GENOTYPIC DIFFERENCES AGAINST PEG SIMULATED DROUGHT STRESS IN TOMATO

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Abstract

Current study describes the effect of artificial drought stress using PEG on different tomato varieties at seedling stage. The performance was assessed by germination, root and shoot length, and seedling biomass. The tomato varieties investigated have shown varying responses for different plant attributes against drought stress which remained significantly different for most of the parameters studied. For a given genotype, the effect of PEG was significant for root length and seedling biomass, whereas it was non-significant in case of germination and shoot length. The genotypic differences among tomato varieties investigated were found significant for all the plant parameters studied. Three different types of genotypic behaviors were observed in this study. The genotypes displayed either enhanced or declined growth in case of root, shoot and seedling biomass representing the two behavior types. Whereas, only in case of germination, in addition it also showed third type of behavior and remained indifferent to control and induced drought stress. Among others Walter, Punjab Chuhara & Kurihara have shown enhanced growth for all the parameters when subjected to PEG as compared to control. Money maker was the only genotype where a decline for all the attributes was recorded against drought stress, reflecting its lower tolerance.

Introduction

Abiotic stresses are governed by various factors the most prominent being the elevated temperatures and scarcity of water (Pena & Hughes, 2007; Narusaka et al., 2003). Unusual dry weather conditions adversely affect germination and seedling growth rates thus enhancing cell elongation sensitivity to damages induced under stressed conditions (Taylor et al., 1982; Delachiave & Pinho, 2003; Hamayun et al., 2010a). Low water potential tends to induce oxidative bursts leading to elevated levels of antioxidant enzymatic activities and high solute concentration and protein accumulations (Zhu, 2002; Khan et al., 2011 & 2012). Drought severely disturbs water balance of the plant body and causes alterations in water uptake patterns of plant (Kmet et al., 2009; Waraich et al., 2011). Deleterious effects of water stress have been reported in different crops such as tomato (Ragab et al., 2007), soybean (Sakthivelu et al., 2008; Hamayun et al., 2010b), corn (Khodarahmpour, 2011) and citrus (Ben-Hayyim, 1987).

Tomato is one of the widely grown vegetables in the world (Aazami *et al.*, 2010). It is the most important vegetable crop of Pakistan, cultivated over an area of 52300 hectares with an annual production of about 529600 tonnes and average yield of 10.1 tonnes/hec (Anon., 2011). Declined water contents tend to reduce leaf area in tomato genotypes (Jurekova *et al.*, 2011) which in turn results in reduced shoot lengths (Unyayar *et al.*, 2005). Changing climatic patterns in Pakistan tend to influence tomato production and quality majorly by water scarcity, frost injury and elevated saline conditions (Abid, 2011).

Polyethylene glycol (PEG), a series of polymers that vary from viscous liquids to waxy solids has been used to induce water stress artificially (Larher *et al.*, 1993). PEG induced osmotic stress is found to reduce cell water potential (Govindaraj *et al.*, 2010). An increase in concentration of PEG-6000, resulted a decrease in germination rate, root length, shoot length and seed vigor in certain crop plants (Khodarahmpour, 2011). Tomato has been selected for better growth under PEG simulated water stress (Bressan *et al.*, 2003). In vitro selection techniques involving the use of PEG, is one of the reliable methods for screening desirable genotypes and to study further the effects of water scarcity on plant germination indices (Kocheva *et al.*, 2003; Sakthivelu *et al.*, 2008).

Regardless of water stress damages on crop yields, a very limited research on drought resistance in Pakistan is carried out on tomato. Also varieties being released are reported to exhibit insufficient tolerance against abiotic stresses (Dhlamini *et al.*, 2005). Tomato genotypes tend to exhibit limited and inadequate genetic variability for drought tolerance. Hence the best way to mitigate the effects of drought stress involves the crossing of cultivated tomato with drought tolerant lines (Pena & Hughes, 2007). The present study aims to evaluate drought tolerant potential and compare the behavior of different tomato genotypes under PEG simulated water stress.

Materials and Methods

The study was conducted at Seed Preservation Laboratory, Plant Genetic Resources Institute (PGRI), NARC, Islamabad during 2011.

Plant material: Ten tomato varieties viz., T-4, Tom-Round, Money Maker, Feston, Ratan, Walter, Punjab Chuhara, Indian, Nagina & Kurihara were investigated against PEG simulated drought stress. The tomato germplasm was obtained from National Gene bank, IABGR, NARC, Islamabad (Table 1).

PEG assay: Assessment of drought stress tolerance of tomato germplasm was conducted using PEG-6000 under lab conditions. Initially a suitable concentration of PEG was optimized through series of experiments. Later on 4% PEG optimized concentration was used to evaluate all the tomato genotypes. Standard germination test was conducted using between paper method of germination as per ISTA rules (Anon., 1993). Twenty five seeds of each variety were grown on paper towels (22 cm x 23cm; Victory brand, Shinbashi Paper Company, Shizuoka,

Japan). Each treatment was replicated three times. Seeds were placed on the surface of double sheets of paper towels which were moistened with distilled water (control) and 25ml of 4% PEG-6000 solution. The sheet was covered with another sheet of paper towel. The sheets were rolled and placed vertically in plastic beaker, covered with polythene bag and placed at $25\pm1^{\circ}$ C in an illuminated germinator. The data was recorded on 10^{th} day.

Germination rate (%): Germination percentage was calculated on the basis of number of normal seedlings (Anon., 1983; Anon., 1993). After ten days the samples were analyzed for their germination percentage. Data for germinated, dead, normal and abnormal seedlings was recorded. Seedlings with both root and shoot were the normal ones, while the seedlings having only root or shoot were considered as abnormal. The number of normal germinated seedlings was used to derive germination percentage.

Root length (cm): Root lengths were measured for randomly selected ten normal seedlings and then mean length was calculated.

Shoot length (cm): Shoot lengths were also measured for same ten normal randomly selected seedlings and then mean length was calculated.

Seedling biomass (g): Fresh weight of above mentioned randomly selected ten normal seedlings was recorded on precision balance in grams.

Relative germination percentage, root and shoot length, and seedling biomass were also calculated.

Statistical analysis: Analysis of variance was carried out as described by Steel *et al.*, (1997). Statistical significance of means was tested by Duncan's Multiple Range Test using MStatC program.

Genotypes	Relative germination (%)	Relative root length (%)	Relative shoot length (%)	Relative seedling biomass (%)
T-4	100.0	170.2	82.0	271.0
Tom-Round	113.6	160.0	133.5	179.7
Money Maker	87.2	91.2	75.7	79.5
Feston	110.0	97.0	74.5	76.4
Ratan	100.0	130.5	92.7	95.6
Walter	107.9	141.3	116.6	106.5
Punjab Chuhara	101.9	145.6	109.1	104.3
Indian	121.4	137.4	99.1	100.5
Nagina	95.6	125.9	113.7	115.4
Kurihara	107.3	126.2	105.9	112.5

Table 1. Relative performance of tomato genotypes at PEG simulated drought stress.

Results

Germination rate (%): The germination response of each tomato genotype to water application or PEG induced water stress remained statistically nonsignificant (Fig. 1A). However, the genotypic differences among tomato cultivars for the same parameter were found significant (Fig. 2A). The mean germination ranged between 88.3 and 39.2% in Ratan and Tom-Round, respectively. The cv. Feston, Ratan and Punjab Chuhara remained statistically at par with each other. The relative germination (%) in tomato ranged from 87.2% in cv. Money Maker to 121.4% in cv. Indian (Table 1). Three types of behaviors for germination were observed in tomato; either germination enhanced, declined or remained indifferent as compared to control conditions. Among tomato varieties assayed Feston, Indian, Walter, Punjab Chuhara, Kurihara and Tom-Round were of type-I behavior having relative germination in the range of 101.9% to 121.4% in Punjab Chuhara and Indian, respectively (Table 1). Nagina and Money Maker were the tomato varieties representing type-II behavior with relative germination of 87.2% and 95.6% in Money

Maker and Nagina, respectively (Table 1). Ratan and T-4 remained indifferent at distilled water as well as on PEG displaying the type-III behavior.

Root length (cm): Effect of PEG on root length displayed significant differences in tomato varieties studied (Fig. 1B). The genotypic differences have also been remained significant among tomato genotypes (Fig. 2B). The mean root length ranged between 8.3cm to 4.5cm in Punjab Chuhara & Kurihara, respectively (Fig. 2). Tom-Round and Money Maker found statistically at par with each other whereas rest of the genotypes had varying root lengths at PEG treatment.

All the tomato varieties, except Money Maker and Feston, had longer root lengths at PEG as compared to distilled water (Fig. 2). The relative root length (cm) observed in different tomato cultivars grouped all the genotypes into two categories on the basis of their performance against PEG. The root length in Money Maker and Feston declined, while rest of the genotypes exhibited varying level of increase at PEG as compared to control. The relative increase in root length was 170.2% in T-4, whereas 125.9% was observed in Nagina (Table 1).

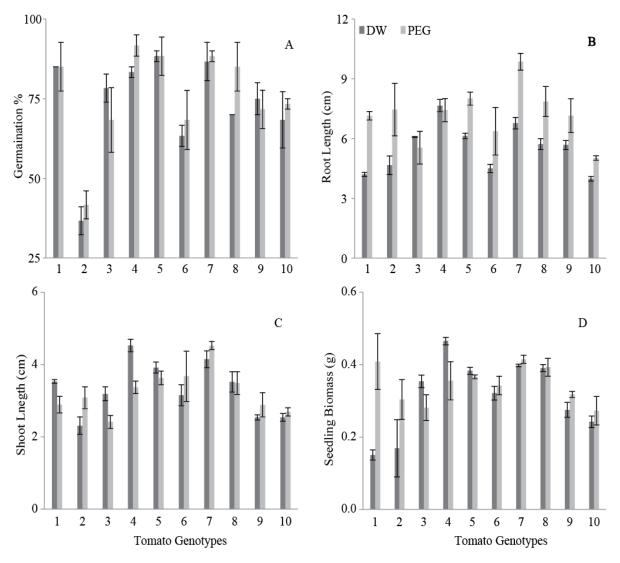


Fig. 1. Comparative response of tomato varieties under control and PEG induced water stress; A) Germination (%), B) Root length (cm), C) Shoot length (cm) and D) Seedling biomass (g). (1=T-4, 2=Tom-Round, 3=Money Maker, 4=Feston, 5=Ratan, 6=Walter, 7=Punjab Chuhara, 8=Indian, 9=Nagina, 10=Kurihara)

Shoot length (cm): The shoot length in response to PEG application did not influence significantly (Fig. 1C), whereas for the same parameter, the genotypic differences observed were found significant (Fig. 2C). The longest shoot length was found in Punjab Chuhara (4.5cm), whilst the shortest was exhibited by Money Maker (2.3cm). Tomato varieties T-4 & Indian, Ratan&Kurihara remained statistically at par with each other. A longer shoot length in Punjab Chuhara, Nagina, Kurihara, Walter and Tom-Round was observed at PEG, displaying a relative increase between 105.9% (Kurihara) to 133.5% (Tom-Round). On the other hand, relative shoot elongation in Feston and Money Maker was decreased by 74.5% and 75.7%, respectively under water stress conditions (Table 1). An irregular pattern of shoot length was also displayed by all the remaining cultivars showing genotypic differences (Table 1).

Seedling biomass (g): The differences in fresh seedling biomass against PEG application were found significant (Fig. 1D). Similarly, tomato genotypes also exhibited significant variability for seedling biomass between genotypes (Fig. 2D). Fresh seedling biomass among tomato genotypes ranged between 0.2g and 0.4g in Tom Round and Feston, respectivley (Fig. 1D). Tomato genotypes T-4 and Nagina, Feston and Punjab Chuhara were found statistically at par with each other. Fresh weight of Nagina, T-4, Walter, Punjab Chuhara, Kurihara and Tom-Round was found elevated when stressed with PEG, however, for rest of the genotypes a decrease in fresh weights was observed (Fig. 1D). Relative fresh weight ranged between 104.3% to 271% in Punjab Chuhara and T-4, respectively (Table 1). Indian was the variety whose behavior was almost similar at distilled water as well as on PEG, whereas a varying seedling biomass in rest of the genotypes was observed.

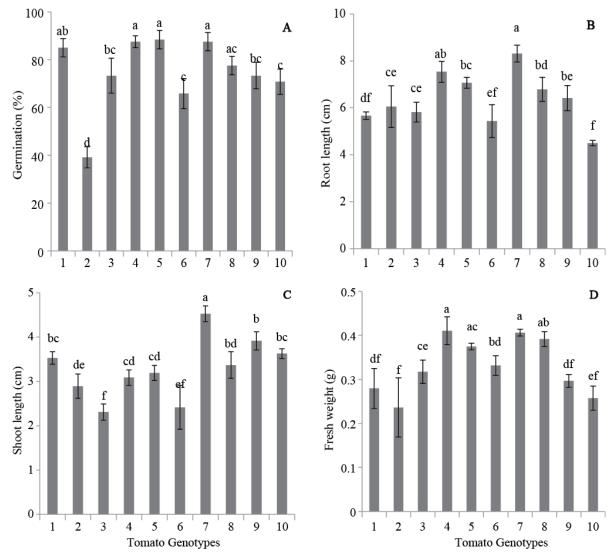


Fig. 2. Genotypic differences in tomato varieties under PEG induced water stress; A) Germination (%), B) Root length (cm), C) Shoot length (cm) and D) Seedling biomass (g). (1=T-4, 2=Tom-Round, 3=Money Maker, 4=Feston, 5=Ratan, 6=Walter, 7=Punjab Chuhara, 8=Indian, 9=Nagina, 10=Kurihara).

Discussion

Current study describes the effect of artificially simulated drought stress using 4% PEG6000 on 10 different tomato varieties at seedling stage; which yielded informative outcome on the tolerance behavior of the genotypes. In general, within genotype; germination percentage was least influenced by stress, while growth was significantly affected (Fig. 1). However, the germination percentage between genotypes was significantly different (Fig. 2); suggesting presence of some physiological attribute that can effect plant population irrespective of drought stress. Soni et al., (2011) reported that Vigna aconitifolia genotypes showed higher level of germination under stressed conditions and were found to be more tolerant at seedling stage. However, in this study, this finding was not applicable to all cases (except var. Money Maker); as highest biomass as well as root length was found in

genotype T-4 whose germination was not influenced by stressful condition (Table 1). It can be suggested that it depends on the material used in screening against stress and a large number of genotypes is recommended to confirm this attribute.

The decline in various plant attributes in response to induced stress is a commonly observed phenomenon which is according to tolerance level in plant. Money maker was the only variety where a decline for all the attributes was recorded against drought stress. This reflects the poor performance of this genotype under stress. Our study suggests that this genotype (Money maker) might be used as susceptible control in future studies. The decline in growth under stress condition is not an unusual phenomenon and this has been reported in different crops (Waseem *et al.*, 2006; Kulkarni & Deshpande, 2007; Jajarmi, 2009; Hamayun *et al.*, 2010b; Sultan, *et al.*, 2012; Shinwari, *et al.*, 2013). Aazami *et al.*, (2010) also recorded reduced growth rate in tomato cultivars at varying PEG

simulated drought stress. Declined shoot growth is reported by Abdel-Raheem et al., (2007) in tomato under osmotic stress conditions induced by PEG. Remarkable decrease in shoot length of tomato has been observed with increasing PEG concentrations (Kulkarni & Deshpande, 2007). Seedling biomass affected by PEG solution in tomato has also been recorded by Nahar & Gretzmacher (2002). Reduced root lengths under osmotic stress conditions have been reported in safflower (Jajarmi, 2009) and pea (Whalley et al., 1998). The tomato genotypes investigated in this study have shown relative increase in growth under drought stress conditions (Table 1); as the root length in 80%, shoot growth/length in 50% and biomass in 70% of genotypes was enhanced as compared to its control. This is because plants have the capacity to survive under water deficit conditions (Oliveira et al., 2011) and those performing better are considered to be drought tolerant. Growth and yield in tomato can be further enhanced by foliar application of minerals (Azeem & Ahmad, 2011).

Among tomato varieties Walter, Punjab Chuhara & Kurihara has shown enhanced growth for all the parameters at PEG as compared to control treatment. Under drought stress, roots are generally affected first then other plant parts (Misra & Dwivedi, 2004; Ghafoor, 2013). The genotypes which showed positive behavior under stressed conditions as compared with control may carry a kind of tolerance mechanism, which makes plants capable of retaining a goods turgor pressure and absolute water level under stressed conditions (Saxena & O'Toole, 2002). Hence, genotypes with the ability of rapid root elongation under stress conditions are likely to be water stress tolerant, and they retain continuous root elongation process by extracting water under stressed conditions (Kulkarni & Deshpande, 2007). Though the genotype T-4 had the highest value of root length and biomass, it seems that the genotypes have advanced the root growth and biomass at the cost of shoot development. Similar behavior was noted in Ratan and Indian genotypes, whereas the genotype Tom-Round showed higher relative germination as well as growth for all parameters. These two genotypes may be used as positive/tolerant controls in future studies. However, the internal physiological investigation is needed for assessing their variable response. Detailed studies focusing level of proline accumulation under stress (Ali et al., 2011) or application of plant growth regulators (Hussain et al., 2010) in these genotypes could render further useful information for selecting suitable genotypes.

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