Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

J P P Journal of Pharmacognosy and Phytochemistry

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2017; 6(5): 2775-2777
Received: 07-07-2017
Accepted: 08-08-2017

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Genetic divergence for thermotolerance based on physiological parameters during germination in Indian mustard (*Brassica juncea* (L.) Czern & Coss.)

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Abstract
The present study was undertaken to analyse the genetic divergence of 35 genotypes of Indian mustard based on physiological parameters during germination for thermotolerance. Mahalanobis D^2 -statistic helped in grouping 35 genotypes of Indian mustard into six clusters. Cluster I comprising of 16 genotypes. Whereas clusters III, IV and V were comprised of one genotypes in each only. On the basis inter-cluster distances, contribution towards total divergence, inter-cluster distance and cluster mean performance, clusters IV; V; VI; III and II genotypes viz. Pusa Mahak, RAURD-78, NRC-DR-2, Varuna, RH-30, PM-25, TPM-1 and NDRE-7 were most promising divergent genotypes of Indian mustard and could be effectively utilized in hybridization –selection breeding programme to develop thermotolerant Indian mustard. The use of diverse genotypes included in these clusters were suggested to give better heterosis and higher variability in the segregating generations.

Keywords: Genetic divergence, Indian mustard, High temperature, Physiological traits

1. Introduction
High temperature is one of the important factor which affects growth, development of plants and also reduces the crop production. Due to current trend in global climate change the seed yield will likely lead to further losses. Breeding for heat tolerance is a major objective in arid and semi-arid regions of the world due to high temperature at seedling stage as well as at its terminal stage. Many physiological traits have been suggested of heat tolerance in other crops, but there has been limited work on genetic divergence of mustard for physiological characteristics because it is a complex phenomenon. Genetic divergence has been measured successfully by many researchers, following Mahalanobis (1936) D^2 -Analysis [5]. It is essential technique to select the parents for future breeding program. If the parent is more diverse, the chances of obtaining high heterotic F1 and broad-spectrum variability in segregating generations are greater (Arunachalam, 1981) [1]
Indian mustard is an important oilseed crops and winter season crop, growing well in diverse agro-ecological condition and accounts for about 90% area under rapeseed- mustard crops. Predominantly, it is used as edible oil and rich source of vitamins, minerals and contains many medicinal properties. It is also used in pharmaceuticals, industries, biodiesel, pet foods and component of many other products. The crops are very sensitive to climatic variables and hence climate change could have significant effect on its production.
Therefore, the present study was undertaken to identify divergent parents for hybridization program for thermotolerance based on physiological parameters during germination under laboratory condition.

2. Materials and Methods
The experiments were conducted in the Plant Breeding and Genetics laboratory, Department of Plant breeding and Genetics, Dr. Rajendra Prasad Central Agricultural University, Pusa (Bihar) during *Rabi*, 2014-15. The material for the present investigation comprised of 35 Indian mustard genotypes from different AICRP-RM research centres were raised in a Randomized Block Design with three replications.

Seed treatment: Seeds were surface sterilized with 0.1 per cent $HgC1_2$ solution for two minutes and then thoroughly washed with distilled water. Healthy seeds were taken for experimental purpose. Mustard seeds (*Brassica juncea* L.) were germinated in 18 cm Petri dishes on a filter paper moistened with distilled water and kept in seed germinator at 35^0C in sterilized petri under controlled conditions (70% relative humidity).

Each petri dish contained twenty-five equidistantly placed seeds. Germination counts were recorded after twenty-four hours.

Observations: The following parameters were recorded after the termination of germination on 8th day viz., Germination per cent (G%), Root length (cm) (RL), Seedling length (SL)(cm), Root: shoot ratio (R/S ratio), Vigour Index (VI), Seedling dry weight (SDW) and Electrolyte leakage (EL). Multivariate analysis of genetic divergence was carried out by using Mahalanobis D^2 -statistic, described by Rao (1952) [7]

3. Results and Discussion

In the present study, an experiment was conducted to study genetic diversity for selecting the thermotolerant diverse parents for hybridization programme to produce desirable segregants for heat stress and other important characters in Indian mustard. 35 genotypes of Indian mustard were grouped into six clusters (Table 1). Cluster I included maximum number of genotypes (16) followed by cluster II (9) while cluster VI accommodated seven genotypes respectively. Cluster III, IV and V were mono-genotypic indicating distinctness from other genotypes. It was also observed that genotypes from the same location were distributed in separate clusters indicating wide genetic diversity among them even though originating from the same geographical region. Thus, the clustering pattern lacked strict correspondence between genetic divergence and geographical distribution. These are in agreement with those obtained by Malik *et al.*, (2006) [6], Chaudhary and Joshi (2001) [3], and Goyal *et al.*, (2012) [2] in Indian mustard.

The values of intra-cluster distance ranged from 0 (cluster III, IV and V) to 26.30 (cluster VI) (Table 2). Cluster VI showed highest intra-cluster distance (26.30) followed by cluster II (19.24). The average inter-cluster distance varied from 4.20 (cluster III and IV) to 56.02 (cluster II and VI). Cluster II and VI showed maximum inter-cluster distance (56.02) followed by cluster IV and V (55.04). Similarly, cluster V and VI exhibited moderate cluster distance (51.95). Therefore, genotypes of Cluster IV, V and VI were more divergent and could be utilized for successful breeding programme. Crosses involving parents belonging to most divergent clusters would be expected to manifest maximum heterosis and wide variability of genetic architecture. Goyal *et al.*, (2012) [2] and Gangapur *et al.*, (2010) [4]

Cluster V reflected high cluster mean for germination percentage, root shoot ratio, and electrolyte leakage in desirable direction. Cluster IV had highest mean for root length, SDW and VI at 35 ^{0}C. Highest seedling length found in cluster III (Table 3).

SDW (33.28%); exhibited maximum contribution towards divergence followed by EL (21.51%) and root shoot ratio (19.50%). Clusters II and VI followed by V and VI were strikingly divergent based on highest inter-cluster distance, indicating greater diversity, subsequently be utilized in intermating for generation of wider variability and utilization in transgressive breeding for development of early –sown mustard under elevated temperature.

Conclusively, based on contribution towards total divergence, inter-cluster distance and cluster mean performance, clusters IV; V; VI; III and II genotypes viz. Pusa Mahak, RAURD-78, NRC-DR-2, Varuna, RH-30, PM-25, TPM-1 and NDRE-7 were most promising divergent genotypes of Indian mustard and could be effectively utilized in hybridization –selection breeding programme to develop thermotolerant Indian mustard under high temperature which would provide superior segregates in Indian mustard genotypes.

Table 1: Grouping of 35 genotypes of Indian mustard in various clusters on the basis of D^2 statistics

Cluster	No. of genotypes	Name of genotypes
I	16	RH-0819, R. Suflam, NDRE-4, Kanti, Pant Rai-19, RH-8814, KMR-10-2, Pusa Agrani, PM-27, DRMR-EJ-902, RH-0116, RAURDE-1001, RAURDE-1002, DRMR-150-35, KMR-10-1, RH-0406
II	9	PKRS-28, RGN-48, RGN-13, TM-2, TPM-1, TM-4, RH-0701, NDRE-7, RAURD-212
III	1	NRC-DR-2
IV	1	RAURD-78
V	1	Pusa Mahak
VI	7	Pusa Bold, Varuna, Pusa Tarak, PM-28, RH-30, Pusa Bahar, Pusa Mustard-25

Table 2: Average intra and inter cluster D^2 values of mustard genotypes for physiological parameters during germination**.**

	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V	Cluster VI
Cluster I	8.39	21.97	16.47	27.98	35.80	35.53
Cluster II		19.24	33.66	49.42	30.47	56.02
Cluster III			0.00	4.20	44.32	27.74
Cluster IV				0.00	55.04	30.81
Cluster V					0.00	51.95
Cluster VI						26.30

Table 3: Per cent contribution of characters towards divergence in 35 mustard genotypes for physiological parameters during germination.

	Source	Contribution%
1	G% 35°C	9.92
2	RL 35°C	5.38
3	R: S Ratio 35°C	19.50
4	SL 35°C	5.21
5	SDW 35°C	33.28
6	VI 35°C	5.21
7	EL 35°C	21.51

Table 4: Mean values of 7 characters in 6 clusters in Indian mustard genotypes

	G% 35 °C	RL 35 °C	R: S Ratio 35 °C	SL 35 °C	SDW 35 °C	VI 35 °C	EL 35 °C
Cluster I	90.33	6.46	1.33	13.08	0.04	1079.39	71.75
Cluster II	88.89	7.17	2.19	12.29	0.03	968.97	51.46
Cluster III	90.67	9.05	1.89	15.39	0.05	1257.93	46.16
Cluster IV	90.67	10.30	2.37	15.33	0.06	1327.40	51.85
Cluster V	92.00	8.72	4.15	14.43	0.03	998.03	80.25
Cluster VI	83.24	7.98	2.05	14.30	0.06	1058.77	67.75

Table 5: Diverse parents in different clusters for physiological parameters during germination

Characters	Clusters	Suitable Parents suggested	Mean *Per se* performance
G% 35°C	V	Pusa Mahak	92.00
RL 35°C	IV	RAURD-78	10.30
R: S Ratio 35°C	V	Pusa Mahak	4.15
SL 35°C	III	NRC-DR-2	13.89
SDW 35°C	IV and VI	RAURD-78, Pusa Bold, Varuna, Pusa Tarak, PM-28, RH-30, Pusa Bahar, Pusa Musrtard-25	0.006
VI 35°C	IV	RAURD-78	1327.40
EL 35°C	III and II	PKRS-28, RGN-48, RGN-13, TM-2, TPM-1, TM-4, RH-0701, NDRE-7, RAURD-212, NRC-DR-2	29.89 and 34.89

4. Acknowledgement

We express our sincere thanks to IARI, New Delhi, DRMR, Bharatpur, Rajasthan, CCSHAU, Hisar, GBPUAT, Pantnagar, BARC Trombay, CSAUAT, Kanpur, NDUAT, Faizabad, SKRAU**,** ARS, Sriganganagar**,** Rajastahan for providing seed materials for the research programme.

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