# Evaluating Cotton Genotypes for Heat Stress Tolerance Using Biochemical Markers and Seedling Traits as Screening Tool

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Heat stress is a prime constraint hampering the attainment of cotton yield potential in Pakistan, while selection of genotypes for high temperature environment using morphological markers often leads to discrepancies. An experiment was conducted in order to characterize thermo-sensitivity of varying cotton genotypes using biochemical markers and to determine the biochemical attributes modulated regulations in biomass accumulation of heat-stressed cotton. The experiment was replicated thrice and laid out in completely randomized design (CRD) with split arrangement during 2016 at the University of Agriculture Faisalabad, Pakistan. Treatments consisted of heat stress in main plots, viz. H<sub>0</sub> = no heat stress imposition, and H<sub>1</sub> = heat imposition for 14 d after emergence at seedling stage, and 15 cultivars of cotton in subplots, viz. FH-Lalazar, FH-142, FH-114, CIM-598, CIM-599, CIM-602, VH-282, VH-326, VH-341, MNH-886, MNH-888, MNH-992, IUB-13, IUB-212, and IUB-222. Decreases in relative leaf water contents and cell membrane thermo-stability were lower for genotypes CIM-598, CIM-599, CIM-602, VH-282, VH-326, VH-341, MNH-888, MNH-992 and IUB-13 compared to other cultivars. Antioxidant enzymes were enhanced under heat stress compared to the control for cultivars CIM-598, CIM-599, CIM-602, VH-282, VH-326, VH-341, MNH-888, MNH-992 and IUB-13, while these were decreased in all other cultivars. Heat-mediated decreases in root, shoot length, fresh and dry weight were lower for cultivars CIM-598, CIM-599, CIM-602, VH-282, VH-326, VH-341, MNH-888, MNH-992 and IUB-13 compared to other genotypes. Essentially, heat stress deleteriously impacted all the evaluated parameters; however, distinct cotton genotypes varied remarkably from each other for antioxidants, membrane stability and biomass accumulation. Moreover, biochemical markers proved to be potential regulators of biomass accumulation and hence can be used as road map in future cotton improvement programs. On the basis of the studied response variables, genotypes CIM-598, CIM-599, CIM-602, VH-282, VH-326, VH-341, MNH-888, MNH-992 and IUB-13 manifested heat tolerance, while FH-114 showed susceptibility to heat stress in cotton.

Key Words: antioxidants, biomass accumulation, cotton, membrane stability, thermotolerance, water relations

Abbreviations: CAT – catalase, CMT – cell membrane thermostability, POD – peroxidase, RDW – root dry weight, RFW – root fresh weight, RL – root length, RLWC – relative water content, SDW – shoot dry weight, SFW – shoot fresh weight, SL – shoot length, SOD – superoxide dismutase

# INTRODUCTION

Worldwide concern in recent years has developed into an urgent need to enhance the yield potential of cotton since its demand is increasing while production is dwindling continually. Production of cotton was diminished by 20.2% in 2015–16 in all major cotton-producing countries around the globe, while growing demands for cotton have added into the import bill of these countries. Adverse weather, increasing costs of inputs, and poor policies have further exacerbated the situation and counterbalanced the demand and supply of cotton (OECD

2017). Cotton production in Pakistan decreased by 1.5% in 2015–2016 compared to the previous year (Government of Pakistan 2017).

Cotton is primarily cultivated to get fiber and is a major cash crop of Pakistan (Iqbal et al. 2012). In agriculture, cotton shares 5.1% in value addition and 1% in GDP. In 2015–16, the area under cotton crop was 2.917 million ha in Pakistan, which was 1.5% less than that of the previous year (Govt. of Pakistan 2017).

The cotton plant (*G. hirsutum* spp.) originated in areas with hot climates; it suffers from extremely high day temperatures greater than 36 °C (Riaz et al. 2013).

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Temperature reaches up to 48-50 °C during the cottongrowing season (Shakoor et al. 2017). High temperature causes loss of 65-70% fruiting points and induces pollen sterility (Baloch and Lakho 2000). Heat stress markedly distresses numerous physiological, biochemical and growth processes in crop plants. According to Reddy et al. (2004), the suitable daily average temperature for proper growth of cotton is 27-29 °C. Heat stress, induced by global warming, is becoming a major threat to crop productivity, and plants change their phenology, anatomy and physio-biochemical features to tackle heat stress (Zhang et al. 2008; Shahid et al. 2017). Heat stress is defined as the magnitude of temperature above the threshold level where it causes permanent damage to the crop (Talukder et al. 2014). The main impact of heat stress is surfeit of reactive oxygen species (ROS) and lipid peroxidation of cellular membranes (Hasanuzzaman et al. 2013). Plants show various agitations in metabolism to cope with high temperature stress and scavenging of ROS (Dietz and Pfannschmidt 2011).

Choosing the right cultivar for high-temperature stress tolerance has been an arduous task universally (Semenov et al. 2014). High-temperature stress can induce oxidative stress through disruption of cell membrane stability and peroxidation of membrane lipids by protein denaturation (Saleem et al. 2018). The production of reactive oxygen species in ample volume under hightemperature stress leads to premature leaf shedding in cotton that is mainly due to proteolysis of proteins and degradation of long-chain proteins into simpler ones (Hemantaranjan et al. 2014). Plants also accumulate compatible solutes and osmoprotectants as a defensive mechanism to regain cellular redox balance and homeostasis. Antioxidants and compatible solutes play a key role in raising heat tolerance potential and thus support growth (Anjum et al. 2017).

Activities of superoxide dismutase, catalase and ascorbate peroxidase have been enhanced under stress conditions up to a temperature of 50 °C which decreased thereafter. However, activities of glutathione reductase and peroxidase decreased with increase in temperature in the range of 20–50 °C (Gong et al. 2012). On the other hand, a boost in the biosynthesis of antioxidant enzyme activity hampers oxidative damage in the cotton crop (Gur et al. 2010). Hence, the biosynthesis of antioxidants has been boosted in tolerant genotypes and impaired in susceptible genotypes under stress conditions (Daud et al. 2012).

Heat stress is the main cause of declining root and shoot characteristics (Abdalla and El-Khoshiban 2007). Traits related to plant growth, such as root, shoot development, flowering and fiber quality, are mostly affected by high temperature (Farooq et al. 2015).

Relative leaf water content is the ratio of actual water content retained by leaf tissues under ambient conditions to the maximum water content that can be retained under excessive availability of water and can make the leaf tissue turgid to its full extent. Leaf water content is an important criterion in screening genotypes for heat stress tolerance (Rahman et al. 2000).

High temperature stress also affects cell membrane thermostability. Collado et al. (2010) observed higher cell membrane thermostability in tolerant genotypes than in susceptible genotypes under stress conditions. Cell membrane thermostability (CMT) has already been used as a screening tool for heat tolerance and heat-sensitive genotypes (Azhar et al. 2009). Moreover, the germination and seedling stages of the plant life cycle have always been found to be more sensitive to heat stress than the adult stages (Lianes et al. 2005).

In general, negative impacts of heat stress are well established, and a lot of efforts have been made to ameliorate these impacts through adjustment of sowing time, foliar spray of nutrients and growth regulators. But no efforts have been made to look into the genetic potential of available cultivars to cope with heat stress. The present study was therefore conducted to arrange promising cotton genotypes in the order of their thermotolerance.

## **MATERIALS AND METHODS**

#### **Experiment Site**

The experiment was conducted in the glass house of the Department of Plant Breeding and Genetics, Faculty of Agriculture, University of Agriculture Faisalabad, Pakistan.

# **Plant Material**

Genotypes FH-Lalazar, FH-142 and FH-114 were obtained from the Ayub Agriculture Research Institute (AARI) Faisalabad, Pakistan. Seeds of varieties CIM-598, CIM-599 and CIM-602 were procured from the Central Cotton Research Institute (CCRI), Multan, Pakistan. Seed material of cultivars VH-282, VH-326 and VH-341 was obtained from the Cotton Research Station, Vehari, Pakistan. Genotypes MNH-886, MNH-888 and MNH-992 were obtained from the Cotton Research Station Multan, Pakistan. Genotypes IUB-13, IUB-212 and IUB-222 were obtained from The Islamia University of Bahawalpur, Pakistan.

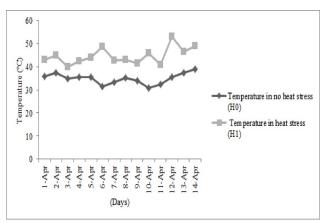
# **Treatments and Agronomic Practices**

The experiment consisted of heat stress [ $H_0$ = No heat stress (pots placed in ambient environment) and  $H_1$ = Heat stress (pots placed in glass house)] and cotton genotypes

(FH-Lalazar, FH-142, FH-114, CIM-598, CIM-599, CIM-602, VH-282, VH-326, VH-341, MNH-886, MNH-888, MNH-992, IUB-13, IUB-212, and IUB-222). Sowing was done in pots on 1st April 2016. Each pot had 8 kg clean, air-dried, sieved soil and the recommended fertilizer dose (N: P: K at the rate of 200: 115: 95 kg ha<sup>-1</sup>) was mixed thoroughly in the soil. In each pot, three seeds were dibbled and then only one plant was maintained. All cultural practices, except treatments, were kept normal and uniform for the whole experiment. Plants were uprooted 14 d after germination to record observations.

#### **Imposition of Heat Stress**

After complete germination of cotton plants, pots were placed in a glass house for 14 d. During imposition of heat stress, temperature was recorded in the morning, at noon and in the evening with the help of a digital temperature and humidity probe (Digital Multimeter-50302). Mean daily temperature in control and heat-stressed main plots are shown graphically in Figure 1.



**Fig. 1.** Mean daily temperature during experimentation on cotton genotypes.

# **Experimental Design**

A completely randomized design (CRD) with split treatment structure having three replications was used to conduct the experiment. The data collected were analyzed statistically ( $P \le 0.05$ ) using Fisher's analysis of variance (Steel et al. 1997) and treatments means were compared by using Tukey's Honestly Significant Difference (Tukey's HSD) test (Gomez and Gomez 1984). Correlation analyses were performed among different recorded parameters.

## **Observations Recorded**

Root and shoot lengths (in centimeters) were measured with the help of a ruler at the time of uprooting. The root length was measured from the point where the root and the shoot join to the end of the roots, while shoot length was measured from the root shoot joint to the top of the shoot. The root and shoot fresh weights were determined after separating the seedlings from the root and shoot joints, while the dry weights were recorded after drying the samples in an oven at 70 °C till the constant weight was achieved.

Relative leaf water contents (RLWC) were determined with some changes in methodology described by Weatherley (1950), as expressed in the formula

$$RLWC\% = \frac{(FW - DW)}{(TW - DW)} \times 100$$

where FW is fresh weight, DW is dry weight, and TW is turgid weight.

Relative cell injury (RCI) and cell and membrane thermostability (CMT) were determined according to the method of Sullivan (1972), as follows

RCI% = 
$$1 - \frac{1 - \left(\frac{T1}{T2}\right)}{1 - \left(\frac{C1}{C2}\right)} \times 100$$

where T<sub>1</sub> is the initial electrical conductivity (EC) value of the heat-treated vial, T<sub>2</sub> is the final EC value of the heattreated vial, C<sub>1</sub> is the initial EC value of the control vial, and C<sub>2</sub> is the final EC value of the control vial. CMT was calculated by deducting RCI (%) from 100.

Superoxide dismutase activity was measured as the amount of enzyme that inhibited photochemical reduction of nitro blue tetrazolium (NBT) and enzymelinked immunosorbent assay (ELISA) plate was used to note the absorbance at 560 nm (Giannopolitis and Ries 1977). Peroxidase activity was recorded as the amount of enzyme required for guaiacol oxidation and absorbance was measured at 470 nm using ELISA plate (Liu et al. 2009). Activity of catalase was determined as the amount of H<sub>2</sub>O<sub>2</sub> consumed by the enzyme and converted to H<sub>2</sub>O and O<sub>2</sub>, and recorded the absorbance at 240 nm wavelengths on ELISA plate (Liu et al. 2009).

#### **RESULTS AND DISCUSSION**

Generally, heat stress produced negative impacts on biomass accumulation, water relations, membrane integrity and antioxidant activities. However, varying responses of genotypes were observed under heatimposed and ambient environment. Thus, a significant heat × genotypes effect was recorded for all attributes (Tables 1 and 2). Negative implications of heat were a consequence of heat-induced transformations in water relations, biomass accumulation and metabolic processes of cotton.

**Table 1.** Analysis of variance for effect of heat stress on young seedlings of cotton genotypes.

SOV Replications	DF	Parameters								
	2	<b>RL</b> 0.5795	<b>SL</b> 0.070	<b>RFW</b> 0.00009	<b>RDW</b> 0.00001	<b>SFW</b> 0.00439	<b>SDW</b> 0.00001			
Heat (H)	1	60.1557**	660.319**	0.03823**	0.0007**	0.41534**	0.02770**			
Error 1	2	0.1436	0.656	0.00005	0.0000005	0.00223	0.00001			
Varieties (V)	14	7.4255**	44.931**	0.00285**	0.0001**	0.02129**	0.00027**			
H×V	14	0.1113**	6.429**	0.00038**	0.000004**	0.00177**	0.00002**			
Error 2	56	0.0282	0.544	0.00006	0.000001	0.00003	0.00001			

SOV, source of variation; DF, degree of freedom; RL, root length (cm); SL, shoot length (cm); RFW, root fresh weight (g); RDW, root dry weight (g); SFW, shoot fresh weight (g); SDW, shoot dry weight (g)

\*\*Highly significant at *P* < 0.01; \*Significant at *P* < 0.05

Table 2. Analysis of variance for effect of heat stress on biochemical characteristics of cotton genotypes.

sov	DF	Parameters								
		RLWC	СМТ	SOD	POD	CAT				
Replications	2	7.2	1.9	3.39	0.41	1.3				
Heat (H)	1	29440.9**	31158.1**	196.01*	1619.09**	5889.8*				
Error 1	2	12.6	5.2	6.58	7.69	95.6				
Varieties (V)	14	78.1**	145.7**	1249.35**	2014.03**	16251.8**				
H × V	14	22.4**	11.1**	535.81**	1053.06**	7650.0**				
Error 2	56	3.7	3.2	2.05	3.30	27.3				

SOV, source of variation; DF, degree of freedom; RLWC, relative leaf water contents (%); CMT, cell membrane thermostability; SOD, superoxide dismutase (U per mg protein); POD, peroxidase (U per mg protein); CAT, catalase (U per mg protein)

\*\*Highly significant at *P* < 0.01; \*Significant at *P* < 0.05

In terms of biomass-accumulating attributes, more shoot length, root fresh weight and shoot dry weights were manifested by genotypes MNH-888 and MNH-992 under no heat and heat-imposed conditions, while the highest shoot fresh weight was produced by genotypes MNH-888 and MNH-992 under no heat. On the other hand, under heat-stress environment, genotypes CIM-598 and CIM-602 manifested high shoot fresh weight. Although level of significance varied, more root length and root dry weight were observed in genotypes CIM-598, CIM-599 and CIM-602 under heat-imposed as well as in ambient environment. The lowest heat-triggered reduction in shoot fresh and dry weights was observed in genotypes CIM-599 and CIM-598. The least decline in root and shoot lengths under heat-imposed conditions (H1) versus ambient conditions (H<sub>0</sub>) was recorded for the genotype MNH-992, while the lowest decline in shoot fresh and dry weights was recorded for CIM-602 under heat stress (H1) versus the control. The highest diminishment in all biomass-accumulating attributes under heat versus control was observed for genotype FH-114 (Fig. 2).

Genotypes CIM-598, CIM-602, MNH-888 and MNH-992 showed the least decrease of 42–43% in relative leaf water contents (RLWC) under heat versus control, whereas the highest heat-mediated diminishment in RLWC was observed for FH-114 (60%) (Fig. 3). The genotype VH-341 exhibited more promising CMT (79.87) under ambient environment (H<sub>0</sub>), yet it was statistically similar to the other cultivars. On the other hand, under stressed conditions, the highest CMT was observed for IUB-13. Genotype IUB-13 was also statistically similar to VH-326, 282, 341, CIM-598, 599, 602, MNH-888 and MNH -992 for CMT. The maximum decline in CMT in stressed environment versus control was shown by FH-114 (63%) while the minimum was shown by VH-326 (44%), closely followed by IUB-13 (42%) (Fig. 3).

Activities of antioxidants such as superoxide dismutase (SOD), peroxidase (POD) and catalase (CAT) declined in genotypes FH-Lalazar, 114, 142, IUB-212, 222

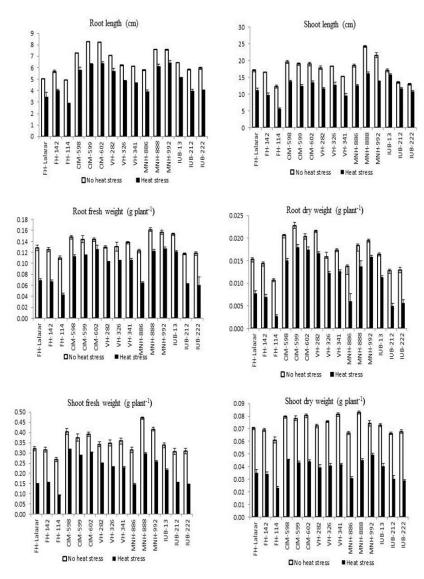


Fig. 2. Effect of heat stress on seedling growth of cotton genotypes.

and MNH-886. Among these, FH-114 manifested more heat-mediated inhibition in SOD, POD and CAT activities (55%, 57% and 56%, respectively) under heat stress condition versus control. However, enhancement in enzymatic activities was recorded for all other cultivars under heat stress versus control (Fig. 3). In terms of SOD activity, CIM-602 and VH-326 showed enhancement of 22% and 19%, respectively, under heat stress versus non-heat stressed environments (Fig. 3).

The cultivars VH-282 and VH-341 exhibited increment in POD activity than the cultivars CIM-598, 599, 602, VH-326, MNH-888, 992 and IUB-13. About 19% and 18% boost in POD activity was observed for VH-282 and VH-341, respectively. Under high temperature, maximum decline (65%) in POD activity was observed for FH-114. Comparatively more increase in CAT activity

under heat versus control was observed for genotypes VH-341 (19%), VH-326, MNH-888 and IUB-13 (18%) (Fig. 3).

## DISCUSSION

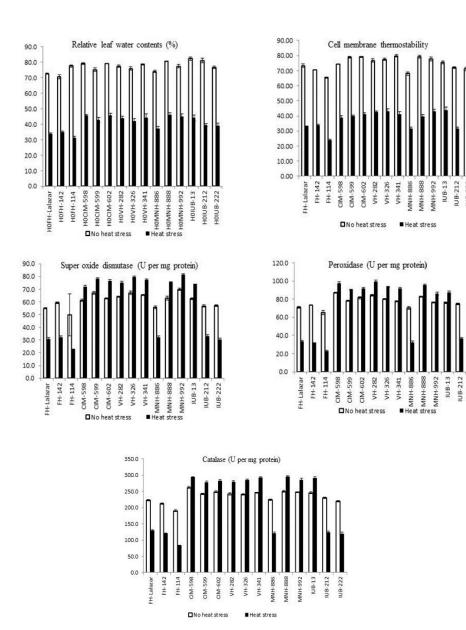
Heat stress generally had negative associations with root/shoot traits and enzymatic antioxidant activities of cotton. These negative implications might have arisen due to motivated ROS production and oxidation of cellular components such as lipids, proteins, DNA and antioxidants. High temperature either caused a decline in antioxidant biosynthesis or on some occasions, genotype-specific response was also marked.

In this trial, heat stress caused a significant reducing effect on growth parameters containing root and shoot fresh biomass. Our results are similar to those of Cheng et al. (2009) who recorded a significant drop in shoot fresh weight due to high temperature in wheat and sugarcane. Abo Hamad (2007) reported that shoot and root growth were visibly inhibited by heat treatment in cotton. However, root biomass decreased was not significantly under heat stress compared to ambient conditions (Kadir and Weigh 2007).

In earlier studies, declining responses of shoot lengths to water stress were observed in millet

(*Pennisetum glaucum*) and cotton (*Gossypium hirsutum*) but genotypes responded differently to these stress conditions (Iqbal et al. 2011). The root is an essential organ for the study of water relations in plants. Root development in cotton was genetically controlled (Riaz et al. 2013), but change in appearance of root traits due to environmental impacts had also been reported (Cooper et al. 2009). Furthermore, a strong positive and remarkable association of RL, SL, RFW, RDW, SFW and SDW with antioxidants resulted in the oxidative stress-mediated decrease in biomass accumulation (Table 3).

Naji and Devaraj (2011) observed increased peroxidase isozyme activity under heat and salt stress. Peroxidase antioxidant enzymes are heme-containing oxidoreductases that take part in a number of metabolic processes, such as cell elongation regulation (Andrews et



**Fig. 3.** Effect of heat stress on activities of antioxidant enzymes of cotton genotypes.

al. 2002), cell wall cross linking, lignification and oxidation of phenolic compounds.

Relative leaf water content (RLWC) is an important indicator of water status. As shown by cultivar 84-S with its higher RLWC than that of cultivar M-503, tolerant varieties did better in maintaining RLWC under stress compared to susceptible genotypes (Cia et al. 2012). Heatinduced diminishment in RLWC and a concomitant decrease in dry matter accumulation was also confirmed from a strong association of RLWC with SOD, POD and CAT under heat stress and ambient conditions (Table 3).

Heat stress/high temperature reduced cell membrane thermostability (CMT), leading to enhanced lipid

peroxidation and protein damage. CMT is considered as an indirect rapid measurement technique for heat stress tolerance in various crops such as cotton and sorghum measuring leaf outflow from leaf pieces (Wahid et al. 2007; Iglesias-Acosta et al. 2010). Intact membranes are crucial for cells to show growth, while CMT is a direct indicator of heat tolerance. Decline in CMT might have promoted cellular that can death ultimately lead to fewer bolls. Our results are similar to those of Ghaffari et al. (2015),that high temperature stress degraded membrane stability and other physiological processes. Moreover, a strong positive association of CMT with antioxidants established the role of antioxidants in sustaining membrane stability (Table 3).

Our results with regard to cell membrane thermostability are also in line with the findings of Yildirim et al. (2009) who reported that cell membrane thermostability of wheat genotypes

decreased and the highest decline was observed in susceptible genotypes. Increment in the activity of the antioxidant defense system under heat stress indicates the ability of the system to tolerate higher temperature. It is well documented that heat stress results in increased production of enzymatic antioxidants in heat-tolerant genotypes to diminish harmful consequences of reactive oxygen species (Iqbal et al. 2015). Some important mechanisms that may induce tolerance against high temperature environment are biosynthesis of late embryogenesis abundant proteins, ion adjustments, antioxidant scavenging system and transcriptional factors to counterbalance reactive oxygen species (Wang et al. 2014). A strong positive association of SOD, POD and

**Table 3.** Correlation matrix showing strength of association among recorded attributes of cotton genotypes under no heat (H<sub>0</sub>) and heat stress (H<sub>1</sub>).

Parame- ter	Treat- ment	RL	SL	RFW	RDW	SFW	SDW	RLWC	CMT	SOD	POD	CAT
RL	H <sub>0</sub>	1.00										
	H <sub>1</sub>	1.00										
SL	$H_0$	0.74**	1.00									
	H <sub>1</sub>	0.80**	1.00									
RFW	$H_0$	0.75**	0.81**	1.00								
	H <sub>1</sub>	0.91**	0.79**	1.00								
RDW	$H_0$	0.87**	0.65**	0.65**	1.00							
	H <sub>1</sub>	0.93**	0.71**	0.90**	1.00							
SFW	$H_0$	0.82**	0.89**	0.86**	0.66**	1.00						
	H <sub>1</sub>	0.94**	0.80**	0.91**	0.93**	1.00						
SDW	$H_0$	0.72**	0.74**	0.82**	0.68**	0.89**	1.00					
	H <sub>1</sub>	0.91**	0.85**	0.96*	0.89**	0.95**	1.00					
RLWC	$H_0$	0.15 <sup>NS</sup>	0.02 <sup>NS</sup>	0.40 <sup>NS</sup>	0.01 <sup>NS</sup>	$0.23^{NS}$	$0.23^{\text{NS}}$	1.00				
	H <sub>1</sub>	0.88**	0.69**	0.91**	0.84**	0.91**	0.90**	1.00				
CMT	$H_0$	0.74**	0.59*	0.75**	0.75**	0.76**	0.90**	0.30 <sup>NS</sup>	1.00			
	H <sub>1</sub>	0.78**	0.62*	0.91**	0.84**	0.78**	0.83**	0.86**	1.00			
SOD	$H_0$	0.69**	0.61*	0.72**	0.74**	0.66**	0.77**	0.06 <sup>NS</sup>	0.87**	1.00		
	H <sub>1</sub>	0.86**	0.71**	0.97**	0.92**	0.89**	0.93**	0.89**	0.94**	1.00		
POD	$H_0$	0.70**	0.60*	0.59*	0.79**	0.70**	0.76**	0.31 <sup>NS</sup>	0.78**	0.62**	1.00	
	H <sub>1</sub>	0.84**	0.70**	0.94**	0.91**	0.91**	0.92**	0.89**	0.91**	0.98**	1.00	
CAT	$H_0$	0.74**	0.67**	0.82**	0.76**	0.76**	0.85**	0.43 <sup>NS</sup>	0.85**	0.78**	0.84**	1.00
	H <sub>1</sub>	0.84**	0.71**	0.97**	0.90**	0.90**	0.93**	0.90**	0.94**	0.99**	0.99**	1.00

Highly significant; NS Non-significant; n (number of pairs of observations) = 90 RL, root length; SL, shoot length; RFW, root fresh weight; RDW, root dry weight; SFW, shoot fresh weight; SDW, shoot dry weight; RLWC, relative leaf water contents; CMT, cell membrane thermostability; SOD, superoxide dismutase; POD, peroxidase; CAT, catalase

CAT with RL, SL, RFW, RDW, SFW and SDW resulted in the biomass-mediated improvements in the defense system of antioxidants (Table 3).

Increment in SOD activity in tolerant cultivars might be due to increased production of superoxide radical (O2-1) under high temperature environment as superoxide radical acts as substrate for SOD (Jiang and Huang 2001). Heat-susceptible cultivars showed decrement in SOD activity under high temperature that can be ascribed to lower efficacy of the scavenging mechanism of these cultivars. Heat-tolerant cultivars recorded increment while susceptible cultivars represented decline in SOD activity under high temperature (Naderi et al. 2014). Almeselmani et al. (2006) recorded statistically significant increments in SOD activity in drought-tolerant Phaseolus acutifolius and heat-tolerant wheat cultivars. Detoxification of hydrogen peroxide (H2O2) to H2O and O2 forms in plants is facilitated both by POD and CAT. Higher H<sub>2</sub>O<sub>2</sub> production under heat stress might have resulted in increment of POD and CAT activity as a defensive mechanism against stress in tolerant cultivars. Our results are in agreement with earlier findings of Iqbal et al. (2015) who observed up-marking in POD and CAT activity under water stress environment in drought-tolerant cultivars of wheat. These results are also comparable to those of Khaliq et al. (2015) who observed increment in POD and CAT activity in salt-stress environment in tolerant cultivars of wheat. However, decrement or no change in CAT activity was recorded for stress-tolerant genotypes by Wang et al. (2014). Kumar et al. (2012) documented increase in SOD, POD and CAT activity under heat-stressed conditions in heat-tolerant genotypes against the decline for heat-susceptible cultivars of crop plants.

# **CONCLUSION**

Heat-induced change in superoxide dismutase, peroxidase, catalase, relative leaf water contents and cell membrane thermostability is a good indicator of change in dry-matter accumulating traits. Heat stress adversely affected root and shoot attributes of all genotypes. Varieties CIM-598, CIM-599, CIM-602, VH-282, VH-326, VH-341, MNH-888, MNH-992 and IUB-13 were

moderately heat-tolerant; FH-Lalazar, FH-142, MNH-886, IUB-212 and IUB-222 showed medium susceptibility while FH-114 was the most susceptible to terminal heat stress.

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