



Multivariate analysis of maize genotypes grown under waterlogging stress

*G. KUMAWAT, J. P. SHAHI, 'K. CHANDRA, M. K. CHOUDHARY,
A. SINGAMSETTI AND G. K. KOLI

Department of Genetics and Plant Breeding, Institute of Agricultural Sciences, Banaras Hindu University,
Varanasi-221005, India.

¹Department of Plant Breeding and Genetics, Sri Karan Narendra Agriculture University, Jobner-303329, India.

Received : 30.03.2021 ; Revised : 06.06.2021 ; Accepted : 09.06.2021

DOI: <https://doi.org/10.22271/09746315.2021.v17.i2.1462>

ABSTRACT

The field experiment was conducted to identify the principal component among fifteen morphological traits of CIMMYT maize hybrids under excess moisture condition. Further, the study also aimed to provide an insight into future studies to select diverse genotypes of maize which could be utilized as parents in maize hybridization program. The study was conducted with 53 CIMMYT maize (*Zea mays* L.) genotypes along with two checks during kharif season, 2017 in an alpha lattice design at experimental field of Institute of Agricultural Sciences, Banaras Hindu University, Varanasi, Uttar Pradesh, India. Excess soil moisture stress was maintained at the knee-high stage (V_6-V_7) of maize and on seventh day draining out of excess water. The data of fifteen morphological traits were recorded and analyzed to find out principal component to reveal the diversity among the genotypes under excess soil moisture stress. It was noted that the first five principal components exhibited more than one eigen values and accounted for 71.97 per cent of the total variation that comprised of 30.97 (PC 1), 14.58 (PC 2), 11.72 (PC 3), 8.09 (PC 4) and 6.61 (PC 5). The genotypes ZH17496 along PCA I axis and ZH17504 along PCA II axis identified on extreme positive side on both the axis were considered to be the best genotypes. These genotypes might be exploited in future hybridization programs. PC1 contributed the maximum towards the total variability (30.97). The characters viz., surface roots, brace roots, days to anthesis and plant population explained the maximum variance in PC1. This study helps in characterizing, reducing redundancy in the data set and assessing diversity among genotypes studied under waterlogging conditions.

Keywords: Diversity, eigen value, principal component analysis, waterlogging stress, *Zea mays* L.

Maize (*Zea mays* L.) is ranked third, among cereals in India, next to rice and wheat (Duvick, 2005; Reddy *et al.*, 2011). Maize fulfils food requirement of human, poultry, animal and is used for manufacturing of alcohol and non-alcohol product, making produce like fuel (Bekriae and Radosavljevic, 2008). The evaluation of characters of a crop in the field experiments is based on the biotic and abiotic stress factors (Ali *et al.* 2011), soil heterogeneity (Laserna *et al.*, 2012) and the variation in the research material (Khorasani *et al.*, 2011). The waterlogging stress or excess soil moisture condition has emerged as a common problem globally, which leads to poor maize harvest (Srivastava *et al.*, 2007; Zaidi *et al.*, 2007; Zubairi *et al.*, 2012; Ren *et al.*, 2014; Lone *et al.*, 2018; Zaidi *et al.*, 2010). In Asia, 18% acreage of the total maize cultivated area gives low yield due to waterlogging problem (Zaidi *et al.* 2007).

Bringing crop improvement in existing released varieties is a sustained selection program in plant breeding (Glenn *et al.*, 2017; Chand *et al.*, 2020); the plant researchers have to select diverse originator with intense variation for accumulating desirable target traits (Chandra *et al.*, 2019). Therefore, sound knowledge on genetic diversity is necessary to exert any recombination breeding process (Govindaraj *et al.*, 2015).

The importance of multivariate analysis for the study of quantitative traits (Sanwal *et al.*, 2015) present in biological experimental populations has been shown in various research areas (Renaud *et al.*, 2015). Multivariate is commonly used to study genetic diversity (Tabachnick *et al.*, 2007). Among the multivariate techniques, principal component analysis (PCA) had been showed very significant in choosing the genotypes for selecting a breeding program (Mohammadi and Prasanna, 2003). The PCA may be used to show counterpart and remove redundancy in research data (Adams, 1977) as morpho-physiological variations showed a high level of correlations (Maji and Shaibu, 2012).

PCA has been applied continuously for analysis of genetic diversity in many crops such as maize (Gana *et al.*, 2013) and millets (Ulaganathan and Nirmalakumari, 2015). The current study was planned to select out the traits that sort the genotypes into different groups and suggest potential genotypes that could be used as parents in the maize improvement program.

MATERIALS AND METHODS

Fifty-three maize genotypes along with two commercial checks (900MG and P3502) (Table 3)

procured from International Maize and Wheat Improvement Center (CIMMYT)-Mexico, regional station, Hyderabad, Asia. The experimental genotypes acquired from CIMMYT maize germplasm majority was collected from Asian tropics under the project entitled “Climate Resilient Maize for Asia (CRMA)”. The genotypes were cultivated in two replications in an alpha lattice incomplete block design (0,1) (Patterson and Williams, 1976), each entry was repeated in incomplete blocks either once or zero times during main season of 2017 at experimental field of Institute of Agricultural Sciences, Banaras Hindu University, Varanasi, Uttar Pradesh, India. Each maize genotype was sown in two rows of 4m length with a recommended 60×20 cm spacing between rows and plants, respectively. Excess soil moisture stress was maintained at the V₆-V₇ growth stage or knee-high stage of maize life span (mostly 35 days after sowing) and managed for continuous seven days at approximately 2–3 inch depth. Superfluous water was removed on the seventh day of waterlogging condition (Zaidi *et al.*, 2016; Zubairi *et al.*, 2012; Kumawat *et al.*, 2020a). Other standard agronomic package of practices and recommended fertilizer doses for maize were followed. Data were recorded from fifteen traits *viz.* pre-harvest traits like-number of nodes bearing brace roots (BR), number of surface roots (SR), days to 50 per cent anthesis (DTA), days to 50 per cent silking (DTS), plant height (PH, cm), ear height (EH, cm) and post-harvest data like-ears per plot (EPP), field weight (FW, t/h), plant population (PP), ear length (EL,cm), ear diameter (ED,cm), number of kernel rows per ear (NRE), number of kernels per row (NKR), 100 seed weight (HSW;g) and yield per plot (YPP;g). Single plant observations

were collected on ten plants chosen at random per genotype to each replication and their mean value was used as input for the PCA analysis and observations on DTA, DTS, EPP, FW, PP, HSW were recorded on plot basis. The scatter plot, heat map, correlation plot were performed using the R package version 4.0.2 and PC analysis by STAR (IRRI, 2014), XLSTAT (Addinsoft, 2020).

RESULTS AND DISCUSSION

The PCA has been exploited in plant breeding selection program for the reduction of variables and clustering of genotypes. The Principal Component Scores (PCs) were exploited for grouping 53 maize genotypes along with two checks into subgroups because few PCs contained major part of the information of the original variables (Syafii *et al.*, 2015; Dudhe *et al.*, 2018). Analysis revealed that first five PCs had contributed 71.97% of the total variance (Table 1). The PCs having the eigen values less than one were considered as non-significant and hence ignored as they are unlikely to have any practical significance (Mustafa *et al.*, 2015). The PC1 was contributed for major portion of the variability in the set of all PCs and the remaining PCs tend to lesser variation. However, the PC6 accounted for 6.24% of the total variation. It indicated that the first 6 traits (BR, SR, DTA, DTS, PH and EH) contributed significantly to the observed variation and these traits were depicting the most of the variability (Kumawat *et al.*, 2020b). This also shows that this type of recorded variation for these traits can be exploited for further crop improvement of genotypes for waterlogging tolerance (Daudo and Olakojo, 2007).

The factor loadings of different variables that were obtained by using PCA are presented in Table 2. The

Table 1: Eigen values and percent variation accounted for the 15 PCs

Statistics	Eigenvalues (Root)	Expression of proportion of variance	Expression of cumulative proportion
PC1	4.6458	30.97%	30.97%
PC2	2.1864	14.58%	45.55%
PC3	1.758	11.72%	57.27%
PC4	1.2134	8.09%	65.36%
PC5	0.9914	6.61%	71.97%
PC6	0.9368	6.24%	78.21%
PC7	0.82	5.47%	83.68%
PC8	0.6961	4.64%	88.32%
PC9	0.4768	3.18%	91.50%
PC10	0.3812	2.54%	94.04%
PC11	0.331	2.21%	96.25%
PC12	0.2277	1.52%	97.76%
PC13	0.1658	1.11%	98.87%
PC14	0.1064	0.71%	99.58%
PC15	0.0633	0.42%	100.00%

traits like FW, PH and YPP signaled considerable positive factor loadings on PC1 while SR and BR decoded negative loadings. The PC2 was related to diversity among genotypes due to EPP and EH with their positive loadings and NKR and DTA with negative loadings. The 3rd PC was explained by variation due to EL and YPP with their positive loadings and negative loadings exhibited by DTS and BR. The PC4 was elucidated by diversity for water tolerance among the maize genotypes for BR and HSW with negative loadings and SR and DTS had a positive value. By using these five PC's, it was recorded that these PCs controlled the total variation for all yield dependent traits (Mustafa *et al.*, 2015).

The PCA scores for 53 maize genotypes with two checks in the first three PCs were analyzed and

considered three axes as X, Y and Z; and squared distance of each genotype from these three axes were computed (Table 3). The genotypes identified on extreme positive side on both the axis were considered to be the best genotypes *i.e.* genotypes (10) ZH17496 (4.45), (26) ZH138269 (3.099), (25) ZH17499 (2.71), (43) ZH17506 (2.61) along PCA I axis and genotypes (39) ZH17504 (2.60), (7) ZH17494 (2.55), (9) ZH138260 (2.32), (11) ZH15553 (2.02) along PCA II axis. These genotypes might be exploited in future hybridization programs.

Fig. 1 depicted the distribution of each variable on the diagonal, the bivariate scatters plots with a fitted line below the diagonal and correlation value above the diagonal. About all the characters of maize genotypes were showing normal distribution. The X-axis shows

Table 2: Factor loading of fifteen traits with respect to the significant PCs

Statistics	PC1	PC2	PC3	PC4	PC5
SR	-0.0262	0.0471	0.0838	0.6241	0.5792
BR	-0.0282	-0.1022	-0.3962	-0.3843	-0.0108
DTA	0.0963	-0.4642	-0.3226	0.1374	-0.103
DTS	0.048	-0.3034	-0.5012	0.3047	-0.1873
PH	0.3873	0.1304	-0.071	0.1499	-0.0523
EH	0.355	0.2535	-0.1673	0.1249	-0.1517
PP	0.1931	0.1165	-0.3662	-0.0322	0.4495
EPP	0.2953	0.3341	-0.2356	-0.1178	-0.0199
FW	0.4112	0.0277	-0.0929	-0.1421	0.119
EL	0.2692	-0.3527	0.2958	-0.209	0.1253
ED	0.2056	0.1551	0.2002	0.2167	-0.5507
NRE	0.2996	0.0983	0.1983	0.2051	-0.0569
NKR	0.2701	-0.4496	0.1948	-0.0733	0.0454
YPP	0.3675	-0.2129	0.2039	-0.0817	0.135
HSW	0.0691	0.2588	0.0548	-0.365	0.184

the column variable and Y-axis shows the row variable in each scatter plot. The correlation values in scatter plots were also supported by correlograms for the traits under investigation (Fig. 2). The large and intense blue circle shows a positively highly significant correlation for the traits, while the small and faint blue circle indicates a non-significant association. The large and intense orange circle shows a significant negative association, whereas the small and faint circles indicate a non-significant association. The YPP shows a high positive significant correlation with FW, EL, NKR, PH and NRE (Begum *et al.*, 2016). The heat map or double dendrogram (Fig. 3) was constructed to support the PCs and factor analysis results (Zhao *et al.*, 2014). Genetic diversity was estimated from 15 traits data by cluster analysis which is based on Euclidean distance and

ward's method and shown as double dendrogram. The rectangular area of the same color in a heat map suggests a group of maize genotypes associated with a corresponding group of traits. Double dendrogram clustered the 53 genotypes with two checks into three main groups with diverged genetic profiles (Aci *et al.*, 2018). The heat map clustered PH in a single cluster while all other morphological traits in other clusters. Similarly, the heat map also supported the finding of PC and factor analysis by clustering the most vigorous and potential genotypes in a single cluster for a morphological trait.

The present investigation of PCA analysis revealed that the genotypes; ZH17496, ZH138269, ZH17499 and ZH17506, ZH17504, ZH17494, ZH138260 and ZH15553 exhibited the maximum divergence and also

Table 3: PCA scores of 53 genotypes with two checks of maize (*Zea mays* L.)

S. No.	Genotype	PC1(X Vector)	PC2(Y Vector)	PC3(Z Vector)
1	ZH15546	-1.79595	2.01516	0.711512
2	ZH15547	0.353916	1.85026	-1.82525
3	ZH15548	-1.23557	1.330219	-1.59822
4	ZH15549	2.069527	1.749835	-2.15432
5	ZH15550	-0.36486	-1.2236	0.309239
6	ZH15551	-2.74765	1.1323	1.090348
7	ZH17494	-0.38731	2.556878	0.924747
8	ZH17495	1.324578	0.192323	-1.77163
9	ZH138260	-0.1118	2.326348	0.688273
10	ZH17496	4.45512	0.434193	0.775
11	ZH15553	2.089438	2.026643	1.26454
12	ZH15554	0.104237	-1.04946	1.114874
13	ZH15555	-0.54343	0.665859	1.262359
14	ZH15556	-0.82034	-0.8545	3.330519
15	ZH15557	-1.95179	-1.0955	-1.6526
16	ZH17497	-3.45617	-1.75337	0.312769
17	ZH17228	1.63866	-0.54926	1.481145
18	ZH138267	0.851733	-1.54536	1.359665
19	ZH17229	2.574351	-1.95111	0.841294
20	ZH15558	-1.23634	1.14478	-0.1825
21	ZH15559	-0.67708	1.436656	1.242451
22	ZH15560	1.362252	-0.28945	0.684456
23	ZH15561	0.79085	-0.5669	0.379617
24	ZH17498	1.02541	1.756295	1.575072
25	ZH17499	2.716932	0.441458	0.629797
26	ZH138269	3.099365	1.645315	-0.38724
27	ZH15562	-0.86292	0.529532	-2.31194
28	ZH15563	-4.59891	0.024988	-0.0659
29	ZH15564	-6.97651	-0.69158	-0.66434
30	ZH17500	-1.2954	-1.02845	0.044385
31	ZH17501	1.424319	-0.14331	-0.79737
32	ZH138294	1.083219	-1.0394	0.036072
33	ZH17230	1.856965	-1.87307	1.193684
34	ZH15565	1.596645	-2.8811	-3.30043
35	ZH15566	-0.95027	1.31116	1.615646
36	ZH17502	0.266395	1.902756	0.012618
37	ZH17503	0.226641	0.008639	-1.91678
38	ZH138303	-2.41016	0.48201	-1.08943
39	ZH17504	-1.53722	2.605197	-0.95308
40	ZH15567	-1.82772	-1.23241	-2.31278
41	ZH17505	-4.23422	-0.39276	1.723602
42	ZH138305	1.027048	1.293149	-0.16091
43	ZH17506	2.61165	-2.41869	2.077873
44	ZH17507	-4.4876	-1.38821	0.5649
45	ZH17508	-0.23282	0.170325	-0.79971
46	ZH138312	0.628025	-0.45816	-0.19335
47	ZH17231	1.206059	-3.00666	-1.6992
48	ZH15568	1.110767	-1.88356	-0.18775
49	ZH138278	0.972918	-0.73433	0.719002
50	ZH17232	-1.22702	-0.51091	0.903723
51	ZH17509	2.505082	1.245162	-1.01302
52	ZH17510	0.710478	-2.31924	-0.79248
53	VH11128	1.078158	-1.49372	0.686337
54	900MG	1.690492	0.46507	-0.33067
55	P3502	1.51783	1.63154	-1.39461

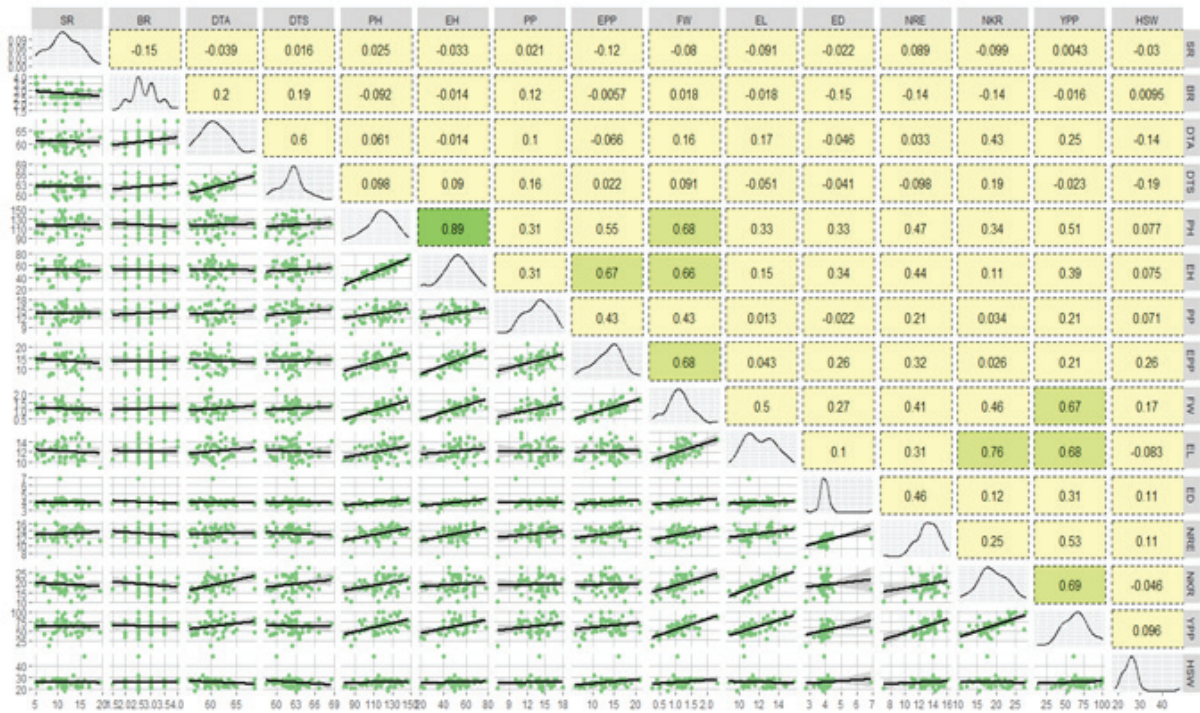


Fig. 1: Scatter plot with correlation value for 15 characters of 53 maize (*Zea mays* L.) genotypes with two checks

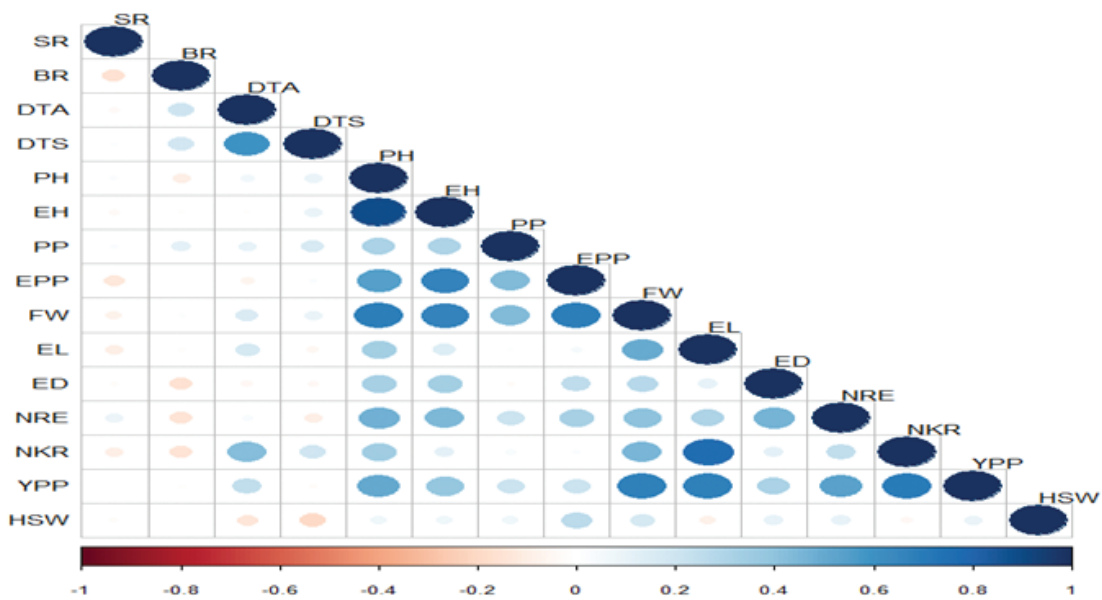


Fig. 2: Correlation plot of 15 morphological traits for 53 maize (*Zea mays* L.) genotypes with two checks

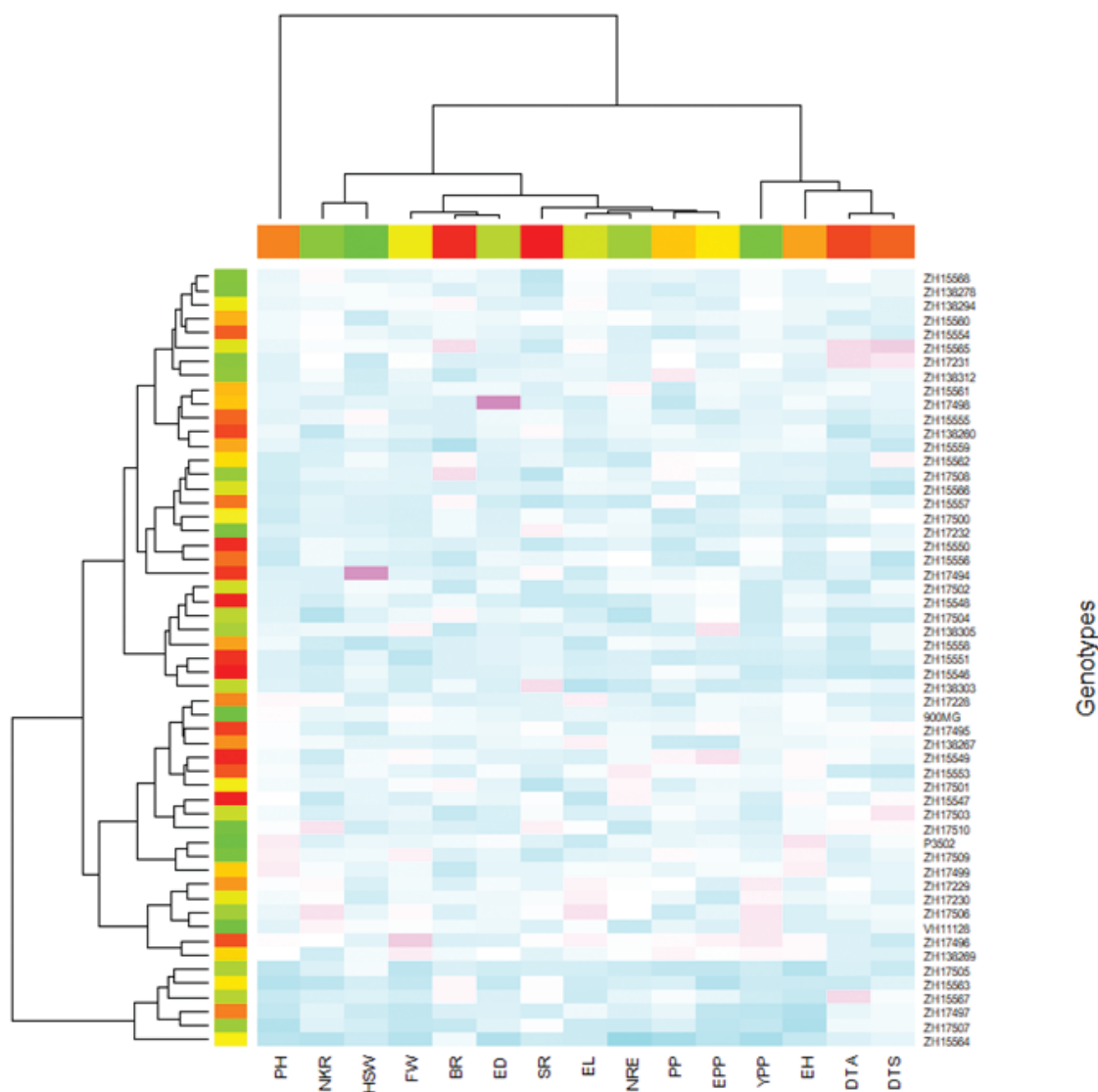


Fig. 3: Heat map cluster diagram of 15 traits and 53 genotypes with two checks of maize (*Zea mays* L.)

recorded better per se performance for the number of SR, BR, DTA and PP. So, these genotypes can be used to develop heterotic hybrids in future crop breeding programs to enhance yield through these traits under excess moisture conditions. The FW, EL, NKR, PH and NRE shows a highly positive significant correlation with YPP. Therefore, to obtain high yield per plant, one should consider these characters in maize breeding programme.

ACKNOWLEDGEMENTS

The authors are thankful to the CIMMYT-Asia, Hyderabad, for providing maize genotypes and the Department of Genetics and Plant Breeding, Institute of Agricultural Sciences, Banaras Hindu University, Varanasi.

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