	2.231	94.702	15.663	10.433	48.848	20.427	12.788	31.100	395.106	1504.808	896.545
Treat MSS	22.103	3.012	131.726	36.177	2.766	6.625	2.739	176.434	31.732	5992.469	156894.947	102687.624
Err MSS	8.894	0.287	25.297	3.889	0.401	2.685	0.638	21.264	7.929	1557.139	28179.760	25788.823
Probability	0.031	0.000	0.000	0.000	0.000	0.032	0.001	0.000	0.002	0.002	0.000	0.002

The bi-plot graph had shown concordance with the cluster analysis showing distribution of genotypes in different environments where the environments represent the clusters. Biplot graph had also shown eight different environments/clusters. The closer the environments in the biplot graph the more is the similarity among them.

Thus the present study can be concluded with the appreciable magnitude if genetic variability and divergence among the lines and ample opportunity for yield improvement combined with spot blotch resistance using these lines.

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