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Determinants of drought tolerance at seedling stage in early and extra-early maize hybrids

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Abstract

Adequate knowledge of response of genotypes to stress and identification of important adaptive traits under stress conditions facilitate rapid progress in a breeding program. The objectives of this study were to evaluate the response of early and extra-early maturing maize hybrids to drought stress imposed at seedling with a view to identifying tolerant hybrids for further breeding studies and identify adaptive traits that could be used as selection criteria for tolerant maize genotypes at this growth stage. This study was conducted in the screenhouse in the Faculty of Agriculture, Obafemi Awolowo University, Ile-Ife, Nigeria. Five seedlings each of 15 hybrids each from early- and extra-early-maturity groups were raised in pots and the pots were arranged in a randomized complete block design with three replicates. The experiment was adequately watered for the first seven days and thereafter watering stopped. The plants were observed for 42 days and data were recorded on emergence and other seedling traits. Data collected were subjected to analysis of variance, principal component analysis and correlation analysis. Results showed significant difference between maturity groups for root length and significant difference among genotypes within maturity were obtained for total number of leaf shed, moisture content of root, fresh shoot weight, total fresh biomass and seedling aspect, indicating there was wide variability in the response of the genotypes from the different maturity groups for tolerance to drought at seedling stage. Principal component analysis also identified these traits as the primary traits contributing to diversity among the hybrids under induced drought at seedling stage. Results of correlation analysis showed differential association among traits of early and those of extra-early maize, indicating that mechanism of tolerance to drought in the two maize maturity groups are different. It was concluded that early and extra-early maize hybrids responded differently under drought stress imposed at seedling stage and that seedling stage total number of leaf shed, moisture content of root, fresh shoot biomass, and total fresh biomass were important drought adaptive traits and the traits should be included in computing base index for selecting drought tolerant maize at seedling stage.

Keywords: biomass, drought, maize, principal component, seedling aspect

Introduction

Maize (*Zea mays* L) is ranked second to wheat in world production of cereal crops. It is widely grown throughout the world in a wide range of agro-ecological environments. Differential demands for maize in different agro-ecologies of West and Central Africa have facilitated the development of maize of different maturity classes, ranging from extra-early maturing, which matures within 85 and 90 days and adaptive to areas of short rainfall duration to extra-late varieties which matures in 120 days and above which are suitable for rainforest conditions with heavy and long duration of rainfall. This makes maize a very versatile crop and it is rapidly replacing traditional cereals such as sorghum (*Sorghum bicolor* L Moench) and millet (*Pennisetum glaucum* L R.Br.), especially in savanna areas with good access to fertilizer inputs and markets (Badu-Apraku and Fakorede, 2003). However, one major production constraint to maize production in sub-Saharan Africa is drought. Drought stress occurs at any period during the growth cycle of maize including seedling establishment, post-emergence growth, flowering or reproduction, and grain filling. Although, drought occurring at flowering and grain-filling stages are considered the most critical in maize growth because they cause gross reduction in grain yield. In addition, Moser (2004) found that pre-anthesis drought significantly reduced the number of kernel rows, number of kernels per row, and 1,000-kernel weight while it consistently increased harvest index. Moreso, when drought occurs during the vegetative stage, it affects the length of the internodes by influencing the cell size development and, thus, the capacity for storing assimilates (Denmead and Shaw, 1960). It has also been reported that prolonged drought at seedling stage causes total crop failure (Edmeades et al, 1989).

The rainforest region of Nigeria and by extension, west Africa is characterized by bimodal rainfall pattern with peaks at June and September. In every year, rainfall starts with a few irregular and unpredictable showers in February and March. Farmers are not ad-

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vised to plant maize at these period because the rains stop when the maize is still at the seedling stage or early vegetative stage and it results in total crop loss. In contrast, this period is characterized with higher insolation than the normal growing season with higher potentials for photosynthesis resulting in higher grain yield (Fakorede and Opeke, 1985). Furthermore, maize produced by crops grown at this period attracts a premium price because it gets to the market earlier with high demand. Earlier studies conducted in this agro-climatic zone had shown that maize planted early significantly out-yielded those planted later in the season primarily because grain-filling coincided with the period of relatively higher incident solar radiation than during the early season. Maize genotype with tolerance to drought at early stage is capable of increasing maize productivity in this agro-ecology of west Africa.

Most research efforts on maize in the world are concentrated on drought at flowering and grain filling periods and adaptive traits for selection for drought tolerance have been identified for tropical maize germplasm. Earlier studies have reported that seedling drought mechanism are independent of drought responses at flowering period (Meeks et al, 2013), indicating that genotypes that are tolerant to drought at flowering stage do not automatically tolerate drought at seedling stage and vice-versa. Information on the response of maize genotypes to drought that occurs at seedling and early vegetative stages and identification of adaptive traits is essential in achieving rapid progress from selection for tolerant maize genotypes.

Efficient phenotyping is of prime importance in successful selection program for a target trait. Effective screening methodology is required to facilitate rapid progress in breeding for tolerance to any stress conditions. Akinwale et al (2016) worked on methodology for screening maize genotypes for tolerance to drought at seedling stage. They reported in a screenhouse study that, withdrawing water supply from seedlings at 7 days after planting and keeping the plants under observation for another 35 days distinguished clearly between susceptible maize hybrids and tolerant hybrids. Bocev (1963) found out that root growth and development in maize could be used as reliable characters that indicated drought tolerance at seedling stage. In practice, however, accurate determination of root number and length in maize seedlings in a potted experiment without cutting or disrupting the length is a difficult task and it cannot be done except by destructive sampling. In addition, Vadez (2014) pointed out that success achieved in breeding crops for improved root is mimimal, signifying that root attributes alone may not suffice as drought adaptive features in crops. Other traits which are more easily measured and capable of discriminating between drought tolerant and susceptible genotypes could significantly facilitate rapid progress from selection for genotypes tolerant to drought at this stage. Scores based on careful observation of symptoms of stress on plants have been used to select for tolerant genotypes under stress and satisfactory progress has been reported based on this scores. Scientists at the International Center for Maize and Wheat Improvement (CIMMYT) and the International Institute of Tropical Agriculture, (IITA) have devised standard scales for scoring tolerance to different stresses. Leaf death scores (or leaf senescence) and stay green characteristics on a scale of 1 to 10 were used for selecting genotypes tolerant to drought at flowering and low soil N stresses, respectively (Banziger et al, 2000; Menkir et al, 2003; Meseka et al, 2013). Similarly, these scores were used for selecting genotypes tolerant to low soil N. Badu-Apraku et al (2011) and Badu-Apraku and Akinwale (2011) found out that a score of general architecture and appeal of plant termed plant aspect, based on visual observation and rated on a scale of 1 to 9, was a reliable predictor of grain yield under drought at flowering and grain filling periods and low soil N stress (Badu-Apraku et al, 2011). The primary objectives of this study were to evaluate the response of early and extra-early maize hybrids to drought stress imposed at seedling stage and to determine the relationship among seedling traits with a view to identifying adaptive traits for drought tolerance at seedling stage.

Materials and Methods

Location

Thirty maize hybrids (15 early and 15 extra-early) obtained from International Institute of Tropical Agriculture (IITA) Ibadan were used for the study. This research was conducted in a screenhouse facility of the Faculty of Agriculture, Obafemi Awolowo University, Ile-Ife, Nigeria.

Screenhouse Evaluation

The pots were filled with 5 kg of soil each and six seeds of each hybrid seeds were sown per pot. The experiment was laid out in a randomized complete block design with three replications. Water was applied to each pot at the rate of 0.6 litre per pot daily for 7 days and the plants were left without watering for the rest of the experiment period. Seedling emergence was assessed starting from 4 days after planting (DAP) till 9 DAP. After watering stopped, plants were observed and readings were taken weekly on seedling height (SHT), number of leaves per plant (NL), leaf area (LA), and number of shed leaves (LSHD). At the end of the experiment, 42nd day, the plants with the ball of soil were carefully removed from each pot and the roots washed free of sand under a gentle running tap. The roots were detached from the shoot at the cotyledonary node. Data were recorded on number of plants per pot, total number of leaves per plant, total number of dead leaves, and length of primary root (RL; in cm) using a meter rule. In addition, fresh shoot weight (g), fresh root weight (g), dry shoot weight (g), and dry root weight (g) were measured using a Metler

Table 1 - Mean squares for germination percentage and some shoot characteristics of 30 early and extra-early maize hybrids tested under drought stress imposed at seedling stage at the screenhouse, Faculty of Agriculture, Obafemi Awolowo University, Ile-Ife, Nigeria in 2014 .

Source	DF	Germination percentage	Seedling height (cm)	Leaf area	Number of shed leaves	Number of leaves	Number of dead leaves	Shoot fresh weight (g)	Shoot dry weight (g)	Shoot moisture content	Seedling aspect
Rep	2	0.15*	4.70	84.64*	3.38	208.31*	265.43*	0.19	0.002	0.022*	3.90
Hybrid	29	355.64	28.18*	52.54	18.98**	41.18	46.10	0.59**	0.01	0.012	12.71**
Extra-early (EE)	*14*	*704.69*	*18.66**	*62.76*	*19.47***	*54.57*	*36.82*	*0.84***	*0.01*	*0.003*	*11.95***
Early (E)	*14*	*360.93*	*36.89*	*44.03*	*19.82***	*22.15*	*45.47*	*0.38***	*0.01*	*0.001*	*14.19***
E vs EE	*1*	*54.44*	*39.56*	*28.66*	*0.28*	*120.18*	*184.90**	*0.01*	*0.01*	*0.014**	*2.50*
Error	*58*	*416.74*	*17.23*	*73.12*	*1.35*	*51.02*	*35.64*	*0.10*	*0.01*	*0.00*	*0.90*
R Square		68	67	62	88	71	90	75	78	60	93
CV		17	8	28	39	27	30	35	40	40	14

*,** Significant at 0.05 and 0.01 levels of probability, respectively

weighing balance. Ratios of weights of root to shoot for both fresh and dry weight was calculated. Fresh biomass (g) and dry biomass (g) were recorded by addition of root and shoot weights based on fresh and dry weights, respectively. Dry weights were determined by subjecting plant tissue samples to oven-drying at 80°C until a constant weight was achieved. Moisture content was determined by subtracting the dry weight of sample from its fresh weight. Seedling aspect was scored on a scale of 1 to 9, where 1 = no visible symptom of stress: vigorous plants, no wilting, no dead leaves, no chlorosis, no height reduction and unrolled turgid leaves; 2 = very mild symptom of stress: vigorous plants, no wilting, no dead leaves, no chlorosis, very slight leaf rolling; 3 = mild symptom of stress: vigorous plants, no wilting, no dead leaves, no chlorosis, leaf rim start to roll; 4 = mild symptom of stress: vigorous plants, no wilting, no dead leaves, slight chlorosis, leaf has shape of a V; 5 = moderate symptom of stress: vigorous plants, no wilting, 10% dead leaves, slight chlorosis, rolled leaf rim covers part of leaf blade; 6 = susceptible plants: less vigorous plants, moderate reversible wilting, 25% dead leaves, severe chlorosis, leaf is rolled like that of an onion; 7 = highly susceptible: severe irreversible wilting with 50% dead leaves; 8 = highly susceptible: severe irreversible wilting with 75% dead leaves, 75% death of seedlings; and 9 = total collapse or 100% death of seedling, dried leaves and stem. Seedling aspect rating scales of 1-5 indicated different levels of tolerance of the seedlings while scales of 6-9 indicated levels of susceptibility of the seedlings.

Statistical Analysis

Data collected were subjected to analysis of variance (ANOVA) using Statistical Analysis Software (SAS) to test for significant effect of the experiment treatments on all the hybrids and means were separated using Least Significant Difference (LSD) at 0.05 level of probability. Correlation analysis and principal component analysis were performed to elucidate interrelationships among traits and identify traits that could be reliably used to select for drought tolerance at this stage.

Results

Genetic variation among hybrids

Results of the analysis of variance of seedling traits showed significant differences among the thirty hybrids for seedling height, number of leaf shed, fresh weight, seedling aspect, root moisture content and total fresh weight (Tables 1 and 2). Other traits such as germination percentage, leaf area, number of leaves and root length were not significantly different among the 30 hybrids tested. When hybrid effect was partitioned into its components, there was significant difference among extra-early hybrids for seedling height, number of shed leaf, shoot fresh weight seedling aspect, root moisture content, Root-Shoot ratio based on fresh weight, Root-Shoot ratio based on dry weight and total fresh weight. On the other hand, only number of shed leaves, shoot fresh weight, seedling aspect, root moisture content and total fresh weight were significant among the early-maturing maize. Result of the orthogonal contrast partitioning revealed that there was significant difference between early and extra-early maturity groups for number of dead leaves, shoot moisture content, root length, fresh root weight, shoot moisture content and total dry weight.

Relationship among seedling traits

Correlation analysis

Results of the correlation analysis of all traits of extra-early maize hybrids (Table 4) revealed significant relationship of root length with only average fresh root weight (r = 0.56*) and average dry root weight (r = 0.55*). For early hybrids, root length had significant correlation only with total dry biomass (r = 0.54*) while across maturity, root length had significant correlation with average fresh root weight (r = 0.51*) average dry root weight (r = 0.49*) and total dry biomass (r = 0.45*). Seedling aspect had significant correlation with average fresh shoot weight (r = -0.88**), shoot moisture content (r = -0.82**), root-shoot ratio based on dry weight (r = -0.78**) and total fresh biomass (r = -0.80) for extra-early hybrids. Similarly, seedling aspect had significant correlation with average fresh shoot weight (r = -0.90**), shoot moisture content (r = -0.91**), root-shoot ratio based on dry weight (r = -0.75**), total fresh biomass (r = -0.88**) and, in addition, number of dead leaves (r = 0.63*) for

Table 2 - Mean squares for some root characteristics and biomass of 30 early and extra-early maize hybrids tested under drought stress imposed at seedling stage at the screenhouse, Faculty of Agriculture, Obafemi Awolowo University, Ile-Ife, Nigeria in 2014.

Source	DF	Root length (cm)	Fresh root weight (g)	Dry root weight (g)	Root moisture content	Root-Shoot Ratio (Fresh weight)	Root-Shoot Ratio (Dry weight)	Total Fresh biomass (g)	Total dry biomass (g)
Rep	2	60.70	0.01	0.04	0.18	2.54*	97.25	0.19	0.01
Hybrid	29	70.62	0.001	0.03	0.54**	1.04	72.05	0.59**	0.01
Extra-early (EE)	*14*	*73.12*	*0.001*	*0.04*	*0.77*****	*1.30*****	*43.07*****	*0.85*****	*0.02*
Early (E)	*14*	*59.13*	*0.001*	*0.02*	*0.35*****	*0.81*	*94.24*	*0.38*****	*0.01*
E vs EE	*1*	*196.54**	*0.02**	*0.01*	*0.07*	*0.51*	*167.00*	*0.01*	*0.06**
Error	58	55.83	0.01	0.03	0.09	0.67	47.24	0.12	0.01
R Square (%)		60	53	40	75	48	45	72	55
CV (%)		26	44	36	44	39	98	32	36

*,** Significant at 0.05 and 0.01 levels of probability, respectively

early hybrids. Across both maturity groups, seedling aspect had significant correlation with average fresh shoot weight (r = -0.83**), shoot moisture content (r = -0.83**), root-shoot ratio based on dry weight (r = -0.75**), root-shoot ratio based on fresh weight (r = -0.44*) and total fresh biomass (r = -0.80**).

Principal component analysis

Results of the principal component analysis (Table 5) showed that the first 4 principal component axes contributed significantly to variation among hybrids in both maturity classes. The first four PC axes accounted for 82% of the total variation among the extra-early hybrids, 79% of the total variation among the 15 early hybrids and 76% of the total variation across the 30 hybrids.

To identify traits that made important contribution to each PC axis, a restriction that stipulates eigenvectors greater than or equal to 0.3 as the logical cut-off points was applied as proposed by Badu-Apraku et al (2006). Based on the restriction, average fresh shoot weight (g), Moisture content of root (%), Root:Shoot ratio based on dry weight, and total fresh biomass were loaded on PC1 axis; emergence (%), number of leaf; average fresh root weight (g), average dry root weight (g), average dry shoot weight (g), and total dry biomass were loaded on the PC2 number of leaf shed and Root:Shoot ratio (Fresh) were loaded on PC3 axis; and Seedling height and Moisture content of shoot (%) were loaded on PC4 axis for extra-early maize. Leaf area, root length, and number of dead leaves were not loaded on any of the four PC axes. For early maize, similar traits were loaded on the PC1 axis, i.e., average fresh shoot weight (g), moisture content of root (%), Root:Shoot ratio based on dry weight, and total fresh biomass were loaded. In contrast, on PC2, number of leaf; average fresh root weight (g), average dry shoot weight (g), moisture content of shoot (%) and total dry biomass were loaded. emergence (%), average dry shoot weight (g) and Root:Shoot ratio (Fresh) were loaded on PC3 axis; while number of leaf shed was loaded on PC4. Seedling height, leaf area, root length, and number of dead leaves were not loaded on any of the four PC axes for early maize. For data combined across maturity, traits loaded on PC1 were the same as those for early and extra-early maize hybrids. On PC2, however, average fresh root weight (g), average dry shoot weight (g), moisture content of shoot (%), and total dry biomass were loaded; emergence (%), seedling height, number of leaves, average dry root weight (g), and Root:Shoot ratio (Fresh) were loaded on PC 3; while number of dead leaves and number of leaves were loaded on PC 4. Leaf area, and root length were not loaded on any of the four PC axes.

Discussion

A primary objective of this study was to find out the response of early and extra-early maize hybrids under drought stress imposed at seedling stage. Results of the analysis of variance revealed significant differences among the 30 hybrids for seedling aspect, number of shed leaves, fresh shoot weight, root moisture content, and total fresh biomass. This indicated that these are reliable traits that could be used as indicators of drought tolerance at seedling stage and it is suggestive that a base index that incorporate these traits should be computed for selecting maize genotypes for drought tolerance at seedling and early vegetative stages. The identification of total fresh biomass is consistent with findings of Farooq et al (2008). Maturity significantly influenced the usefulness of the traits to select drought tolerant genotypes. For instance, the result showed that seedling height was significant only for extra-early hybrid indicating that seedling height can only be used for selecting drought tolerant extra-early hybrids but not early hybrid. In the same vein, traits such as Root-Shoot ratio based on both fresh and dry weights were useful traits to select for drought tolerant extra-early genotypes but not useful for early genotypes. Root-shoot ratio has also been identified as important trait indicator of drought tolerance in maize. Li et al (2014) and Naveed et al (2014) explained that although both shoot and root growth were inhibited by drought stress, shoot growth was more sensitive than root growth, thus shoot-root ratio was typically reduced. This further implied that under drought stress, plants allocate more resources to root than to shoot growth in order to enhance water acquisition and limit evaporation (Lynch and Ho, 2005). Traits such as number of dead leaves, shoot moisture content, root length, fresh root weight, and total dry biomass were useful only for discriminating between

Table 3 - Mean values of selected seedling traits of 30 early and extra-early maize hybrids evaluated at the screenhouse under drought induced at seedling stage at Obafemi Awolowo University, Ile-Ife, Nigeria in 2014.

S.No	Hybrid	Maturity	Hybrid Type	Endosperm colour	Seedling aspect	Seedling height (cm)	Number of shed leaves	Fresh shoot biomasst (g)	Root moisture content	Total fresh biomass (kg)
1	TZEI11 × TZEI24	Early	Single cross	White	3.3	56	0	1.0	0.8	1.2
2	TZEI3 × TZEI26	Early	Single cross	White	4.7	52	8	1.0	0.8	1.1
3	TZEI3 × TZEI4	Early	Single cross	White	8.3	48	0	0.2	0.1	0.4
4	TZEI5 × TZEI98	Early	Single cross	White	2.0	41	2	1.4	1.3	1.6
5	TZEI7 × TZEI26	Early	Single cross	White	3.3	51	0	1.4	1.2	1.5
6	TZEI87 × TZEI2	Early	Single cross	White	3.7	52	0	1.2	1.0	1.4
7	TZEI9 × TZEI16	Early	Single cross	White	2.7	47	0	1.2	1.1	1.3
8	TZEI11 × TZEI136	Early	Single cross	Yellow	6.7	53	3	0.6	0.5	0.8
9	TZEI129 × TZEI16	Early	Single cross	Yellow	8.7	50	7	0.3	0.2	0.5
10	TZEI14 × TZEI25	Early	Single cross	Yellow	7.3	54	3	0.6	0.4	0.7
11	(TZEI108 × TZEI63) × (TZEI59 × TZEI87)	Early	Double cross	White	3.3	51	0	1.2	0.9	1.3
12	(TZEI2 × TZEI63) × (TZEI108 × TZEI87)	Early	Double cross	White	6.3	53	0	0.9	0.7	1.1
13	(TZEI129 × TZEI125) × (TZEI157 × TZEI16)	Early	Double cross	Yellow	5.7	53	3	0.7	0.5	0.9
14	(TZEI17 × TZEI129) × (TZEI157 × TZEI16)	Early	Double cross	Yellow	7.3	53	1	0.9	0.7	1.2
15	(TZEI108 × TZEI59) × TZEI63	Early	3-way cross	White	6.7	50	0	0.9	0.7	1.0
16	TZEEI29 × TZEEI21	Extra-early	Single cross	White	7.7	54	0	0.7	0.4	0.9
17	(TZEEI29 × TZEEI14)	Extra-early	Single cross	White	7.7	53	0	0.4	0.2	0.6
18	TZEEI29 × TZEEI49	Extra-Early	Single cross	White	6.3	52	3	0.9	0.7	1.0
19	TZEEI3 × TZEEI46	Extra-early	Single cross	White	4.3	57	5	0.9	0.7	1.2
20	TZEEI39 × TZEEI90	Extra-early	Single cross	White	5.7	52	1	0.5	0.3	0.6
21	TZEEI63 × TZEEI95	Extra-Early	Single cross	White	6.3	55	0	0.5	0.3	0.6
22	TZEEI9 × TZEEI79	Extra-early	Single cross	White	9.0	48	0	0.4	0.2	0.6
23	TZEEI79 × TZEEI82	Extra-early	Single cross	Yellow	4.3	51	4	0.8	0.7	1.0
24	TZEEI82 × TZEEI95	Extra-early	Single cross	Yellow	3.7	55	4	1.1	0.8	1.2
25	(TZEEI29 × TZEEI21) × (TZEEI14 × TZEEI39)	Extra-early	Double cross	White	3.0	54	0	2.1	1.9	2.3
26	(TZEEI82 × TZEEI63) × (TZEEI79 × TZEEI58)	Extra-early	Double cross	Yellow	2.7	53	0	1.9	1.5	2.1
27	(TZEEI95 × TZEEI58) × (TZEEI82 × TZEEI79)	Extra-early	Double cross	Yellow	6.7	49	0	0.8	0.6	1.0
28	(TZEEI129 × TZEEI37) × TZEEI13	Extra-early	3-way cross	White	3.7	50	0	1.4	1.2	1.5
29	(TZEEI79 × TZEEI163) × TZEEI78	Extra-early	3-way cross	Yellow	8.3	51	3	0.4	0.3	0.6
30	(TZEEI9 × TZEEI79) × TZEEI63	Extra-early	3-way cross	Yellow	5.7	53	0	0.5	0.3	0.7
	Grand mean				5.5	52	2	0.9	0.7	1.1
	LSD				1.3	6.1	1.8	0.51	0.5	0.6

maturity groups, and they were not good indicators of drought tolerance at seedling stage. Contrary to our findings, some earlier studies identified root length and density as good indicators of drought tolerance in cultivated crops (Ober et al, 2005; Gowda et al, 2011; Iqbal et al, 2011). However, our findings is in agreement with the reports of other studies who reported no relationships between length and density of root with drought tolerance (Kashiwagi et al, 2006; Jongrungklang et al, 2012). Avramova et al (2016) identified total root length and shoot dry weight as reliable measurements of drought tolerance at the seedling stage under field conditions in maize. Vadez (2014) observed that although roots have long been thought of as a major avenue to improve crop adaptation to water limitations based on the assumption that deeper and more profuse root systems could tap extra water from the soil profile and alleviate drought effects, success in breeding cultivars with improved root systems is limited. He reported that the role that roots play in drought adaptation may not necessarily be associated with their density or depth, but rather from their hydraulic characteristics.

Based on analysis of data from a lysimetric system that allows monitoring and comparing plant water use over the entire crop life cycle and yield, Vadez (2014) reported that the role of roots as adaptive features under drought may not be on the basis of their length, density or depth, but rather on their hydraulic characteristics (ability to effectively trap and utilize available water into their tissues). In our study, moisture content of the root, which is a measure of the effectiveness of the roots to draw moisture into its tissues, was found to be more important rather than root length under drought at seedling stage. Also, Cantao et al (2008) reported significant differences for root morphological attributes and root and canopy growth. They observed that drought tolerant inbred lines showed distinct root system than susceptible lines, by presenting longer root, surface area, and volume under drought stress at seedling stage.

Plant productivity under drought stress is strongly related to the process of dry matter partitioning and temporal biomass distribution (Kage et al, 2004). Fa-

Table 4 - Correlation for 30 early and extra-early maize hybrids evaluated in screenhouse under induced drought at seedling stage at Obafemi Awolowo University, Ile-Ife, Nigeria in 2014.

Trait	Extra-early maturity class				Early maturing class				Across maturity classes			
	Root length (cm)	Seedling aspect	Total fresh biomass (g)	Total dry biomass (g)	Root length (cm)	Seedling aspect	Total fresh biomass (g)	Total dry biomass (g)	Root length (cm)	Seedling aspect	Total fresh biomass (g)	Total dry biomass (g)
Emergence (%)	-0.25	-0.48	0.01	-0.44	-0.14	-0.52*	0.25	0.15	-0.22	-0.49	0.08	-0.27
Seedling height	-0.14	-0.44	0.23	0.37	0.06	0.34	-0.27	0.13	0.04	0.05	0.00	0.30
Leaf area (cm^2)	0.34	-0.16	0.22	0.37	-0.27	-0.18	0.02	-0.07	0.12	-0.15	0.15	0.26
Number of leaf shed	-0.33	-0.27	-0.02	-0.06	-0.40	0.30	-0.38	-0.04	-0.35	0.02	-0.16	-0.06
Number of leaves	-0.10	-0.47	0.13	-0.30	0.29	0.08	0.04	0.25	0.14	-0.20	0.10	0.01
Number of dead leaves	-0.06	0.09	-0.32	-0.42	0.07	0.63*	-0.45	-0.04	0.12	0.39	-0.33	-0.06
Average Fresh root weight (g)	0.56*	0.21	0.06	0.82**	0.30	0.31	0.01	0.58*	0.51*	0.27	0.04	0.77**
Average Fresh shoot weight (g)	0.09	-0.82**	1.00**	0.35	0.36	-0.90**	1.00**	0.54*	0.17	-0.83**	1.00**	0.36
Average dry root weight (g)	0.55*	0.19	0.07	0.67**	0.33	-0.06	0.31	0.59**	0.49*	0.10	0.15	0.65**
Average dry shoot weight (g)	0.08	-0.31	0.49*	0.92**	0.45	-0.40	0.53*	0.85**	0.31	-0.27	0.46*	0.92**
Moisture content of shoot (%)	0.21	0.09	0.001	0.48	0.11	0.44	-0.26	0.23	0.26	0.29	-0.09	0.46*
Moisture content of root (%)	0.08	-0.82**	0.98**	0.26	0.33	-0.91**	0.99**	0.47	0.15	-0.83**	0.98**	0.27
Root-Shoot ratio (Fresh)	-0.43	-0.45	0.36	0.14	-0.08	-0.48	0.28	0.08	-0.23	-0.44*	0.33	0.16
Root-Shoot ratio (Dry)	-0.12	-0.78**	0.85**	-0.03	0.05	-0.75**	0.67**	-0.11	-0.11	-0.75**	0.67**	-0.17
Root length, cm	-	0.15	0.13	0.30	-	-0.18	0.39	0.54*	-	0.01	0.22	0.45*
Seedling aspect	0.15	-	-0.80**	-0.16	-0.18	-	-0.88**	-0.35	0.01	-	-0.80**	-0.17
Total fresh biomass	0.13	-0.80**	-	0.41	0.39	-0.88**	-	0.59*	0.22	-0.80**	-	0.43*
Total dry biomass	0.30	-0.16	0.41	-	0.54*	-0.35	0.59*	-	0.45*	-0.17	0.43*	-

*,** Significant at 0.05 and 0.01 levels of probability, respectively

rooq et al (2008) reported that water stress on crop plants generally cause significant reduction in fresh and dry biomass production. In general, the result of this study identified more shoot characteristics as indicators of drought tolerance than root characteristics. The results further identified that fresh weights are better distinguishing traits under drought than dry weights.

Based on seedling aspect, there were 13 tolerant hybrids (scores < 6). Six were extra-early and seven were early maturing. This indicated that seedling drought tolerance is not maturity specific or maturity does not condition drought at seedling stage. Moreover, nine of the 13 tolerant hybrids were single cross hybrid. This is not in agreement with Hallauer et al (2010) who opined that single cross hybrids are more sensitive to environmental stress conditions than 3-way cross and double cross hybrids. This may be due to the fact that Hallauer et al (2010) based their conclusion on temperate maize germplasm in contrast with tropical maize germplasm used in this study. The best tolerant maize hybrids identified in this study were TZEI5 × TZEI98, (TZEEI82 × TZEEI63) × (TZEEI79 × TZEEI58), TZEI9 × TZEI16, (TZEEI29 × TZEEI21) × (TZEEI14 × TZEEI39), (TZEI108 × TZEI63) × (TZEI59 × TZEI87), TZEI11 × TZEI24, and TZEI7 × TZEI26 (Table 3). Any of these hybrids could be advanced to F_2 singly or after crosses with one or more hybrids and a new set of inbred lines with higher level of tolerance to drought at seedling stage can be extracted. In addition, there were more white maize genotypes among the tolerant hybrids than yellow maize, suggesting that white maize showed more tolerant to drought at seedling stage than yellow maize. The identification of TZEEI 29 x TZEEI 21 among parents of a double cross hybrid with tolerance to drought at seedling stage is consistent with the report of Badu-Apraku et al (2011), who identified TZEEI 29 x TZEEI 21 as an ideal genotype under drought at flowering and grain filling periods. Similarly, TZEI7 and TZEI11 which are among parents for some of the tolerant early maize hybrids in this study had been identified to be among ideal inbreds under drought at flowering period in an earlier study (Badu-Apraku and Akinwale, 2011). Even though it has been reported in an earlier study that drought tolerant genes at seedling and flowering periods are mutually independent, it could be inferred from this study that TZEEI29, TZEEI21, TZEI7 and TZEI11, possess in them genes that confer tolerance to drought at both stages. In addition to tolerance to drought at flowering stage, TZEI11 was also identified as an outstanding inbred under low N (Badu-Apraku and Akinwale, 2011). This implies that TZEI11 is a very versatile and multipurpose tropical inbred line which could be used to introgress genes for multiple stress into other maize populations. In contrast, inbreds TZEI2, TZEI3 and TZEI17, which were found to be promising under drought at flowering in an earlier study (Badu-Apraku and Akinwale, 2011) were identified as parents of some susceptible hybrids in this study.

Another important objective of this study was to elucidate relationships among seedling traits with a view to identify novel traits for measuring drought tolerance at seedling traits among the hybrids. Across maturity classes, correlation analysis showed that root length had non-significant relationship with other traits except other root attributes. This implies that root length is not an important adaptive trait among early and extra-early maize. Seedling aspect on the other hand had significant negative correlation with average fresh shoot weight ($r = -0.83^{**}$), shoot moisture content ($r = -0.83^{**}$), root-shoot ratio based on dry weight ($r = -0.75^{**}$), root-shoot ratio based on fresh weight ($r = -0.44^{*}$), and total fresh biomass ($r = -0.80^{**}$). Many of these traits with significant correlation with seedling aspect had been identified as important traits in earlier studies (Moser, 2004; Vadex,

Table 5 - Eigenvectors of the first four principal components (PC1, PC2, PC3, and PC4) axes for 30 early and extra-early maize hybrids evaluated in screenhouse under induced drought at seedling stage at Obafemi Awolowo University, Ile-Ife, Nigeria in 2014.

	Extra-early maturity class				Early maturing class				Across maturity classes			
Trait	PC1	PC2	PC3	PC4	PC1	PC2	PC3	PC4	PC1	PC2	PC3	PC4
Emergence (%)	0.13	**-0.34**	0.02	0.32	0.11	0.05	**0.34**	0.29	0.11	-0.06	**0.39**	-0.22
Seedling height	0.24	0.04	-0.17	**0.33**	-0.17	0.28	0.29	-0.10	0.01	0.26	**0.33**	0.07
Leaf area (cm^2)	0.11	0.23	-0.07	0.04	0.04	-0.04	0.16	0.14	0.09	0.09	-0.02	0.25
Number of leaf shed	0.13	-0.02	**-0.39**	0.09	-0.16	-0.08	0.15	**0.55**	-0.05	-0.03	0.26	**0.39**
Root length	-0.06	0.22	0.29	0.10	0.12	0.24	-0.17	-0.15	0.06	0.26	-0.21	-0.17
Number of leaf	0.09	**-0.31**	0.27	0.26	-0.06	**0.31**	0.27	-0.11	0.05	0.15	**0.31**	-0.41
Number of dead leaves	-0.16	-0.26	0.25	0.19	-0.28	0.22	0.09	-0.08	-0.19	0.19	0.21	**-0.32**
Seedling Aspect	**-0.38**	0.10	-0.09	-0.04	**-0.37**	-0.01	-0.11	-0.06	**-0.40**	0.07	-0.08	0.06
Average fresh root weight (g)	-0.04	**0.39**	0.10	0.24	-0.12	**0.35**	-0.27	0.10	-0.05	**0.42**	-0.16	0.07
Average fresh shoot weight (g)	**0.36**	0.05	0.19	-0.16	**0.38**	0.10	-0.02	0.03	**0.42**	0.02	-0.07	-0.04
Average dry root weight (g)	-0.09	**0.30**	0.27	0.06	0.06	0.11	**-0.48**	0.30	0.00	0.24	**-0.38**	-0.02
Average dry shoot weight (g)	0.21	**0.32**	-0.04	0.12	0.14	**0.34**	0.16	0.14	0.19	**0.34**	0.01	0.23
Moisture content of shoot (%)	0.05	0.25	-0.19	**0.31**	-0.20	**0.34**	0.06	-0.12	-0.07	**0.37**	0.14	0.12
Moisture content of root (%)	**0.35**	0.02	0.21	-0.18	**0.38**	0.07	-0.03	0.02	**0.41**	-0.02	-0.07	-0.07
Root:Shoot ratio (Fresh)	0.27	0.01	**-0.32**	-0.04	0.14	0.05	**0.50**	-0.02	0.21	0.001	**0.30**	0.27
Root:Shoot ratio (Dry)	**0.32**	-0.10	0.15	-0.30	**0.31**	-0.15	0.06	-0.06	**0.33**	-0.24	-0.07	-0.07
Total fresh biomass	**0.35**	0.08	0.20	-0.14	**0.37**	0.13	-0.04	0.04	**0.41**	0.05	-0.08	-0.03
Total dry biomass	0.13	0.38	0.08	0.12	0.15	0.34	-0.13	0.27	0.15	**0.37**	-0.15	0.17
Proportion	0.28	0.24	0.16	0.14	0.31	0.24	0.15	0.10	0.25	0.22	0.16	0.12
Cumulative	0.28	0.52	0.68	0.82	0.31	0.54	0.69	0.79	0.25	0.48	0.64	0.76

values in bold lettering indicate eigenvectors with values higher than or equal to 0.3

2014). This indicates that seedling aspect could serve singly as a selection criterion for drought tolerance at seedling stage or in combination with other traits in a selection index. A careful look into the relationship among traits for each of the maturity group implied that maturity affected relationship among traits. Root length of extra-early hybrids had significant relationship with average fresh and dry root weight while for early hybrids, it had significant positive relationship with total dry biomass only. This indicate that root contributed significantly to dry matter accumulation only for early hybrids in spite of the fact that extra-early had significantly longer roots than the early hybrids. It has been reported earlier that under drought stress, roots elongate in search of water for survival and that deeper and more profuse root systems could tap extra water from the soil profile and alleviate drought effects while shoots are reduced to conserve moisture (Vadez, 2014). Premised on this, root has been considered as a major avenue to improve crop adaptation to water limitations at seedling stage. Similarly, seedling aspect had significant relationship with the same traits for both early and extra-early maturing hybrids but early maturing class had significant relationship with emergence percentage and number of dead leaves in addition. Furthermore, the pattern of relationship between other traits and fresh and dry biomass for each of the maturity classes were different.

The patterns of relationship among traits for extra-early were different from the early-maturing hybrids, indicating that mechanism for tolerance to drought at seedling stage for the two maturity classes are not the same. For instance, seedling aspect had significant correlation with emergence and number of dead leaves for early hybrids but it had no significant relationship with these two traits for extra-early (Table 4). Similarly, total fresh biomass had significant relationship with total dry biomass for early hybrids but the relationship was not significant for extra-early hybrids. Root length had significant relationship with total dry biomass for early hybrids but the relationship was not significant for extra-early. In addition, results of the principal component analysis also indicated this trend. Although, traits that were identified as contributing to variation under PC1 were the same for both maturity groups, the traits with high loadings on under PC2 and PC3 for both groups were not the same, indicating that response of hybrids from the two groups was different. This differential response might not be unconnected with the significant differences in their root length, fresh root weight and total dry biomass.

Cultivars are considered for cultivation not on the basis of one good trait but a combination of important traits (Yan and Kang, 2003). Base index is a novel approach to develop cultivars that combine many good traits. The identification of seedling aspect, average fresh shoot weight, moisture content of root, Root:Shoot ratio (Dry), and total fresh biomass as traits with high loading under the first principal component axis revealed that the traits are of primary importance in determining maize genotypes with tolerance to drought at seedling stage. These traits should be included in computation of base index for selecting for drought tolerant genotypes at seedling stage. The traits with high loading on the PC1 should be assigned a greater weight than those loaded on other PCs axes. Unlike root attributes, selecting drought tolerant genotypes based on seedling aspect is faster because seedling aspect is scored between 16 and 20 days after planting or between 9 and 13 days after water supply is withdrawn; scoring is easier and does not involve destructive sampling. The fact that it was significantly correlated with some of the adaptive traits reported with other studies such as Root-Shoot ratio (Fresh), Root-Shoot ratio (Dry) and total fresh biomass indicated that it is a reliable trait that can represent these other traits in a base index for seedling drought tolerance. However, field studies is needed to validate the efficiency of seedling aspect in predicting drought tolerance at seedling stage under field conditions. Another trait with high loading on PC1 that should be considered in computing the base index is moisture content of root. Other potential traits that could be included in computing the base index are the traits with high loadings on PC2 such as average fresh root weight, average dry shoot weight, moisture content of shoot and total dry biomass but less weights should be attached to these group of traits. There is the need to validate the use of the identified traits and resulting base index by well-planned field studies under drought before the early planting season.

Conclusion

Early and extra-early hybrids responded differently to drought conditions at seedling stage in terms of interrelationship among adaptive seedling traits under drought stress at seedling stage. It was also concluded from the study that reliable traits that could be used to select for drought tolerance at seedling stage were seedling aspect, average fresh shoot weight, total number of dead leaves at sampling time, shoot-root ratio of dry matter, moisture content of the root, and total fresh biomass.

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