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Identification of heat tolerant barley (*Hordeum vulgare* L.) genotypes based on heat susceptibility index

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Abstract
The current research was planned to understand the effect of high temperature on grain yield and its contributing characters in order to select heat tolerant genotypes for future breeding programmes. Thirty six (eight parents and twenty-eight F_1's) barley genotypes were evaluated under two environments created by different dates of sowing *i.e*. timely sown and late sown, during *rabi* 2022-23. In the present study, the heat susceptibility index value of parents and crosses for yield and its contributing characters were calculated and genotypes were classified into four different categories *i.e.* highly heat tolerant, heat tolerant, moderately heat tolerant and heat susceptible. From the result of heat susceptibility index (HSI), it could be visualized that the parents RD-2660, RD-2052, RD-2715 and RD-2794 were found to be desirable for grain yield and its contributing characters under heat stress environment. Among the crosses namely; RD-2035 x RD-2660, RD-2715 x RD-2899, RD-2035 x RD-2624, RD-2035 x RD-2794, RD-2624 x RD-2907 and RD-2660 x RD-2907 showed the superiority under heat stress environment for more than two characters. These parents and crosses could be utilized as a promising breeding material for the development of new set of heat stress tolerant barley varieties.

Keywords: Barley, heat susceptibility index (HSI), heat stress, heat tolerant

Introduction
Barley (*Hordeum vulgare* L.) is a self-pollinated, annual, winter season diploid valuable cereal crop having chromosome number 2n=2x=14 and a member of grass family Poaceae. It is originated from the Near East and Ethiopian High lands. Barley grain can be utilized as animal feed (60%), for malt production (30%), seed production (7%) and for human food (3%) [Baik and Ullrich, 2008] [3]. The major components of barley grains are carbohydrate (78%), protein (10%), fat (1%), β-glucan (3.9%), crude fiber (5%) and 2% of calcium (Shaveta *et al*., 2019) [12]. In India, it is mainly grown in Rajasthan, Uttar Pradesh, Bihar, Haryana, Punjab, Madhya Pradesh, Himachal Pradesh and Uttarakhand states and covering a total area of about 6.09 lakh hectares and total production of 18.18 lakh tonnes with an average grain productivity of 2988 kg per hectare (Anonymous 2020-21) [2]. In light of climate change and global warming, which indicate a slow rise in the atmospheric temperature as well as an increase in the frequency and duration of heat stress incidents in the near future, high temperature is considered as a vital stress factor with a significant potential influence on crop productivity. Furthermore, prolonged periods of drought are frequently followed by high temperatures, and the co-occurrence of these two abiotic stresses in the field might have a detrimental effect on crop production globally and have severe consequences on food security. Heat stress is an important abiotic stress that globally causes significant crop losses. Heat tolerance is a complex polygenic character involving epistatic interactions among loci and powerful genotype × environment interactions (Abou-Elwafa and Amein, 2016) [1]. The different growth and developmental stages of the plants differ in their sensitivity to heat stress. For example, in barley, the period of spike initiation to flowering is very sensitive to high temperature stress and acceleration of this phase seems to be the main cause of yield reduction under hot condition. Heat stress during post anthesis or grain filling stage affects availability and translocation of photosynthates to the developing kernels and starch synthesis and deposition within the kernel, consequently resulting in lower grain weight and altered grain quality (Mohammadi *et al.*, 2004) [10].

Materials and Methods

The experimental material consisted of 36 diverse genotypes of barley including eight parents and twenty-eight F_1's. The material was grown under two different dates of sowing *i.e.* timely sown (14th November) and late sown (14th December) at Instructional Farm, Sri Karan Narendra College of Agriculture, Jobner (Rajasthan) during *rabi* 2022-23. The experimental location is situated at 435 meters above mean sea level on latitude 26º5' N and 75º20' E. The experimental materials were evaluated in randomized block design with three replications under both the environments. Rows were planted in 3 m length spaced at 30 cm with 10 cm inter plant distance under both the environments and the recommended cultural practices were adopted to raise the crop. Observations were recorded on 15 quantitative characters *viz*., days to anthesis, days to maturity, duration from anthesis to maturity, flag leaf area, spike length, grains per spike, 1000-grain weight, biological yield per plant, grain yield per plant, harvest index, proline content, total soluble sugars, relative water content, membrane stability index and chlorophyll content. Ten randomly selected competitive plants in each replication were recorded for all the traits under study except days to anthesis, days to maturity and 1000-grain weight which were recorded on plot basis. Heat susceptibility index (HSI) was calculated for grain yield and other attributes by using the formula as suggested by Fischer and Maurer (1978) [6].

$$HSI = \left[1 - \left(\frac{Y_D}{Y_P}\right)\right] / D$$

Where,

Y_D = Mean value of the genotype in stress environment (E_2)

Y_P = Mean value of the genotype under non-stress environment (E_1)

D = Heat stress intensity

D = 1- [mean value of all genotypes in stress environment (E_2) / mean value of all genotypes under non-stress environment (E_1)]

The heat susceptibility index values were utilized to characterize the relative tolerance of genotypes based on minimization of yield losses compared to normal environmental conditions.

Results and Discussion

Heat stress is a common abiotic factor responsible for reducing yield and there is a necessity to identify heat stress tolerant genotypes. Modhej *et al*. (2005) [9] displayed that the higher heat stress tolerance of barley after anthesis was due to its shorter crop growth duration compared to wheat. Heat stress is a key task to barley productivity in India. The effect of climate change is clearly evident from recent vagaries across areas in India. Therefore, breeding aimed at selecting genotypes with high temperature stress tolerance is one of the most vital objectives of the barley breeder. Keeping the above facts in view, the current study was conducted to magnify the yield level of barley in high temperature areas by selecting stress tolerant parents and cross combinations for future breeding programme. The results of current study confirmed that in comparison to normal sown condition (E_1), mean performance of parents and F_1's declined under late sown or heat stress condition (E_2) for all the character except proline content and total soluble sugars. Similar findings were also reported by Sultan *et al*. (2016) [14], Parashar (2019b) [11], Thakur *et al.* (2020) [15] and Hebbache *et al.* (2021) [7]. It is fairly accepted that yield is a complex character and a final product of the action and interaction of a number of component characters. Thus, selection based on yield *per se* will not be much effective. Therefore, in order to determine the tolerance of different parents and crosses for heat stress, the heat susceptibility index was estimated based upon the values and direction of desirability of different characters used in the study. The genotypes with high positive HSI values are susceptible to higher temperature and *vice versa* (Devi *et al*. (2021) [5].

On the basis of HSI, the parents and F_1's were classified into four different categories *i.e.* highly heat tolerant (HSI < 0.50), heat tolerant (HSI: 0.51-0.75), moderately heat tolerant (HSI: 0.76 – 1.00) and heat susceptible (HSI > 1.00).

On the basis of HSI, it appeared that the parents RD-2052 and RD-2624 for days to anthesis; RD-2715, RD-2052 and RD-2907 for days to maturity; RD-2715, RD-2052 and RD-2907 for duration from anthesis to maturity; RD-2052 for flag leaf area; RD-2715, RD-2660, RD-2794 and RD-2907 for spike length; RD-2899 for grains per spike; RD-2907, RD-2052 and RD-2660 for 1000-grain weight; RD-2052, RD-2794 and RD-2035 for biological yield per plant; RD-2794 and RD-2660 for grain yield per plant; RD-2660 and RD-2794 for harvest index; RD-2035, RD-2715 and RD-2907 for proline content; RD-2660, RD-2035 and RD-2794 for total soluble sugars; RD-2715, RD-2660 and RD-2624 for relative water content; RD-2715, RD-2660 and RD-2794 for membrane stability index; RD-2035 for chlorophyll content were least affected under heat stress. An overall assessment of parents indicated that RD-2660, RD-2052, RD-2715 and RD-2794 were found to be desirable for grain yield and most of its attributes on the basis of heat susceptibility index (Table 1-2).

Perusal of Table 1 and 2 revealed that the crosses RD-2715 x RD-2899, RD-2052 x RD-2899 and RD-2794 x RD-2907 for days to anthesis; RD-2715 x RD-2899, RD-2660 x RD-2907 and RD-2624 x RD-2907 for days to maturity; RD-2624 x RD-2794, RD-2624 x RD-2899 and RD-2660 x RD-2794 for duration from anthesis to maturity; RD-2794 x RD-2899, RD-2052 x RD-2794 and RD-2052 x RD-2660 for flag leaf area; RD-2035 x RD-2907, RD-2624 x RD-2899 and RD-2624 x RD-2715 for spike length; RD-2035 x RD-2660, RD-2660 x RD-2907 and RD-2624 x RD-2794 for grains per spike; RD-2052 x RD-2624, RD-2035 x RD-2660 and RD-2052 x RD-2660 for 1000-grain weight; RD-2035 x RD-2794, RD-2052 x RD-2715 and RD-2035 x RD-2624 for biological yield per plant; RD-2035 x RD-2624, RD-2035 x RD-2660 and RD-2035 x RD-2794 for grain yield per plant; RD-2035 x RD-2660, RD-2715 x RD-2899 and RD-2899 x RD-2907 for harvest index; RD-2660 x RD-2907, RD-2624 x RD-2899 and RD-2624 x RD-2907 for proline content; RD-2052 x RD-2624, RD-2035 x RD-2907 and RD-2035 x RD-2660 for total soluble sugars; RD-2715 x RD-2794, RD-2035 x RD-2715 and RD-2624 x RD-2907 for relative water content; RD-2052 x RD-2907, RD-2624 x RD-2715 and RD-2035 x RD-2794 for membrane stability index; RD-2715 x RD-2794, RD-2715 x RD-2899 and RD-2035 x RD-2624 for chlorophyll content exhibited comparatively more tolerance among 28 F_1's crosses under heat stress environment.

Perusal of Table 1 and 2 revealed that among the parents in general, RD-2794 and RD-2660 were least affected under heat stress. Crosses RD-2035 x RD-2624, RD-2035 x RD-2660, RD-2035 x RD-2794, RD-2052 x RD-2794 and RD-

2715 x RD-2794 showed HSI value less than 0.50 and were least affected under heat stress environment (E_2) for grain yield per plant while among the crosses, RD-2035 x RD-2715 and RD-2052 x RD-2715 revealed HSI value 0.50-0.75, therefore, these were considered as heat tolerant. An overall assessment of parents indicated that RD-2660, RD-2052, RD-2715 and RD-2794 were found to be desirable for most of the grain yield and its attributes on the basis of heat susceptibility index. The crosses namely; RD-2035 x RD-2660, RD-2715 x RD-2899, RD-2035 x RD-2624, RD-2035 x RD-2794, RD-2624 x RD-2907 and RD-2660 x RD-2907 showed the superiority under heat stress environment for more than two characters.

The magnitudes of heat stress intensity (D-value) are presented in Table 3. Low value of heat stress intensity (D-value) *i.e.* less than 0.20, indicated that the characters *viz.*, days to anthesis (0.10), days to maturity (0.15), spike length (0.14), 1000-grain weight (0.13), biological yield per plant (0.11), harvest index (0.15), proline content (-0.16), total soluble sugars (-0.13), relative water content (0.14), membrane stability index (0.10) and chlorophyll content (0.08) showed more tolerance, whereas, duration from anthesis to maturity (0.24), flag leaf area (0.35), grains per spike (0.29) and grain yield per plant (0.25) with high heat stress intensity (D-value) *i.e.* 0.21 to 0.35, suffered more under heat stress environment (E_2). Similar results were also observed by Singh *et al.* (2011) [13], Bhardwaj *et al.* (2017) [4], Kumar *et al.* (2018) [8] and Devi *et al.* (2021) [5].

Table 1: Heat susceptibility index (HSI) for days to anthesis, days to maturity, duration from anthesis to maturity, flag leaf area, spike length, grains per spike, 1000-grain weight and biological yield per plant

Parents	Days to anthesis	Days to maturity	Duration from anthesis to maturity (days)	Flag leaf area (cm^2)	Spike length (cm)	Grains per spike	1000-grain weight (g)	Biological yield per plant (g)
RD-2035	1.26	1.04	0.93	1.09	0.75	1.14	1.33	0.49
RD-2052	0.96	0.69	0.49	0.99	0.94	1.21	0.30	0.17
RD-2624	0.99	1.06	1.14	1.28	0.75	1.37	0.91	0.72
RD-2660	1.36	1.28	1.26	1.20	0.64	1.22	0.67	1.25
RD-2715	1.12	0.61	0.20	1.19	0.61	1.23	1.63	1.04
RD-2794	1.37	0.92	0.58	1.14	0.69	1.13	1.65	0.23
RD-2899	1.21	0.99	0.83	1.13	0.94	0.77	0.87	1.25
RD-2907	1.08	0.78	0.57	1.23	0.69	1.09	0.28	1.15
F_1 crosses								
RD-2035 × RD-2052	1.40	1.36	1.33	1.16	0.95	1.13	2.08	1.08
RD-2035 × RD-2624	0.72	0.96	1.19	1.09	1.17	0.83	0.95	0.39
RD-2035 × RD-2660	0.79	1.35	1.91	1.11	0.90	0.84	-0.37	1.79
RD-2035 × RD-2715	0.81	1.25	1.61	1.17	0.94	1.26	0.93	1.11
RD-2035 × RD-2794	0.83	1.38	1.93	0.82	0.91	1.16	0.77	-0.76
RD-2035 × RD-2899	1.41	1.30	1.23	0.56	1.49	0.83	0.90	0.90
RD-2035 × RD-2907	0.80	0.84	0.93	0.88	0.16	0.96	0.26	1.77
RD-2052 × RD-2624	0.96	1.38	1.60	1.10	0.90	1.07	-0.60	1.45
RD-2052 × RD-2660	1.26	1.09	0.96	0.53	0.93	0.27	0.13	0.41
RD-2052 × RD-2715	0.64	1.31	1.84	0.82	1.22	0.69	0.63	-0.43
RD-2052 × RD-2794	0.99	0.81	0.67	0.41	1.86	0.89	1.39	1.10
RD-2052 × RD-2899	0.37	1.02	1.49	1.30	1.18	1.16	0.87	1.21
RD-2052× RD-2907	1.40	0.94	0.58	1.17	1.03	0.92	0.75	0.63
RD-2624 × RD-2660	1.05	0.87	0.78	1.33	0.91	0.88	2.11	0.90
RD-2624 × RD-2715	0.88	1.31	1.70	1.13	0.60	1.07	0.46	0.79
RD-2624 × RD-2794	1.72	0.88	0.19	0.68	0.86	0.65	0.97	1.39
RD-2624 × RD-2899	1.42	0.87	0.44	0.98	0.44	1.02	1.12	1.56
RD-2624 × RD-2907	0.70	0.70	0.78	1.04	0.71	1.17	1.66	1.11
RD-2660× RD-2715	0.55	1.06	1.43	1.13	0.85	1.28	1.13	0.97
RD-2660 × RD-2794	1.03	0.73	0.53	1.17	0.87	0.98	0.80	0.81
RD-2660 × RD-2899	0.87	0.77	0.74	1.05	0.87	0.95	0.90	0.89
RD-2660 × RD-2907	0.87	0.70	0.61	1.08	0.83	0.64	0.39	1.04
RD-2715 × RD-2794	1.23	0.87	0.62	0.58	0.95	0.93	1.69	1.00
RD-2715 × RD-2899	0.14	0.42	0.67	0.97	1.05	0.98	0.98	3.14
RD-2715 × RD-2907	0.67	1.22	1.69	1.09	2.34	1.04	0.17	1.13
RD-2794 × RD-2899	1.61	1.10	0.73	0.30	2.55	0.98	2.28	1.17
RD-2794 × RD-2907	0.48	0.71	0.93	1.03	0.96	0.88	2.04	0.74
RD-2899 × RD-2907	0.94	0.86	0.86	0.83	2.66	1.26	0.91	3.44

Table 2: Heat susceptibility index (HSI) for grain yield per plant, harvest index, proline content, total soluble sugars, relative water content, membrane stability index and chlorophyll content

Parents	Grain yield per plant (g)	Harvest index (%)	Proline content (µg/100 mg fresh weight)	Total soluble sugars (mg/gm fresh weight)	Relative water content (%)	Membrane stability index (%)	Chlorophyll content (SPAD)
RD-2035	1.03	1.47	0.51	0.51	1.15	1.31	0.74
RD-2052	1.02	1.59	1.16	1.11	1.15	1.31	1.23
RD-2624	1.41	1.99	0.86	0.83	0.79	0.96	1.62
RD-2660	0.88	0.62	1.18	0.43	0.62	0.74	1.44
RD-2715	1.26	1.53	0.71	1.08	0.62	0.69	1.52
RD-2794	0.54	0.72	0.86	0.73	1.01	0.86	1.34
RD-2899	1.24	1.37	2.30	1.90	1.22	1.49	1.73
RD-2907	1.45	1.78	0.84	1.71	1.19	1.01	1.51
F_1 crosses							
RD-2035 × RD-2052	1.35	1.63	1.15	1.16	1.08	0.79	1.40
RD-2035 × RD-2624	-0.01	-0.28	1.22	0.24	0.81	1.14	0.44
RD-2035 × RD-2660	0.02	-1.60	1.13	0.19	1.14	1.37	0.88
RD-2035 × RD-2715	0.58	0.14	0.58	0.65	0.65	0.87	0.88
RD-2035 × RD-2794	0.18	0.79	0.62	1.28	1.12	0.47	1.21
RD-2035 × RD-2899	1.36	1.78	1.33	1.24	1.37	1.64	1.15
RD-2035 × RD-2907	1.35	1.19	0.61	0.12	0.98	1.69	0.69
RD-2052 × RD-2624	1.21	1.12	0.89	-0.14	0.93	0.71	0.58
RD-2052 × RD-2660	1.11	1.63	1.16	0.23	1.10	0.58	0.69
RD-2052 × RD-2715	0.58	1.27	1.15	0.89	1.11	0.76	0.50
RD-2052 × RD-2794	0.45	-0.09	0.76	0.87	1.34	1.13	0.58
RD-2052 × RD-2899	0.99	0.85	1.40	1.10	0.98	1.01	1.25
RD-2052 × RD-2907	1.25	1.78	0.75	1.23	1.40	0.32	1.44
RD-2624 × RD-2660	1.69	1.54	1.49	2.05	0.97	0.88	0.86
RD-2624 × RD-2715	1.50	2.16	1.31	2.39	0.98	0.37	0.94
RD-2624 × RD-2794	1.00	0.75	0.75	1.15	0.76	1.15	1.00
RD-2624 × RD-2899	1.20	1.06	0.29	1.39	0.77	1.75	1.02
RD-2624 × RD-2907	0.96	0.87	0.41	0.83	0.68	1.00	1.85
RD-2660 × RD-2715	1.13	1.29	0.84	0.50	0.90	0.98	0.95
RD-2660 × RD-2794	1.41	1.92	2.34	0.27	0.98	0.84	0.95
RD-2660 × RD-2899	0.86	0.77	3.34	0.83	0.79	0.71	0.72
RD-2660 × RD-2907	0.87	0.70	0.26	0.29	0.87	1.32	0.52
RD-2715 × RD-2794	0.38	-0.10	0.79	1.09	0.55	0.93	0.37
RD-2715 × RD-2899	0.93	-1.33	0.87	1.00	0.97	0.62	0.37
RD-2715 × RD-2907	0.91	0.74	2.06	1.66	1.09	2.11	0.56
RD-2794 × RD-2899	0.99	0.83	1.00	1.25	0.83	1.04	1.17
RD-2794 × RD-2907	1.28	1.67	0.80	1.01	1.26	0.73	1.21
RD-2899 × RD-2907	1.28	-0.75	0.95	1.70	1.21	0.98	1.11

Table 3: Mean of all genotypes under normal (E_1) and heat stress (E_2) environments and heat stress intensity (D-value) for different characters

Characters	Mean of all genotypes in		D-value
	E_1	**E_2**	
Days to anthesis	75.73	68.14	0.10
Days to maturity	113.31	96.54	0.15
Duration from anthesis to maturity (days)	37.58	28.40	0.24
Flag leaf area(cm^2)	20.59	13.31	0.35
Spike length (cm)	8.01	6.85	0.14
Grains per spike	42.49	30.04	0.29
1000-grain weight (g)	36.98	32.27	0.13
Biological yield per plant (g)	29.99	26.56	0.11
Grain yield per plant (g)	12.20	9.14	0.25
Harvest index (%)	40.44	34.45	0.15
Proline content (µg/100mg fresh weight)	65.03	75.49	-0.16
Total soluble sugars (mg/gm fresh weight)	4.77	5.37	-0.13
Relative water content (%)	80.58	69.45	0.14
Membrane stability index (%)	73.70	66.26	0.10
Chlorophyll content (SPAD)	50.35	46.31	0.08

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