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BIOTECHNOLOGY

Response of Segregating and Non Segregating Generations of Four Tomato Crosses to Environmental Variations for Physiological Attributes Related to Heat Tolerance

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Abstract

Six generations obtained from each of the four tomato crosses viz. Cross-I (H7997 x CLN 1621 E), Cross-II (H7997 x BL 337), Cross - III (H7997 x Nagcarlan) and Cross- IV (H7997 x CLN 2366A) were evaluated over four environments. The analysis of variance exhibited significant differences among the genotypes for all the characters except for saturation deficit. The G x E interaction (linear) component was significant for all the characters except for relative stress injury. Pooled deviation was found to be significant for senescence index, membrane stability percentage, relative water content and saturation deficit. From stability analysis it was seen that hybrids H7997 x CLN 1621E, H7997 x Nagcarlan and H7997 x CLN2366A , F2 of cross H7997 x CLN 2366A together with B2 generation of cross H7997 x BL337 exhibited average stability for fruit yield per plant.

Highlights

- Six generations obtained from four crosses were evelauted for yield and physiological characters
- The generations were evaluated in four environments
- GxE interactions were found significant for most of the characters
- Among hybrids, H7997 x CLN 1621E, H7997 x Nagcarlan and H7997 x CLN2366A were found promising

Keywords: Tomato, environments, G x E interaction, stability analysis

Tomato is a very versatile vegetable for culinary and processed products. Tomato is an annual typical day neutral and self pollinated plant. Although tomato has a good potential to be cultivated in a spectrum of environments it confronts lots of abiotic stress and heat stress due to increased temperature is a matter of concern in many areas in the world. Transitory or constantly high temperatures cause an array of morpho-anatomical, physiological and biochemical changes in plants, which affect plant growth and development and may lead to a drastic reduction in economic yield (Wahid *et al*. 2007). Plants continuously struggle for survival under various environmental stress conditions including heat tolerance (Hasanuzzamam *et al*. 2013). At very high temperatures, severe cellular injury and even cell death may occur within minutes, which could be attributed to a catastrophic collapse of cellular organization (Schoffl *et al.* 1999). Direct injuries due to high temperatures include protein denaturation and aggregation, and increased fluidity of membrane lipids. Indirect or slower heat injuries include inactivation of enzymes in chloroplast and mitochondria, inhibition of protein synthesis, protein degradation and loss of membrane integrity (Howarth 2005). A thorough understanding of physiological responses of plants to high temperature, mechanisms of heat tolerance and possible strategies for improving crop thermotolerance is imperative (Wahid *et al.* 2007). Yield is a complex character and is influenced by many other characters so evaluation of morpho-physiological characteristics helps to describe the qualitative and quantitative attributes of the accessions of a given species to differentiate them; determine their usefulness and also to identify the genes that promote their use in crop production or improvement. Environment plays a crucial role in the expression of any character and genotype- environment interactions pose major problem in developing new cultivars and in choosing suitable cultivars to grow in specific region/location. Identification of yield-contributing traits and knowledge of GE interactions and yield stability are important for breeding new cultivars with improved adaptation to the environmental constraints prevailing in the target environments. When the backcross generations were combined it could be seen that out of the four crosses, average stability for fruit yield per plant was evident in B_2 generation of cross H7997 x BL337.

Materials and methods

The experiment was conducted at three different environments during offseason and one in *rabi* season, 2012-2013 at the Experimental Farm, Department of Horticulture, Assam Agricultural University, Jorhat, Assam, India. The weekly meteorological data obtained from the Department of Agricultural Meteorology, Assam Agricultural University, Jorhat, Assam, India on monthly mean maximum and minimum day temperatures during the period of investigation showed that mean maximum ranged from 21.90- 44.00^{o} Cand mean minimum temperature ranged from 9.50 – 30.00 ^{O}C. Four heat tolerant tomato genotypes *viz*., CLN 1621E, BL 337, Nagcarlan and CLN 2366A, and one heat sensitive genotype H 7997 were utilised to generate four crosses. *viz*. Cross-I (H7997 x CLN 1621 E), Cross- II (H7997 x BL 337), Cross - III (H7997 x Nagcarlan) and Cross- IV (H997 x CLN 2366A) by attempting crosses during *rabi*, 2012 and these along with the parental lines H7997, CLN 1621 E, BL 337, Nagcarlan and CLN 2366A comprised the entries for experiment on generation mean analysis. H7997 was used as a recurrent parent in backcross I (B_1) and the heat tolerant genotypes were used as recurrent parent in backcross II (B_2).Two rows of each parent, $F_1$and backcross generations and 8 rows of each F_2 were planted in randomized block design with two replications. Inter and intra row was kept as 50 cm and 30 cm respectively. Observations were recorded on five randomly sampled plants in each of P_1, P_2, 10 plants of $F_1$and 40 plants in F_2 and 20 plants that of $B_1$and B_2 in each of the replications on pollen viability percentage(Norton,1966), chlorophyll stability index (Chetty *et al*. 2002) relative water content(Wetherly and Barrs, 1962), saturation deficit, membrane stability percentage (Premachandra *et al.* 1990), relative stress injury (Goyal *et al*. 2002), senescence index and lipid peroxidation (Heath and Packer, 1968). In addition data were recorded on sampled plants on each entry for fruit yield per plant (kg/plant). Six generations of each of the three crosses were screened in four planting dates *viz.*, 5th March (E_1), 10th April (E_2), 5th June (E_3) and 15th October (E_4). In E_3, the experiment was conducted inside polyhouse. The collected data were subjected to statistical analyses using Microsoft Excel 2007. The mean data of each environment was subjected to pooled analysis of variance over environments to study genotype - environment (GE) interaction and phenotypic stability by using the model given by Eberhart and Russell (1966). The three stability parameters were calculated to compare the genotypes: Mean (m) = The ideal genotype should have high mean over environments , regression coefficient (b_i) = The ideal genotype should have regression coefficient equal to 1, deviation mean square (S^2_{di}) = The ideal genotype should have deviation mean square from linear regression equal to zero ($S^2_{di} = 0$). To test the significance of difference of 'b_i' value from unity the procedure given by Gomez (1968).

Results and discussion

Gene expression is an interplay of genotype and environment and this genotype -environment interaction poses major problem in the development of stable varieties. Breeding of cultivars with desired characteristics such as high economic yield, tolerance or resistance to biotic and abiotic stresses, traits that add value to the product, and the stability of these traits in target environments. Inconsistent genotypic responses to environmental factors such as temperature, soil moisture, soil type

Table 1: Pooled analysis of variance of generations of tomato crosses over environments for physiological characters

Source of variation	D.F	Mean sum of squares								
		SI	MS(%)	LP	RWC	SD	RSI	CSI	PV(%)	FY/P
Genotypes	20	11.24**	34.35**	7.60**	33.46**	33.46	44.01**	49.42**	22.19**	29956.61**
Genotype + (Genotype x Envt.)	63	18.01	77.09**	15.48**	99.67**	99.67**	10.03	63.76**	287.60**	182965.2
Environment (linear)	1	857.31**	3795.15**	565.45**	5055.14**	5055.14**	327.58**	2923.53**	16261.00**	10267435**
Genotype x environment (linear)	20	49.28**	205.85**	10.56**	269.60**	269.60**	19.03	165.93**	844.22**	524884.70**
Pooled deviation	48	2.24*	12.08*	3.66	13.98**	13.98**	5.24	12.04	8.82	11433.74**
Pooled error	84	6.12	7.38	2.66	7.32	3.48	7.44	11.08	7.70	8661.44

**Significant at 1 % level of probability, * Significant at 5 % level of probability ; SI-Senescence index, MS%- Membrane stability percentage, LP- Lipid peroxidation, RWC-Relative water content, SD- Saturation deficit, RSI- Relative stress injury, PV%- Pollen viability percentage,FY/P- fruit yield per plant

Table 2: Estimates of stability parameters of generations of tomato crosses for physiological characters

Genotypes	SI				MS%				LP				RWC%			
	m	b_i	S^2_{di}	Stability	m	b_i	S^2_{di}	Stability	m	b_i	S^2_{di}	Stability	m	b_i	S^2_{di}	Stability
P_1 (H7997)	19.63	2.28**	2.14	-	64.08	0.81	-0.21	-	23.19	2.72**	-1.39	-	59.90	1.77	4.14	-
P_2 (CLN1621E)	13.75	0.32**	0.10	-	73.62	1.71**	2.33	-	17.31	1.26	1.18	AVS	69.25	0.83	7.28	-
P_3(BL337)	13.00	1.11	0.11	AVS	73.37	0.84	41.17**	-	19.46	1.28	0.21	-	67.00	1.15	34.31**	-
P_4(Nagcarlan)	14.50	0.68	6.90	AVS	73.25	1.00	10.78**	-	19.99	1.60	-1.93	-	69.50	1.49	41.33**	-
P_5 (CLN2366A)	16.00	0.79	-0.77	-	75.00	1.25	0.68	-	20.73	2.00**	6.50	-	69.37	1.12	13.02	-
P_1XP_2(Cross I)	17.63	1.33	0.14	-	78.50	1.04	10.34**	-	16.98	1.54	4.01	AVS	71.63	0.91	18.52**	-
P_1XP_3(Cross II)	15.13	1.54**	0.40	BAVS	77.62	1.41	1.90	AVS	17.90	0.68	4.08	AVS	69.75	1.10	-0.78	AVS
P_1XP_4(Cross III)	15.13	0.85	-0.62	AVS	80.75	1.02	-0.01	AVS	17.47	0.77	0.55	AVS	70.50	1.10	9.97	AVS
P_1XP_5(Cross IV)	15.50	1.05	-0.09	-	79.12	0.93	29.87**	-	19.00	0.78	-1.06	-	65.50	0.97	45.56**	-
F_2 (Cross I)	15.13	0.95	-0.89	AVS	77.62	0.67	53.20**	-	18.88	0.45	-2.48	AVS	69.25	1.12	0.67	-
F_2 (Cross II)	14.25	0.90	-1.23	AVS	77.12	1.18	0.74	AVS	19.01	-0.08	-2.91	-	69.88	1.00	5.30	AVS
F_2 (Cross III)	17.38	0.74	7.75	-	80.62	1.00	2.97	AVS	18.62	0.55	-3.32	AVS	70.88	0.81	-1.77	AVS
F_2 (Cross IV)	16.38	0.96	2.22	-	75.75	1.04	3.20	-	18.94	0.96	-0.96	-	69.25	0.77	-0.30	-
B_1 Cross I	14.00	1.20	-0.84	AVS	76.37	0.78	12.79*	-	20.34	0.96	2.38	-	70.63	0.82	1.38	AVS
B_2 CrossI	16.63	0.95	3.45	-	78.00	1.13	4.22	AVS	19.87	1.32	-1.35	-	67.63	0.95	42.92**	-
B_1Cross11	15.88	0.77	-1.52	-	79.75	0.82	0.36	-	19.24	0.84	4.89	-	70.75	0.81	1.83	AVS
B_2Cross II	14.63	1.11	0.11	AVS	79.25	1.27	-0.50	AVS	18.35	0.55	-2.54	AVS	73.50	1.04	-3.36	AVS
B_1 Cross III	17.38	0.72	-0.91	-	78.50	0.78	6.95	AVS	19.22	1.16	-2.95	-	71.88	0.73	-1.09	AVS
B_2 Cross III	14.38	0.97	-1.27	AVS	78.75	0.40*	6.54	-	18.08	0.70	-0.42	AVS	73.63	0.53	0.43	AVS
B_1 Cross IV	12.63	0.59	-1.15	AVS	73.87	1.32	50.74*	-	18.55	0.77	2.45	AVS	70.75	0.99	2.26	AVS
B_2 Cross IV	14.75	1.18	0.33	AVS	78.25	0.60	2.58	AVS	17.49	0.19**	-1.79	AAVS	68.63	0.98	-1.24	-

**Significant at 1 % level of probability, * Significant at 5 % level of probability; AV- average stable, BAVS- below average stable, AAVS- above average stable

Genotypes	SD%				RSI%				CSI				PV%				FY/P			
	m	b_i	S^2_{di}	Stability	m	b_i	S^2_{di}	Stability	m	b_i	S^2_{di}	Stability	m	b_i	S^2_{di}	Stability	m	b_i	S^2_{di}	Stability
P_1 (H7997)	40.10	0.83	7.22	-	25.13	1.33	10.00	-	61.87	1.75*	-0.53	-	67.62	1.08	3.70	-	0.78	1.20	-0.18	-
P_2 (CLN1621E)	32.25	1.77	4.08	-	17.00	2.07	31.72**	-	67.25	1.47	78.19**	-	76.00	1.27*	0.11	-	1.07	1.24	-0.16	-
P_3(BL337)	31.50	1.15	34.25**	-	17.50	0.20	-1.28	-	68.09	1.09	19.58	-	78.50	1.27	-1.48	-	0.85	0.47*	-0.17	-
P_4(Nagcarlan)	30.38	1.49	41.27**	-	17.63	1.07	-1.41	-	67.42	1.56	4.68	-	73.38	1.29**	-2.60	-	1.07	0.76	0.18	
P_5 (CLN2366A)	30.38	1.12	12.96	AVS	17.38	0.88	26.00**	-	69.62	0.50	1.89	AVS	79.25	1.51**	26.87	BAVS	1.09	0.93	-.0.18	AVS
P_1XP_2(Cross I)	28.38	0.91	18.46**	-	14.88	0.62	-2.92	AVS	73.00	0.90	3.78**	-	82.88	0.96	34.26**	-	1.32	0.93	0.18	AVS
P_1XP_3(Cross II)	30.25	1.10	-0.84	AVS	15.75	0.88	-2.87	AVS	76.13	0.79	6.49	AVS	81.88	0.97	5.98	AVS	0.96	0.62**	-0.17	-
P_1XP_4(Cross III)	29.50	1.10	9.91	AVS	15.75	0.67	0.85	AVS	72.38	1.08	1.86	AVS	80.38	0.92	1.99	AVS	1.38	0.92	-0.37	AVS
P_1XP_5(Cross IV)	34.50	0.97	45.50**	-	14.75	0.76	-1.26	AVS	71.25	1.41	4.44	AVS	81.88	0.70**	-2.31	AAVS	1.45	0.98	-0.48	AVS
F_2 (Cross I)	30.75	1.12	0.61	-	15.50	0.92	-0.52	AVS	70.75	0.66	-2.84	AVS	79.63	1.06	-2.25	AVS	1.01	0.70*	0.17	-
F_2 (Cross II)	30.13	1.00	5.24	AVS	15.73	1.64	-2.59	AVS	66.75	0.67	-5.21	-	78.50	0.78	-2.30	-	1.01	0.98	0.17	-
F_2 (Cross III)	29.13	0.81	-1.83	AVS	15.61	1.21	-3.29	AVS	67.38	0.71**	-2.14	-	81.00	0.80	-1.66	AVS	1.04	1.13	0.16	-
F_2 (Cross IV)	30.75	0.77	-0.36	-	15.75	0.92	-3.00	AVS	68.00	0.82	-4.70	-	80.25	0.89	7.19	AVS	1.10	1.17	-0.18	AVS
B_1 CrossI	29.38	0.82	1.32	AVS	15.63	1.09	-1.24	AVS	68.00	0.76	6.33	-	78.25	0.89	-1.18	-	1.02	1.15	-0.18	-
B_2 CrossI	32.38	0.95	42.86**	-	14.69	1.42	-2.63	AVS	67.63	0.89	8.18	-	76.25	1.02	-2.35	-	1.20	1.27	0.17	AVS
B_1Cross11	29.25	0.81	1.77	AVS	15.88	0.87	-1.71	AVS	68.63	0.54	-0.75	AVS	80.38	0.89	3.90	AVS	1.00	0.97	-0.17	-
B_2 Cross II	26.50	1.04	-3.42	AVS	15.25	1.19	-0.21	AVS	75.00	0.80	2.25	AVS	80.25	0.89	0.45	AVS	1.11	1.04	0.18	AVS
B_1 Cross III	28.13	0.73	-1.15	AVS	15.88	1.18	-3.15	AVS	66.25	1.15	-4.09	-	79.63	0.80	31.86**	-	1.04	1.10	0.15	-
B_2 Cross III	26.38	0.53	0.37	AVS	13.75	0.59	-0.21	AVS	68.25	1.33	9.61	-	76.00	0.93	11.68	-	0.98	1.17	0.17	-
B_1 Cross IV	29.25	0.99	2.20	AVS	15.54	1.08	-1.54	AVS	66.13	1.55	-1.54	-	76.75	1.11	-3.02	-	1.02	1.34*	0.17	-
B_2 Cross IV	31.38	0.98	-1.30	-	15.38	0.41	-3.04	AVS	67.75	0.59	16.70	-	79.75	0.96	-0.59	AVS	1.04	0.94	0.16	-

**Significant at 1 % level of probability, * Significant at 5 % level of probability; AV- average stable, BAVS- below average stable, AAVS- above average stable or fertility level from location to location and year to year are a function of genotype - environment (GE) interactions. The present investigation with six generations obtained from each of the four crosses was undertaken to study the GE interaction with the objective of obtaining suitable varieties which could perform well over a spectrum of environment and also to identify suitable types suited to particular environment.

The pooled analysis of variance for stability revealed that all the generations differed significantly for almost all the characters except for saturation deficit. The highly significant environmental variance for almost all the characters suggested considerable difference among environments and their predominant effect on characters.

The linear genotype x environment interactions were significant for all the characters except for relative stress injury. Non significance of GE linear components for these characters signifies that they do not show genetic difference for their regression on environmental index revealing the absence of divergent genetic response to the linear effect of the environment. It was further observed that the characters differed in respect to the contribution of linear components towards GE variance. In this investigation, for most of the characters the GE interaction is due to the linear and non linear components. However, linearity was more pronounced for most of the characters. This indicated that variation among the genotypes can be largely explained by differences in regression slopes for these characters. Thus response of the genotypes to the changing environments can be portrayed as orderly and predictable with respect to these characters. This obviously indicated that the accurate prediction of the phenotypic performances of the genotypes could be deduced for these characters. The importance of linear and non linear components of GE interaction in tomato were also reported by Ortiz and Lzquido (1994) and Kallo *et al*. (1998). Among the off seasons, E_1, E_2 and E_3, E_1 (mean average day temperature 30.86°C) was found to cause decisive improvement in most of the characters including yield.

regression slope as a measure of stability was considered by Finlay and Wilkinson (1963). They suggested that a genotype was maximum stable, when its mean performance was high and regression of its performance over the environmental mean approached zero. Eberhart and Russell (1966) observed that the deviation from regression, which is the non linear parameter, should also be taken into consideration along with the linear parameter, i.e. the regression coefficient while examining a variety for phenotypic stability. They observed that an ideal variety should possess regression coefficient equal to unity (b_i=1). This variety would have average response to the changes in environments. Regression value larger than unity indicates the sensitivity of the variety to the changes in environmental condition. Such a variety is termed as 'below average stable' and performs much better than its inherent potentially in high yielding environmental conditions, but the performance is poor in stress condition. Regression values less than unity signifies the insensitivity of the variety to changes in the environment and such an 'above average stable' variety is suitable specifically for stress environments. They further suggested that, a genotype should exhibit the least deviation from regression (S^2_{di}), to be stable one. The variance due to deviation from regression coefficient is primarily due to the uncontrollable causes and depends on the environment (Bains and Gupta, 1972). In most of the studies on regression analysis of genotype x environment interaction, a linear relationship between genotype-environment interaction and environmental index has been reported (Freeman, 1973).

In the present investigation, the hybrids H7997 x CLN 1621E, H7997 x Nagcarlan and H7997 x CLN2366A exhibited average stability for fruit yield per plant. Further, hybrid, H7997 x CLN 1621E exhibited average stability for relative stress injury. H7997 x Nagcarlan exhibited average stability for senescence index, membrane stability percentage, lipid peroxidation, relative water content, saturation deficit, relative stress injury and pollen viability percentage while hybrid H7997 x CLN 2366A also exhibited average stability for relative stress injury and chlorophyll stability index. It was evident that the parents of the hybrids also exhibited average stability for few or more characters. Parent H7997 exhibited average stability for days to fruit maturity. Parent CLN 1621E exhibited average stability for number of primary branches per plant and number of fruits per plant. Nagcarlan exhibited average stability for senescence index; CLN2366A exhibited average stability for senescence index, saturation deficit and chlorophyll stability index. Thus it can be seen that the average stability exhibited by the hybrids H7997 x CLN 1621E, H7997 x Nagcarlan and H7997 x CLN2366A for various characters including fruit yield per plant could be due to transmission of linear and non linear stability from their respective parents. With respect to F_2 generations, the crosses except H7997 x CLN 2366A did not show average stability for fruit yield per plant. It also exhibited average stability for relative stress injury and pollen viability percentage. When the backcross generations were combined it could be seen that out of the four crosses, average stability for fruit yield per plant was evident in B_2 generation of cross H7997 x BL337 .This cross further exhibited average stability for few or more characters like senescence index, relative water content, saturation deficit, relative stress injury, chlorophyll stability index and pollen viability percentage. The recovery of the genotype of the recurrent parent's average stability for these characters might have induced average stability of the respective backcross. The evidence of stability in segregating generations like F_2 and backcross generations suggest transmission of stability and scope of subsequent selection facilitating development of phenotypically stable genotype.

Conclusion

The present investigation has provided some useful information regarding the performance of the six generations *viz*. P_1, P_2, F_1, F_2, B_1 and B_2 of four tomato crosses involving normal and heat tolerant parents and the information generated shapes a framework for the development of heat tolerant tomato genotypes that could be successfully grown during the off season.

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