



Journal of Biological Sciences

ISSN 1727-3048

science
alert

ANSI*net*
an open access publisher
<http://ansinet.com>



Research Article

Morphological Responses of Maize to Drought, Heat and Combined Stresses at Seedling Stage

^{1,2}Liliane Ngoune Tandzi, ¹Graeme Bradley and ¹Charles Mutengwa

¹Department of Agronomy, Faculty of Science and Agriculture, University of Fort Hare, P. bag X1314, 5700 Alice, South Africa

²Institute of Agricultural Research for Development (IRAD), P.O. Box 2123, Messa, Yaounde, Cameroon

Abstract

Background and Objective: Drought and heat stresses are major abiotic constraints causing limitations to plant growth worldwide and limited focus has been devoted to the combination of the two stresses. The objectives of this study were to: (1) Identify maize genotypes tolerant to heat, drought and combined drought and heat stress and (2) Identify some secondary traits associated these stresses at seedling stage. **Materials and Methods:** Twenty maize genotypes were evaluated in a randomized complete block design with three replicates and stresses were imposed in a growth chamber. The leaf stress response percentage, leaf area, plant height, plant aspect and some indices (STI, HTI, DTI and MSTI) were measured. The variances in traits among genotypes were performed using SAS software and Turkey's test was used for mean separation. **Results:** There were significant differences between genotypes for all the traits assessed under stress environments. Three inbred lines (L6-Y, L24-Y and Sweety 015) expressed relatively good performance across environments and could be potentially useful genotypes in breeding maize for tolerance to combined drought and heat stress and for tolerance to the individual stresses. Shoot weight, plant height and chlorophyll content showed significant relationships with stress tolerance indices (STI, HTI, DTI and MSTI) and could therefore, be used as secondary traits in maize screening at seedling stage under combined drought and heat stress environments. Heat stress environment was highly and positively correlated with combined drought and heat stress environment (+0.79) for stress tolerance index. **Conclusion:** Result of study demonstrates that the heat tolerant genotypes are likely to tolerate combined drought and heat stress conditions. The identified stress tolerant genotypes need to be evaluated in open environment for confirmation of the results.

Key words: Heat, drought, combined drought and heat, maize, seedling, tolerance

Received: February 17, 2018

Accepted: October 22, 2018

Published: December 15, 2018

Citation: Liliane Ngoune Tandzi, Graeme Bradley and Charles Mutengwa, 2019. Morphological responses of maize to drought, heat and combined stresses at seedling stage. *J. Biol. Sci.*, 19: 7-16.

Corresponding Author: Liliane Ngoune Tandzi, Department of Agronomy, Faculty of Science and Agriculture, University of Fort Hare, P. bag X1314, 5700 Alice, South Africa Tel: (27)0634594323/(237)678729005

Copyright: © 2019 Liliane Ngoune Tandzi *et al.* This is an open access article distributed under the terms of the creative commons attribution License, which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.

Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Maize (*Zea mays* L.) is a globally important staple crop for food, livestock feed and biofuels, even though it is very sensitive to abiotic stresses, including high temperature, which leads to considerable yield loss in crop production. Of the various abiotic stresses such as light intensity, salinity, drought, temperature (freezing/heat) is the most prevalent that considerably retard not only plant production but also the quality of crops¹⁻⁴. Heat stress is defined as the rise in temperature beyond a threshold level for a period sufficient to cause permanent damage to plant growth and development⁵. The disturbance in cellular homeostasis is due to high temperature stress which can cause drastic reduction in growth, development and even death of plants^{6,7}. The growth and development optimum temperature is specific to each genotype. The temperature stress occurs when the environmental temperature increases beyond the critical limit. Heat stress is responsible for 1.0-1.7% maize yield loss per day, for every degree rise in temperature⁸ above 30°C. Rahman *et al.*⁹ found that heat stress provided at the time of anthesis and subsequent developmental stages of grain formation was more devastating when temperature fluctuated between 40-45°C and sometimes up to 48°C in Pakistan. The local hybrids YH-1898 and YH-1921 showed reasonable tolerance against high temperature with 40-50% seed setting as compared to commercial hybrids (DK-6525, DK-6142 and NK-8441) with 20-25% seed setting⁹. According to Guy¹⁰ growth and development of maize is directly proportional to temperature increase until the optimum temperature is reached; therefore it becomes harmful to the plant. The stages of maize growth are differently affected by high temperature stress. Heat stress during germination is associated with impaired emergence¹¹, reduced plant stand and plant density¹². Sanchez *et al.*¹³ found that optimum temperature for maize growth from sowing to emergence is 29.3°C, with a threshold maximum of 40.2°C. Whereas, an increase in temperature above 30°C could reduce yield by 1% under optimal rain-fed condition and by 1.7% under drought conditions¹⁴.

Several research studies have been conducted to screen maize for tolerance to drought stress all over the world¹⁵⁻¹⁹. Drought is defined as a condition whereby there is inadequate moisture in the soil at a particular time to meet the needs of the plant²⁰. Maize yield losses due to drought stress range between 17-60% in southern Africa²¹⁻²². Meeks *et al.*²³ evaluated maize inbred lines and their hybrid testcross progeny at seedling stage for germination, survival and recovery after a series of drought cycles and concluded

that seedling stress response is more useful as secondary screening parameter for maize genotypes.

Maize plants usually develop different mechanisms to counteract the environmental stresses. They need to adapt quickly to overcome these stresses during their short life cycle. From simulation models, an average increase in temperature of up to 2.5-5.4°C can be expected by year 2100 coupled with a decrease in precipitation of about^{24,25} 15%. Warming is projected to occur during the 21st century, with plausible increases of 4-6°C over the sub-tropics and 3-5°C over the tropics by the end of the century under low mitigation scenario²⁶. Stress as it is understood today is a factor that alters normal functioning of a number of mechanisms in an organism²⁷. The majority of research on abiotic stresses has focused on individual stresses while in farmers' fields, plants are regularly subjected to a combination of stresses^{28,29}. With the general warming of the world, developing cultivars of maize that can perform well under heat stress, drought stress and combined heat and drought stress should be taken into consideration. The tolerance of plants to a combination of different stress conditions, especially those that mimic the field environment should be the focus of future research³⁰. The aim of the study was to identify maize genotypes which could express tolerance to heat, drought and combined heat and drought stress at seedling stage. Moreover, the study also sought to identify some secondary traits associated with these stresses, which could be utilized for maize selection during the seedling stages of development.

MATERIALS AND METHODS

Ten yellow Quality Protein Maize (QPM) inbred lines and 10 introduced varieties from the Institute of Agricultural Research for Development (IRAD) in Cameroon were used in the study (Table 1). The introduced varieties were composed of six inbred lines (two white and four yellow) and four open pollinated varieties (two white and two yellow). The experiment was conducted in the department of Biochemistry and Microbiology in a growth chamber during the month of August, 2017.

Experimental design and management: Maize genotypes were laid out in a randomized complete block design with three replicates. Three viable seeds were planted per pot for each genotype using Hygromix as growing media (commercial potting mix). The pots were placed in a tray of about 4 cm deep. The plants were kept at field capacity until two weeks after planting. The growth chamber was set at 25°C day and 22°C night, humidity 40% day and 60% night

Table 1: List of inbred lines and OPVs used for the evaluation

Genotype	Color	Origin	Particularity
L16-Y	Yellow	UFH	QPM
L17-Y	Yellow	UFH	QPM
L18-Y	Yellow	UFH	QPM
L24-Y	Yellow	UFH	QPM
L3-Y	Yellow	UFH	QPM
L32-Y	Yellow	UFH	QPM
L33-Y	Yellow	UFH	QPM
L34-Y	Yellow	UFH	QPM
L5-Y	Yellow	UFH	QPM
L6-Y	Yellow	UFH	QPM
87036	White	IRAD	Good combiner
88069	Yellow	IRAD	Good combiner
ATP S6 Y-1	Yellow	IRAD	Tolerant to low soil pH
ATP S8 30Y-3	Yellow	IRAD	Tolerant to low soil pH
ATP SR Y	Yellow	IRAD	Commercial acid tolerant OPV
CMS 8704	Yellow	IRAD	Commercial OPV
EVDT-99-W	White	IRAD	QPM
Exp1 24	White	IRAD	Good combiner
Obatampa	White	IRAD	QPM
Sweetly 015	Yellow	IRAD	Sweet corn

UFH: University of Fort Hare, IRAD: Institute of Agricultural Research for Development, OPV: Open Pollinated Variety, QPM: Quality Protein Maize

with 12 h photoperiod. The experiment was repeated two after following the same procedure.

Treatments: Maize genotypes were exposed to controlled conditions (no stress), water stress, combined drought and heat stress and heat stress alone treatments. Control and heat stressed plants were irrigated once after two days to maintain field capacity at 75%. Plants were maintained at 25% field capacity using a SM300 soil moisture meter for 5 days in drought environment and 3 days in combined drought and heat environment. The control and water stressed genotypes were kept in the same growth chamber at optimum temperature of 25°C during the day and 22°C at night. Heat stress and drought and heat stress were imposed at a high temperature regime (40°C/25°C) with 60% humidity for 3 days/nights. Temperature was increased gradually from 25-40°C with 5°C increments per hour during the 3 days. Two days of recovery were allowed at normal temperature provided in the control environment.

Data collection: The leaf number per genotype and the damaged leaf (leaf with any injury or wiltiness) number per genotype were counted. The leaf length, leaf width, plant height were measured using a ruler. A vernier caliper was used to access the stem diameter. An infrared thermometer was used to measure the leaf temperature. The chlorophyll content data were collected using a chlorophyll meter. The plant aspect was scored from 1-5 (with 1 being the best) and shoot weight was collected using a sensitive weighing balance.

Leaf stress response and leaf area were also calculated using the following equation:

$$\text{Leaf stress response} = \frac{\text{Damaged leaf}}{\text{Total number of leaves}} \times 100$$

$$\text{Leaf area} = \text{Leaf length} \times \text{leaf width} \times k$$

where, k = 0.75 as the coefficient determination of leaf area³¹.

The shoot weight of genotypes was used to calculate tolerance indices using the following equation:

$$\text{Stress Tolerance Index (STI)} = \frac{A_s A_c}{\bar{A}_c^2}$$

$$\text{Heat Tolerance Index (HTI)} = \frac{A_s(A_s / A_c)}{\bar{A}_s^2}$$

$$\text{Modified Stress Tolerance Index (MSTI)} = k_i \text{STI with } k = A_i^2 / \bar{A}^2$$

$$\text{Drought Tolerance Index (DTI)} = \frac{A_s / \bar{A}_s}{A_c / \bar{A}_c}$$

where, A_s and A_c represent shoot weight under stress and control conditions respectively, \bar{A}_s and \bar{A}_c represent the mean shoot weight under stress and control conditions respectively, A_i represents shoot weight of a given genotype³²⁻³⁴.

Data analysis: Analysis of the variance and correlations were computed using SAS package version 9.2 and the Turkey's test was performed to separate significantly different means of genotypes for a given trait. Cluster analysis was conducted using JMP.

RESULTS AND DISCUSSION

Analysis of variance revealed significant differences between genotypes in response to drought, heat and a combination of drought and heat stresses. Under drought stress, significant differences were observed for leaf area ($p < 0.05$), leaf stress response, stem diameter ($p < 0.01$), plant height, leaf temperature and shoot weight ($p < 0.001$) (Table 2). Genotypes showed significant differences for all the parameters under heat and in the combination of heat and drought stress (Table 2). There were no significant differences among genotypes for leaf stress response, chlorophyll content and plant aspect in the control environment (Table 2). The differences in growth potential makes the use of stress indices to be effective in distinguishing

Table 2: Mean squares for various traits recorded for maize genotypes evaluated under drought, heat, combined drought and heat stress and control environments

Parameters	df	Leaf stress (%)	Leaf area (cm ²)	Plant height (cm)	Chloro content (%)	Stem diameter (cm)	Plant aspect	Temperature (°C)	Shoot weight (g)
Drought									
Rep	2	11688 ^{NS}	103 ^{NS}	0.3 ^{NS}	29.8 ^{NS}	0.15 ^{***}	1.1 ^{NS}	0.0 ^{NS}	0.0 ^{NS}
Genotype	19	11874 ^{**}	118 [*]	5.7 ^{***}	29.7 [*]	0.05 ^{**}	0.5 ^{NS}	0.8 ^{***}	0.4 ^{***}
Error	38	3903	54	1.1	13	0.01	0.7	0.0	0.0
Heat									
Rep	2	734 ^{NS}	1.7 ^{NS}	0.06 ^{NS}	25.7 ^{NS}	0.004 ^{NS}	2.1 [*]	0.0 ^{NS}	0.02 ^{NS}
Genotype	19	1434 ^{**}	151.3 ^{***}	8.5 ^{***}	70.9 ^{***}	0.06 ^{***}	2.04 ^{***}	14.6 ^{***}	11.3 ^{***}
Error	38	484	35.2	2.4	18.8	0.02	0.5	0.0	0.02
Drought and heat									
Rep	2	2.3 ^{NS}	421 ^{NS}	1.1 ^{NS}	53.6 ^{NS}	0.08 [*]	1.2 ^{NS}	0.0 ^{NS}	0.0 ^{NS}
Genotype	17	2049 ^{***}	4250 ^{**}	10 ^{***}	177.3 ^{***}	0.09 ^{***}	2.5 ^{**}	13.4 ^{***}	0.08 ^{***}
Error	34	167	1384	2.3	36.3	0.02	0.75	0.0	0.0
Control									
Rep	2	1707 ^{***}	10.8 ^{NS}	1.8 ^{NS}	127 ^{**}	0.1 ^{NS}	0.8 ^{NS}	0.0 ^{NS}	0.02 ^{NS}
Genotype	19	204 ^{NS}	271 ^{***}	9.3 ^{**}	42 ^{NS}	0.08 [*]	0.7 ^{NS}	68.8 ^{***}	17 ^{***}
Error	38	168	65.9	3.5	24.2	0.04	0.6	0.0	0.02

***p<0.001, **p<0.01, *p<0.05, NS: Non-significant, Rep: Replication, Leaf stress: Leaf stress response (%), Chloro content: Chlorophyll content, Temperature: Leaf temperature

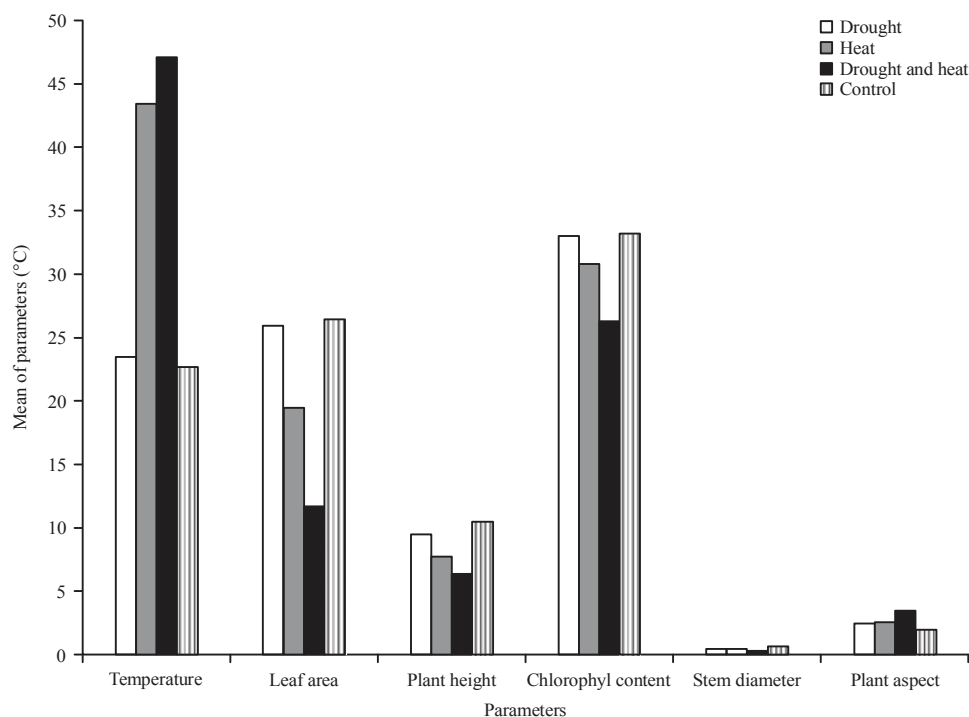


Fig. 1: Mean value of parameters measured under drought, heat, combined drought and heat and control condition

resistant from susceptible genotypes, when the differences in response of genotype is observed under stressed relative to non-stressed environments. Previous research studies resulted in significant differences among morphological traits collected on the genotypes evaluated under heat stress, drought stress and combined heat and drought stress at various growing stage of maize^{14,35,36}. Significant differences were observed on plant height, leaf area and leaf temperature among genotypes evaluated under the

control environment. This is due to the fact the evaluated maize genotypes were genetically different.

Performance of genotypes for traits recorded under stress and control environments:

Among the parameters collected under drought, heat, combined drought and heat stress and control environments, leaf temperature of plants was very high under combined drought and heat stress followed by heat stress alone (Fig. 1). The height of plants and the

chlorophyll content of maize leaves were lower under combined drought and heat stress followed by heat stress alone, than under drought stress alone. The reduced accumulation of chlorophyll content observed under high temperature stress may be due to either a decrease of biosynthesis of the chlorophyll or due to its increased degradation or the combined effect as stated by Fahad *et al.*³⁷ Higher temperatures were characteristic of susceptible maize genotypes under both combined drought and heat stress and under heat stress alone. These observations are in agreement with findings by Pfunde³⁶, who used maize inbred lines only. The stem diameter was quite high under control environment only (ranging from 0.3-0.9 cm) compare to the stress environments. These results are similar to the findings of Lipiec *et al.*⁵ who found that the effects of drought and high temperature were reflected in reducing mass accumulation in plants and caused early senescence and premature death. However, Yadav *et al.*³⁸ found that high temperatures during vegetative growth improved net photosynthetic rate resulting in higher total stover yield (+28%) at maturity. This could be possible in the field environment where all agro-climatic factors are not under control.

Leaf stress response: Many genotypes expressed very severe leaf stress responses (>60%) under drought, heat and combined drought and heat stress conditions (Fig. 2). The general behaviour of maize plants under combined drought and heat stress varied from one genotype to another. According to Suzuki *et al.*³⁹ recent studies have revealed that the response of plants to combinations of two or more stress conditions is unique and cannot be directly extrapolated from the response of plants to each of the different stresses applied individually. However, Obata *et al.*³⁵ reported that the combination of drought and heat evoked relatively few

specific responses and most of the metabolic changes were predictable from the sum of the responses to individual stresses. The results could be specific to the traits collected in the study. Based on leaf stress response, the most tolerant genotype was L6-Y in all the stress environments which gave high percentage of yield response across environments. The most susceptible genotype was ATP SR Y. The tolerance of a genotype to heat and drought stresses alone did not confer tolerance to combined drought and heat stress. On the contrary, tolerance to combined drought and heat stress of maize genotypes was genetically distinct from tolerance to individual stresses but with some similarities. These results differed from the findings of Cairns *et al.*²⁹ who did not find any similarities among maize inbred lines screened under the same three stresses. The difference could be due to the fact that the behaviour of a given maize genotype under a stress environment is not predictable and it differs from one genotype to another. More advanced research on proteomics and or metabolomics could provide clarification on the genotype response to each of the environmental stress.

Stress tolerance index: The stress tolerance index was common to all stress environments and was useful in estimating the tolerance of genotypes. This index was very low in combined heat and drought stress compared to heat stress and drought stress alone (Fig. 3), meaning that the stress effect was more severe in the combined drought and heat stress environment. The effect of combined drought and heat stress was considerably stronger than those of the individual stress factors. Similar results have been obtained with combined drought and heat stress under different environments in previous researches^{5,39-41}. Mahrookashani *et al.*⁴² found in his study

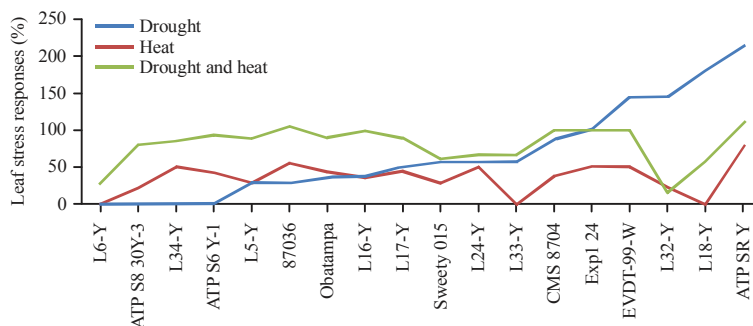


Fig. 2: Percentage variation of maize genotypes under drought, heat and combined drought and heat stress for leaf stress response

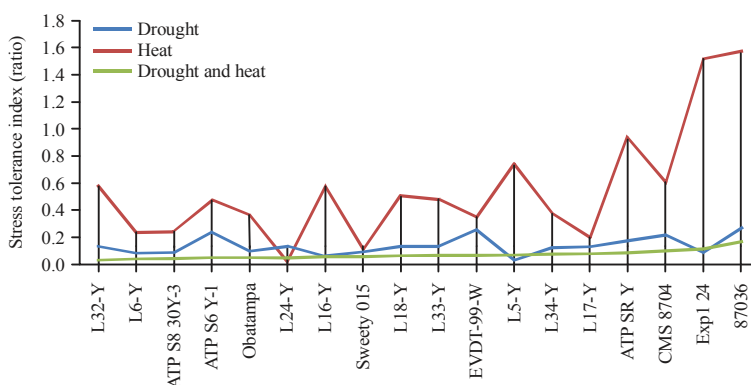


Fig. 3: Variation of stress tolerance indices of maize genotypes in different stress conditions at seedling stage

that the effects of combined heat and drought on traits were considerably stronger than those of the individual stress factors but the magnitude of the effects varied for specific growth traits. In the current study, maize varieties were less affected by heat stress alone compared to water stress and combined drought and heat stress.

Correlation between environments based on stress tolerance index: The stress tolerance index under heat stress alone was significantly high and positively correlated with the stress tolerance index under combined drought and heat stress (+0.79). A tolerant genotype under heat stress is likely to tolerate the combined drought and heat stress environment. However, the relationship of the stress tolerance index was not significant in heat stress alone, drought stress alone and between drought stress and combined drought and heat stress. This was similar to the findings by Cairns *et al.*²⁹ who reported no significant relationship between grain yield under drought stress and combined heat and drought stress among maize genotypes.

Correlation coefficients of stress indices with some parameters: Under combined drought and heat stress environment, shoot weight was significantly and positively correlated with leaf stress response (+0.55), plant height (+0.50), stress tolerance index (+0.73), heat stress index (+0.83) and drought tolerance index (+0.71) and modified stress tolerance index (+0.83) (Table 3). Leaf stress response was significantly and negatively correlated with chlorophyll content (-0.58). Under heat stress, shoot weight was significantly and positively correlated with plant height (+0.66), chlorophyll content (+0.46), stress tolerance index

(+0.91), heat tolerance index (+0.92) and modified stress tolerance index (+0.83) (Table 3). In the drought stress environment, shoot weight was significantly and positively correlated with stress tolerance index (+0.78), drought tolerance index (+0.44) and modified stress tolerance index (0.88) (Table 3). Shoot weight was highly and significantly correlated with all the indices in all the stress environments. Therefore, shoot weight and all the indices estimated in the study could be useful in maize screening maize at seedling stage under drought, heat and combined drought and heat stress. The significant and positive correlation between STI, HTI, DTI and MSTI indicated that these indices had the same capability in determining tolerance under stress. Similar results were obtained by Pfunde³⁶. The very high correlation between STI and MSTI showed that there is no need of using the two indices in one experiment.

Ranking of genotypes based on stress indices: The ranking of maize genotypes was conducted based on DTI in drought environment, HTI in heat stress condition, STI and MSTI in combined drought and heat stress condition (Table 4). L3-Y was found to be the most drought tolerant genotype, L5-Y was the most heat tolerant variety, whereas 87036 was the most tolerant genotype in combined drought and heat stress environment. According to Naghavi *et al.*⁴³ the identification of tolerant cultivars based on a single criterion may be contradictory. The indices of the current research study were estimated based on the shoot weight trait only. The use of indices involving all the parameters statistically significant in their calculation could be more reliable. The ranking of genotypes using the stress indices will then be more efficient and will reflect the observation in the field.

Table 3: Correlation coefficients with some parameters (leaf stress, plant height, chlorophyll content and shoot weight) and four indices under combined drought and heat stress, heat stress alone and drought stress condition

Parameters	Leaf stress	Plant height	Chlorophyll content	Shoot weight	STI	HTI	MSTI
Drought and heat							
Plant height(cm)	0.79***						
Chlorophyll_content (%)	-0.58*	-0.50*					
shoot_weight (g)	0.55*	0.50*	-0.14 ^{NS}				
STI	0.58*	0.52*	-0.24 ^{NS}	0.73***			
HTI	0.29 ^{NS}	0.18 ^{NS}	0.004 ^{NS}	0.83***	0.27 ^{NS}		
MSTI	0.41 ^{NS}	0.34 ^{NS}	-0.10 ^{NS}	0.83***	0.89***	-0.52*	
DTI	0.23 ^{NS}	0.14 ^{NS}	0.03 ^{NS}	0.71***	0.07 ^{NS}	0.96***	0.32 ^{NS}
Heat							
Plant_height (cm)	0.24 ^{NS}						
Chlorophyll_content(%)	-0.04 ^{NS}	0.25 ^{NS}					
Shoot_weight (g)	0.16 ^{NS}	0.66***	0.46*				
STI	0.22 ^{NS}	0.68***	0.32 ^{NS}	0.91***			
HTI	0.13 ^{NS}	0.56**	0.45*	0.92***	0.74***		
MSTI	0.30 ^{NS}	0.54**	0.31 ^{NS}	0.83***	0.93***	0.73***	
Drought							
Plant_height (cm)	0.49*						
Chlorophyll_content (%)	0.04 ^{NS}	0.37 ^{NS}					
Shoot_weight (g)	0.08 ^{NS}	-0.03 ^{NS}	-0.31 ^{NS}				
STI	0.30 ^{NS}	0.31 ^{NS}	0.02 ^{NS}	0.78***			
MSTI	0.11 ^{NS}	0.05 ^{NS}	-0.36 ^{NS}	0.88***	0.86***		
DTI	-0.29 ^{NS}	-0.23 ^{NS}	-0.26 ^{NS}	0.44*	*0.1 ^{NS}	-	0.17 ^{NS}

***p<0.001, **p<0.01, *p<0.05, NS: Non-significant, STI: Stress tolerance index, HTI: Heat tolerance index, MSTI: Modified stress tolerance index, DTI: Drought tolerance index

Table 4: Ranking of maize genotypes based on stress indices

Genotype	HRI	R1	DTI	R2	STI	R3	MSTI	R4
L3-Y	0.01	18	4.54	1	/	/	/	/
L5-Y	0.62	1	0.39	18	0.07	7	0.12	3
L6-Y	0.04	15	0.58	17	0.04	17	0.01	17
L16-Y	0.18	10	0.37	19	0.05	12	0.03	14
L17-Y	0.03	17	1.01	11	0.08	5	0.11	5
L18-Y	0.18	9	1.01	8	0.06	9	0.06	9
L24-Y	0.00	20	1.01	12	0.05	13	0.02	15
L32-Y	0.23	7	1.01	7	0.03	18	0.01	18
L33-Y	0.16	11	1.00	9	0.06	10	0.06	10
L34-Y	0.30	5	2.00	3	0.07	6	0.20	2
87036	0.52	2	0.91	13	0.17	1	0.53	1
88069	0.03	16	1.01	10	/	/	/	/
Exp1 24	0.33	3	0.25	20	0.11	2	0.11	4
Sweety 015	0.01	19	0.68	16	0.05	11	0.04	13
ATP S6 Y-1	0.13	12	1.67	5	0.04	15	0.02	16
ATP S8 30Y-3	0.21	8	2.17	2	0.04	16	0.06	11
CMS 8704	0.07	14	0.69	15	0.10	3	0.10	6
ATP SR Y	0.31	4	0.85	14	0.08	4	0.08	7
Obatampa	0.26	6	1.42	6	0.05	14	0.05	12
EVDT-99-W	0.08	13	1.83	4	0.07	8	0.07	8

DTI: Drought tolerance index, R: Ranking, HTI: Heat tolerance index, STI: Stress tolerance index, MSTI: Modified stress tolerance index

Clustering of genotypes based on morphological response under stress environments: The maize genotypes evaluated were grouped based on the morphological data collected under drought, heat and combined drought and heat stress conditions (Fig. 4). In each stress environment, genotypes were clustered in three groups and there was variation of

genotypes within each group. Group I consisted of the most susceptible genotypes, group II had intermediate responses while group III consisted of the tolerant genotypes. The most tolerant genotypes in all environments were L6-Y, L24-Y and Sweety 015. On the other hand, the most susceptible genotypes were 87036 and ATP SR Y, all of which were

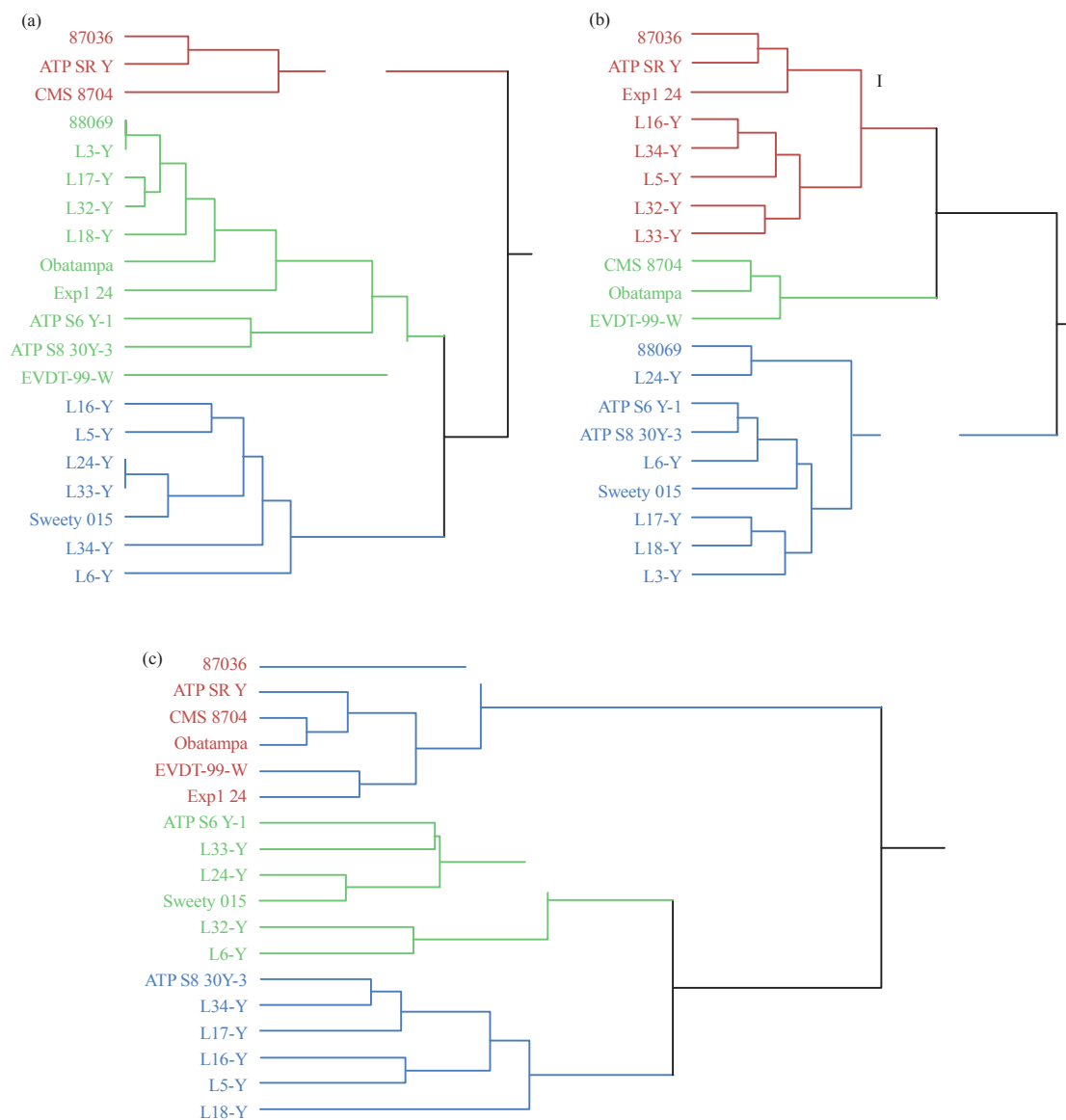


Fig. 4: Hierarchical clustering of maize genotypes evaluated under (a) Drought stress, (b) Heat stress and (c) Combined drought and heat stress at seedling stage

I: Susceptible genotypes; II: Moderately tolerant genotypes; III: Tolerant genotypes

introduced from Cameroon. These introduced genotypes were not bred for tolerance to environmental stresses investigated in this study.

CONCLUSION

Three inbred lines (L6-Y, L24-Y and Sweety 015) expressed relatively good performance across the stress environments and their tolerance should be confirmed under field conditions. Shoot weight, plant height and chlorophyll content could be used as secondary traits for maize screening

at early stages under stress conditions. Tolerant genotypes to heat stress are likely to tolerate combined drought and heat stress. Therefore, confirmation need to be done in open environment.

SIGNIFICANCE STATEMENT

The present study which gives an overview of maize responses to drought, heat and combined drought and heat stresses at early stage of plant development showed three inbred lines (L6-Y, L24-Y and Sweety 015) expressing good

performance under stress conditions and this was not yet explored by previous study. The selected stress tolerant genotypes must be screened in open stress environments and the tolerance mechanisms need to be clearly defined by researchers.

ACKNOWLEDGMENT

The National Research Foundation (NRF) of South Africa is acknowledged for funding this research through the Research and Technology Fund (RTF) grant number 98706.

REFERENCES

1. Ashraf, M., N.A. Akram, F. Al-Qurainy and M.R. Foolad, 2011. Drought tolerance: Roles of organic osmolytes, growth regulators and mineral nutrients. *Adv. Agron.*, 111: 249-296.
2. Ahmad, P., M. Ashraf, M. Younis, X. Hu, A. Kumar, N.A. Akram and F. Al-Qurainy, 2012. Role of transgenic plants in agriculture and biopharming. *Biotechnol. Adv.*, 30: 524-540.
3. Ashraf, M., 2014. Stress-induced changes in wheat grain composition and quality. *Crit. Rev. Food Sci. Nutr.*, 54: 1576-1583.
4. Qadir, S., S. Jamshieed, S. Rasool, M. Ashraf, N.A. Akram and P. Ahmad, 2014. Modulation of plant growth and metabolism in cadmium-enriched environments. *Arch. Environ. Contam. Toxicol.*, 229: 51-88.
5. Lipiec, J., C. Doussan, A. Nosalewicz and K. Kondracka, 2013. Effect of drought and heat stresses on plant growth and yield: A review. *Int. Agrophys.*, 27: 463-477.
6. Hasanuzzaman, M., K. Nahar, M.M. Alam, R. Roychowdhury and M. Fujita, 2013. Physiological, biochemical and molecular mechanisms of heat stress tolerance in plants. *Int. J. Mol. Sci.*, 14: 9643-9684.
7. Brosche, M., T. Blomster, J. Salojarvi, F. Cui and N. Sipari *et al.*, 2014. Transcriptomics and functional genomics of ROS-induced cell death regulation by radical-induced cell death. *PLoS Genet.*, Vol. 10, No. 2. 10.1371/journal.pgen.1004112.
8. Lobell, D.B., M. Banziger, C. Magorokosho and B. Vivek, 2011. Nonlinear heat effects on African maize as evidenced by historical yield trials. *Nat. Climate Change*, 1: 42-45.
9. Rahman, S.U., A. Muhammad, K. Hussain, M. Arshad, S. Hussain, T. Mukhtar and A. Razaq, 2015. Breeding for heat stress tolerance of maize in Pakistan. *J. Environ. Agric. Sci.*, 5: 27-33.
10. Guy, C., 1999. Molecular responses of plants to cold shock and cold acclimation. *J. Mol. Microbiol. Biotechnol.*, 1: 231-242.
11. Laude, H.M. and B.A. Chaugule, 1953. Effect of stage of seedling development upon heat tolerance in bromegrasses. *J. Range Manage.*, 6: 320-324.
12. Buriro, M., F.C. Oad, M.I. Keerio, S. Tunio, A.W. Gandahi, S.W.U. Hassan and S.M. Oad, 2011. Wheat seed germination under the influence of temperature regimes. *Sarhad J. Agric.*, 27: 539-543.
13. Sanchez, B., A. Rasmussen and J.R. Porter, 2014. Temperatures and the growth and development of maize and rice: A review. *Global Change Biol.*, 20: 408-417.
14. Meseka, S., A. Menkir, O. Bossey and O. Azeez, 2015. Evaluation of advanced drought tolerant maize (*Zea mays* L.) hybrids for grain yield and other agronomic traits under combined drought and heat stress. Proceedings of the Annual Meeting on Synergy in Science: Partnering for Solutions, November 15-18, 2015, Minneapolis, MN.
15. Willenborg, C.J., R.H. Gulden, E.N. Johnson and S.J. Shirliffe, 2004. Germination characteristics of polymer-coated canola (*Brassica napus* L.) seeds subjected to moisture stress at different temperatures. *Agron. J.*, 96: 786-791.
16. Rauf, M., M. Munir, M. Hassan, M. Ahmad and M. Afzal, 2007. Performance of wheat genotypes under osmotic stress at germination and early seedling growth stage. *Afr. J. Biotechnol.*, 6: 971-975.
17. Khayatnezhad, M., R. Gholamin, S. Jamaati-e-Somarin and R. Zabihi-e-Mahmoodabad, 2010. Effects of peg stress on corn cultivars (*Zea mays* L.) at germination stage. *World Applied Sci. J.*, 11: 504-506.
18. Adebayo, A.M., A. Menkir, E. Blay, V. Gracen and E. Danquah, 2017. Combining ability and heterosis of elite drought-tolerant maize inbred lines evaluated in diverse environments of lowland tropics. *Euphytica*, 213: 43-43.
19. Partheeban, C., C.N. Chandrasekhar, P. Jeyakumar, R. Ravikesavan and R. Gnanam, 2017. Effect of PEG induced drought stress on seed germination and seedling characters of maize (*Zea mays* L.) genotypes. *Int. J. Curr. Microbiol. Applied Sci.*, 6: 1095-1104.
20. FAO., 2013. Crop water information: Maize. Food and Agriculture Organization. http://www.fao.org/nr/water/crop_info-maize.html
21. Fischer, K.S., G.O. Edmeades and E.C. Johnson, 1989. Selection for the improvement of maize yield under moisture-deficits. *Field Crops Res.*, 22: 227-243.
22. Rosen, S. and L. Scott, 1992. Famine grips Sub-Saharan Africa. *Agric. Outlook*, 191: 20-24.
23. Meeks, M., S.C. Murray, S. Hague and D. Hays, 2013. Measuring maize seedling drought response in search of tolerant germplasm. *Agronomy*, 3: 135-147.

24. Tadross, M., P. Suarez, A. Lotsch, S. Hachigonta and M. Mdoka *et al.*, 2007. Changes in growing-season rainfall characteristics and downscaled scenarios of change over Southern Africa: Implications for growing maize. Proceedings of the IPCC Regional Expert Meeting on Regional Impacts, Adaptation, Vulnerability and Mitigation, June 2007, Nadi, Fiji, pp: 193-204.
25. Ciscar, J.C., 2012. The impacts of climate change in Europe (the PESETA research project). *Climatic Change*, 112: 1-6.
26. Engelbrecht, F., J. Adegoke, M.J. Bopape, M. Naidoo and R. Garland *et al.*, 2015. Projections of rapidly rising surface temperatures over Africa under low mitigation. *Environ. Res. Lett.*, Vol. 10, No. 8. 10.1088/1748-9326/10/8/085004/meta.
27. Ahmad, P., A.A.H. Abdel Latef, S. Rasool, N.A. Akram, M. Ashraf and S. Gucl, 2016. Role of proteomics in crop stress tolerance. *Front. Plant Sci.*, Vol. 7. 10.3389/fpls.2016.01336.
28. Voesenek, L.A.C.J. and R. Pierik, 2008. Plant stress profiles. *Science*, 320: 880-881.
29. Cairns, J.E., J. Crossa, P.H. Zaidi, P. Grudloyma and C. Sanchez *et al.*, 2013. Identification of drought, heat and combined drought and heat tolerant donors in maize. *Crop Sci.*, 53: 1335-1346.
30. Mittler, R., 2006. Abiotic stress, the field environment and stress combination. *Trends Plant Sci.*, 11: 15-19.
31. Musa, U.T. and T.H. Usman, 2016. Leaf area determination for maize (*Zea mays* L.), okra (*Abelmoschus esculentus* L.) and cowpea (*Vigna unguiculata* L.) crops using linear measurements. *J. Biol. Agric. Healthc.*, 6: 103-111.
32. Fischer, R.A. and R. Maurer, 1978. Drought resistance in spring wheat cultivars. I. Grain yield responses. *Aust. J. Agric. Res.*, 29: 897-912.
33. Fernandez, G.C.J., 1992. Effective selection criteria for assessing plant stress tolerance. Proceedings of the International Symposium on Adaptation of Vegetables and other Food Crops in Temperature and Water Stress, August 13-16, 1992, Shanhua, Taiwan, pp: 257-270.
34. Farshadfar, E., M. Zamani, M. Motallebi and A. Imamjomeh, 2001. Selection for drought resistance in chickpea lines. *Iran. J. Agric. Sci.*, 32: 65-77.
35. Obata, T., S. Witt, J. Lisec, N. Palacios-Rojas and I. Florez-Sarasa *et al.*, 2015. Metabolite profiles of maize leaves in drought, heat and combined stress field trials reveal the relationship between metabolism and grain yield. *Plant Physiol.*, 169: 2665-2683.
36. Pfunde, C.N., 2016. Investigation into the genetic diversity, physiology, proteomics and combining ability of quality protein maize inbred lines under drought and heat stress. Ph.D. Thesis, University of Fort Hare, South Africa.
37. Fahad, S., A.A. Bajwa, U. Nazir, S.A. Anjum and A. Farooq *et al.*, 2017. Crop production under drought and heat stress: Plant responses and management options. *Front. Plant Sci.*, Vol. 8. 10.3389/fpls.2017.01147.
38. Yadav, S.K., Y.K. Tiwari, D.P. Kumar, A.K. Shanker, N.J. Lakshmi, M. Vanaja and M. Maheswari, 2016. Genotypic variation in physiological traits under high temperature stress in maize. *Agric. Res.*, 5: 119-126.
39. Suzuki, N., R.M. Rivero, V. Shulaev, E. Blumwald and R. Mittler, 2014. Abiotic and biotic stress combinations. *New Phytol.*, 203: 32-43.
40. Dreesen, F.E., H.J. de Boeck, I.A. Janssens and I. Nijs, 2012. Summer heat and drought extremes trigger unexpected changes in productivity of a temperate annual/biannual plant community. *Environ. Exp. Bot.*, 79: 21-30.
41. Rollins, J.A., E. Habte, S.E. Templer, T. Colby, J. Schmidt and M. von Korff, 2013. Leaf proteome alterations in the context of physiological and morphological responses to drought and heat stress in barley (*Hordeum vulgare* L.). *J. Exp. Bot.*, 64: 3201-3212.
42. Mahrookashani, A., S. Siebert, H. Hugging and F. Ewert, 2017. Independent and combined effects of high temperature and drought stress around anthesis on wheat. *J. Agron. Crop Sci.*, 203: 453-463.
43. Naghavi, M.R., A.P. Aboughadareh and M. Khalili, 2013. Evaluation of drought tolerance indices for screening some of corn (*Zea mays* L.) cultivars under environmental conditions. *Not. Scient. Biol.*, 5: 388-393.