

Research Article

Development of reference set for *Fusarium* wilt resistance using principal component scores in pigeonpea (*Cajanus cajan* Millsp L.) germplasm

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Abstract

A total of one hundred early duration germplasm accessions of pigeonpea were analyzed to understand the genetic behavior and form a reference set by measuring twentyfive yield related traits and *Fusarium* wilt resistance parameters. Significant mean sum of squares obtained for all the studied traits indicated the existence of variation among the genotypes. Principal Component Analysis (PCA) identified 7 principal components which explained 77.6% of the total variation. Using the principal Component Score strategy (PCS), a reference set of 17 germplasm accessions which represent 17 per cent of the entire germplasm was identified. The genotypes viz., ICP 4019, T 1424, ICP 970, ICP 3620, ICP 1836, T 1411 and ICP 2502 possessed both resistance to *Fusarium* wilt and good yield. These genotypes can be screened further over locations for confirmation of resistance and can be utilized as donors for resistance to *Fusarium* wilt during breeding programmes.

Keywords

Principal Component Analysis, Reference set, Diversity, Pigeonpea

Introduction

Pigeonpea ($2n=2x=22$) is the second most important pulse in India, largely preferred crop in rainfed systems due to its wide adaptability to varied climatic conditions and drought tolerance capacity. Narrow genetic base of the crops due to continuous selection for genotypes suitable for cultivation have the risk of being disposed to biotic and abiotic stresses (Mc Couch *et al.*, 2013; Guarino and Lobell, 2011; Xiao *et al.*, 1996). This emphasizes the importance to collect, conserve, document and maintain the germplasm of crops. Among the diseases affecting pigeonpea, *Fusarium* wilt caused by *Fusarium udum* causes heavy yield loss since it infects the crop at any stage, maximum damage being caused during flowering and podding stages. The economic losses due to *Fusarium* wilt in pigeonpea was reported to be more than 99 per cent before pod production and more than 70 per cent after pod production in India (Nene *et al.*, 1981). Principal Component Score strategy (PCS) can be used to develop reference collections for reducing redundancy within the collection and to identify diverse potential genotypes for biotic and abiotic stresses. It eliminates collinearity between variables and selects individual genotypes based on their cumulative relative contribution (Noirot *et al.*, 1996). Analyzing diversity within the germplasm helps to create useful variations by crossing diverse genotypes to improve the crop yields along with resistance. The Principal Component Analysis

(PCA) helps to identify the traits contributing largely for variation in a germplasm. In the present study, one hundred early duration pigeonpea germplasm was subjected to PCA analysis to know the pattern of similarity of the genotypes and the traits. PCS strategy was used to develop a reference set in pigeonpea germplasm based on principal component scores of the germplasm accessions.

Materials and Methods

The experiment was conducted at glass house of Centre for Plant Breeding and Genetics, Tamil Nadu Agricultural University, Coimbatore during *Kharif* 2017 involving 100 genotypes sown in two replications. Eleven yield and yield contributing traits viz., days to fifty per cent flowering (DFF), days to maturity (DTM), number of primary branches per plant (NPBPP), plant height (PH), pod bearing length (PBL), number of pods per plant (NPPP), pod weight per plant (PWPP), number of seeds per pod (NSPP), hundred seed weight (HSW), shelling percentage (SP), single plant yield (SPY) and six *Fusarium* wilt parameters measured during seedling stage viz., plant height (PH), percentage of affected leaves (PAL), root length (RL), stem diameter (SD), disease intensity index (DII) and per cent disease incidence (PDI) at 20, 35 and 50 days after inoculation were recorded in each genotype for characterization of the germplasm. Mean values of accessions were computed for determining analysis of variance. Principal

component analysis (PCA) was performed to assess genetic diversity among pigeonpea accessions using minitab 18. The PCS strategy described by Noiro *et al.*, (1996) employs PCA to eliminate collinearity between variables and selects individuals based on their cumulative relative contribution. The generalized sum of square (GSS) for all the accessions was calculated from the PCS (Lebart *et al.*, 1977).

The contribution P_i of the individual i to the total GSS was calculated as

$$P_i = \sum_{j=1}^k x_{ij}^2$$

The CR_{*i*} of the individual i to the total GSS was calculated as

$$CR_i = P_i / NK$$

where N is the number of individuals and K the variables.

The genotype with the highest CR_{*i*} (in %) to the total GSS was considered for the reference set. This was repeated up to 50% of GSS, and all the genotypes with the highest CR_{*i*} to 50% of GSS were considered for the final reference set.

Results and Discussions

Principal components analysis identified seven principal components with eigen value more than one accounting for 77.6 per cent of the original variability. Principal Component Scores and relative contribution of the individual genotypes (CR_{*i*}) for these seven PCs were estimated for separating the reference collection. By this strategy, 17 germplasm accessions were identified to form the reference collection which had maximum CR_{*i*}. The first PC explains 29.4 per cent of the variation in the germplasm due to positive loadings of 0.326 for per cent disease incidence, 0.314 for percentage of affected leaves, 0.296 for disease intensity index and negative loadings of -0.286 for seedling height and -0.264 for root length. The second PC summarizes 15.2 per cent of the variation accounted by positive loadings of 0.469 for single plant yield, 0.454 for number of pods per plant, 0.415 for number of primary branches per plant and 0.414 for pod weight per plant. The third PC explains 8.5 per cent of the variation due to positive loadings of 0.643 and 0.642 for days to fifty per cent flowering and days to maturity respectively. The fourth PC explains 8 per cent variation occurred due to negative loadings of -0.419 for plant height, -0.418 for pod bearing length, -0.377 for stem diameter and positive loadings of 0.274 for disease intensity index and 0.247 for per cent disease incidence. The fifth PC summarizes 6.1 per cent of the variation into positive loadings of 0.455

for shelling percentage and 0.392 for per cent disease incidence and negative loading of -0.319 for pod weight per plant. Negative loadings of -0.401 for shelling percentage, -0.394 for number of seeds per pod, -0.233 for stem diameter and positive loadings of 0.326 for plant height, 0.325 for pod bearing length and 0.265 for per cent disease incidence accounts for 5.9 per cent of the variation in the germplasm as detailed by the sixth PC. The seventh PC explains 4.4 per cent of the variation occurred due to positive loadings of 0.125 for per cent disease incidence and 0.278 for disease intensity index and negative loadings of -0.314 for hundred seed weight, -0.313 for shelling percentage and -0.183 for single plant yield.

PCA analysis explains 77.6 per cent of the variation in the germplasm in seven principal components. Reduction in seedling height and root length occurs due to wilt resulting in higher number of affected leaves and increased per cent disease incidence (PDI). Plant height is considerably affected by the pathogen since it blocks the vascular bundles denying the passage of nutrients through them subsequently limiting number of productive branches resulting in stunted plants. Jalander and Gachande (2011) observed differences for plant height among the genotypes infected with *F.udum*. Srivastava *et al.*, (2014) reported the production of acid detergent fibre (ADF), lignin in leaves and the activity of phenylalanine ammonia lyase and increased polyphenol oxidase and phenylalanine ammonia lyase activity in the roots. Phenolic compounds increase the cell wall thickening of roots that blocks the root pathogen in resistant genotypes. These act as modulator of pathogenicity and activators of plant defense genes (Datta and Lal, 2012). Per cent disease incidence also increases with increase in disease intensity index, shelling percentage and lesser stem diameter. Plant height, pod bearing length, pod weight per plant resulting in yield increase when there is lesser PDI. Yield increases due to the production of more number of primary branches per plant which as a result produces more number of pods per plant and pod weight per plant. The increase in source activity increases the sink due to more energy production. Hence, the trait number or primary branches per plant should be given importance during breeding programmes. While selecting genotypes for more number of primary branches per plant, selection for leaf shape also can be practised since erect leaves with more surface area can increase yield through source – sink relationship. Therefore, genotypes with maximum number of primary branches and leaf surface area for efficient photosynthesis can be selected for breeding programmes.



Yield decreases with increase in PDI as the result of decreased hundred seed weight and shelling percentage as explained by the seventh PC. The genotypes identified with resistance combined with good yield can be screened further over locations for confirmation of resistance and can be utilized as donors for resistance to *Fusarium* wilt during breeding programmes.

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Table 1. Eigen vectors, principal components for quantitative traits in pigeonpea accessions

	Principal Component Analysis						
	Principal Components						
	1	2	3	4	5	6	7
DFE	-0.036	0.029	0.643	-0.127	0.098	0.132	0.14
DTM	-0.036	0.034	0.642	-0.127	0.105	0.127	0.136
NPBPP	0.054	0.415	-0.096	0.082	-0.161	0.138	0.06
PH	-0.033	0.129	-0.232	-0.419	0.291	0.326	0.238
NPPP	0.056	0.454	-0.019	0.097	-0.167	0.109	-0.017
PBL	0.008	0.193	-0.242	-0.418	0.237	0.325	0.179
NSPP	0.076	0.162	0.045	-0.181	0.057	-0.394	0.143
PWPP	0.033	0.414	0.095	0.121	-0.319	0.169	-0.003
HSW	0.059	0.255	0.15	-0.02	0.17	-0.18	-0.314
SP	0.097	0.156	-0.033	-0.15	0.455	-0.401	-0.313
SPY	0.077	0.469	0.037	0.018	-0.021	-0.091	-0.183
PH (20 DAI)	-0.254	0.108	-0.019	0.274	0.155	-0.157	0.277
PH (35 DAI)	-0.286	0.117	-0.026	0.194	0.171	-0.196	0.258
PH (50 DAI)	-0.278	0.137	-0.027	0.119	0.119	-0.163	0.338
PAL (20 DAI)	0.296	0.023	-0.06	-0.021	0.009	-0.187	0.2
PAL (35 DAI)	0.314	0.009	-0.031	0.036	-0.043	-0.13	0.202
PAL (50 DAI)	0.296	-0.016	0	0.044	-0.111	-0.075	0.293
RL	-0.264	0.071	-0.025	0.143	0.228	-0.005	0.087
SD	-0.027	0.01	0.022	-0.377	-0.132	-0.233	-0.109
DII (20 DAI)	0.227	-0.014	-0.039	0.274	0.128	0.03	0.132
DII (35 DAI)	0.265	-0.03	-0.005	0.207	0.241	0.078	0.093
DII (50 DAI)	0.296	0.001	0.066	-0.097	0.007	-0.139	0.278
PDI (20 DAI)	0.158	0.065	0.03	0.213	0.392	0.189	-0.192
PDI (35 DAI)	0.236	-0.061	0.029	0.247	0.254	0.265	-0.137
PDI (50 DAI)	0.326	-0.023	0.039	-0.055	-0.026	-0.074	0.125
Eigen Value	7.3561	3.8028	2.1277	1.9953	1.5227	1.4864	1.1107
Per cent variation (%)	0.294	0.152	0.085	0.08	0.061	0.059	0.044
Cumulative variance (%)	0.294	0.446	0.531	0.611	0.672	0.732	0.776

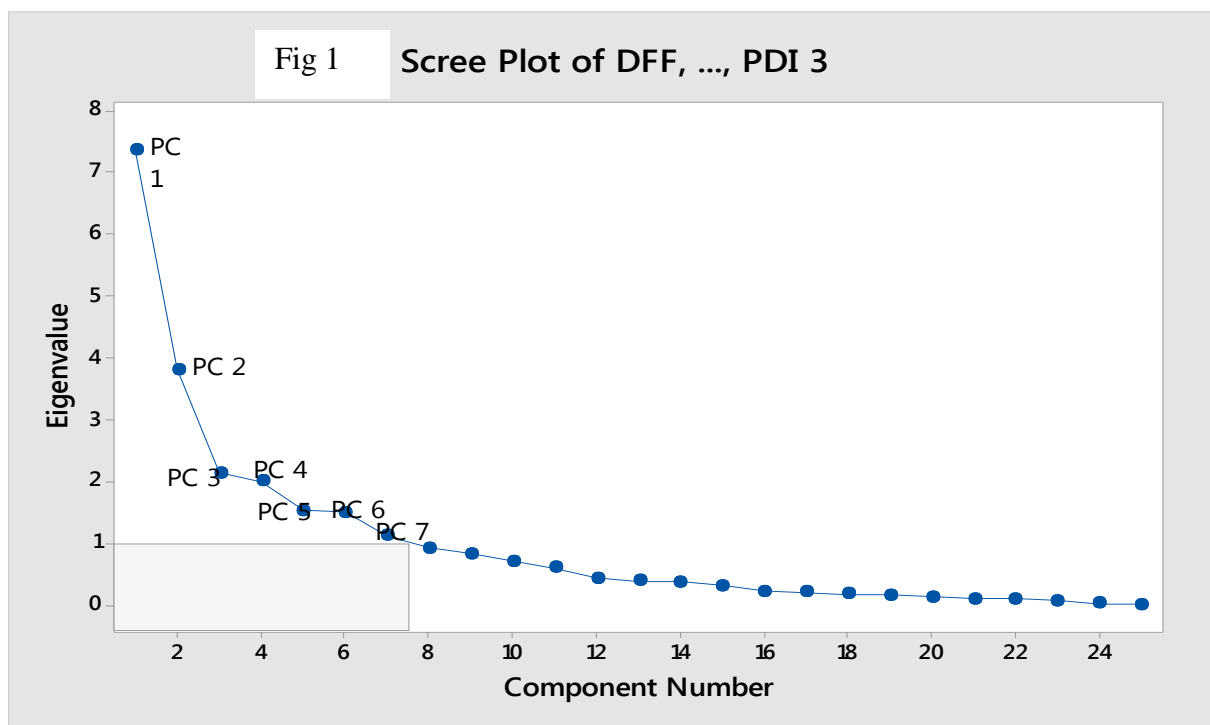


Fig 1. The scree plot displays the principal components and their corresponding eigen values. The first seven principal components have eigen values more than one. Eigen values indicate the amount of variation. Those PCs which equals eigen value of one or more than one explains maximum variation and diversity in the germplasm. The first seven PCs contribute for maximum of 77.6 per cent of the total variation. The first PC explains 29.4 per cent of the variation in the germplasm. The second and third PCs summarize 15.2 and 8.5 per cent of the total variations respectively. The fourth and fifth PCs explain 8 and 6.1 per cent of the variation. The sixth and seventh PCs explain 5.9 and 4.4 per cent of the total variation.

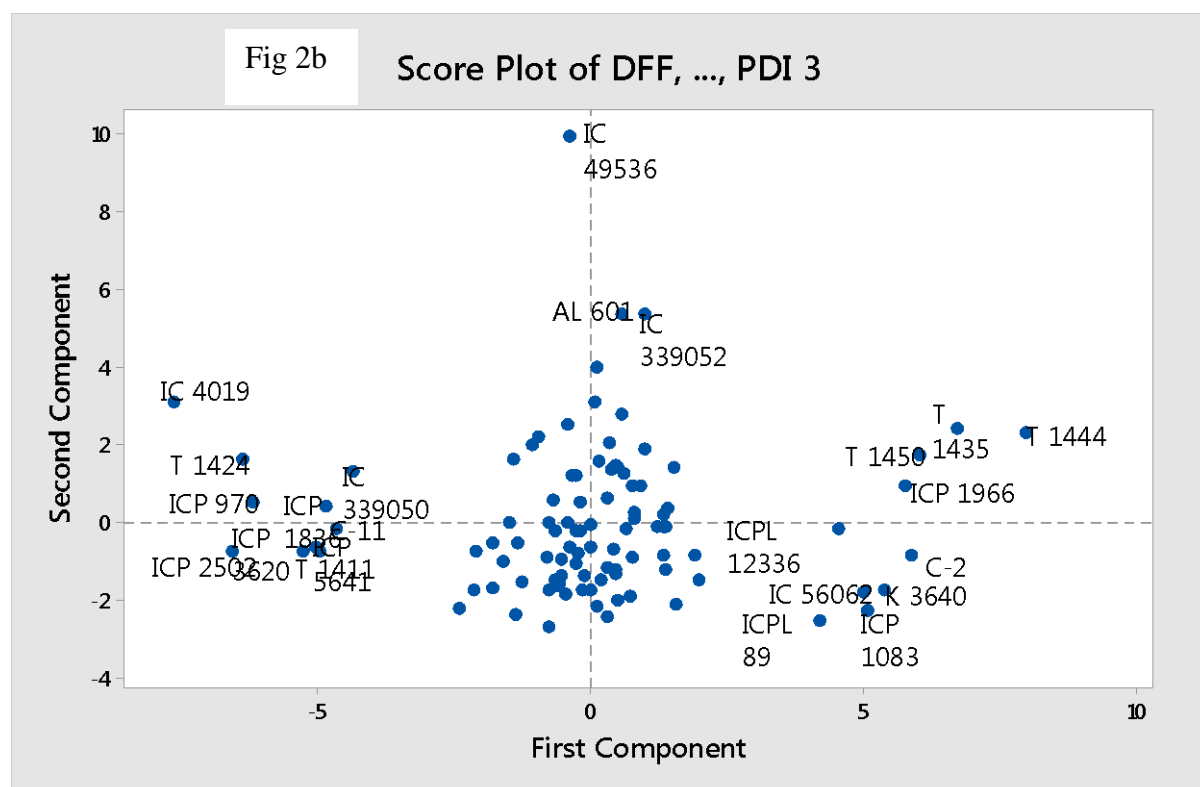
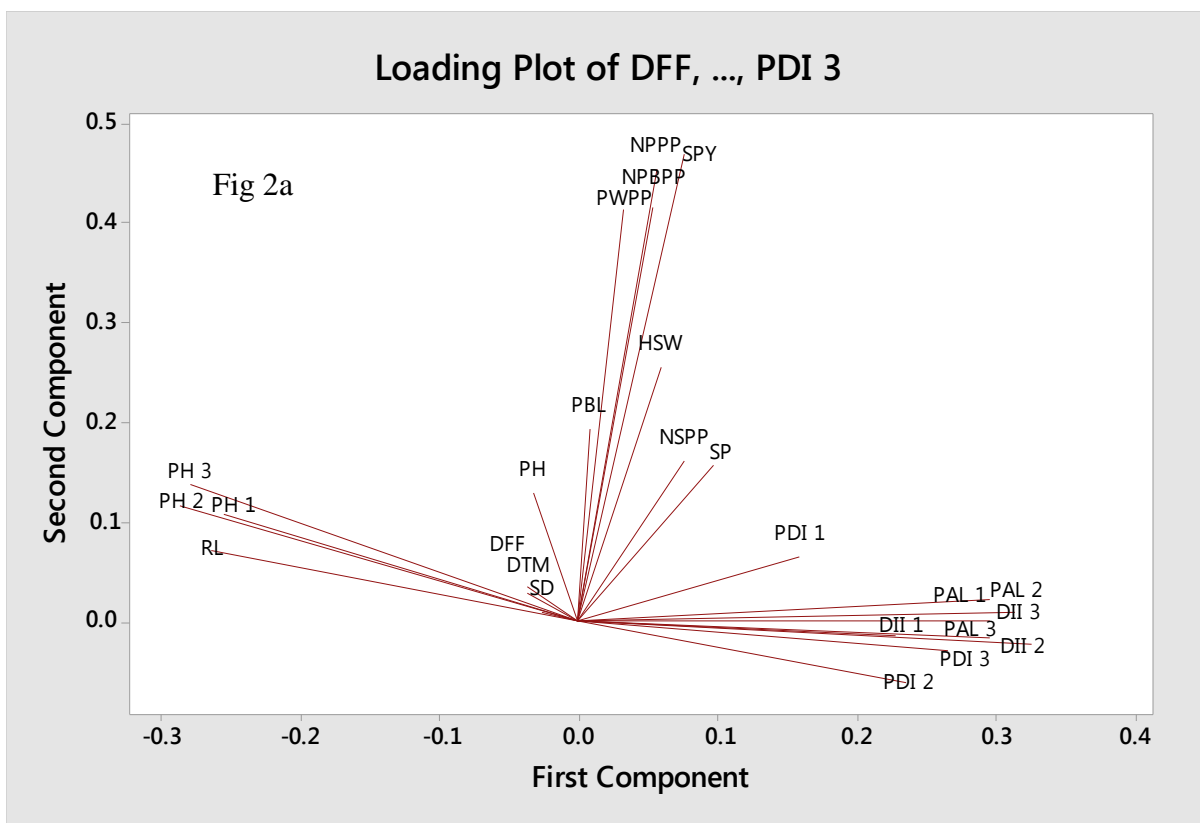


Fig 2a and Fig 2b. The score and loadings plot explain the individual genotypes in the first principal component against the individual genotypes second principal component based on their scores in these components. The genotypes around the point of origin show average performances for all the traits. The genotypes IC 4019, T 1424, ICP 970, IC 339050, IC 1836, ICP 5641, T 1411, ICP 2502, ICP 3620, C-11 in the left corner have low values for decreased root length and plant height whereas the genotypes in the right side have high values for decreased root length and reduced plant height. The genotypes above the average viz., IC 49536, AL 601, IC 339052 show high performance for yield contributing traits. The genotypes IC 4019 and T 1444 yields good with the former being the least affected and the latter being the most affected by the wilt disease respectively among all the hundred genotypes.

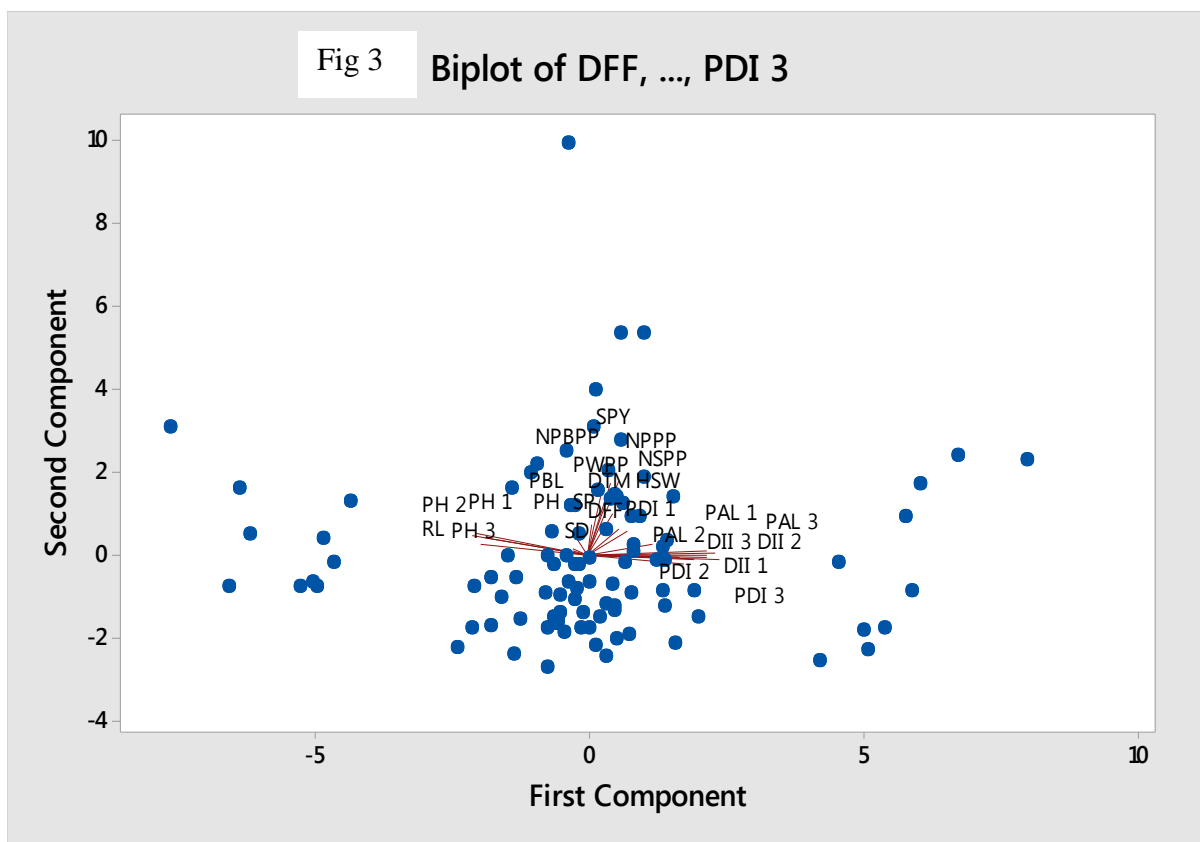


Fig 3. PCA biplot explains the traits and the corresponding genotypes which have high values for the corresponding traits. The characters Percentage of affected leaves, disease intensity index, per cent disease incidence are explained by PC 1. The genotypes T 1444, T 1435, C-2, K 3640, ICPL 89, T 1450, ICPL 12336, IC 56062, ICP 1083, ICP 1966 have high values for these traits (Fig 2b). The traits seedling height and root length are represented by PC 2. The genotypes ICP 4019, T 1424, ICP 970, ICP 2502, ICP 3620, T 1411, ICP 5641, C-11, IC 339050, ICP 1836 has maximum seedling height and root lengths (Fig 2b). The traits represented in PC 1 and PC 2 are in opposite directions therefore, the genotypes which has high root lengths and seedling height have lesser percentage of affected leaves, lower disease intensity index and lesser per cent disease incidence. The genotypes IC 49536, AL 601, IC 339052, IC 52937, C-6, T 1444, T 1447, T 1432, T 1435, T 1450 have high values for yield and yield contributing traits (Fig 2b).

