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Research Article

Early Stage Evaluation of Root Traits Associated with Drought Tolerance in Maize (*Zea Mays* L.) Genotypes

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Abstract

Drought stress is one of the most important environmental issues that reduce growth, development and yield of the plants. Developing maize cultivars that can perform well in drought and other abiotic stress is an important goal throughout the world. The present study was planned to explore the variations among the maize genotypes and determine the performance of root traits under drought conditions. The research was established in a completely random design with five replications. The seedling characters viz., number of seminal roots, primary root length, number of lateral roots, root fresh mass, root dry mass and root tissue density were evaluated under four levels of polyethylene glycol (PEG) 6000 (0, -2, -4 and -6 bar). Significant varietal differences for all characters were found ($p \leq 0.05$). On the basis of mean values, the maize genotypes VIM147 and VIM396 showed best performance under the drought conditions. Principal component analysis was also used to assess the contribution of root traits which were attributing maximum variations among maize genotypes. Root dry mass was recognized as the best indicator and easiest typical to determine the drought tolerance of maize.

Keywords: Maize, Polyethyleneglycol-6000 (PEG-6000), Drought stress, roll towel, Root

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Introduction

Root architecture is strongly linked to plant survival under abiotic and biotic stress conditions. The development of a healthy root system is an important part of the overall plant development programme. Three main functions are commonly attributed to root systems, i.e., absorption of water and nutrients, as well as plant anchorage [1]. Maize is the third most important cereal crop after wheat and rice. It is necessary for global food security [2]. But abiotic stresses including water stress, salinity and temperature in which drought and salinity are major limiting factors for crop yield, badly affect the plant growth and ultimately yield [3]. A maize root consists of two main systems, one system is formed during embryogenesis and developed during germination, whereas the second root system structure is formed during postembryonic development. The embryonic root system is the dominant system for about the first two weeks after germination. Following this early root development the postembryonic structure becomes dominant. In early stages of growth the embryonic roots play an important role in the absorption of nutrients and water by increasing the root's surface area [4]. The capacity of a root system to overcome challenging environmental conditions will determine plant performance. Complex root systems are characterized by a high number of branching points, having a higher probability of finding adequate resources by exploring a large portion of the soil face than root systems with less complex root systems [5].

Assessment of drought tolerance at seedling stage is necessary to predict a good crop stand at maturity [6]. Maize plants with more roots at seedling stage subsequently developed stronger root system, producing more green matter and high seed yield [7]. Significant genotypic differences in root growth and development under both normal as well as drought exist among various crop plants including maize [8] and therefore, could be used as selection criteria for improved drought tolerance in various crops. However, root growth in cultivars intrinsically capable of avoiding drought through enhanced water uptake. Nevertheless, reduction in root growth and development in response to drought has also been reported [9]. Therefore, it is necessary to identifying breeding lines for rooting traits under drought conditions. For the development of elite line having drought tolerance, the existence of variability in the available germplasm of maize is a key to success for the maize breeders. This current study was planned to explore the variation and to determine the root traits conferring drought tolerance in maize.

Materials and Methods

Description of Genetic Materials

The present research work was carried out during 2016 at the Department of Crop Physiology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu. Four maize genotypes (VIM455, VIM147, VIM213 and VIM396) were used to study the effect of drought stress by using PEG-6000 on root traits. The seed materials obtained from the Maize research station, Vagarai. The seeds should be selected in uniform size.

Experimentation

All seeds were surface sterilized with 1% (v/v) sodium hypochlorite or a commercial surfactant solution for 10 minutes followed by three washes with distilled and sterilized water. For each genotype, five seeds were placed in the upper third of a non-toxic germination paper with the embryo facing the bottom of the germination paper. The space between the seeds was adjusted to prevent contact between different root systems. Each germination paper was moisturized with a Captan® (BAYER) 2.5 g l^{-1} solution, and then rolled up vertically. Five rolled germination papers were placed vertically in a 2.5L plastic container with 750 ml of distilled water plus 20 ml of Captan solution. Water stress is induced by varying concentrations (0, -2, -4 and -6 bars) of PEG-6000. All experiments were conducted in a germination chamber without illumination at 28ºC and 100% relative humidity. According to the field experiment terminology, each germination paper was regarded as a single plot and each plastic container as an incomplete block.

Data Collection and statistical analysis

Recording of data regarding various seedling traits was started after 9 days after sowing, seedlings from each germination paper were carefully opened and divided at the cotyledonary node into their respective root and shoot portions. The data on the number of seminal roots, seminal root length (cm), primary root length (cm), number of lateral roots, shoot length (cm), root fresh mass (g), root dry mass (g) and Root tissue density (RTD) is ratio between root dry mass (RDM) and root fresh mass (RFM) were recorded. The data collected were subjected to analysis of variance technique [10] using Minitab7 and SPSS V.20 statistical software and numerical taxonomic techniques following the procedure of principal component analysis [11].

Results and Discussion

The results of this study reveal that different concentrations of PEG-6000 such as -2 bars, -4 bars and -6 bars along with the control had significant ($P \leq 0.01$) effect on the germination of maize genotypes (VIM455, VIM147, VIM213 and VIM396). Analysis of variance and mean comparison showed that there were significant differences between drought stress levels and genotypes (**Table 1**).

Table 1 Analysis of variance on mean of squares of measured traits in maize genotypes under drought stress

S.O.V	Df	SRL	PRL	SRN	LRN	SL	RFM	RDM	RTD
G	3	254.58	261.15	16.85	462.17	66.76	0.35	0.28	0.08
T	3	443.21	320.48	2.82	3369.22	1645.02	1.12	1.61	0.42
G×T	9	26.46	30.40	3.04	114.11	18.13	0.09	0.03	0.03
Error	48	0.07	0.10	0.01	0.07	0.08	0.01	0.01	0.01
C.V (%)		1.36	1.37	1.30	1.87	1.47	1.42	1.51	1.45

**=Non-significant, significant at 1% probability levels, G- Genotypes, T- PEG Levels

Seminal root length (cm)

Seminal root length eight days after germination (SRL) varied between 11.53 and 30.71 cm (Table 4). The mean for the maize genotypes VIM147 and VIM213 were 27.02 and 14.57 cm, respectively (**Table 2**). Seminal root length under drought (-6 bars) ranged between 22.01 and 14.44 cm was observed in the maize genotypes VIM147 and VIM213, respectively. So, we may select this trait as selection criteria for the evaluation of hybrids against drought. These results are similar to the results of Khan *et al*. [12].

Table 2 Mean comparison of main effects of drought stress levels

Drought stress	SRL	PRL	SRN	LRN	SL	RFM	RDM	RTD
Control	27.02	28.00	5.17	34.22	29.29	1.21	1.05	0.87
-2 bars	21.77	24.39	4.38	13.72	27.53	1.12	0.81	0.72
-4 bars	18.54	20.78	4.62	6.73	17.98	1.05	0.65	0.62
-6 bars	14.57	17.65	4.21	0.96	7.25	0.61	0.29	0.48

Number of seminal roots (SRN)

Under normal conditions (0 bars), highest number of seminal roots was shown by VIM147 followed by VIM455 (**Table 3**). Under drought condition (-4 bars), the best performance regarding number of seminal roots were showed by VIM147 (6.74), VIM455 (4.37), VIM396 (4.19) followed by VIM213 (3.18); while under drought condition (-6 bars) best performance regarding number of seminal roots were shown by VIM147 (6.99), VIM455 (3.44), VIM396 (3.41) followed by VIM213 (3.03) (**Table 4**) but the better performances under the normal conditions by genotypes (VIM213, VIM396) regarding this trait are less performances under drought conditions. These results are similar to the results of Khan *et al*. [13].

Table 3 Mean comparison of main effects of maize genotypes

Drought stress	SRL	PRL	SRN	LRN	SL	RFM	RDM	RTD
VIM455	21.48	21.43	4.45	9.93	21.26	0.92	0.68	0.71
VIM147	23.26	26.85	6.048	20.41	22.64	1.20	0.85	0.70
VIM213	14.59	17.66	3.63	9.08	17.79	0.86	0.53	0.56
VIM396	22.57	24.89	4.25	16.21	20.36	1.00	0.75	0.71

Table 4 Mean performances of maize genotypes under different levels of moisture stress for various plant traits

GENOTYPES	PEG LEVELS	SRL	PRL	SRN	LRN	SL	RFM	RDM	RTD
VIM455	0 bars	26.58	25.34	5.13	32.29	28.52	1.21	1.04	0.85
VIM 147	0 bars	30.71	30.31	5.25	36.22	32.54	1.31	1.19	0.90
VIM 213	0 bars	26.61	25.12	5.12	32.19	27.85	1.09	0.93	0.85
VIM 396	0 bars	28.11	27.32	5.21	36.19	28.25	1.23	1.06	0.86
VIM 455	-2 bars	22.91	22.23	4.87	5.27	27.88	1.15	0.79	0.69
VIM 147	-2 bars	29.11	23.31	5.22	28.92	28.21	1.21	0.96	0.79
VIM 213	-2 bars	18.11	15.91	3.22	2.62	27.54	0.97	0.65	0.67
VIM 396	-2 bars	27.51	25.69	4.22	18.13	26.55	1.17	0.87	0.74
VIM 455	-4 bars	19.07	20.46	4.37	1.17	19.41	1.01	0.68	0.67
VIM 147	-4 bars	25.62	20.23	6.74	15.22	20.63	1.18	0.66	0.56
VIM 213	-4 bars	11.53	12.75	3.18	1.01	11.46	0.87	0.46	0.53
VIM 396	-4 bars	26.92	20.72	4.19	9.52	20.43	1.14	0.83	0.73
VIM 455	-6 bars	17.19	17.91	3.44	1.01	9.24	0.34	0.22	0.65
VIM 147	-6 bars	22.01	19.24	6.99	1.32	9.24	1.13	0.61	0.54
VIM 213	-6 bars	14.44	4.61	3.03	0.51	4.33	0.52	0.11	0.21
VIM 396	-6 bars	17.02	16.55	3.41	1.01	6.23	0.47	0.25	0.53
Grand Mean		22.72	20.48	4.60	13.91	20.52	1.00	0.71	0.67

Primary root length (cm)

Primary root length eight days after germination (PRL) varied between 4.61 and 30.31 cm (Table 4). The mean for the maize genotypes VIM147 and VIM213 were 26.85 and 17.66 cm, respectively (Table 2). Primary root length under drought (-6 bars) ranged between 19.24 and 4.61 cm was observed in the maize genotypes VIM147 and VIM213, respectively. These results are similar to the results of Taiz and Zeiger [14]. Drought drastically affected all the root traits except primary root length which was affected very little under drought condition.

Number of lateral roots (LRN)

Under normal conditions (0 bars), highest number of seminal roots was shown by VIM147 followed by VIM455 (Table 3). Under drought condition (-4 bars), the best performance regarding number of lateral roots were showed by VIM147 (15.22), VIM455 (1.17), VIM396 (9.52) followed by VIM213 (1.01); while under drought condition (-6 bars) best performance regarding number of lateral roots were shown by VIM147 (1.32), VIM455 (1.01), VIM396 (1.01) followed by VIM213 (0.51) (Table 4). Therefore, lateral root phenotypes to optimize mobile resources should be long and dispersed along the axial roots. Greater lateral root branching increases the rate at which a soil domain is depleted of resources, especially for immobile resources like P [1].

Shoot length (cm)(SL)

The mean of shoot length for high level PEG (-6 bars) concentration is 7.25 cm, while the genotypes presented the best behavior in relation to this characteristic, with means between 22.64 cm (VIM147) and 17.79 cm (VIM213) (Tables 2 and 3). Water stress during the vegetative growth stage lowers shoot length in maize genotypes and consequently it will affects the yield. It thus appears that vigorous shoot growth corresponds to vigorous root growth under a wide range of environmental conditions (included drought) and that either variable can be used to select for seedling vigour [15].

Root fresh mass (g)

Under drought (-6 bars), there were significant differences in root fresh weight among genotypes (Table 4). VIM213 exhibited the lowest RFM (0.47 g), while the other genotype VIM147 (1.13 g) presented higher amounts, with noticeable statistical differences among them. In case of 0 bars and -2 bars of PEG, there was little significant difference was observed among the genotypes. Drought drastically declined RFM in winter wheat genotypes grown in greenhouse container culture for three weeks [16]. The trend demonstrated by VIM213 may be indicative of sensitivity to drought stress.

Root dry mass (g)

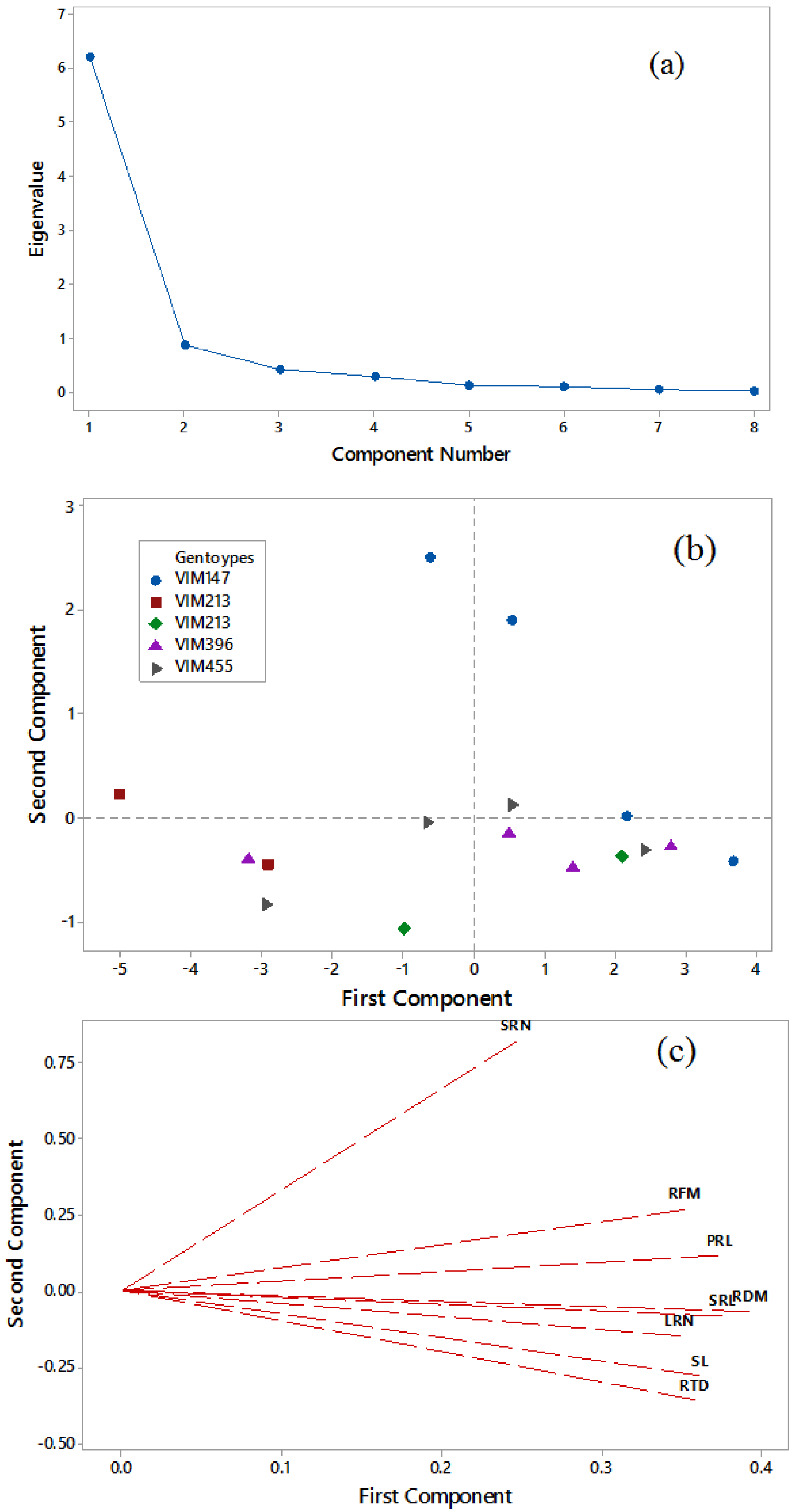
When screening the maize genotypes responses for root dry mass under drought stress, it was found that all genotypes, except for VIM147 and VIM396, performed well. Hughes *et al*., [17] reported that maize genotypes with low root dry weight are less tolerant to drought stress. As was the case with RDM and RFM, VIM213 again showed a low RDM (Table 4).

Root tissue density (RTD)

As to the RDM/RFM ratio (Table 2), one homogeneous group of genotypes included the following genotypes: VIM455 (0.71), VIM147 (0.70) and VIM399 (0.71). The other genotype presenting the lower value was VIM213 (0.56). Genotypic ability for high RDM/RFM ratio contributes to drought tolerance. It seems that maize crops are less tolerant to drought due to their high shoot dry weight and low root dry weight. Nour *et al*., [18] correlated high root dry mass of young plants with superior drought resistance in sorghum genotypes.

Principal component analysis

The genotypes were highly significant (Table 2) with respect to all the measured parameters [19] Drought drastically affected all the root traits except primary root length which was affected very little and number of lateral roots also were less affected under drought condition. The variation for roots traits was distributed among the PCs (**Figure 1a** and **1b**). Positive and highly significant correlation was observed between the pairs of traits that is, number of seminal roots with number of lateral roots and primary root length with number of lateral roots, fresh root weight, dry root weight and number of lateral roots with, fresh root weight and dry root weight, and fresh root weight with dry root weight at genotypic level (**Figure 1c** and **1d**). These findings suggest that positive performance of number of seminal roots along with roots mass is very important to tolerate the drought conditions. These results are similar to the results of Rashid *et al*. [20] who identified the major characters that is, days to 50% flowering, plant height (cm), productive tillers/plant, panicle length (cm), panicle fertility %, 1000-seed weight (g) and yield (kg/ha) accounting variation among Basmati rice mutants.


(a)
Eigenvalue
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Component Number
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Gentoypes
● VIM147
■ VIM213
◆ VIM213
▲ VIM396
▶ VIM455
Second Component
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2
1
0
-1
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-4
-3
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First Component
(c)
SRN
RFM
PRL
SRL
RDM
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Second Component
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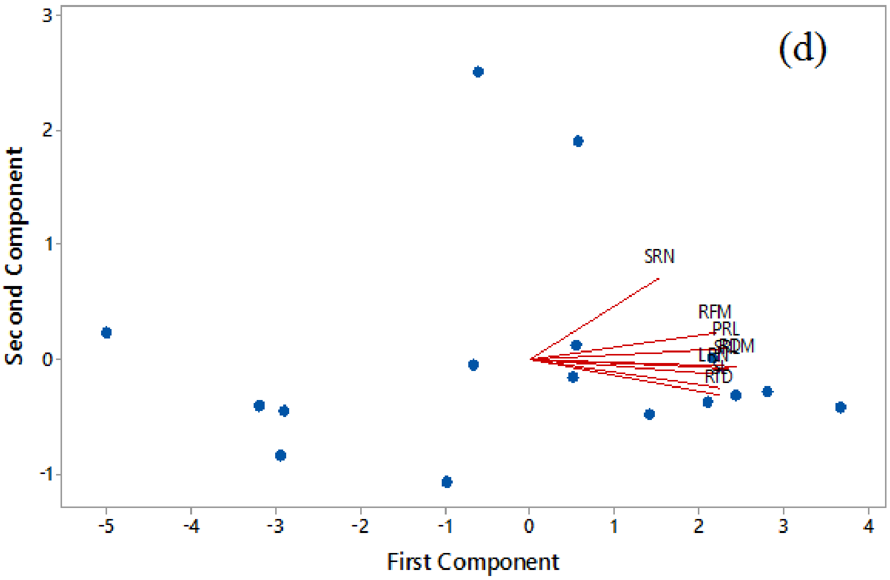

Figure 1 Principal component analysis for root traits under drought stress in maize genotypes. (a) Scree plot of root traits of maize genotypes under drought stress. (b) Score plot of root traits of maize genotypes under drought stress. (c) Loading plot of root traits of maize genotypes under drought stress. (d) Biplot of root traits of maize genotypes under drought stress

Conclusion

Accordingly to the PCA and mean values data, best performance under varying drought levels were observed in VIM455, VIM147, VIM213 and VIM396. Among these, the maize genotypes VIM147 and VIM396 performed well under drought stress. Our results further validate that screening is an effective tool to exploit genetic variation among maize hybrids. To develop high yielding drought tolerant maize genotypes through selection, these technique and genetic variations in root traits assessment can be used Root dry mass was recognized as the best indicator and easiest typical to determine the drought tolerance of maize. This benchmark can also be utilized for other agricultural crops to establish high yielding drought tolerant genotypes.

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