



## EVALUATION OF MAIZE ACCESSIONS FOR DROUGHT TOLERANCE THROUGH PRINCIPAL COMPONENT ANALYSIS

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### SUMMARY

Drought is among the severe abiotic stresses that reduces crop yield. It greatly affects the growth and development at both vegetative and reproductive stages and the yield processes of crops. Maize is the third most important and widely distributed crop, suffering from drought stress, resulting in final kernel yield losses. The conduct of a screening experiment selected drought-sensitive and drought-tolerant maize accessions against water stress applied via calculating field capacity. This experiment used two treatments, T<sub>0</sub> and T<sub>1</sub> (T<sub>0</sub> with 100% field capacity and T<sub>1</sub> with 50% field capacity), in a completely randomized design (CRD) with a factorial arrangement and two replications at the wirehouse, Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan. Recording data on seedling traits ensued. Drought-tolerant and drought-sensitive accessions' selection resulted from principal component analysis, with a conclusion that the accessions H45C × H6B, H21 × H2B, H17 × H16, H23 × H21, H17 × H16A were drought tolerant, while H20 × H7C, H47A × H6C, and H9 × H21 were drought sensitive. This research will provide information in the future for comparing drought-sensitive and drought-tolerant accessions and help identify drought-tolerant maize accessions benefitting future breeding programs.

**Keywords:** Drought stress, screening, production, genetic variability, selection, principle component analysis, drought tolerant, drought-sensitive

**Key findings:** Among 50 accessions screened against drought stress, the accessions H45C × H6B, H21 × H2B, H17 × H16, H23 × H21, and H17 × H16A were drought tolerant, while H20 × H7C, H47A × H6C, and H9 × H21 were drought sensitive. These accessions will benefit future breeding programs for comparing drought-sensitive and drought-tolerant accessions to develop drought-tolerant maize accessions.

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### INTRODUCTION

Abiotic stresses, such as, insufficient or surplus water, low or high temperatures, heavy

metals, high salinity, and ultraviolet radiation, are antagonistic to plant growth and development, leading to enormous losses in crop yield worldwide. Among abiotic stresses,

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drought is a major natural disaster that can affect large areas. It severely affects livestock and crops, including essential commodities, such as, maize, soybeans, and wheat (Kumar *et al.*, 2015; Mazhar *et al.*, 2021). The rapid increase in population and, ultimately, high consumption growth rate will result in immense challenges to food security globally for at least another 40 years (Ma *et al.*, 2016). In 1947, Pakistan's per capita available water was 5000 m<sup>3</sup>, expected to decline to 1200 m<sup>3</sup> by 2025. Agriculture utilizes approximately 95% of Pakistan's total available water reserves. Although Pakistan has the largest glaciers in the world, it is among the world's 36 (Khan and Qayyum, 2015) countries facing severe water scarcity issues. Water scarcity can impart drastic economic losses to agriculture (FAOSTAT, 2021, Lozano *et al.*, 2019).

Maize emerged as the highest-yielding crop among cereals worldwide. It is an essential staple for countries like Pakistan, where the food supplies and available resources continuously decline due to a rapidly growing population. It is a crop of utmost importance globally due to its many uses; it provides food to humans and livestock and is a vital component for various industrial products (Hossain *et al.*, 2016). Globally, it ranks as the third most cultivated crop, occupying an area of more than 118 million ha, with an annual production of 1016.73 million MT (FAOSTAT, 2020). Nowadays, maize production faces serious threats due to severe climatic pressures.

Maize breeders and growers are facing a foremost challenge due to the rapid climate changes. Swift climate changes are causing an increase in desertification that leads to drought problems on the ground and atmospheric heat (FAO, 2021, IPCC, 2019). Among several factors that contribute to low yield, one frequently limits maize production is drought (Beyene *et al.*, 2017). It can occur at any stage of maize growth, but the most sensitive stages for water scarcity are the flowering and grain-filling periods (Meseka *et al.*, 2013). Water stress affects the maize crop, resulting in low plant density, poor germination, wilting, silk delay, stunted growth, top firing, tassel blast, and, eventually, reduced grain yield. Drought stress affects seedling growth, including leaf area, size, shoot length, and dry leaf weight (Pettigrew, 2004). Drought limits plant growth and development, making it difficult for the plant to complete its life cycle

(Chohan *et al.*, 2012). The problem of low productivity in maize caused by drought stress needs action with great care. It is urgent to develop drought-tolerant maize hybrids with a high-yield potential (Amin *et al.*, 2014).

Selection is highly significant in plant breeding and genetics. Selecting desirable accessions for a particular breeding program is a difficult task. Several tools like cluster analysis (Jain and Dubes, 1988), discriminate analysis (Johnson and Wichern, 2007), principal component analysis (Gabriel, 1980; Yan and Kang, 2011), and multivariate regression analysis (Hair *et al.*, 1998) have been discovered to deal with the selection of plants. This research using principal component analysis as a vital statistical method helped select the drought-tolerant and drought-sensitive accessions (Gabriel, 1980; Yan and Kang, 2011). This statistical tool helps reveal the hidden and simplified structures that often underlie a complex data set by lowering the dimensions of the said data set. The amount of variation in traits, which is crucial in plant breeding, is depicted by the eigenvalue of a particular principal component (Jolliffe, 2002).

Genetic variability refers to individual differences because of genetic makeup (Sumanth *et al.*, 2017). The presence of variability plays a key role in the success of different plant breeding programs, depending upon the genetic variability and selection skills of the plant breeder (Adhikari *et al.*, 2018). Selection becomes effective only if a significant amount of variability appears in the parent population. In the presented research, 50 maize accessions screened at the seedling stage against drought stress helped identify drought-tolerant maize accessions in the existing germplasm with principal component analysis.

## MATERIALS AND METHODS

The screening experiment at the seedling stage proceeded in the wirehouse area, Department of Plant Breeding and Genetics, University of Agriculture, Faisalabad, Pakistan. Fifty single-cross hybrids, selected for screening against drought, used two replications for each entry (three plants per replication) with two levels of irrigation (Table 1). Sowing of 50 single-cross hybrids ensued in polythene bags (23 cm × 8 cm) filled with sand and holed for aeration.

**Table 1.** The maize accessions used in the presented study.

No.	Accessions	No.	Accessions	No.	Accessions	No.	Accessions	No.	Accessions
1	H4 × H54	11	H38 × H36E	21	H53 × H8B	31	H33A × H3G	41	H2B × H3A
2	H30 × H15	12	H29 × H32	22	H44 × H14D	32	H9G × H3B	42	H21 × H6B
3	H22 × H4A	13	H36 × H14C	23	H51C × H14	33	H17 × H16A	43	H9 × H29
4	H29 × H26B	14	H3 × H8B	24	H26C × H10	34	H9 × H21	44	H19 × H3D
5	H18 × H26A	15	H8 × H23	25	H33D × H16	35	H20 × H6B	45	H20 × H7C
6	H25 × H28B	16	H2 × H1C	26	H45C × H6B	36	H48B × H9B	46	H6 × H3D
7	H24 × H16	17	H17 × H1D	27	H21N × 3B	37	H23 × H21	47	H14G × H7
8	H23 × H18F	18	H38 × H3G	28	H25B × H3B	38	H21 × H2B	48	H15 × H8G
9	H37 × H6B	19	H17D × H11	29	H14A × H9D	39	H23 × H2B	49	H17 × H4F
10	H38 × H98	20	H17 × H16	30	H25B × H3B	40	H47A × H6C	50	H31 × H6G

Each number indicates a specific hybrid, the name of each hybrid is given in Table 1 against each No. The name of the accession is written in the next column.

**Table 2.** Analysis of variance for seedling traits in maize.

Source of variation	Accessions (A)	Treatment (T)	A × T	Error
Degree of freedom	49	1	49	100
Leaf temperature	7.05*	19.04*	16.37*	0.4372
Fresh shoot length	42.21*	4175.70*	57.67*	1.4
Fresh root length	80.45*	4716.63*	59.29*	5.77
Fresh shoot-to-root length ratio	0.36*	1.17*	0.34*	0.018
Fresh shoot weight	0.50*	120.04*	0.65*	0.022
Dry shoot weight	0.05*	9.41*	0.06*	0.0008
The fresh shoot-to-dry shoot weight ratio	1.55*	0.004	3.799*	0.105
Fresh root weight	0.25*	74.88*	0.23*	0.04
Dry root weight	0.02*	7.78*	0.03*	0.009
Fresh root to dry root weight ratio	1.83*	101.33*	2.00*	1.2
Root density	0.59*	197.30*	0.65*	0.35

\*= Significant at 0.05 probability level

Field capacity measurement optimized levels of irrigation (Brady and Weil, 2016). The research used two treatments ( $T_0$  and  $T_1$ ) to evaluate the maize accessions at the seedling stage.

The irrigation in  $T_0$  comprised 100% of the field capacity, while  $T_1$  gained only 50% field capacity. Applying the same amount of irrigation to single crosses under both treatments transpired at the time of sowing. After 10 days of sowing, plants under  $T_0$  gained irrigation with 100% field capacity, while plants under  $T_1$ , with 50% field capacity. Fresh root and shoot lengths recording in cm, followed after 20 days of sowing, as with fresh shoot and root weights. Calculating data for seedling dry weights proceeded after drying the seedling in an oven (Fisher Scientific, Model 655 F) at  $65\text{ }^\circ\text{C} \pm 5\text{ }^\circ\text{C}$  for 72 h. Recording the leaf temperature of seedlings employed a digital IR thermometer (Model RAYPRM 30). Attaining root density of already sharpened roots occurred after 20 days of sowing. The obtained ratios of the fresh shoot-to-root length and fresh root-to-dry root weight followed these formulas:

$$\text{Fresh shoot-to-root length ratio} = \frac{\text{Fresh shoot length}}{\text{Fresh root length}}$$

$$\text{Fresh root-to-dry root weight} = \frac{\text{Fresh root weight}}{\text{Dry root weight}}$$

### Statistical analysis

The variability in recorded data assessment used an analysis of variance (Steel *et al.*, 1997), with the selection of genotypes performed via principal component analysis (Yan and Kang, 2011).

## RESULTS AND DISCUSSION

### Genetic variability

The genetic variability of 50 single cross-maize hybrids, assessed in the screening experiment, obtained values of the mean sum of squares from the analysis of variance for all the studied traits (Table 2). Significant differences occurred for all the studied features among

accessions. Treatments showed highly substantial differences for all the characteristics, except fresh shoot weight to dry shoot weight ratio, which showed non-significant differences. Accession by treatment interactions was also amply meaningful for all the traits, except fresh root weight to dry root weight ratio, which showed non-significant differences.

### Effect of drought stress on various traits in maize accessions

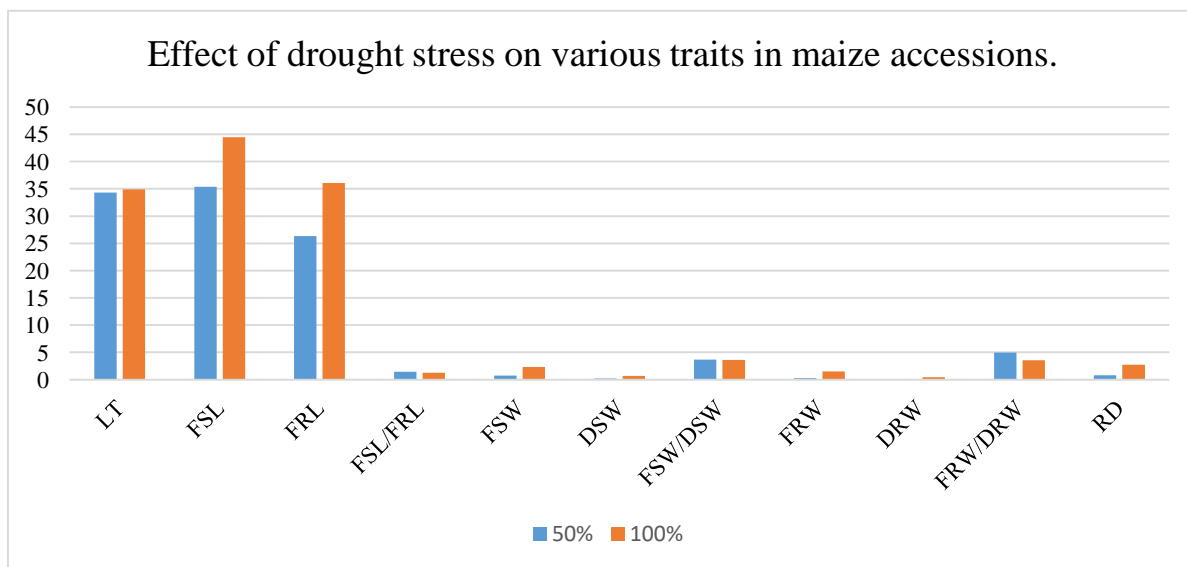
Drought affected the performance of almost all the studied traits, with leaf temperature recorded as slightly high in the accessions treated with normal irrigation. The effect of drought stress on various features in maize accessions appears in Figure 1. Fresh shoot and root length of seedlings showed high seedling length with 100% irrigation. The traits of the fresh and dry shoot and root weight provided with higher values with 100% irrigation. Ratios of the fresh shoot to fresh root length, fresh shoot to dry shoot weight, and fresh root to dry root weight were also higher under 100% irrigation.

### Principal component analysis

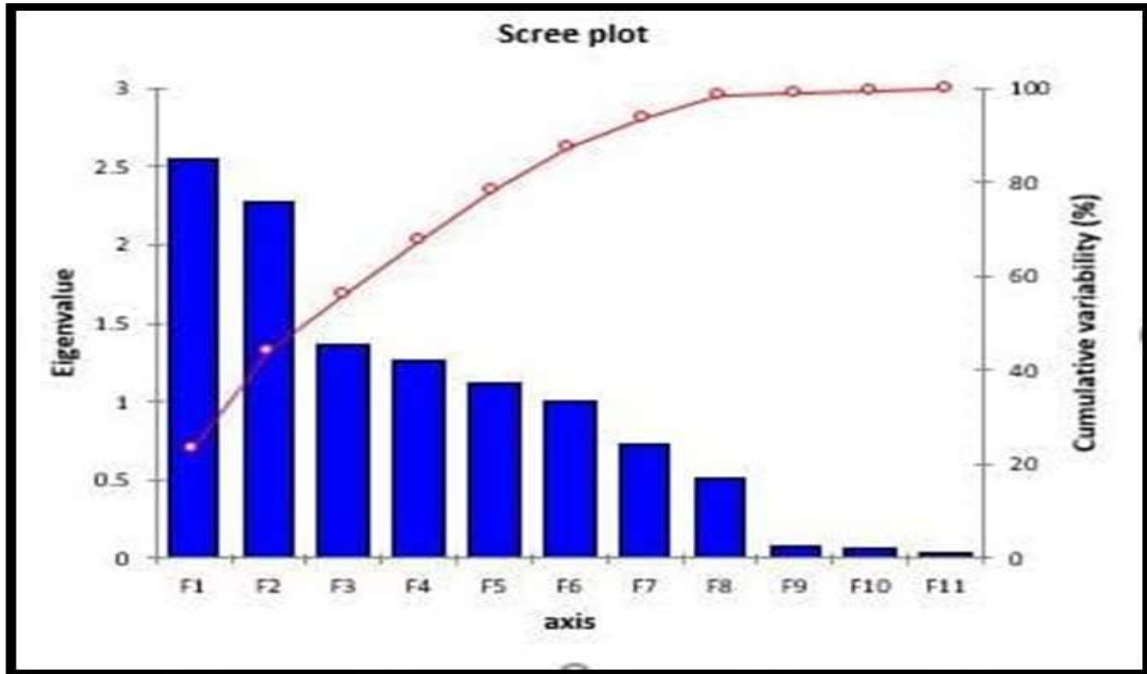
Principal component analysis (PCA) helps select genotypes when various accessions are available for selection and several traits to be studied. Under drought conditions, eigenvalues for all the studied characters were greater than

one, except for root density, fresh and dry root weight, fresh shoot weight to dry shoot weight ratio, and fresh root weight to dry root weight ratio. All the studied traits showed total variance higher than 4%, except root density, fresh root to dry root weight ratio, and dry root weight (Figure 2). Eigenvalue for all the studied traits reveals higher than one, except root density, fresh and dry root weight, dry shoot weight, fresh root weight to dry root weight ratio, and fresh and dry shoot weight. All the traits' total variance is higher than 5%, except for root density, fresh root weight to dry root weight ratio, and dry root weight (Figure 3). Biplot for PCA components, i.e., PCA-1 for drought and PCA-2 for normal irrigation, is shown in Figures 4 and 5, respectively. Principal component analysis has four quadrates. Details of the accessions present in each quadrate of PCA-1 and PCA-2 are in Table 3.

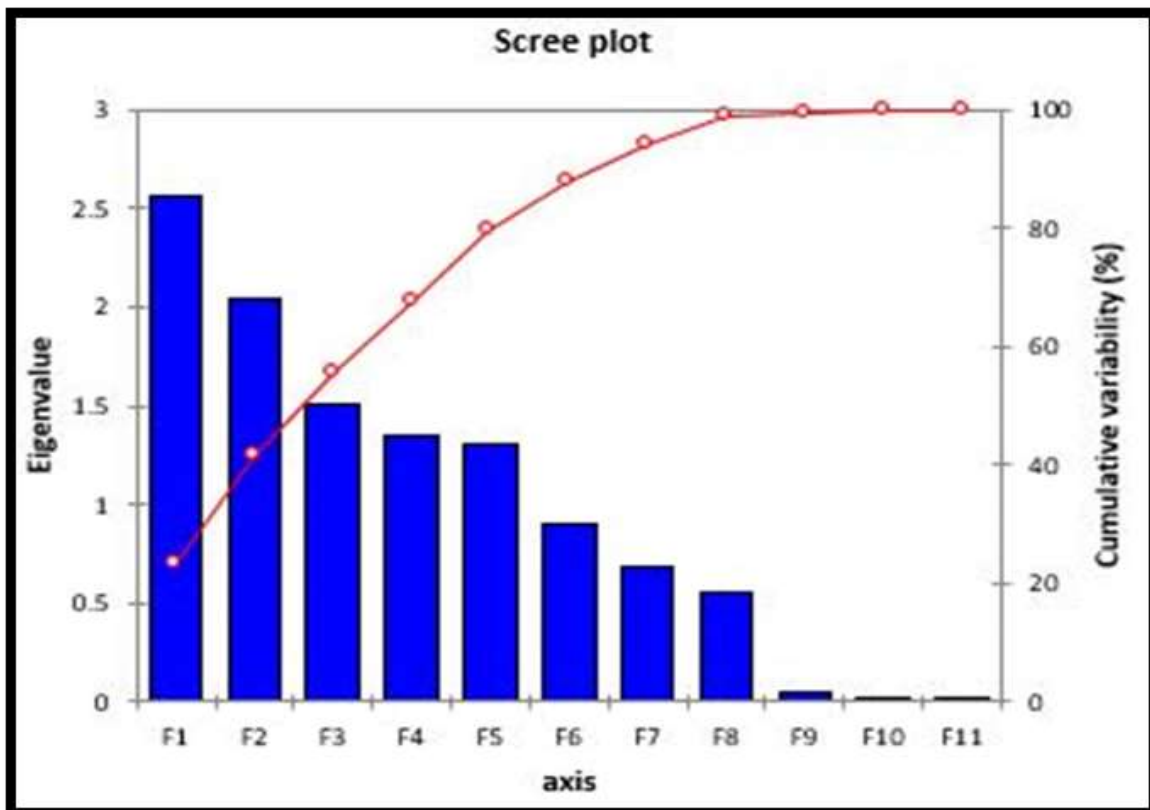
Based on the results of principal component analysis, accessions 26 (H45C × H6B), 38 (H21 × H2B), 20 (H17 × H16), 37 (H23 × H21), and 33 (H17 × H16A) emerged as drought-tolerant maize accessions, and 45 (H20 × H7C), 40 (H47A × H6C), and 34 (H9 × H21) as drought-sensitive maize accessions. The biplot showed that almost all the variables and genotypes gave a high degree of variation. Previous studies enunciated that various crop genotypes revealed greater variation for most yield-related traits (Ravi *et al.*, 2018; Worknesh *et al.*, 2019; Ahmed *et al.*, 2020).



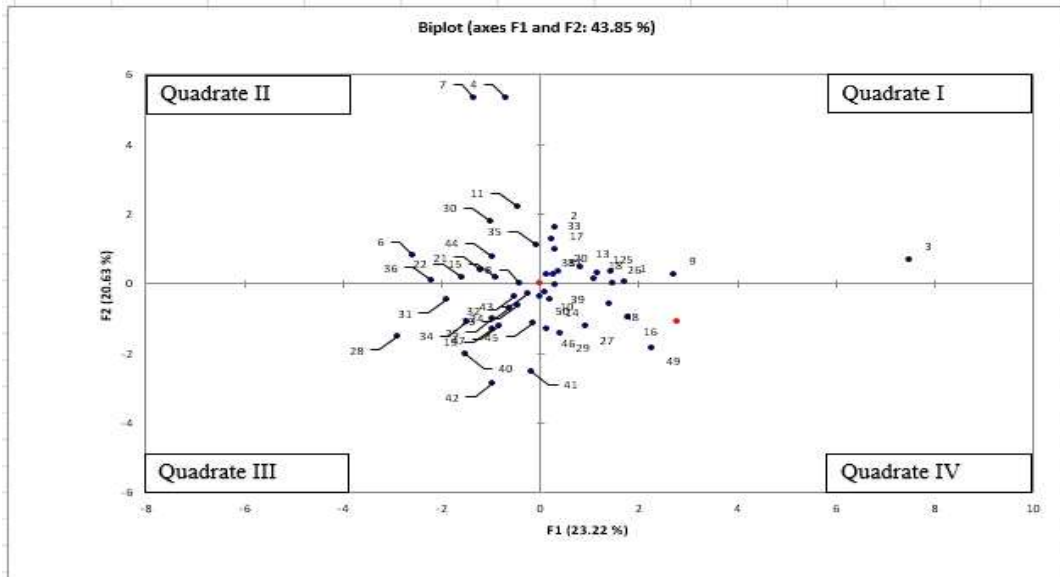
**Figure 1.** Effect of drought stress on various traits in maize accessions.



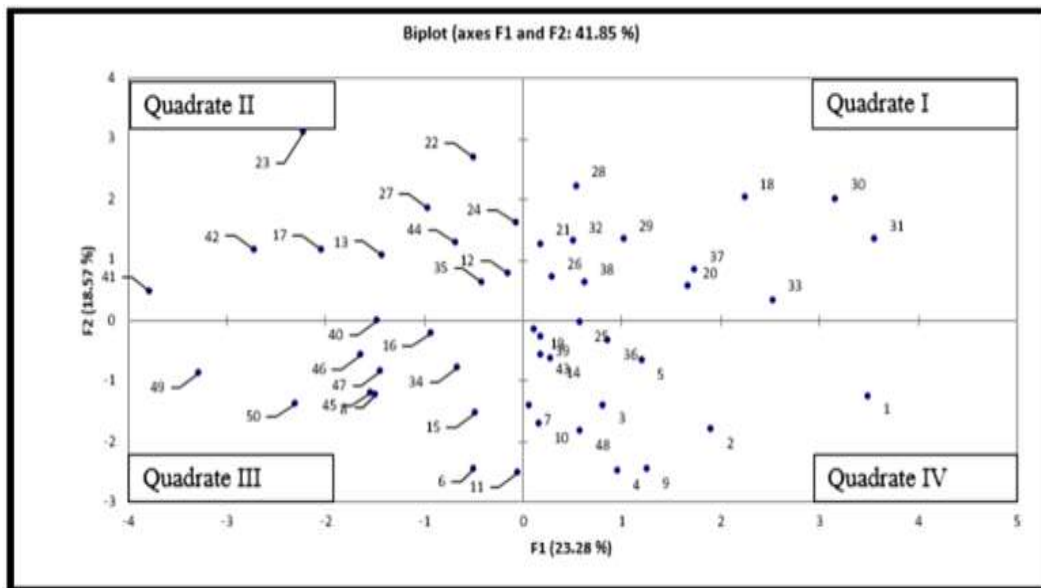
**Figure 2.** Scree plot for maize accessions under drought stress.



**Figure 3.** Scree plot of maize accessions under normal irrigation.



**Figure 4.** PCA-1 of maize accessions under drought stress.



**Figure 5.** PCA-2 of maize accessions under normal irrigation.

**Table 3.** Accessions found in four quadrates of PCA-1 and PCA-2.

Quadrates	PCA-1	PCA-2
Quadrate I	13, 5, 18, 1, 12, 33, 11, 2, 26, 9, 17, 26, 38, 37, 20, 39, 3*	28, 29, 18, 30, 31, 33, 27, 20, 32, 21, 26, 3*
Quadrate II	11, 30, 44, 6, 21, 15, 22, 35, 36*	22, 24, 23, 27, 44, 35, 12, 13, 17, 42, 41*
Quadrate III	23, 34, 31, 28, 25, 40, 32, 19, 24, 45, 47, 42*	16, 34, 15, 6, 46, 47, 45, 8, 50, 49*
Quadrate IV	43, 10, 14, 50, 48, 16, 49, 27, 29, 46, 45, 41*	25, 19, 39, 43, 14, 5, 3, 7, 10, 48, 11, 4, 9, 2, 1*

\*Each number indicates a specific hybrid; the name of each hybrid is given in Table 1 against each S.No. The name of the accession is written in the next column.

The research results showed that genetic variability occurred among all the studied accessions for all the studied traits. Treatments also showed significant differences for all the traits under study, except fresh shoot weight to dry shoot weight ratio. Accession  $\times$  treatment was also highly significant for all the studied traits, except the fresh root weight to fresh root weight ratio. Various studies in the past also observed broader genetic variability among seedling traits (Ali *et al.*, 2013; Badar *et al.*, 2020; Naseem *et al.*, 2020).

The results indicated that mean values for leaf temperature, fresh shoot length, fresh root length, fresh shoot to fresh root length ratio, fresh shoot weight, fresh root weight, fresh shoot weight to dry shoot weight ratio, fresh root weight, dry root weight, fresh root weight to dry root weight ratio, and root density ranged from 32.00 °C to 37.56 °C, 31.25 to 46.24 cm, 21.33 to 39.71 cm, 0.96 to 2.50, 0.72 to 2.28 g, 0.26 to 0.76 g, 5.67 to 4.62, 0.29 to 1.42 g, 0.13 to 0.59 g, 2.56 to 5.49, and 1.00 to 2.60 g ml<sup>-1</sup>, respectively. Earlier studies also revealed similar ranges for seedling traits in maize (Khan *et al.*, 2010; Radic *et al.*, 2018; Badar *et al.*, 2020). At the seedling stage, the root-to-shoot ratio can serve as an effective selection tool against drought stress.

A better way to study the drought-tolerant traits of maize accessions is to conduct effective screening for identifying drought-tolerant and drought-susceptible genotypes using easily measured and evaluated traits. A clear negative impact of drought stress treatments existed on germination and seedling performance of all maize accessions by reducing shoot and root traits. A drought-induced moisture deficit leads to impaired germination, resulting in poorly established plants and poor standing at the early seedling stage (Ghani *et al.*, 2015; Badar *et al.*, 2020). Maize plants absorb moisture and nutrients through seminal roots, but selecting a deeper root system is also critical for efficiency in nutrient acquisition (Cao and Wj, 2004). Besides root characters, drought stress affects all seedling traits, such as shoot length and fresh and dry weights of shoots and roots (Kaydan and Yagmur, 2008). A reduction in shoot length is due to decreased water absorption and a water deficit created by external osmosis (Anjum *et al.*, 2017). The development of well-developed root systems and the characteristics of early seedlings in cereals have contributed to improved stress tolerance. Most accessions showed significantly

reduced root length and fresh and dry mass under simulated drought (Chloupek *et al.*, 2010; Avramova *et al.*, 2016; Ahmed *et al.*, 2019).

Evaluation of drought tolerance in maize accessions has employed principal component analysis (Arisandy *et al.*, 2017). The principal component analysis partitioned the total variance into four principal components representing the highest diversity among genotypes. The PC analysis ultimately confirmed the degree of variation between the materials in hand, which could benefit the design of the breeding programs to improve drought tolerance and grain yield, since the theory holds that maximum variation leads to maximum heterotic effects.

## CONCLUSIONS

Under drought stress, mean values of studied traits are most suitable to develop selection criteria for drought-tolerant accessions. But the urgent need is to enhance the selection-efficiency, a major challenge in plant breeding programs. When the number of genotypes and studied traits is high, selecting genotypes is better using principal component analysis (Gabriel, 1981; Yan and Kang, 2011). The accessions H45C  $\times$  H6B, H21  $\times$  H2B, H17  $\times$  H16, H23  $\times$  H21, and H17  $\times$  H16A were drought tolerant, while H20  $\times$  H7C, H47A  $\times$  H6C, and H9  $\times$  H21 were drought sensitive.

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