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HEAT TOLERANCE IS VARIABLE IN COTTON (*GOSSYPIUM HIRSUTUM* L.) AND CAN BE EXPLOITED FOR BREEDING OF BETTER YIELDING CULTIVARS UNDER HIGH TEMPERATURE REGIMES

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Abstract

Fifty accessions of *Gossypium hirsutum* L., were evaluated for heat tolerance based on relative cell injury level (RCIL %) and stomatal conductance. Accessions showed highly significant differences for both the parameters and ranged in relative cell injury level between 44.8 to 74.9% and from 58.1 μms^{-1} to 120.6 μms^{-1} in stomatal conductance. The breeding material (NIAB 111/2 followed by MNH 553) showing the least mean Relative cell injury level also showed the highest stomatal conductance and both these traits showed negative correlation (r= -0.70). The data on the basis of both the above mentioned criteria were used to sort out heat tolerant and sensitive cotton accessions for further breeding program.

Introduction

Cotton belt of Pakistan by virtue of its geographical position is in a zone of high temperature wherein during summer it approaches 50°C in some areas and experiences stress due to heat. Heat stress usually occurs in conjunction with other environmental stresses such as drought and high light intensity that aggravate the impact in terms of low plant population per unit area, reduced fibre yield and the quality of cotton fiber (Rahman, 2006). It has been estimated that crops achieve only about 25% of their potential yield because of the damaging effects of environmental stress (Boyer, 1982) including heat. These stresses are location-specific, exhibiting variation in frequency, intensity and duration and can occur at any stage of plant growth and development.

The accurate methodology to screen for stress tolerance under natural conditions is expensive as well as time consuming (Blum & Eberconn, 1981). Field evaluation of cotton under high temperatures with appropriate irrigations is a practical approach to evaluate heat responses (Hall, 2001). Timing of heat stress might affect the ability to screen for heat tolerance. There is a need to develop the screening procedure for identifying levels of acquired thermotolerance led to the evaluation of different cellular constituents that might be used as an *In vivo* indicator of heat injury or cell viability (Burke, 2001). Progress has been achieved with whole plant screening approaches in a few crop species including tomato (Stevens, 1979), cotton (Feaster & Turcotte, 1985) common bean (Dickson & Petzoldt, 1989) and cow pea (Hall, 1992).

Relative cell injury level from leaf disks at high temperature has been suggested as a screening technique for heat tolerance in plants (Sullivan, 1972). Several studies suggested the effectiveness of cell membrane thermostability in terms of relative cell injury level in detecting genetic variability in heat tolerance in warm season crops (Ismail & Hall, 1999). This technique is simpler, quicker and less expensive than the whole plant screen. Potentially it can be used with early vegetative stage leaf tissue from plants grown in field nursery environments (Ismail & Hall, 1999).

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A substantial increase in yield potential has been accompained by increases in stomatal conductance (Cornish *et al.,* 1991; Lu & Zeiger, 1994, Lu *et al.,* 1994; Radin *et al.,* 1994; Pettigrew & Meredith, 1994; Percy *et al.,* 1996). Lu *et al.,* (1994) reported that advanced lines of Pima cotton (*Gossypium barbadense* L.) bred for higher yield potential and heat resistance have higher stomatal conductance indicating that selection for high yield potential and heat resistance has imposed a selection pressure for higher stomatal conductance. Stomatal conductance could also be a useful selection criterion for higher yields in irrigated crops grown at supra-optimal temperature. The present studies intended to evaluate cotton germplasm for genetic variability and to identify the suitable stock(s) to be used in breeding for heat tolerance based both on Relative cell injury level and stomatal conductance.

Materials and Methods

The studies were conducted during year 2003-07 in the Department of Plant Breeding and Genetics and Centre of Agricultural Biochemistry and Biotechnology (CABB) at the University of Agriculture, Faisalabad. The plant material consisting of 50 varieties/lines of *G. hirsutum* L., was screened against heat stress in glass house using the following parameters.

i. Cell membrane thermostability (CMT)
ii. Stomatal conductance

The day and night temperature was maintained between 45-50°C and 20-30°C respectively. All the 50 entries were sown in earthen pots (11cm × 12cm) filled with alluvial soil having pH 7.5 and EC 1.4 dS m^{-1} and were placed in glass house. There were 10 pots for each entry. Five seeds were sown in each pot. After 25 days of sowing, the seedlings were thinned to one plant per pot. Five plants were tagged to collect the samples to be used for the test of Relative Cell Injury percentage (RCI %) which was estimated following the method proposed by Sullivan (1972) as given hereunder. The models of instruments used were MEMMERT-WB1, Germany for water bath, SANYO-MIR253 for incubator, EYELA-MMS, RIKAKIKAI CO., Ltd for mechanical shaker, EC meter (TOA-CM-14P, Japan for EC meter.

$$RCI\ \% = 1- [\{1- (T_1/T_2)\} / \{1 - (C_1/C_2)\}] \times 100$$

where,

T_1= EC of sap in 50°C before autoclaving.
T_2= EC of sap in 50°C after autoclaving.
C_1= EC of sap in 25°C before autoclaving.
C_2 =EC of sap in 25°C after autoclaving.

Data on stomatal conductance were recorded using diffusion porometer (AP4, Delta-T Devices, Cambridge, UK) between 1.00 and 2.00 pm. The data on 46 accessions were used to perform analysis of variance through SAS Procedure GLM and the data in percentage were log transformed.

Results

Four accessions showed complete burning at germination stage whereas death of some seedlings was observed in spite of enough water in the rooting media. Results indicated significant differences ($\alpha = 0.05$) among various accessions for relative cell injury level (Table 1). The accessions ranged between 44.8 to 74.9%. Relative cell injury level was recorded to be more than 70% in fourteen accessions, whereas it ranged between 60 to 70% in 18 lines and 50-60% in 7 accessions. However, only 3 accessions showed Relative cell injury level less than 50% i.e., 40-50% (Fig. 1). The tolerant accessions having comparatively low relative cell injury level included NIAB-111/2, BH-160, MNH-554, N-313, BH-163 and Mutant-94. The sensitive accessions having comparatively high levels comprised of FH-945, CIM-496, VH-142, CIM-707 and NIAB-801/2.

Stomatal conductance: Analysis of variance indicated highly significant differences ($\alpha = 0.0001$) among various accessions of cotton for stomatal conductance (Table 2). Accessions varied from 58.1 to 120.6μms^{-1} for stomatal conductance. Ten accessions showed stomatal conductance greater than 100μms^{-1}, 25 ranged between 70 to 100μms^{-1}, whereas 11 had less than 70μms^{-1} (Fig. 2).

The accessions NIAB-111/2, BH-160, MNH-554, N-313, BH-163 and MUTANT-94 were sorted as tolerant and accessions FH-945, CIM-496, VH-142, CIM-707 and NIAB-801/2 were regarded as sensitive to heat (Table 3).

Discussion

Cotton (*Gossypium hirsutum* L.) crop in Pakistan is sown in May and matures in September-November primarily in regions where air temperature between 45-50 °C is a common feature throughout the growing season. At high temperature, plant development rate is much increased (Hodges *et al*., 1993; Reddy *et al*., 1997 a & b) reducing overall life period besides other detrimental effects like denaturing of membranous structures. High temperature had a strong negative correlation with lint yields (Rawson, 1992; Hodges *et al*., 1993; Ziska *et al.,* 1997; Singh *et al.,* 2007) and lint quality. In any crop-improvement program, the first and foremost requirement is to identify the suitable stock (s) to be used in breeding.

A few accessions in the present studies showed complete burning at germination stage whereas, death of some seedlings was observed in spite of enough water in the rooting media. This clearly indicated the impact of high temperature on growth and development in cotton. Burke (2001) reported that seedling heat tolerance is essential in most dry land cotton production areas. Under heat conditions, emerging cotton seedlings poorly develop root system and show burning effects on the leaves particularly the younger leaves are adversely affected (Lather *et al.,* 2001). According to Hall (2001), when plants growing in pots are subjected to high air temperatures, both the shoots and the roots are faced with hot conditions.

Reddy *et al.,* (1992 a & b) observed that optimum temperature for leaf area development to be 26°C. According to Burke (2001) when seedling temperature increases above optimal levels, acquired thermo-tolerance system is induced. Maximum protection levels are induced when plant temperature reaches 37–40°C, but at higher temperatures (beyond 45°C) protection levels decline rapidly which is the case under prevailing conditions of Pakistan and finally plant death may occur as in the present studies. Relative cell injury level (%) is reported to be helpful for the estimation of damage caused by high temperature. Bibi *et al*., (2003) demonstrated that membrane leakage was the most sensitive technique for quantifying temperature tolerance in cotton under field conditions.

Table 1. Analysis of variance for relative cell injury level (%).

SOV	DF	SS	MS	Pr > F
Reps	2	87.59	43.79	0.66
Varieties	45	7924.66	176.10	0.0249
Error	90	9713.36	107.92	

Table 2. Analysis of variance for stomatal conductance.

SOV	DF	SS	MS	Pr > F
Reps	2	1061.68	530.84	0.0070
Varieties	45	32436.66	720.81	0.0001
Error	90	9095.745	101.06	

Table 3. List of the germplasm after screening.

S. No.	Heat tolerant germplasm	S. No.	Heat susceptible germplasm
1.	NIAB-111/2	1.	FH-945
2.	BH-160	2.	CIM-496
3.	MNH-554	3.	VH-142
4.	N-313	4.	CIM-707
5.	BH-163	5.	NIAB-801/2
6.	Mutant-94		

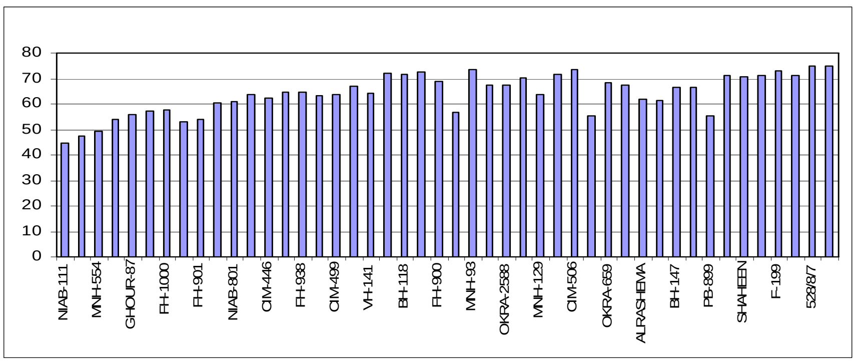

Fig. 1. Mean Relative cell injury level (%) of cotton germplasm.

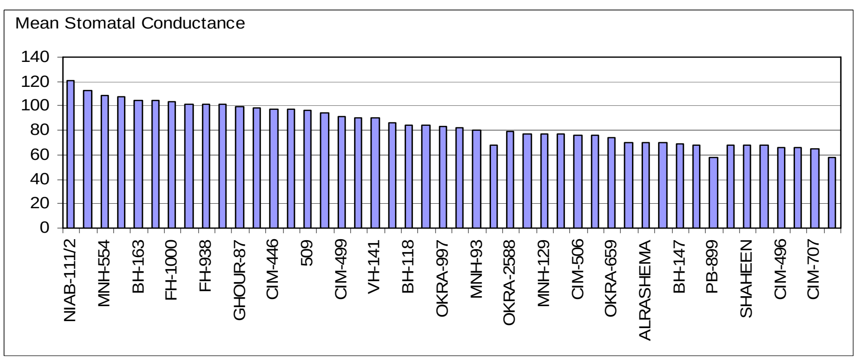

Fig. 2. Mean Stomatal conductance of cotton germplasm.

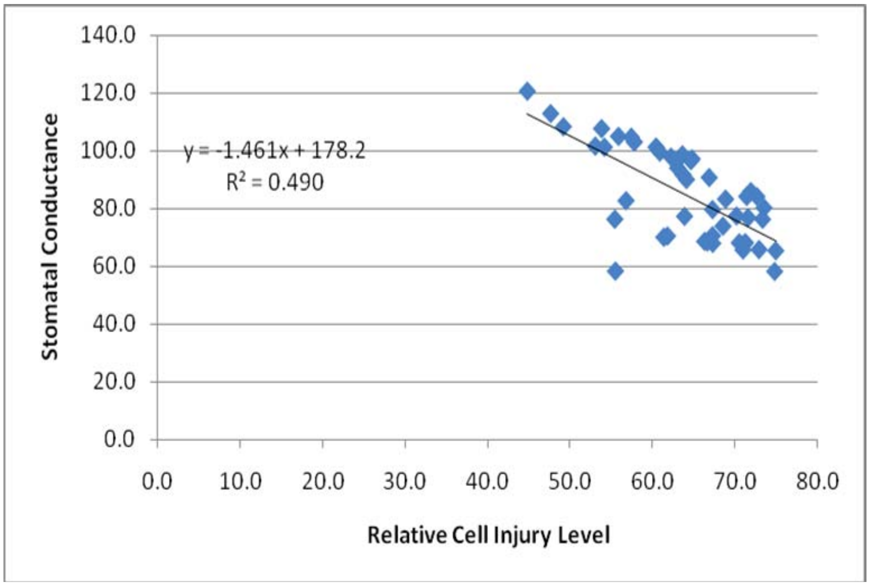

Fig. 3. Relationship between relative cell injury level and stomatal conductance.

Significant differences among accessions for both the parameters indicated the presence of a good deal of genetic variability and that these vary with the breeding material. Leaf stomatal conductance was negatively correlated (r= -0.70) with leaf water potential in the present studies (Hatfield *et al*., 1987), which indicated that leaf conductance differences among the varieties were influenced by the leaf water status. Keeping water availability a constant factor, those accessions having comparatively high stomatal conductance would tolerate heat through increased transpiration thereby producing more cooling effect and those having comparatively low stomatal conductance would be regarded as sensitive to heat.

It was concluded on the basis of the results from the present studies that heat tolerance in terms of relative cell injury level and stomatal conductance is variable in cotton accessions and both are negatively correlated and that with the development of heat tolerant cotton cultivars the yields and the quality of fibre could be improved.

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