Electronic Journal of Plant Breeding, 6(1): 341-349 (Mar 2015)
ISSN 0975-928X

Research Note

Screening for heat tolerance in wheat germplasm by applying physiological and biochemical indices

Mamta Arya[1] and J.P. Jaiswal
Department of Genetics and Plant Breeding, GBPUA&T, Pantnagar, Uttarakhand
[1]Present Address: Directorate of Soybean Research, Khandwa Road, Indore
* E mail: mamta.gpb@gmail.com

(Received: 12 Aug 2014; Accepted:22 Jan 2015)

Abstract
In the present study 102 indigenous germplasm lines were used and the effect of sudden heat shock was studied on the basis of six physiological and biochemical characters *viz.*, chlorophyll content, Membrane thermostability (MSI), Superoxide dismutase (SOD) activity, proline content , malondialdehyde (MDA) content and hydrogen peroxide (H_2O_2) content. Some promising lines were selected for each trait so that these can be used for the accumulation of favorable alleles in the high yielding wheat cultivar background.

Key words:
Wheat, germplasm, heat tolerance, physiology, biochemical

Wheat yields can be severely reduced in moisture-stressed environments (Morris *et al*., 1991), which affect at least 15 million ha of spring wheat alone in the developing world. The optimum temperature for wheat growth and development varies from 1.8 to 34.3^{0}C while for anthesis and grain filling its 12-22^{0}C (Farooq *et al.,* 2011). Exposure to temperatures above this can significantly reduce grain yield (Tewolde *et al.,* 2006). The current estimates indicate that in India alone 13.5 million hectare of wheat cropping area are reported to be under heat stress (Joshi *et al*., 2007). Despite advances in our understanding of genes of major effect conferring disease resistance in wheat, the genetic basis of heat adaptation is poorly understood. In present scenario, physiological traits associated with heat adaptation constitute the best available tool for genetic improvement of crops, since they represent *de facto* favorable combinations of alleles. When followed by hybridization between genotypes with different but complementary physiological traits, the probability of cumulative gene action increases (Reynolds and Rebetzke, 2011). An advantage to the process of screening through physiological and/or biochemical traits is that the screening can be performed in seedling stage thus saving a lot of time and providing opportunities to handle a large number of population effectively. In this context the present study was carried out to screen germplasm lines for heat tolerance using physiological and biochemical traits.One hundred two indigenous germplasm lines were procured in January 2011 from National Bureau of Plant Genetic Resources, New Delhi, India, and were used in the study. The experiment was laid out in Completely Randomized Design with two factors. This was planned by germinating six seeds from each accession in Petri plates. Each accession replicated thrice. Two set of such plates (total 6 for each accession) were kept in BOD incubator under normal temperature (20^{0}C, 70% RH) and supplied with Hoagland's nutrient medium (Hoagland and Snyder, 1933) to ensure proper supply of nutrients to each accession. After 10 days, one set was transferred to another incubator and a heat shock of 37^{0}C was given for 8 hrs (Efeoglu and Terzioglu, 2009). The samples were directly used for analysis and physiological and biochemical assays were performed with control as well as treated samples and data were recorded for further analyses.Observations on six physiological and biochemical characters were recorded *viz.*, Chlorophyll content (Hiscox and Israelstam, 1979), Membrane Stability Index (Sairam *et al*., 1997), Super Oxide Dismutase activity (Giannopolitis and Ries, 1977), Proline content (Bates *et al*., 1973) , Malondialdehyde content (Heath and Packer, 1968) and Hydrogen peroxide content (Alexieva *et al.*, 2001).Analysis of variance (ANOVA) of the data was performed for two factor CRD and means and variances were ruled out.

The mean sum of square for genotypes, treatment and interaction (genotype x treatment) was found to be significant for all the six characters, under study. Thus the analysis of variance revealed significant difference among these genotypes (Table 1).

Physiological and biochemical screening **:-** Characterization of desirable traits is very much essential for utilization of any genotype in breeding for heat tolerance. When the above germplasm accessions were subjected to heat stress there was a change in the patterns of traits studied in the terms of relative percentage of decrease. In most of the genotypes, there was a reduction in chlorophyll content, membrane stability index and hydrogen peroxide content, whereas there was an increase in values of superoxide dismutase, malondialdehyde content and proline content (Table 2). There was variation seen among the genotypes for their response to heat stress and on this basis few accessions were screened out which have shown better performances (Table 3).

*Changes in chlorophyll content:*Chlorophyll content in plants is an important trait to access photosynthetic efficiency under stressed conditions. High temperature treatment is known to influence the total chlorophyll content of a leaf. Marked inhibition in chlorophyll accumulation, even chlorosis occurs at high temperature (Mohanty and Mohanty, 1988). Amount of total chl was reduced in most of the accessions under heat stress. However accession number IC 273946 showed an increase from 0.89 mg/g (c) to 3.86 mg/g (t), followed by IC 47012 (c- 0.80 mg/g; t- 2.23 mg/g). Thus, total chl changed significantly in the 37^{0}C, 8 h treatment in most of the accessions (Table 2). Similar results were also obtained by Efeoglu and Terzioglu (2009) while dealing with photosynthetic response of wheat varieties to heat stress.

*Changes in MSI content:*Data shows that imposition of heat stress to the seedlings significantly reduced the level of MSI. Most reduction in MSI was reported in accession number IC 28564. Thus it can be considered as most heat labile and can be rejected for further use. The results are in accordance with Ibrahim and Quick (2001) and Bhutta (2011). However some accessions were found to be having increased MSI after heat stress treatment like IC 258215 (c- 16.22; t- 31.68), IC 290195 (c- 28.7; t-

36.75), IC 290249 (c- 27.27; t- 50.95) and IC 310584 (c- 16.27; t- 36.42) (Table 2).

*Changes in SOD activity:*Significant change in SOD activity (an increase) was observed in all the accessions by imposing heat stress. The increase in activity ranged from a minimum of 0.03 for IC 535396 to a maximum of 2.4 for IC 290222. (Table 2). This shows that the accession IC 290222 has the potential of tolerating stress by producing large amount of SOD. This was followed by IC 279707 (2.15) and IC 290195 (2.14).

Heat stress increased the superoxide levels in cells. If this radical is not scavenged by SOD, it disturbs the vital biomolecules (Mittler, 2000). SODs are metallo-protein which catalyzes dismutation of the superoxide free radicals (O^-_2) to molecular oxygen and H_2O_2. In this study, differences in SOD activity of the accessions were observed. Some accessions showed higher SOD activity compared to another. It could be stated that the higher SOD activity is part of the ability of the genotype to tolerate high temperature. Increase in SOD activity as a result of heat stress was reported by Jiang and Huang (2001); such increase could be due to the accumulation of Cu/Zn-SOD mRNA (Tsang *et al.*, 1991).

Changes in Proline accumulation: Under the different abiotic stressed conditions, proline is accumulated in cells as an osmoprotectant. Increased amount of proline was found under heat stress. The maximum increase from that under control was found in accession IC 47454 (12.95) followed by IC 535475 (12.12) and IC 316104 (11.89). accession IC 281558 has shown minimum increase in proline content after stress (1.73). There was also recorded a decline in the proline levels under heat stressed condition, out of which IC 28547 showed maximum decrease from 32.18 µ mol (c) to 30.29 µ mol (t) (Table 2). Higher proline content in wheat plants after water stress application has been reported by Vendruscolo *et al.* (2007). Increased amount of proline was also established in several stress conditions such as salinity (Poustini *et al.*, 2007), cold (Charest and Phan, 1990) and UV (Tian and Lei, 2007).

Changes in MDA content: Malondialdehyde (MDA) content was estimated in the leaf sample of 102 wheat accessions growing under controlled as well as heat stressed conditions. There was a significant change in the MDA levels of the plants subjected to heat stress. MDA value increases under heat stress but its magnitude is a criteria to select genotypes for relatively lower increase. Minimum increase from control was found for accession IC 310102 (0.02) followed by IC 279336 (0.51) and IC 290195 (0.56) (Table 2). High level of MDA content indicates more lipid peroxidation and more membrane permeability and species are comparatively more susceptible for stress than those which produce less MDA content at higher magnitude of stress. Reddy *et al.* (1998) observed rice lines showed less accumulation of MDA were found superior to other lines in terms of stress tolerance. Reduced contents of MDA is an important indicator of stress tolerance as shown in some earlier studies (Liang *et al.*, 2003; Braukova *et al.*, 2005; Ruiz *et al.*, 2005).

Changes in H_2O_2 content: Hydrogen peroxide (H_2O_2) is produced from β-oxidation of fatty acids and photorespiration. A decreased level of H_2O_2 in the cell under stress corresponds to higher activity of CAT and APX and increased stability of membranes and CO_2 fixation (Bhutta, 2011). In this study H_2O_2 content was estimated in control as well as heat stressed plants of 102 accessions. The value during control condition varied between 0.98 µ mol for IC 316093 and 3.05 µ mol for IC 290219, whereas, during stressed conditions the range of the hydrogen peroxide content varied between 0.46 µ mol for IC 47014 and 3.23 µ mol for IC 258214 and IC 290185 (Table 2). Maximum decrease was found in accession IC 279159 (1.9) which means that this accession performs better in terms of H2O2 and further leading to increased CAT and APX activities. The results are in accordance with previous observations of Esfandiari *et al.* (2007) and Shao *et al.* (2005). On the basis of the performances shown by the accessions under study, some promising ones were selected for each character (Table 3). Thus these accessions can be used in breeding programme for development of heat tolerant varieties so that the mitigating effect of global warming can be tackled.

Acknowledgement
The authors sincerely thank NBPGR, New Delhi, for experimental material and Director, Experiment Station, GBPUAT, Pantnagar for providing necessary facilities for the investigation and the author Mamta A acknowledges the financial support from UGC.

References

Alexieva, V., Sergiev, I., Mapelli, S. and Karanov, E. 2001. The effect of drought and ultraviolet radiation on growth and stress markers in pea and wheat. *Plant Cell Environ.*, 24:1337–1344.

Bates, L. S., Waldren, R. P. and Teare, I.D. 1973. Rapid determination of free proline for water stress studies. *Plant and Soil*, 39:205-208.

Bhutta, W.M. 2011. Antioxidant activity of enzymatic system of two different wheat (*Triticum aestivum*

L) cultivars growing under salt stress. *Plant Soil Environ.,* 57(3): 101-107.

Braukova, L., Ivanov, S., Alexieva, V. and Karanov, E. 2005. Salt induced alterations in the levels of some oxidative parameters and unspecific defence compounds in leaves of two plant species (cotton and bean) with different sensitivity to salinity. *Compt Rend De J'Acad Bulgare des Sci* 58(11): 1307-1312.

Charest, C. and Phan, C.T. 1990. Cold acclimation of wheat (*Triticum aestivum* L): Properties of enzymes involved in proline metabolism. *Physiol Plant* 80(2): 159-168.

Efeoglu, B. and Terzioglu, S. 2009. Photosynthetic responses of two wheat varieties to high temperature. *Eur Asia J Bio Sci.,* 3: 97-106.

Esfandiari, E., Shakiba, M.R., Mehboob, S., Alyari, H. and Toorchi, M. 2007. Water stress antioxidant enzyme activity and lipid peroxidation in wheat seedlings. *J. Food Agri and Environ.,* 5: 149-153.

Farooq, M., Bramley, H., Jairo, A., Palta and Siddique, K.H.M. 2011. Heat Stress in Wheat during Reproductive and Grain-Filling Phases. *Critical Rev. in Plant Sci.,* 30:1–17.

Giannopolitis, C.N. and Ries, S.K. 1977. Superoxide dismutases: I Occurrence in higher plants. *Plant Physiol.,* 59(2):309–314.

Heath, R.L. and Packer, L. 1968. Photoperoxidation in isolated chloroplasts I Kinetics and stoichiometry of fatty acid peroxidation. *Archives in Biochem. and Biophy.,*125:189–198.

Hiscox, J.D. and Israelstam, G.F. 1979. A method for the extraction of chlorophyll from leaf tissue without maceration. *Canadian J. Bot.,*57: 1332-1334.

Hoagland, D.R. and Snyder, W.C. 1933. Nutrition of strawberry plants under controlled conditions: (a) Effects of deficiencies of boron and certain other elements: (b) Susceptibility to injury from sodium salts. *Proc. Am Sot Hort Sci*., 30: 288-294.

Ibrahim, M., Amir, H. and Quick, J.S. 2001. Heritability of heat tolerance in winter and spring wheat. *Crop Sci.,* 41:1401–1405.

Jiang, Y. and Huang, B. 2001. Effect of calcium on antioxidant activities and water relation associated with heat tolerance in two cool season grasses. *J. Exp Bot.,* 52: 341-349.

Liang, Y., Chen, Q., Liu, Q., Zhang, W. and Ding, R. 2003. Exogenous silicon (Si) increases antioxidant enzymes activity and reduces lipid peroxidation in roots of salt stressed barley (*Hordeum vulgare* L). *J. Plant Physiol.,* 160(10): 1157-1164.

Mittler, R. 2000. Oxidative stress antioxidants and stress tolerance. *Trends in Plant Sci.,* 7: 405-410.

Morris, M.L., Belaid, A. and Byerlee, D. 1991. Wheat and Barley production in rainfed marginal environments of the developing world World Wheat Factors and Trends Mexico DF CIMMYT.

Poustini, K., Siosemardeh, A. and Ranjbar, M. 2007. Proline accumulation as a response to salt stress in 30 wheat (*Triticum aestivum* L) cultivars differeing in salt tolerance. *Genet. Resour. Crop. Evol.,* 54(5): 925-934.

Reddy, C.P., Vajranabhaian, S.N. and Kumar, M.U. 1998. Lipid peroxidation as a mechanism of stress tolerance in upland rice (*Oryza sativa* L) calli. *Indian J. Plant Physiol.,* 3: 68-70.

Reynolds, M.P. and Rebetzke, G. 2011. Application of plant physiology in wheat breeding. In AP Bonjean WJ Angus M Van Ginkel eds: The World Wheat Book: A History of Wheat Breeding Vol 2 Editions TEC Paris pp 877–906.

Ruiz, J.M., Blasco, B., Rivero, R.M. and Romero, L. 2005. Nicotine-free and salt-tolerant tobacco plants obtained by grafting to salinity resistant root stocks of tomato. *Physiol Plant.,* 124: 465-475.

Shao, H.B., Liang, Z.S. and Shao, M.A. 2005. Changes of antioxidative enzymes and MDA content under soil water deficits among 10 wheat (*Triticum aestivum* L) genotypes at maturation stage Colloids and Surfaces. *Biointerfaces,* 45: 7-13.

Tewolde, H., Fernandez, C.J. and Erickson, C.A. 2006. Wheat cultivars adapted to post-heading high temperature stress. *J. Agron. Crop Sci.,* 192: 111–120.

Tian, X.R. and Lei, Y.B. 2007. Physiological responses of wheat seedlings to drought and UV-B radiations effect of exogenous sodium nitroprusside application. *Russian J. Plant Phys.,* 54(5): 676-682.

Tsang, W.T., Bowler, C., Herouert, D., Van Camp, W., Villarred, R., Genetello, C., Van Montagu, M. and Inze, D. 1991. Differential regulation of superoxide dismutase in plants exposed to environmental stress. *The Plant Cell,* 3: 783-792.

Vendruscolo, A.C.G., Schuster, I., Pileggi, H., Scapim, C.A., Molinari, H. Bc., Marur, C.J. and Vieira, L.G.C. 2007. Stress-induced synthesis of proline confers tolerance to water deficit in transgenic wheat. *J. Plant Physiol.,* 164(10): 1367-1376.

Table 1. Analysis of variance for different characters

Source of variation	Degree of Freedom	Mean squares					
		Total chl	MSI	SOD	Proline	MDA	H_2O_2
Genotyopes (a)	101	0.0925**	793.02**	0.399**	517.95**	151.95**	0.9532**
Heat stress (b)	1	0.167**	12509.42**	67.209**	5651.05**	404.99**	4.027**
a x b	101	0.839**	364.101**	0.4279**	11.5077**	129.99**	0.9599**
error	408	0.0049	2.5079	0.0022	1.663	0.0841	0.00023

** Significant at 1% level of probability

Table 2a. Means for different characters

	Chlorophyll (mg/g)			MSI (%)			SOD (u/ml)		
	Control	Stress	Difference	Control	Stress	Difference	Control	Stress	Difference
IC 258211	2.36	1.72	0.64	36.63	34.48	2.15	0.56	1.51	0.95
IC 258212	1.98	0.47	1.51	35.50	14.55	20.95	1.35	1.90	0.55
IC 258214	2.48	0.93	1.55	29.18	15.34	13.84	0.42	1.26	0.84
IC 258215	2.51	1.74	0.77	16.22	31.68	-15.46	0.41	1.02	0.61
IC 260944	1.12	2.14	-1.02	27.65	12.37	15.28	0.41	1.76	1.35
IC 273946	0.89	3.86	-2.97	44.07	32.88	11.19	1.55	1.04	-0.51
IC 273954	1.25	0.92	0.33	25.80	10.40	15.4	0.41	0.93	0.52
IC 274004	2.16	2.52	-0.36	48.68	21.47	27.21	1.29	1.54	0.25
IC 279526	1.62	1.64	-0.02	19.59	21.03	-1.44	1.59	1.54	-0.05
IC 279642	2.65	2.03	0.62	36.86	22.22	14.64	0.42	1.19	0.77
IC 279707	1.76	2.85	-1.09	30.53	35.95	-5.42	0.09	2.24	2.15
IC 281554	1.72	2.02	-0.3	62.55	30.56	31.99	0.28	0.92	0.64
IC 290195	2.13	2.30	-0.17	28.70	36.75	-8.05	0.15	2.29	2.14
IC 290197	1.47	1.76	-0.29	25.31	12.24	13.07	0.69	1.43	0.74
IC 290205	2.05	2.02	0.03	18.20	27.19	-8.99	0.35	0.63	0.28
IC 290219	0.92	1.50	-0.58	21.38	15.18	6.2	0.49	1.23	0.74
IC 290220	1.06	2.15	-1.09	34.73	34.66	0.07	0.12	1.28	1.16
IC 290221	1.58	2.30	-0.72	17.21	12.64	4.57	0.94	1.86	0.92
IC 290222	1.48	1.43	0.05	34.13	24.14	9.99	0.59	2.99	2.4
IC 290230	2.31	3.62	-1.31	14.58	23.82	-9.24	0.82	1.06	0.24
IC 290236	1.75	2.31	-0.56	24.92	26.63	-1.71	0.53	1.19	0.66
IC 290243	2.14	2.99	-0.85	24.67	28.98	-4.31	0.56	1.64	1.08
IC 290249	1.49	1.60	-0.11	27.27	50.95	-23.68	0.09	0.86	0.77
IC 290264	1.91	2.04	-0.13	37.07	26.56	10.51	0.16	1.73	1.57
IC 290331	2.05	2.02	0.03	32.21	57.37	-25.16	0.63	0.95	0.32
IC 290337	1.50	2.86	-1.36	38.13	19.53	18.6	0.73	1.55	0.82
IC 47011	1.33	1.47	-0.14	23.57	39.92	-16.35	0.63	1.64	1.01
IC 47012	0.80	2.23	-1.43	17.67	32.12	-14.45	0.86	0.92	0.06
IC 47014	1.59	2.23	-0.64	19.62	74.33	-54.71	1.03	0.19	-0.84
IC 47017	0.41	2.12	-1.71	52.67	32.05	20.62	0.53	0.74	0.21
IC 47454	1.10	0.59	0.51	14.26	9.65	4.61	0.82	1.28	0.46
IC 535195	1.78	1.28	0.5	16.46	19.38	-2.92	0.48	1.21	0.73
IC 535212	1.85	1.69	0.16	27.45	13.87	13.58	0.78	0.03	-0.75
IC 535274	1.45	1.26	0.19	22.87	25.41	-2.54	1.06	0.97	-0.09
IC 35029	1.66	2.04	-0.38	34.26	26.58	7.68	1.30	1.55	0.25
IC 35032	1.15	1.18	-0.03	34.19	23.81	10.38	0.48	0.80	0.32
IC 290061	1.51	1.09	0.42	31.25	0.76	30.49	0.44	1.21	0.77
IC 41590	1.24	1.61	-0.37	10.62	11.92	-1.3	0.32	1.76	1.44
IC 41589	1.35	0.83	0.52	20.21	13.21	7	0.49	1.21	0.72
IC 535396	1.78	2.18	-0.4	22.15	18.11	4.04	0.52	0.55	0.03
IC 535414	1.28	1.84	-0.56	27.39	19.62	7.77	0.55	1.09	0.54
IC 535417	1.18	1.09	0.09	28.25	25.65	2.6	0.71	0.54	-0.17
IC 535443	0.87	0.96	-0.09	21.87	2.76	19.11	0.69	1.39	0.7

Table 2a. Contd..

	Chlorophyll (mg/g)			MSI (%)			SOD (u/ml)		
	Control	**Stress**	**Difference**	**Control**	**Stress**	**Difference**	**Control**	**Stress**	**Difference**
IC 535448	1.50	1.61	-0.11	31.75	24.07	7.68	0.56	0.74	0.18
IC 535449	1.55	1.24	0.31	10.75	18.87	-8.12	0.53	1.13	0.6
IC 535462	1.38	1.78	-0.4	33.33	35.37	-2.04	0.47	2.40	1.93
IC 535474	1.47	1.86	-0.39	27.85	37.20	-9.35	0.43	1.28	0.85
IC 535475	2.04	1.50	0.54	35.58	27.57	8.01	0.38	1.86	1.48
IC 535489	1.79	2.13	-0.34	37.14	46.45	-9.31	0.34	1.04	0.7
IC535259	1.17	2.03	-0.86	78.12	47.83	30.29	0.35	0.23	-0.12
IC 260946	1.38	2.71	-1.33	66.27	39.72	26.55	0.30	0.82	0.52
IC 279159	1.55	1.94	-0.39	45.41	25.79	19.62	0.31	1.19	0.88
IC 279317	2.23	2.31	-0.08	39.18	32.62	6.56	0.23	2.24	2.01
IC 279331	2.11	1.84	0.27	63.94	26.23	37.71	0.35	0.92	0.57
IC 279336	2.02	0.48	1.54	39.79	35.37	4.42	0.38	1.19	0.81
IC 279480	1.78	1.00	0.78	32.43	37.20	-4.77	0.42	2.24	1.82
IC 279866	1.86	1.69	0.17	65.47	27.57	37.9	0.62	0.92	0.3
IC 279868	2.33	2.10	0.23	57.11	46.45	10.66	0.45	0.90	0.45
IC 28086	2.02	0.48	1.54	68.74	42.52	26.22	0.58	1.09	0.51
IC 281555	1.08	1.05	0.03	37.12	29.23	7.89	1.19	0.77	-0.42
IC 281558	1.94	1.72	0.22	37.84	34.02	3.82	1.04	1.11	0.07
IC 281565	2.13	2.12	0.01	35.56	31.54	4.02	0.74	1.44	0.7
IC 28518	2.16	1.79	0.37	52.43	17.03	35.4	0.58	1.52	0.94
IC 28547	2.20	2.27	-0.07	39.11	11.59	27.52	0.54	0.92	0.38
IC 28552	1.46	1.80	-0.34	22.96	13.94	9.02	0.62	1.32	0.7
IC 28557	1.19	2.27	-1.08	53.17	37.76	15.41	0.64	1.64	1
IC 28564	2.06	1.95	0.11	73.21	7.88	65.33	0.67	1.28	0.61
IC 290030	2.01	2.28	-0.27	34.06	10.88	23.18	0.53	1.21	0.68
IC 36882	1.60	1.75	-0.15	55.99	33.16	22.83	0.72	0.97	0.25
IC 290068	1.41	2.11	-0.7	46.24	17.73	28.51	0.78	1.55	0.77
IC 290095	1.95	2.14	-0.19	39.00	38.01	0.99	0.63	1.32	0.69
IC 290156	1.97	2.71	-0.74	39.49	27.65	11.84	0.49	1.64	1.15
IC 290181	2.34	1.52	0.82	64.95	33.33	31.62	0.44	1.47	1.03
IC 290185	2.55	2.18	0.37	69.68	62.84	6.84	0.34	1.13	0.79
IC 290196	1.68	1.51	0.17	56.91	54.45	2.46	0.43	0.86	0.43
IC 290256	2.25	2.20	0.05	66.49	55.22	11.27	0.32	1.21	0.89
IC 290296	2.58	2.38	0.2	28.20	35.83	-7.63	0.27	0.43	0.16
IC 310102	2.32	1.54	0.78	37.78	44.01	-6.23	0.33	1.19	0.86
IC 310120	2.37	2.27	0.1	32.19	37.40	-5.21	0.41	2.01	1.6
IC 310584	1.61	2.30	-0.69	16.27	36.42	-20.15	0.42	1.39	0.97
IC 310586	1.42	1.61	-0.19	73.37	49.16	24.21	0.34	1.37	1.03
IC 310588	2.27	0.94	1.33	57.52	36.42	21.1	0.53	0.76	0.23
IC 310590	2.35	2.20	0.15	29.67	29.50	0.17	0.56	1.15	0.59
IC 313156	2.43	1.75	0.68	23.00	32.50	-9.5	0.62	1.55	0.93
IC 31529	2.32	1.12	1.2	41.63	45.79	-4.16	0.63	0.80	0.17
IC 315921	2.43	1.15	1.28	44.93	40.81	4.12	0.68	0.37	-0.31
IC 316085	2.03	1.69	0.34	60.88	35.43	25.45	0.68	0.78	0.1
IC 316087	1.97	1.05	0.92	40.52	28.76	11.76	0.61	0.92	0.31
IC 316088	2.33	1.65	0.68	40.19	37.77	2.42	0.53	0.76	0.23
IC 316091	2.46	2.12	0.34	19.97	23.68	-3.71	0.53	1.76	1.23
IC 316092	2.58	1.50	1.08	27.64	17.73	9.91	0.48	2.06	1.58
IC 316093	2.41	2.22	0.19	28.71	21.83	6.88	0.54	1.28	0.74
IC 316099	2.19	1.87	0.32	56.86	39.35	17.51	0.41	1.43	1.02
IC 316102	2.18	1.74	0.44	25.48	29.06	-3.58	0.41	0.64	0.23
IC 316104	1.89	2.12	-0.23	26.29	23.16	3.13	0.39	0.92	0.53

Table 2a. Contd..

	Chlorophyll (mg/g)			MSI (%)			SOD (u/ml)		
	Control	Stress	Difference	Control	Stress	Difference	Control	Stress	Difference
IC 31614	2.05	1.94	0.11	41.33	32.28	9.05	0.37	0.43	0.06
IC 31615	2.08	1.70	0.38	36.50	34.95	1.55	0.29	1.09	0.8
IC 317422	2.07	1.01	1.06	59.69	49.75	9.94	0.29	1.79	1.5
IC 317529	2.56	2.27	0.29	40.99	41.83	-0.84	0.28	1.19	0.91
IC 317610	1.65	1.68	-0.03	45.74	28.95	16.79	0.33	1.63	1.3
IC 317632	2.24	1.22	1.02	39.87	25.64	14.23	0.37	0.92	0.55
IC 31774	2.02	1.15	0.87	38.07	30.84	7.23	0.33	1.35	1.02

Tot chl- total chlorophyll; MSI- Membrane Stability Index; SOD- Super Oxide Dismutase; MDA- Malondialdehyde; H_2O_2- Hydrogen peroxide

Table 2b . Means for different characters

	Proline (μ mol)			MDA (μ mol)			H2O2 (μ mol)		
	Control	Stress	Difference	Control	Stress	Difference	Control	Stress	Difference
IC 258211	16.39	25.56	9.17	6.69	8.28	1.59	1.98	2.48	-0.5
IC 258212	25.72	33.87	8.15	9.20	15.51	6.31	2.64	2.47	0.17
IC 258214	5.36	12.68	7.32	6.69	8.73	2.04	1.06	3.23	-2.17
IC 258215	19.62	26.99	7.37	13.85	17.57	3.72	2.93	1.93	1
IC 260944	13.48	17.62	4.14	13.14	9.25	-3.89	2.64	2.02	0.62
IC 273946	17.78	25.93	8.15	8.69	13.31	4.62	1.41	1.32	0.09
IC 273954	33.57	41.44	7.87	18.04	13.51	-4.53	2.82	2.35	0.47
IC 274004	13.34	21.36	8.02	7.78	9.18	1.4	2.23	1.56	0.67
IC 279526	18.47	26.53	8.06	8.56	14.54	5.98	1.43	1.87	-0.44
IC 279642	23.92	31.38	7.46	13.78	11.96	-1.82	1.67	1.44	0.23
IC 279707	33.02	40.75	7.73	16.56	17.51	0.95	2.85	3.03	-0.18
IC 281554	13.62	21.40	7.78	8.62	13.51	4.89	2.16	1.05	1.11
IC 290195	32.69	30.08	-2.61	14.88	15.44	0.56	1.41	0.85	0.56
IC 290197	18.29	25.93	7.64	3.66	16.73	13.07	1.93	1.58	0.35
IC 290205	42.39	50.08	7.69	16.04	11.44	-4.6	2.11	1.54	0.57
IC 290219	22.60	30.25	7.65	9.44	18.15	8.71	3.05	1.47	1.58
IC 290220	15.86	23.55	7.69	7.51	16.15	8.64	1.27	1.29	-0.02
IC 290221	30.18	37.91	7.73	8.80	10.47	1.67	1.73	2.21	-0.48
IC 290222	17.99	25.77	7.78	9.83	19.18	9.35	1.06	1.73	-0.67
IC 290230	21.36	29.18	7.82	7.31	7.25	-0.06	1.87	1.57	0.3
IC 290236	41.49	49.36	7.87	15.76	11.89	-3.87	1.44	2.57	-1.13
IC 290243	31.88	37.40	5.52	4.15	11.57	7.42	1.75	1.24	0.51
IC 290249	24.87	29.74	4.87	16.28	9.44	-6.84	2.03	1.47	0.56
IC 290264	15.03	18.84	3.81	11.44	10.09	-1.35	2.02	1.08	0.94
IC 290331	37.19	42.81	5.62	12.22	15.25	3.03	1.46	1.30	0.16
IC 290337	26.16	32.37	6.21	7.31	9.70	2.39	1.63	1.21	0.42
IC 47011	19.92	25.44	5.52	16.34	8.22	-8.12	2.21	1.66	0.55
IC 47012	29.02	33.28	4.26	18.99	13.44	-5.55	1.29	1.40	-0.11
IC 47014	16.09	19.95	3.86	12.47	8.02	-4.45	1.77	0.46	1.31
IC 47017	8.94	13.73	4.79	3.05	18.09	15.04	1.86	0.92	0.94
IC 47454	7.23	20.18	12.95	2.92	7.25	4.33	1.82	0.78	1.04
IC 535195	17.71	24.43	6.72	9.57	5.70	-3.87	2.24	1.67	0.57
IC 535212	28.74	35.32	6.58	1.44	5.96	4.52	1.54	2.02	-0.48
IC 535274	33.13	40.16	7.03	1.70	15.70	14	1.08	1.43	-0.35
IC 35029	18.63	22.80	4.17	5.96	4.34	-1.62	1.62	1.44	0.18
IC 35032	28.42	31.08	2.66	3.96	5.05	1.09	1.78	1.14	0.64
IC 290061	22.65	26.78	4.13	3.57	6.34	2.77	1.61	1.36	0.25
IC 41590	27.68	30.28	2.6	2.80	12.28	9.48	1.25	1.21	0.04
IC 41589	12.26	20.87	8.61	2.41	7.38	4.97	1.81	2.54	-0.73
IC 535396	8.94	18.53	9.59	9.51	14.09	4.58	1.30	1.77	-0.47
IC 535414	42.87	49.96	7.09	11.25	10.02	-1.23	1.37	3.07	-1.7

Electronic Journal of Plant Breeding, 6(1): 341-349 (Mar 2015)
ISSN 0975-928X

Table 2b . Contd..

	Proline (μ mol)			MDA (μ mol)			H2O2 (μ mol)		
	Control	**Stress**	**Difference**	**Control**	**Stress**	**Difference**	**Control**	**Stress**	**Difference**
IC 535417	22.88	25.12	2.24	3.05	9.44	6.39	1.14	0.64	0.5
IC 535443	7.32	14.41	7.09	7.63	14.02	6.39	1.53	1.50	0.03
IC 535448	10.41	19.38	8.97	6.41	11.89	5.48	2.47	0.80	1.67
IC 535449	35.21	44.14	8.93	13.96	12.86	-1.1	2.55	1.86	0.69
IC 535462	8.45	19.32	10.87	1.38	10.54	9.16	1.78	0.98	0.8
IC 535474	29.97	41.12	11.15	12.99	5.96	-7.03	1.77	0.90	0.87
IC 535475	28.54	40.66	12.12	12.92	20.28	7.36	2.64	1.68	0.96
IC 535489	32.92	39.61	6.69	0.32	18.02	17.7	1.41	1.66	-0.25
IC535259	18.42	22.23	3.81	37.31	19.31	-18	2.82	1.40	1.42
IC 260946	28.44	31.47	3.03	17.25	15.63	-1.62	1.41	0.46	0.95
IC 279159	41.28	46.48	5.2	4.09	11.38	7.29	2.82	0.92	1.9
IC 279317	7.02	11.66	4.64	22.80	17.12	-5.68	2.23	2.32	-0.09
IC 279331	17.50	21.73	4.23	17.76	7.18	-10.58	1.86	2.31	-0.45
IC 279336	28.54	34.10	5.56	5.25	5.76	0.51	1.82	3.07	-1.25
IC 279480	18.47	22.97	4.5	17.12	11.57	-5.55	1.43	1.51	-0.08
IC 279866	23.92	27.66	3.74	4.22	17.83	13.61	1.67	2.21	-0.54
IC 279868	33.02	36.02	3	20.28	17.57	-2.71	1.73	1.73	0
IC 28086	24.66	29.06	4.4	8.15	18.22	10.07	1.27	1.57	-0.3
IC 281555	14.82	21.42	6.6	11.53	7.57	-3.96	1.73	2.57	-0.84
IC 281558	40.36	42.09	1.73	6.22	15.16	8.94	1.06	2.48	-1.42
IC 281565	29.32	27.15	-2.17	17.57	16.19	-1.38	1.41	2.47	-1.06
IC 28518	23.09	22.28	-0.81	4.37	17.29	12.92	1.98	1.21	0.77
IC 28547	32.18	30.29	-1.89	21.87	7.48	-14.39	2.64	1.01	1.63
IC 28552	18.10	17.29	-0.81	4.26	10.06	5.8	1.86	1.74	0.12
IC 28557	10.39	9.70	-0.69	13.51	19.87	6.36	1.82	1.73	0.09
IC 28564	20.87	19.95	-0.92	1.46	13.81	12.35	2.24	2.73	-0.49
IC 290030	31.91	35.83	3.92	38.41	17.68	-20.73	1.54	1.82	-0.28
IC 36882	36.29	42.09	5.8	9.20	7.74	-1.46	1.98	1.21	0.77
IC 290068	21.80	27.57	5.77	16.19	9.61	-6.58	2.64	1.01	1.63
IC 290095	36.29	43.68	7.39	3.42	28.71	25.29	1.44	1.74	-0.3
IC 290156	44.65	52.55	7.9	20.75	10.77	-9.98	1.75	2.48	-0.73
IC 290181	35.05	43.68	8.63	2.02	12.65	10.63	2.03	2.47	-0.44
IC 290185	28.03	31.23	3.2	16.04	17.29	1.25	1.86	3.23	-1.37
IC 290196	15.03	18.52	3.49	3.59	14.97	11.38	1.82	1.21	0.61
IC 290256	34.03	40.13	6.1	22.15	16.41	-5.74	2.24	1.01	1.23
IC 290296	23.00	31.03	8.03	3.91	10.47	6.56	1.54	1.74	-0.2
IC 310102	16.76	24.24	7.48	15.03	15.05	0.02	1.43	1.21	0.22
IC 310120	15.52	22.81	7.29	0.95	15.96	15.01	1.67	1.01	0.66
IC 310584	20.96	29.60	8.64	22.37	4.28	-18.09	2.23	1.74	0.49
IC 310586	30.06	40.68	10.62	0.56	11.38	10.82	1.54	1.51	0.03
IC 310588	14.55	24.98	10.43	7.87	14.22	6.35	1.72	1.05	0.67
IC 310590	25.58	36.06	10.48	1.51	16.67	15.16	1.36	0.85	0.51
IC 313156	15.52	26.27	10.75	38.58	11.76	-26.82	1.92	1.58	0.34
IC 31529	20.96	31.77	10.81	8.19	14.54	6.35	1.78	2.21	-0.43
IC 315921	30.06	40.91	10.85	9.81	17.96	8.15	1.38	1.73	-0.35
IC 316085	38.33	48.95	10.62	13.03	4.09	-8.94	1.72	1.57	0.15
IC 316087	28.72	39.39	10.67	7.25	11.51	4.26	1.36	2.57	-1.21
IC 316088	21.70	32.42	10.72	18.17	17.05	-1.12	1.92	2.32	-0.4
IC 316091	11.87	22.63	10.76	6.75	19.25	12.5	1.38	2.31	-0.93
IC 316092	18.47	26.44	7.97	16.65	18.56	1.91	1.65	3.10	-1.45
IC 316093	23.92	31.93	8.01	20.90	19.59	-1.31	0.98	1.08	-0.1
IC 316099	33.02	41.07	8.05	15.63	17.85	2.22	1.90	1.08	0.82
IC 316102	28.81	40.20	11.39	2.34	16.24	13.9	2.56	0.88	1.68

Table 2b . Contd..

	Proline (µ mol)			MDA (µ mol)			H2O2 (µ mol)		
	Control	**Stress**	**Difference**	**Control**	**Stress**	**Difference**	**Control**	**Stress**	**Difference**
IC 316104	7.02	18.91	11.89	38.47	16.56	-21.91	1.79	1.61	0.18
IC 31614	17.50	24.19	6.69	9.10	15.33	6.23	1.36	2.24	-0.88
IC 31615	18.47	22.28	3.81	14.95	14.56	-0.39	1.67	1.76	-0.09
IC 317422	18.47	21.73	3.26	37.70	10.04	-27.66	1.19	1.60	-0.41
IC 317529	23.92	29.11	5.19	1.59	17.33	15.74	1.65	0.95	0.7
IC 317610	33.02	37.66	4.64	2.69	10.43	7.74	1.53	1.08	0.45
IC 317632	31.68	36.13	4.45	9.51	17.72	8.21	1.54	0.88	0.66
IC 31774	33.57	39.37	5.8	15.48	9.46	-6.02	1.32	1.61	-0.29

Tot chl- total chlorophyll; MSI- Membrane Stability Index; SOD- Super Oxide Dismutase; MDA- Malondialdehyde; H_2O_2- Hydrogen peroxide

Table 3. Promising accessions for different characters

Character	Promising accessions
Total Chlorophyll	IC 281565, IC 290205, IC 290331, IC 281555, IC 290256, IC 290222, IC 535417, IC 310120
MSI	IC 290220, IC 310590, IC 290095, IC 31615, IC 258211, IC 316088, IC 290196
SOD	IC 290222, IC 279707, IC 290195, IC 279317, IC 535462, IC 279480, IC 310120, IC 316092, IC 290264, IC 317422, IC 535475
Proline	IC 47454, IC 535475, IC 316104, IC 316102, IC 535474, IC 535462, IC 315921, IC 31529, IC 316091, IC 313156, IC 316088, IC 316087, IC 316085, IC 310586, IC 310590, IC 310588
MDA	IC 310102, IC 279336, IC 290195, IC 279707, IC 35032, IC 290185, IC 274004
H_2O_2	IC 279868, IC 535443, IC 310586, IC 41590, IC 273946, IC 28557, IC 28552, IC 316085, IC 290331, IC 258212, IC 316104, IC 35029, IC 310102, IC 279642, IC 290061, IC 290230, IC 313156, IC 290197, IC 290337, IC 317610, IC 273954, IC 310584, IC 535417

MSI- Membrane Stability Index; SOD- Super Oxide Dismutase; MDA- Malondialdehyde; H_2O_2- Hydrogen peroxide