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Phenotypic evaluation of a set of selected exotic maize inbred lines for drought stress tolerance

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Abstract

Increased rainfall variability accentuated by climate change will have severe effects on production of maize which is grown predominantly as a rainfed crop in India. A set of 31 exotic inbred lines were analysed under both well-watered (controlled) and water-deficit stress (at flowering stage) conditions at the IARI Experimental Farm, New Delhi (during *Kharif* 2007), and IARI Regional Station, Pusa-Bihar (during *Rabi* 2007-08). Significant variation among the genotypes were observed for grain yield per plant, anthesis-silking interval (ASI), number of ears per plot, total number of leaves per plant and leaf senescence, under both well-watered and drought-stressed conditions. Significant effects of locations/seasons and environments, besides location/season x genotype and environment x genotype interactions for most of the drought component traits were analysed. The study led to the identification of a few inbreds, such as DTPYC9-F46-1-2-1-2-B, CML341, CML340 and CMLP2 at Delhi, and CML340, LPSC7-F64-2-6-2-2-B-B, LPSC7-F71-1-2-1-1-B-B, CMl360 and CML341 at Pusa (Bihar) as promising drought-tolerant genotypes. The study also established the negative association of ASI with ear per plot as well as with grain yield per plant under water stress conditions at both the locations. Leaf senescence was also found to be negatively correlated with ear per plot and grain yield per plant, thereby confirming the usefulness of lower ASI and leaf senescence as the potential secondary traits for breeding under drought stress conditions in rainfed maize.

Key words: Maize, drought stress, anthesis-silking interval (ASI), leaf senescence, grain yield

Introduction

The most important abiotic stress affecting maize crop production worldwide is water deficit which in turn is becoming one of the topmost constraints affecting the production and productivity of maize in several developing countries worldwide, especially in Africa and Asia [1]. Over 90% of maize grown in the tropics is managed without irrigation either due to unavailability of irrigation or due to financial constraints. Demand for maize in Asia is increasing significantly because of per capita GDP (gross domestic product) growth and also with increasing demand for feed and biofuel [2]. India, being the second largest maize growing country amongst "Asia-7", offers innumerable challenges and opportunities to increase regional maize production [3]. One of the major limiting factors for maize production and productivity in India is inadequate soil moisture particularly during flowering and grain filling stages [4]. In India, ~80% of total area under maize is rainfed and prone to drought stress conditions. In comparison to the national and global averages, yield levels of rainfed maize is significantly lower in the traditional maize growing areas of India, particularly in the states of Rajasthan, Madhya Pradesh, Uttar Pradesh and Bihar. The unpredictability of normal rainfall, across seasons and regions, thus increased the importance of drought tolerance as a major breeding objective.

Breeding for drought tolerance in maize is primarily aimed at identifying genotypes with optimal reproductive capacity with low yield penalty under drought-stress conditions (especially at flowering stage) as compared to well-watered conditions; this is further evaluated by assessing the stability of promising genotypes by conducting multi-environment drought trials. Selection for grain yield alone under drought has also often been considered inefficient because of the increase in

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environmental variance relative to genetic variance, which decreases yield heritability as yield decreases. Under these conditions, selection for secondary traits which are correlated to grain yield and have relatively high heritability may increase selection efficiency [5]. Progress in grain yield under drought stress has been achieved through selection for component traits such as reduced anthesis-silking interval (ASI), reduced barrenness and improved kernel set [6]. Banziger *et al.* [7] recommended the use of grain yield, ears per plant, ASI, leaf senescence, tassel size and leaf rolling as important criteria for breeding for drought tolerance in maize. Putative drought tolerance traits have been reviewed extensively, although very few have proven useful in drought breeding programmes. Generally, number of ears per plant, kernels per plant and anthesis-silking intervals (ASI) are considered as the most important drought adaptive traits, followed by tassel branch number, leaf senescence and plant height [8]. The present study, therefore, was aimed at evaluating a set of exotic genotypes at two different locations in India to identify potential drought tolerant genotypes, analyse the role of genotype x environment (G x E) interactions and assess the utility of specific secondary traits in breeding for drought tolerance.

Materials and methods

The genetic materials selected for phenotyping under drought stress included a set of 31 inbreds, including lines obtained from CIMMYT (International Maize and Wheat Improvement Center), Mexico; INRA, Montpellier, France; and Kenya Agricultural Research Institute (KARI), Nairobi. The drought trials were carried out in the IARI Experimental Farms at two locations namely New Delhi [during *Kharif* (monsoon season)-2007] and Pusa-Bihar [during *Rabi* (dry/winter) 2007-08], each having two different water treatments/environments (i) well-watered (WW) and (ii) water-stressed (WS) conditions. The trials were undertaken under a randomized block design (RBD) with two replicates under WW and WS conditions at each location. Each plot consisted of single row of 3m length, with 75 cm row-to-row spacing and 20 cm plant-to-plant spacing. An average of 15 plants per row was maintained by thinning**.** The irrigation schedules for the well-watered (WW) and water stressed (WS) blocks were same till three weeks before flowering. The irrigation was stopped three weeks before flowering in the stress block, and no irrigation was applied until mid- or late grain filling stages, when an additional irrigation was given in the stress block. The procedure employed for imposing water deficit stress was same as that suggested by Banziger *et al.* [7].

The meteorological data, including minimum and maximum temperatures, relative humidity (RH) and rainfall, were collected throughout the experimental period at both locations (data not presented), as suggested by Banziger *et al.* [7]. The *Rabi* or dry season at Pusa-Bihar was free of any rainfall. At Delhi, there was no rainfall just before or during the flowering during *Kharif* season, and thus, there was no interference from rainfall from the viewpoint of managed stress trial.

For statistical analysis, the effects of ‘location’ and ‘environment’ were considered as ‘fixed’ as Delhi and Pusa-Bihar do not completely represent the range of drought-prone maize growing-environments in the country, and the term ‘environment’ here is specific, since drought at ‘pre-flowering to flowering stage’ is different from drought at ‘seedling stage’ or ‘early vegetative stage’ in case of maize. The effect of ‘genotypes’, in contrast, was considered as ‘random’ as the genotypes analysed in the study include exotic inbred lines that have been derived from diverse pedigrees and selected on the basis of their phenotypic responses under managed drought stress trials at different locations (Mexico and African countries) by the Global Maize Program of CIMMYT.

The traits recorded in WW and WS blocks included (i) total number of leaves, (ii) anthesis-silking interval (ASI), which is the differential of the days to 50% anthesis and days to 50% silking, (iii) senescence at two different dates - first at flowering stage and second at grain filling stage, (iv) number of ears per plot and (v) grain yield per plant. The border 2-3 plants in each plot were not considered for data recording. The total number of green leaves was counted at the time of flowering, particularly when the anthesis has taken place. The days to 50% anthesis was recorded as the number of days taken from the date of sowing to the date of first appearance of anthers in 50% of plants in a plot. In case of days to 50% silking, number of days taken from sowing till emergence of silks in 50 % of the plants in a plot, were noted. Leaf senescence was recorded at two different dates, first at flowering stage and second at grain filling stage, following the protocol described by Banziger *et al.* [7]. The number of ears per plot was measured on the basis of total number of ears harvested in each plot, excluding the border plants. For grain yield per plant, all the plants in the plot except the border plants were harvested and fresh dehusked ears were weighed. Data thus generated were analyzed using WINDOSTAT version 8.5 software for various statistical parameters. Pearson's simple correlation coefficients were estimated among various morpho-physiological traits, as per standard procedure.

Results and discussion

Analysis of Variance (ANOVA) revealed significant differences among the inbred lines for majority of the traits at both Delhi and Pusa-Bihar (Table not presented), indicating the presence of wide genetic variation amenable for breeding for drought tolerance. Several researchers [9-11] also reported presence of significant genetic variation for traits such as grain yield and its component traits under drought stress conditions. The results also showed the importance of locations/ seasons, environments, location/season x treatment and environment x treatment interaction for almost all the characters (Table 1). This is evident from the fact that, LPSC7-F64-2-6-2-2-B-B and LPSC7-F71-1-2-1-1-B-B produced 11.5g and 10.5g yield/plant, respectively, under stress conditions at Pusa-Bihar, while the same genotypes were found to be highly susceptible to drought stress at Delhi, with only 3.0g and 2.8g yield/ plant, respectively. In contrast, DTPYC9-F46-1-2-1-2-B performed well under drought stress conditions at Delhi, while the same showed high susceptibility at Pusa-Bihar. Significant effect of G x E for grain yield and other important traits such as days to 50% anthesis and number of ears per plant under drought stress conditions were also reported earlier [11-13].

The mean grain yield per plant varied from 2.7 to 61.3 g under normal conditions at Delhi, while it was found to be 0 to 11.2 g under water stress conditions (Table 2); the same for Pusa-Bihar experiment was found to be 3.2 to 171.00 g and 0 to 14.5 g under well-watered and drought stress conditions, respectively. The mean grain yield per plant at Delhi under well-watered conditions was recorded to be 19.6 g, whereas the same was 3.0 g under water-stressed conditions. The effect of drought stress on grain yield was also prominent at Pusa-Bihar, as the average grain yield per plant reduced to 3.9 g under drought conditions, as compared to 33.0 g under normal conditions.

Although majority of the genotypes experienced high grain yield loss (as high as 100%) due to water stress conditions at both the locations, DTPYC9-F46-1-2-1-2-B at Delhi was found to be the most promising genotype, producing 11.2g grain yield per plant under drought stress conditions, followed by CML341 (10.0g) and CML340 (9.1g). Among the genotypes analyzed at Pusa-Bihar, CML340 (14.5g), LPSC7-F64-2-6-2-2-B-B (11.5g) and LPSC7-F71-1-2-1-1-B-B (10.5g) were identified as the best genotypes under water stress conditions. However, some genotypes like CML340 and CML341 were found to be stable and promising at both the locations under stress conditions.

The effect of drought stress was pronounced in almost all the genotypes since the ear formation was drastically reduced due to water stress conditions. The average number of harvested ears per plot was found to be 13.1 under well-watered conditions at Delhi, while it was 2.0 under the drought stress conditions (Table 2). The range was found to vary from 11 to 15 and 0 to 5.5 ears per plot under normal and water-stress conditions, respectively. Similar trend was also observed in experiment conducted at Pusa-Bihar. Although many of the genotypes produced nearly no ears (barren plants) at harvest at both the locations, CML254, CML247, CML340, CML341 and CMLP2 at Delhi, and DTPWC9-F104-5-4-1-1-B-B and DTPWC9-F115-1-2-1-2-B-B at Pusa-Bihar, were found to produce moderate number of ears per plot. The number of ears per plant is generally

Table 1. ANOVA of different traits under both controlled and drought stressed conditions at Delhi and Pusa-Bihar

Source of variation	d.f.	Total No. of leaves	Anthesis silking interval	Senescence during flowering	Senescence during grain filling	Ear per plot	Grain yield per plant
Locations/Seasons	1	37.01**	543.10**	1892.04**	3992.04**	1.17	3140.42**
Environments	1	67.31**	820.17**	7186.39**	40647.68**	6350.90**	32434.50**
Genotypes	30	2.61**	41.77**	245.91**	1129.11**	5.29**	714.03**
Replications	1	0.47	0.68	4.94	304.94	3.88	10.44
Location/Season x Treatment	30	2.07**	50.64**	198.70**	1247.87**	6.46**	451.18**
Environment x treatment	30	0.95**	17.72**	95.14**	226.85**	6.07**	637.20**
Error	154	0.52	5.02	41.54	117.44	3.21	101.74

,* Significant at P = 0.05 and P = 0.01, respectively.

Table 2. Mean performance of the selected maize inbreds at Delhi and Pusa-Bihar under control (well-watered) and drought stress conditions

Genotypes	TNL				ASI				SF				SG				EPP				GYP			
	Control		Stress		Control		Stress		Control		Stress		Control		Stress		Control		Stress		Control		Stress	
	D	B	D	B	D	B	D	B	D	B	D	B	D	B	D	B	D	B	D	B	D	B	D	B
CML245	**12.8**	9.1	**11.2**	9.2	**3.0**	6.0	**15.5**	6.5	**7.5**	25.0	**52.5**	22.5	**45.0**	47.5	**100.0**	75.0	**11.5**	9.0	**0.0**	0.0	**61.3**	60.2	**0.0**	0.0
CML247	**13.8**	12.2	**12.4**	11.3	**3.5**	3.5	**5.5**	6.5	**2.5**	15.0	**5.0**	22.5	**20.0**	55.0	**47.5**	75.0	**14.0**	6.5	**4.0**	0.0	**14.6**	8.8	**2.6**	0.0
CML254	**14.7**	12.2	**12.0**	10.8	**6.5**	2.0	**8.5**	5.0	**2.5**	15.0	**10.0**	20.0	**30.0**	35.0	**47.5**	75.0	**13.0**	13.5	**4.5**	3.5	**24.7**	18.5	**6.1**	1.3
CML287	**12.6**	11.6	**14.3**	11.2	**3.5**	5.0	**3.0**	6.5	**0.0**	15.0	**5.0**	22.5	**10.0**	35.0	**20.0**	75.0	**14.0**	13.5	**1.5**	2.0	**30.1**	171.0	**1.8**	4.5
CML312	**13.5**	13.2	**11.8**	11.3	**20.0**	2.5	**23.0**	7.5	**12.5**	15.0	**30.0**	25.0	**50.0**	35.0	**85.0**	47.5	**11.0**	13.5	**0.0**	0.5	**6.6**	7.6	**0.0**	0.7
CML333	**12.5**	12.3	**11.5**	12.1	**2.0**	1.5	**2.0**	4.5	**5.0**	10.0	**22.5**	25.0	**20.0**	35.0	**50.0**	50.0	**12.5**	10.5	**1.0**	4.0	**24.6**	36.0	**1.5**	2.7
CML339	**13.7**	12.1	**12.9**	11.8	**2.0**	2.5	**3.0**	8.5	**0.0**	15.0	**0.0**	25.0	**10.0**	52.5	**17.5**	75.0	**14.0**	13.0	**2.0**	0.0	**9.5**	10.2	**1.7**	0.0
CML340	**13.2**	12.0	**12.3**	11.7	**4.5**	2.0	**8.5**	5.5	**0.0**	5.0	**5.0**	20.0	**10.0**	25.0	**20.0**	67.5	**13.5**	11.5	**5.0**	5.0	**33.7**	27.9	**9.1**	14.5
CML341	**12.6**	12.2	**12.8**	11.8	**5.0**	2.5	**5.0**	3.5	**0.0**	15.0	**5.0**	20.0	**10.0**	32.5	**27.5**	40.0	**12.5**	11.0	**4.0**	5.5	**33.3**	23.5	**10.0**	7.0
CML344	**13.6**	14.1	**11.0**	13.1	**5.0**	1.0	**27.5**	4.5	**17.5**	15.0	**40.0**	22.5	**75.0**	37.5	**100.0**	50.0	**14.5**	14.0	**0.0**	4.0	**12.6**	12.1	**0.0**	3.3
CML360	**13.5**	11.9	**12.6**	9.9	**4.5**	4.5	**7.5**	5.5	**2.5**	15.0	**42.5**	22.5	**15.0**	45.0	**72.5**	72.5	**14.0**	13.5	**1.0**	5.0	**10.3**	33.3	**3.3**	8.3
CML389	**11.5**	12.6	**10.6**	12.1	**10.0**	2.0	**21.5**	4.5	**0.0**	15.0	**10.0**	20.0	**27.5**	20.0	**35.0**	60.0	**12.5**	13.5	**0.0**	2.5	**18.3**	19.8	**0.0**	3.5
CML442/CML197/TUXPSEQ	**13.7**	12.6	**12.3**	12.2	**3.0**	5.5	**4.0**	5.0	**0.0**	15.0	**0.0**	20.0	**10.0**	47.5	**10.0**	75.0	**13.5**	14.5	**2.5**	4.5	**33.3**	32.9	**3.8**	6.3
CML444	**14.3**	13.6	**11.5**	10.4	**5.0**	3.5	**5.0**	5.0	**0.0**	10.0	**7.5**	15.0	**5.0**	35.0	**32.5**	47.5	**13.5**	13.0	**3.0**	3.0	**6.3**	18.7	**4.6**	4.2
CML69	**13.2**	12.7	**11.2**	12.0	**4.5**	2.0	**27.0**	5.5	**2.5**	15.0	**47.5**	20.0	**25.0**	45.0	**100.0**	70.0	**14.5**	11.5	**0.0**	2.5	**8.7**	39.6	**0.0**	2.7
CMLP1	**14.2**	13.0	**14.7**	12.2	**5.0**	2.5	**7.0**	5.5	**10.0**	10.0	**27.5**	15.0	**50.0**	35.0	**77.5**	75.0	**13.0**	14.5	**0.0**	3.5	**6.9**	6.9	**0.0**	0.3
CMLP2	**12.9**	12.7	**11.5**	12.0	**4.5**	2.0	**6.5**	7.5	**5.0**	5.0	**7.5**	25.0	**25.0**	17.5	**50.0**	45.0	**12.5**	14.0	**5.5**	1.5	**22.9**	28.0	**7.7**	1.5
DTPWC9-F104-5-4-1-1-B-B	**13.0**	11.7	**11.4**	11.0	**4.0**	2.5	**11.5**	3.5	**0.0**	15.0	**32.5**	17.5	**10.0**	35.0	**85.0**	40.0	**14.5**	12.0	**0.0**	6.5	**20.3**	16.8	**0.0**	5.8
DTPWC9-F115-1-2-1-2-B-B	**12.9**	13.8	**10.6**	12.2	**5.5**	2.0	**10.5**	3.5	**2.5**	5.0	**5.0**	25.0	**20.0**	15.0	**30.0**	55.0	**13.5**	8.0	**1.0**	6.5	**33.0**	41.7	**1.8**	6.3
DTPWC9-F31-1-3-1-1-B-B	**12.7**	12.1	**11.1**	11.7	**4.5**	1.5	**9.5**	4.5	**0.0**	5.0	**7.5**	25.0	**20.0**	15.0	**40.0**	50.0	**13.0**	8.5	**3.5**	2.5	**13.0**	20.3	**3.3**	3.2
DTPYC9-F46-1-2-1-2-B	**13.4**	12.1	**12.0**	11.6	**3.5**	8.5	**6.5**	8.5	**2.5**	5.0	**47.5**	10.0	**15.0**	30.0	**75.0**	50.0	**11.5**	12.0	**4.0**	2.0	**26.9**	29.6	**11.2**	1.7
DTPYC9-F74-1-1-1-1-B-B	**12.0**	12.0	**11.4**	11.2	**2.5**	1.5	**4.0**	7.5	**15.0**	15.0	**27.5**	25.0	**85.0**	40.0	**100.0**	50.0	**13.5**	11.5	**0.0**	3.5	**41.4**	36.5	**0.0**	5.8
H16	**13.5**	12.4	**12.4**	11.1	**3.5**	1.5	**6.0**	6.5	**5.0**	15.0	**10.0**	20.0	**30.0**	52.5	**50.0**	70.0	**11.5**	12.5	**3.5**	1.5	**5.4**	34.4	**3.4**	0.8
K64R	**13.7**	13.9	**12.9**	12.0	**3.5**	3.0	**2.5**	6.5	**5.0**	15.0	**32.5**	25.0	**30.0**	35.0	**62.5**	50.0	**11.0**	14.5	**0.0**	1.5	**18.5**	28.5	**0.0**	0.7
KU13	**12.0**	12.2	**10.7**	9.8	**3.5**	1.0	**5.0**	4.5	**5.0**	5.0	**5.0**	10.0	**20.0**	35.0	**27.5**	50.0	**13.5**	13.5	**3.0**	0.0	**2.7**	3.2	**2.4**	0.0
LPSC7-F64-2-6-2-2-B-B	**13.9**	12.9	**13.2**	12.0	**13.5**	3.0	**14.0**	4.5	**20.0**	15.0	**37.5**	15.0	**75.0**	22.5	**75.0**	45.0	**12.0**	12.5	**1.5**	5.0	**11.2**	76.7	**3.0**	11.5
LPSC7-F71-1-2-1-1-B-B	**14.3**	12.0	**13.2**	11.7	**3.5**	1.5	**8.5**	4.5	**0.0**	5.0	**10.0**	20.0	**10.0**	27.5	**52.5**	75.0	**12.5**	9.5	**3.5**	4.5	**25.0**	36.0	**2.8**	10.5
LPSC7-F86-3-1-1-1-B-B-B	**14.2**	12.5	**12.9**	11.8	**5.0**	2.0	**8.0**	7.5	**0.0**	15.0	**10.0**	22.5	**25.0**	35.0	**47.5**	75.0	**12.0**	12.5	**1.0**	2.0	**7.6**	37.5	**1.2**	5.0
MAS[206/312]-23-2-1-1-B*6-B-B	**13.8**	11.7	**12.3**	10.8	**6.0**	2.0	**9.5**	6.5	**2.5**	15.0	**2.5**	17.5	**25.0**	32.5	**27.5**	57.5	**13.5**	11.0	**3.5**	1.0	**18.9**	26.2	**5.0**	3.0
SCMALAWI	**13.7**	12.6	**12.3**	11.8	**5.0**	2.5	**8.0**	6.5	**0.0**	15.0	**2.5**	15.0	**10.0**	20.0	**25.0**	62.5	**15.0**	13.5	**2.5**	1.0	**17.0**	29.6	**6.3**	0.6
ZM621A-10-1-1-1-2-B*7-B-B	**12.2**	12.2	**12.7**	11.5	**3.0**	1.5	**5.5**	7.5	**5.0**	17.5	**7.5**	25.0	**25.0**	42.5	**27.5**	75.0	**14.0**	10.0	**1.5**	2.5	**10.4**	51.5	**1.3**	4.8
Mean	**13.2**	12.4	**12.1**	11.4	**5.1**	2.7	**9.3**	5.8	**4.2**	12.7	**17.9**	20.5	**27.0**	34.6	**52.2**	60.7	**13.1**	12.0	**2.0**	2.8	**19.6**	33.0	**3.0**	3.9
LSD	**1.91**	0.47	**2.21**	0.39	**3.25**	2.21	**3.07**	1.02	**11.01**	6.37	**5.82**	4.54	**26.93**	7.50	**9.47**	13.10	**2.96**	3.00	**3.63**	1.54	**5.78**	13.19	**0.84**	3.08

TNL: total no. of leaves per plant; ASI: anthesis silking interval; SF: Senescence at flowering stage; SG: Senescence at grain filling stage; EPP: ear per plot and GYP: grain yield per plant; D: Delhi; B: Bihar; LSD: least significant difference determined by the supply of C and N prior to and during flowering stage. Determination of the number of kernels per ear occurs during the lag phase between pollination and the onset of the linear phase of grain biomass [14]. Both the number of potential ears per plant and kernels per ear are particularly sensitive to environmental stresses [15].

Selection for drought tolerance based on grain yield has often been considered inefficient [16], therefore selection based on secondary traits having high heritability increases efficiency of selection [17]. Among the various secondary traits, anthesis silking interval (ASI) is one of the most important traits associated with grain yield under drought stress in maize, and has been a leading choice linked to stress tolerance and reproduction in maize [18]. Maize under drought stress exhibits delay in silking resulting in an increase in the anthesis-to-silking interval (ASI), thereby resulting into incomplete or nil fertilization, and decreased or nil kernel development [19]. The ASI was found to vary from 2.0 to 20.0 days under normal conditions at Delhi, with mean ASI across genotypes being 5.1 (Table 2). The range became broader (2.0 to 27.5 days) under stress condition and the average ASI was found to be 9.3 days. The ASI varied from 1.0 to 8.5 days at Pusa-Bihar under control-conditions, while it was found to be 3.5 to 8.5 days under the stress conditions. The mean ASI under well-watered conditions was found to be 2.7 days and it was enhanced to 5.8 days under stress conditions. CML245 at Delhi, recorded ASI of 3 days under normal conditions, while it was 15.5 days under stressed conditions. At Pusa-Bihar, genotypes such as CML339 experienced highest increase in ASI value with 2.5 days under normal conditions and 8.5 days under stress conditions. Although, in general, majority of genotypes experienced enhanced ASI under drought stress conditions, genotypes such as CML287, CML333, CML339, CML442/CML197/TUXPSEQ, CML444 and K64R at Delhi and CML341, CML360, CML442/CML197/TUXPSEQ and DTPWC9-F104-5-4-1-1-B-B at Pusa-Bihar were identified as the promising genotypes showing relatively lesser effects of drought stress on ASI. Edmeades *et al*. [20] also reported that tropical maize populations improved for mid-season drought tolerance showed reduced ASI under drought.

An increase in ASI in maize indicates that by the time silk emerges, the pollen shedding is essentially over [21]. It was reported earlier that ASI with less or more than five days, reduces grain yield due to poor pollen supply [22]. In the present study too, most of the genotypes showing susceptibility to drought stress conditions had an ASI difference of >±5 days under drought condition. It has been observed by researchers worldwide that drought stress just before or during the flowering period leads to late silking and thereby increases length of the anthesis-silking interval (ASI). This asynchrony between male and female flowering has been recognized as a major source of low grain yield [23].

The mean leaf senescence during flowering stage was observed to be 4.2% under well-watered conditions, while it was 17.9% under the stress conditions at Delhi (Table 2). Genotypes such as CML287, CML339, CML340, CML341, CML389, CML442/CML197/TUXPSEQ, CML444, DTPWC9-F115-1-2-1-2-B-B, DTPWC9-F31-1-3-1-1-B-B, H16, KUI3, LPSC7-F71-1-2-1-1-B-B, LPSC7-F86-3-1-1-1-B-B-B, MAS[206/312]-23-2-1-1-B*6-B-B, SCMALAWI and ZM621A-10-1-1-1-2-B*7-B-B showed little leaf senescence under both the conditions. Some genotypes such as CML245 exhibited 7.5% leaf area having senescence under normal conditions, while it got enhanced to 52.5% under drought stress conditions. In Pusa-Bihar, the range of leaf senescence varied from 10-15% under drought stress conditions, with majority of genotypes showing parity between well-watered and drought stress conditions. Leaf senescence at grain filling stage further increased the leaf area with senescence. The range for leaf senescence was 5 to 85% under normal conditions, while it was 10 to 100% under drought stress conditions at Delhi. The mean leaf senescence area across genotypes was observed to be 27% under well-watered conditions, while it was 52.2% under drought stress conditions. Genotypes such as CML245, CML344, CML69 and DTPYC9-F74-1-1-1-1-B-B experienced 100% leaf area having senescence, while CML360, CMLP1, K64R, LPSC7-F71-1-2-1-1-B-B, LPSC7-F86-3-1-1-1-B-B-B, CML247 and CML254 exhibited nearly 50 to 80% leaf senescence. On the other hand, CML287, CML339, CML340 and CML442/CML197/TUXPSEQ were identified as the promising genotypes having lesser leaf senescence. Delayed leaf senescence and stay-green characters facilitates kernel growth and assimilate accumulation during later grain filling stage, resulting in more number of fully developed kernels per ears and higher kernel weight, and therefore, improved yields in drought tolerant lines across moisture regimes [24-26]. However, in the Pusa-Bihar experiment, the leaf area senescence at grain filling varied from 15 to 55% under well-watered conditions and it was 40 to 75% under the drought stress conditions. The mean leaf senescence was found to increase from 34.6% (under control conditions) to 60.7% (under water stress conditions). Genotypes such as CML245, CML247, CML254, CML287, CMLP1, LPSC7-F71-1-2-1-1-B-B, LPSC7-F86-3-1-1-1-B-B-B and ZM621A-10-1-1-1-2-B*7-B-B exhibited as high as 75% of the leaf area having senescence, with majority of the genotypes having more than 50% leaf senescence under drought stress conditions.

Maize varieties that produce more number of leaves and maintain them longer may yield higher, particularly under drought stress conditions. The significant difference in the mean number of leaves of a genotype under well-watered and water-stressed conditions indicates the effect of water deficit or nutrient supply to the plant under drought condition. No significant difference was observed in any of the genotypes with respect to number of leaves. CML254 recorded 14.7 numbers of leaves under control condition, while the same got reduced to 12.00 under the stress condition at Delhi (Table 2). Majority of the genotypes recorded decrease in leaf number under stress conditions. However, CMLP1 showed similar number of leaves 14.2 and 14.7, under both the conditions. Similar trend was also observed in case of CML340, CM341, LPSC7-F64-2-6-2-2-B-B and ZM621A-10-1-1-1-2-B*7-B-B. At Pusa-Bihar, CML344 was identified as the best genotype under both the conditions. Besides, genotypes such as CML287, CML333, CML340, CML341, CML389, CML442/CML197/TUXPSEQ, DTPYC9-F46-1-2-1-2-B and CML245 also showed similar results under both the conditions.

Although majority of these exotic lines were ranked highly in experiments carried out at CIMMYT under drought stress conditions, many of the lines did not respond favorably under drought stress conditions in India, due to significant differences in the local climatic factors, including temperature, relative humidity, etc., besides soil-related factors. However, considering the performance for various traits, CML341, CML340, DTPYC9-F46-1-2-1-2-B and CMLP2 were identified as the highly promising drought tolerant genotypes at Delhi under drought stress conditions. In case of Pusa-Bihar experiment, CML340, CML341, LPSC7-F64-2-6-2-2-B-B, and LPSC7-F71-1-2-1-1-B-B were found to be the promising genotypes. However, based on the data of both the locations/seasons, CML340, CML341 and DTPYC9-F46-1-2-1-2-B proved to be promising in terms of their responses under drought stress conditions and could be potentially utilized in the breeding programmes.

The correlation of a secondary trait (particularly those expressing at or before flowering stage) with grain yield is of importance for indirect selection of genotypes for higher grain yield under abiotic stresses like drought. The study revealed positive correlations between ASI and leaf senescence under stress conditions at Delhi (Table 3). Importantly, ASI showed negative association with EPP (-0.35*) and grain yield per plant (-0.36*) under the stress conditions. Similar trend was also observed in case of experiment conducted at Pusa-Bihar during *Rabi* 2007-08. Since grain yield is directly proportional to short ASI, selection against silk delay is the most effective method of breeding for drought tolerance because it is well correlated with improved yields under drought stress [6, 23, 27]. Besides, positive correlations (0.69** at Delhi and 0.70** at Pusa-Bihar) were observed between ear per plot and grain yield per plant [12, 23, 28]. Interestingly, correlations between grain yield with ASI and EPP at both the locations/seasons were found to be weak under optimum management conditions while it is very strong under stress. Similar observations were also reported by others [13, 23, 26], while experimenting with a set of CIMMYT genotypes under drought stress conditions.

In contrast, leaf senescence (during grain filling stage) at Delhi, exhibited significant negative

Table 3. Phenotypic correlation coefficients among different traits under well-watered and drought-stress conditions at Delhi and Pusa-Bihar

Traits	TNL	ASI	SF	SG	EPP	GYP
TNL	-	0.12 (-0.32)	0.10 (-0.30)	0.02 (-0.26)	0.04 (-0.26)	-0.19 (-0.28)
ASI	-0.33 (-0.11)	-	0.32 (0.11)	0.31 (0.17)	-0.24 (0.36)	-0.25 (0.29)
SF	-0.09 (0.19)	0.47** (0.13)	-	0.81** (0.49**)	-0.21 (0.09)	-0.06 (0.20)
SG	-0.14 (-0.15)	0.53** (0.28)	0.89** (0.14)	-	-0.29 (-0.02)	0.03 (-0.04)
EPP	0.02 (0.30)	-0.35* (-0.63**)	-0.44* (-0.01)	-0.45* (-0.21)	-	-0.03 (0.01)
GYP	0.06 (0.14)	-0.36* (-0.35)	-0.28 (-0.02)	-0.40* (-0.06)	0.69** (0.70**)	-

,*Significant at P = 0.05 and P = 0.01, respectively.
TNL: Total no. of leaves per plant; ASI: Anthesis silking interval; SF: Senescence at flowering stage; SG: Senescence at grain filling stage; EPP: ear per plot and GYP: grain yield per plant. Above diagonal represents controlled condition, while below diagonal represents drought stress conditions. Values without parenthesrs represent Delhi (*Kharif* 2007) and values in parentheses represent Pusa-Bihar (*Rabi* 2007-08).

correlations with EPP (-0.45*) and grain yield per plant (-0.40*) under stress conditions (Table 3). These traits can also be used as secondary traits during indirect selection for grain yield. Above all, the genotypes showing higher yield under low N and drought tend to show lower ASI, delayed senescence and a higher number of ears per plant [29]. The results thus substantiate the need for study of secondary traits [such as ASI, EPP, anthesis date and rate of leaf senescence] as important criteria for selection under drought stress conditions [30].

Importantly, when one compares the performance of promising genotypes, such as DTPYC9-F46-1-2-1-2-B, CML341, CML340, and CMLP2 at Delhi, the interrelationships among grain yield, EPP, leaf senescence (at grain filling) and ASI was found to be satisfactory and correlated too. ASI for these genotypes ranged 3.5 days under control to 6.5 days under stress; thus, an average of 3 days ASI difference was observed (Table 2). Leaf senescence was also delayed/low ranging from 20% to 50% under stress, except for DTPYC9-F46-1-2-1-2-B, which showed 75% leaf senescence. It is known that N accumulation decreases under both drought and N stress [31, 32]. Due to this, the growing ear fulfills its demand by remobilized N from leaves and stems under both types of stresses [33]. Therefore, under drought stress, selection for delayed senescence has not only delayed foliar senescence [5], but also has resulted in more grain production by efficient utilization of leaf N, particularly in the DTP populations developed at CIMMYT, Mexico. In general, the potential use of the ASI to identify stress tolerant genotypes at flowering coupled with EPP as a measure of barrenness provides perhaps more complete information at final harvest [28]; and are the best drought adaptive traits for drought tolerance improvement [12].

In conclusion, the present study led to the identification of some highly promising CIMMYT (Mexico) genotypes with drought tolerance at flowering stage at different locations/seasons in India. The study also revealed a few genotypes with low ASI, delayed senescence, high EPP and grain yield per plant under both well-watered and drought stress conditions, which could be potentially used in breeding for drought tolerance. Improved maize cultivars developed using such lines could be valuable for subsistence maize farmers in stress-prone areas.

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