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RESEARCH ARTICLE



Study on heat stress indices and their correlation with yield in Indian mustard genotypes under diverse conditions

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Abstract

Increasing temperatures are becoming a major challenge for agro-systems. Late mustard planting often results in overall growth reduction, poor metabolism, less productivity and yield due to heat stress at the terminal stage. A set of 45 genotypes comprising introgression lines, landraces, wild species, induced mutants, advance breeding lines and the released varieties/cultivars were evaluated under normal (Yp) and stressed environment (Ys) for yield stability during 2016-2018. The higher value of heat tolerance index, geometric mean productivity, yield stability index, relative stability index and yield production score index with lower heat susceptibility index indicated tolerance to high temperature in some genotypes. Based on the yield performance and derived traits, six genotypes namely, HLM-41-13-2 and ELM-38 with zero erucic acid quality, MCN-05-8 an induced mutant, germplasm lines PCR-3 and CSR-1163, while MCN-08-2 is released variety RB50 were rated as promising for heat stress tolerance at both the locations. Biplot analysis indicated that genotypes under group I and IV are tolerant of high yield potential and stability in a stressed environment irrespective of location.

Keywords: Biplot, correlation, heat tolerance, indian mustard, stress indices, yield potential

Introduction

Brassica being major oilseed crop in India, occupy third position after groundnut and soybean with an annual average yield of about 29 million tons. Rapeseed and mustard contribute about 28.6% of total oilseeds production in India (https://economictimes.indiatimes.com/) standing as the fourth leading oilseeds producing country, next to the USA, China, and Brazil. India's mustard output is set to hit a record 10 million tonnes this year (https://www. financialexpress.com) from about 8.5 million hectares of area. Although the country has made a significant paradigm in the total oilseeds production from 5.26 million metric tons in 1949-50 to 36.10 million metric tons in 2020-2021(https:// www.statista.com) but the country still is not able to fulfill the demand of the growing population and the gap between demand-supply and therefore, has necessitated huge imports of edible oil. About 56.02% of edible oil was imported from other countries (Anonyms 2021). The area planted and area harvested for crops differ, which accounted for crop loss due to several environmental stresses. Besides biotic and abiotic stresses, cultivation under rainfed or with conserved/limited irrigation in marginal areas causes differences in the overall productivity of mustard growing regions of India. Further, due to intensive cropping patterns and late harvest of cotton and rice, mustard planting is often delayed, resulting in the heat stress at the terminal stage, causing yield losses. Heat stress affects the overall growth, metabolism and productivity of crops worldwide. Tolerance of plant toheat stress is always a thrust area for breeders. The yield potential is higher under a favorable environment,

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but gaining genetic increment under stress conditions is always challenging (Snowdon et al. 2021). Stress indices have been widely used in different crops for screening tolerant genotypes based on yield reduction under stress conditions (Banerjee et al. 2020). Several evidences reported response of stress indices based on yield under drought stress (Meena et al. 2013; Nowosad et al. 2017; Sandhu et al. 2019). However, limited information is available for these responses in Indian mustard under heat stress. The mathematical relationship between stress and optimum conditions based on seed yield was anticipated by many scientists (Pandey et al. 2015; Kumar et al. 2020; Bahrami et al. 2021; Abou-Elwafa and Shehzad 2021). Therefore, this experiment aimed to determine promising cultivars based on heat stress indices prevailing at two locations (Ludhiana and Abohar in Punjab state), as the region has high potential for expanding rapeseed cultivation suitable for crop diversification, economical use of land and resources.

Materials and methods

The field experiments were laid in winter (rabi) season 2016-17 and 2017-18 at two locations, Ludhiana (Punjab Agricultural University), and Abohar (Dr. J.C. Bakhshi Regional Research Station, Abohar, Punjab). The meteorological observations (T_{max} and T_{min}) were recorded at both the locations (Figs. 1a and 1b). A set of 45 genotypes comprising introgression lines, landraces, wild species, induced mutants and breeding lines (Table 1) were sown under timely (last week of October to first week of November) and late planting (last week of November) conditions along with three checks, PBR- 375, JD-6, DRMRIJ-31(Giriraj) and a natural registered germplasm line, BPR-541-4 for terminal heat stress. The crop was sown in 5 x 0.3m² plot size in Randomized Block Design with three replications. The stress indices, namely, heat susceptibility index (HSI), relative stress index (RSI) (Fisher and Maurer 1978), heat tolerance index (HTI) (Fernandez 1992; Schneider et al. 1997), yield index (YI) (Gavuzzi et al. 1997), yield stability index (YSI) (Bouslama and Schapaugh et al. 1984) and tolerance (TOL) were calculated using yield of stressed and non-stressed genotypes. The geometric mean

productivity (GMP), mean productivity (MP), harmonic mean (HM) (<u>Rosielle</u> and Hamblin 1981), yield susceptibility score index (YSSI) and yield production score index (YPSI) (<u>Thiry</u> et al. 2016) were derived from the above-mentioned indices.

Statistical analysis

Yield data recorded in the two crop seasons were statistically analyzed using CPCSI software developed by the Department of Statistics, Punjab Agricultural University, Ludhiana (Cheema and Singh 1990). Means for treatment effects were separated based on the critical difference (CD) using Analysis of variance (ANOVA) technique for seed yield. The CD values were tested at 5% (p \leq 0.05) significance level. Pooled data over the years was used to compute correlation coefficient by OPSTAT software (Sheoran et al. 1998), principle component analysis (PCA) and dendrogram clustering using MINITAB 16 (version 16.2.1).

Results and discussion

Significant variability existed between the genotypes when planted at two dates and two locations. The range of yield reduction was maximum at PAU, Ludhiana. Maximum yield was observed at Abohar under both the sowing conditions (Table 2). The results are in agreement with Sharma et al. (2022) who recorded an overall 33.92% reduction in seed yield plant⁻¹ of Brassica juncea under late-planted conditions. The results also agree with the wheat cultivars where lower grain number was due to lesser reproductive spikes and shorter grain-filling duration, leading to decreased grain weight (Impa et al. 2019). Heat stress at seedling and at flowering stages tends to enhance vegetative growth and flower abortion in Indian mustard causing a decline in seed yield (Sandhu et al. 2020). However, the seed yield showed significant positive association with all the stress indices except, HSI, which had negative correlation with Ys at PAU, Ludhiana (r=-0.350*) and at RRS, Abohar (r= -0.075). YSI and RSI had negative correlation with Yp at both locations (Table 3). Rad and Abbasian (2011) had studied drought tolerance in winter rapeseed genotypes and found that stress susceptibility index (SSI) had a positive correlation with



Fig. 1. Minimum and maximum air temperature recorded during two-crop seasons (a) PAU, Ludhiana and (b) RRS, Abohar

SI.No.	Genotype	Origin/Source	S.no	Genotype	Origin/source
1	ELM-85	Zero erucic acid line of <i>B. juncea</i>	26	MSC-3	B. juncea introgression line
2	CM-9-2	<i>B. juncea</i> germplasm line, landrace	27	MCN-05-16	<i>B. juncea</i> germplasm line, landrace
3	MCN-09-18	B. juncea germplasm line, landrace	28	MCN-05-8	Induced mutant
4	CRL-1359-13-6-2	Zero erucic acid line of <i>B. juncea</i>	29	HLM-40-33-11	Zero erucic acid <i>B. juncea</i> line
5	HLM-9906	Zero erucic acid line of <i>B. juncea</i>	30	Bio-QM-1	Zero erucic acid <i>B. juncea</i> line
6	MCN-08-14	B. juncea germplasm line, landrace	31	MCN-13-23	B. juncea introgression line
7	NR-4	Exotic material	32	JMG-244	<i>B. juncea</i> germplasm line, landrace
8	MCN-09-40	Natural germplasm line released as variety	P 33	GMCN-167	B. juncea germplasm line, landrace
		BR-357 under timely sown irrigated condit	ions		
9	MCN-06-08	B. juncea germplasm line, landrace	34	CSR-1037	<i>B. juncea</i> germplasm line, landrace
10	CM-11-7	Zero erucic acid line of <i>B. juncea</i>	35	DNA-(wf) 8-10	Induced mutant
11	DAR-3	B. juncea introgression line	36	MCN-09-36	Variety released as RH-761 for timely sown rainfed condition
12	MCN 13-19	<i>B. juncea</i> germplasm line, landrace	37	CM-16	Zero erucic acid line of <i>B. juncea</i>
13	CM-21-11	Zero erucic acid line of <i>B. juncea</i>	38	CRL-1359-175- 13-99	Zero erucic acid line of <i>B. juncea</i>
14	MCN-09-38	B. juncea germplasm line, landrace	39	CM-21-1	Zero erucic acid line of <i>B. juncea</i>
15	HLM-41-13-2	Zero erucic acid line of <i>B. juncea</i>	40	CM-21-16	Zero erucic acid line of <i>B. juncea</i>
16	ELM-38	Zero erucic acid line of <i>B. juncea</i>	41	MCN-09-29	B. juncea germplasm line, landrace
17	B-326	Natural B. juncea germplasm line	42	JMG-02-01	B. juncea germplasm line, landrace
18	NPJ-2-4	B.juncea germplasm line, landrace	43	CSR-158	B. juncea germplasm line, landrace
19	MCN-08-2	Variety released as RB-50 for rain fed condition under AICRP	44	B-272	<i>B. juncea</i> germplasm line, landrace
20	MCN-09-39	Variety RL-1359 for irrigated condition	45	CRL-1359-19	Zero erucic acid line of <i>B. juncea</i>
21	DAR-8	B.juncea introgression line	46	BPR-541-4	<i>B. juncea</i> germplasm line for heat tolerance, landrace
22	CSR-45	<i>B. juncea</i> germplasm line, landrace	47	DRMRIJ 31	Variety released as Giriraj
23	PTJ-3-79	B. juncea introgression line	48	JD-6	Varietyfor early and late sowing under irrigated conditions
24	PCR-3	B. juncea germplasm line, landrace	49	PBR-378	Variety released for rain fed condition under AICRP
25	CSR-1163	<i>B. juncea</i> germplasm line, landrace			

Table 1. A list of genotypes studied and their source/origin

Table 2. Range and mean of seed yield and stress indices in *B. juncea* genotypes at two locations (Pooled mean)

Traits		PAU, Ludhiana		RRS, Abohar		
		Range	Mean	Range	Mean	
Yield under nor	n stress (Yp)	675.9-2102.6	1134.17	1510.9-4275.0	2194.7	
Yield under stre	ess (Ys)	390.8-1161.3	669.76	949.7-2385.6	1489.8	
Heat susceptibi	ility index (HSI)	index (HSI) 0.20-1.56 0.96 0.35		0.35-1.80	0.95	
Geometric mea	an productivity (GMP)	513.9-1444.0	867.09	1255.1-2952.1	1801.4	
Mean Productiv	vity (MP)	533.4-1478.6	902.0	1304-3041.4	1842.2	
Harmonic mear	n (HM)	495.2-1410.4	834.2	1207.9-2886	1762.2	
Tolerance (TOL))	93.9-1339.6	464.4	181.2-2467.7	704.9	
Heat tolerance	index (HTI)	0.21-1.62	0.61	0.33-1.81	0.70	
Yield index (YI)		0.58-1.73	0.58-1.73 1.00		1.00	
Yield stability ir	ndex (YSI)	0.36-0.92	0.61	0.42-0.89	0.70	
Relative stabilit	y index (RSI)	0.62-1.55	1.03	0.62-1.31	1.03	
Yield susceptib	ility score index (YSSI)	0.10-2.21	0.38	0.12-1.15	0.40	
Yield productio	on score index (YPSI)	46.6-491.8	218.78	218.78 90.8-1234-7		
CD ≤0. 05	Y=32.9, E=32.9, G=163.3, Y> Y×E×G=326.6	KE= NS, Y×G=230.9, E×G=230.9,	Y=59.6, E=59.6, G=295.1, Y×E= NS, Y×G=417.3, E×G=417.3, Y×E×G=590.2			

Table 3. Correlation of seed yield with stress indices at PAU, Ludhiana (below diagonal) and RRS, Abohar (above diagonal)

	Yp	Ys	HSI	GMP	MP	HM	TOL	HTI	ΥI	YSI	RSI	YSSI	YSPI
Yp	1	0.709**	0.622**	0.942**	0.969**	0.904**	0.888**	0.943**	0.708**	-0.620**	-0.622**	0.284*	0.887**
Ys	0.637**	1	-0.075	0.904**	0.861**	0.940**	0.304*	0.888**	1.000**	0.077	0.075	0.791**	0.304*
HSI	0.471**	-0.350*	1	0.346*	0.422**	0.263	0.889**	0.355*	-0.076	-1.000**	-1.000**	-0.494**	0.889**
GMP	0.911**	0.897**	0.088	1	0.995**	0.995**	0.682**	0.993**	0.903**	-0.345*	-0.346*	0.538**	0.682**
MP	0.955**	0.838**	0.198	0.992**	1	0.981**	0.747**	0.990**	0.860**	-0.421**	-0.422**	0.481**	0.747**
НМ	0.852**	0.944**	-0.027	0.992**	0.968**	1	0.607**	0.985**	0.940**	-0.261	-0.263	0.593**	0.607**
TOL	0.841**	0.118	0.852**	0.543**	0.641**	0.434**	1	0.693**	0.303*	-0.888**	-0.889**	-0.134	1.000**
HTI	0.899**	0.892**	0.077	0.991**	0.982**	0.984**	0.531**	1	0.887**	-0.354*	-0.355*	0.526**	0.693**
YI	0.636**	1.000**	-0.352*	0.897**	0.837**	0.944**	0.117	0.891**	1	0.078 ^{NS}	0.076	0.791**	0.303*
YSI	-0.468**	0.353*	-1.000**	-0.085	-0.195	0.030	-0.851**	-0.074	0.354*	1	1.000**	0.491**	-0.889**
RSI	-0.466**	0.355*	-1.000**	-0.082	-0.193	0.032	-0.849**	-0.072	0.356*	1.000**	1	0.491**	-0.889**
YSSI	0.256	0.778**	-0.585**	0.548**	0.482**	0.609**	-0.217	0.544**	0.778**	0.588**	0.587**	1	-0.134
YSPI	0.033	0.792**	-0.827**	0.441**	0.329*	0.549**	-0.513**	0.444**	0.792**	0.829**	0.829**	0.805**	1

** and * indicates significant at 1% and 5% significance level,

Yp= Yield under non stressed condition; Ys=Yield under stress conditions; HIS= Heat susceptibility index; GMP= Geometric mean productivity; MP= Mean productivity; HM- Harmonic mean; TOL= Tolerance; HTI= Heat tolerance index; YI= Yield index; YSI= Yield stability index; RSI= Relative stability index; YSSI= Yield susceptibility score index; YPSI- Yield productivity score index

irrigated yield/non-stress (Yp) and negative correlation with yield under stress (Ys) based on sensitivity to drought stress.

Tolerance is attributed to lower values of HSI and TOL. Genotypes with least reduction in yield had lower value of above indices. During the two crop seasons, ELM-38 having zero erucic acid along with natural germplasm lines, PCR-3, CSR-1163, CM-21-16, JMG-02-01 and CSR-158 showed tolerance at PAU Ludhiana, while an introgression line DAR-3, zero erucic acid quality line ELM-38, natural germplasm lines, MCN-13-19 and CSR-1163 were identified as tolerant at RRS, Abohar. ELM-38 and CSR-1163 were considered as elite genotypes since they exhibited drought tolerance at both locations. The negative correlation of HSI with Ys, YI, YSI, RSI, YSSI and YSPI and positive correlation with TOL, GMP and MP was recorded at both the locations. The earlier research conducted in wheat found that that MP, GMP and Stress tolerance index (STI) are the most suitable stress indices in wheat (Poudel et al. 2021; Devi et al. 2021). A significant correlation of yield under stress with YSI, YI, MP, STI and GMP was reported by Sharma et al. (2022), who suggested using these indices to select high yielding and heat tolerant lines in Brassica juncea. The results support the findings and advocate that these indices could be used to select high yielding genotypes under both the conditions.

HSI represents the genotypic yield potential under heat stress as reported by <u>Koscielny</u> et al. (2018) but later it was concluded that HSI does not account for differences in yield potential among genotypes. Previously the negative correlation of HSI with Ys, YI, YSI, RSI, YSSI and YSPI and positive with TOL, GMP and MP exists at both locations as reported by Singh and Choudhary (2003) in *B. juncea*. It provides a measure of yield stability based on the minimization of yield loss under stressed conditions (Bahrami et al. 2021). Therefore, a stress tolerant genotype as identified by HSI need not necessarily have high yield potential, but should have minimum yield reduction. <u>Ajay</u> et al. (2021) proposed that the ideal genotype should be high yielding under any environmental condition, but the genetic effects are independent of environmental effects, most genotypes do not perform satisfactorily in all environments.

Higher value of GMP, MP and HM indicated yield potential of genotypes under stress and non-stress environment irrespective of yield reduction. During the two crop seasons at both the location two genotypes were promising, the induced mutant MCN-05-8 and MCN-08-2, has been released as RB50 variety under moisture stress (rainfed) conditions under All India Co-ordinated Research Program (AICRP).GMP and MP had significant and positive correlation with all stress indices while non-significant and negative with YSI and RSI at the two studied locations. Several researchers have advocated that a suitable selection index must significantly relate to seed yield in stressed and non-stressed conditions (Anshori et al. 2021; Farshadfar and Sutka 2002; Darvishzadeh et al. 2010). In the present investigation the GMP, MP and HM were highly correlated with each other as well as with Ys and Yp.

Heat tolerance index (HTI) was recorded maximum at Abohar (Table 2) and least at PAU Ludhiana. Higher HTI during the two planting times and locations rated MCN-08-2, a released variety, RB50 and PCR-3 a natural germplasm line tolerant to heat stress. The non-significant correlation of HTI existed for HSI, YSI and RSI but highly significant with YSSI and YSPI at both the locations. Based on the higher value of YI, YSI and RSI promising genotypes at two locations were ELM-38 along with two natural germplasm lines PCR-3 and CSR-1163. YI was significantly but negatively correlated with HSI and non-significant with TOL at PAU. Non-significant association of YI existed with HSI at Abohar. Farshadfar and Sukta (2002) and Nayyeripas (2019) proposed the most appropriate index for selecting stress-tolerant cultivars which had partly or high association with seed yield under stress and non-stress conditions and later these results were supported by Poudel et al. (2021). However, in the present study, cultivars with the highest YSI exhibited yield reduction ≤30.0% in the late planting over optimum sowing. Correlation coefficients and principal component analysis performed by Devi et al. (2021) and Poudel et al. (2021) in wheat revealed the importance of mean productivity, geometric mean productivity, stress tolerance index and stress susceptibility index in selecting heat tolerant genotypes.

Higher value for both YSSI and YPSI were recorded in

two natural germplasm lines PCR-3 and CSR-1163 at both the locations. Thiry et al. (2016) proposed two indices i.e. YSSI and YPSI for better understanding of genotypic behaviour under stress, indicating if a high yield under stress is due to tolerance (resilience) or due to a high genotypic production capacity(mean yield performance) or both. This can be achieved by analyzing the components of YSSI and YPSI, where high resilient/tolerant and high productive genotypes should have a high value of both the predicted indices

Multivariate analysis

The first dimension/component represents high yielding potential and the second indicates tolerant dimensions. Genotypes possessed high values of PC1 could be high yielding under stressed and non-stressed environments. The second component can be named as a stress-tolerant dimension and it separates the stress-tolerant genotypes from non-stress tolerant ones. At PAU, PC1 contributes 67.1% variation and PC2 contribute 32.1% variation. However, the respective contributions were 72.9 % and 26.3% at RRS,



Fig. 2. Scoring plot of genotypes (a) and loading of stress indices (b) at PAU, Ludhiana



Fig. 3. Scoring plot of genotypes (a) and loading of stress indices (b) at RRS, Abohar

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Abohar (Figs. 2 and 3). The studied genotypes were divided into four groups based on the component analysis. Group I (High PC1 and PC2) consists of genotypes with high yield potential and tolerance under both environments. Group II (Low PC1 and High PC2) comprises low yielding genotypes under stress conditions. Group III (Low PC1 and Low PC2) contain genotypes with low yield under both environment and Group IV (High PC1 and Low PC2) had high yielding genotypes under stress condition. Genotypes in group I and IV are suitable for both environments according to Thiry et al. (2016) classification. Cluster analysis based on heat stress indices, grouped genotypes into four separate clusters. At PAU cluster I involved forty five genotypes, cluster II had two genotypes and cluster III and IV had one genotype each. Genotypic variability could be exploited in crossing program. Kaya et al. (2002) stated that high PCA1 and low PCA2 genotypes were high yielding and stable, whereas genotypes with low PCA1 and high PCA2 were low yielding and unstable. Cultivars with PCA1 scores nearly zero are stable for all the environments with high grain yield across environments. The results of biplot agree with the analysis of the screening indices, which supported the accuracy and efficiency of the assessment based on the tolerance and susceptible indices. Biplot analysis in Indian mustard (Singh and Bhajan 2016; Saroj et al. (2021)), in sunflower hybrid (<u>Tyagi</u> and Dhillon 2019), in wheat by Poudel et al. (2021) and safflower (Bahrami et al. 2014) revealed the similar trends. Multivariate analysis in B. napus indicated presence of adequate genetic variation for useful selections to identify stable genotypes and environmental interaction that has more influence on the overall yield performance of the genotypes. Thus, the identified genotypes can cultivate in heat prone areas and can also be used as genetic resources in crop improvement programs.

Overall, the outcome of the present investigation was to facilitate the identification of heat-tolerant genotypes viz., HLM-41-13-2 and ELM-38 are zero erucic acid lines, MCN-08-2 is released variety RB50 for cultivation under moisture stress/rainfed conditions, MCN-05-8 is the induced mutant and two natural germplasm lines PCR-3 and CSR-1163based on two years evaluation at two locations. These selected genotypes can be used as donors in the breeding programs to develop heat tolerant mustard varieties to sustain production in changing climatic conditions. In terms of resilience and production, score indices quickly revealed and characterised the best or the worst genotypes within a population.

Author's contribution

Conceptualization of research (PS) Designing of the experiments (PS, MS); Contribution of experimental materials (PS); Execution of field/lab experiments and data collection (PC); Analysis of data and interpretation (PC,PS); Preparation of manuscript (PC, RS).

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