

Assessment of Genetic Divergence for Exploitation of Dual Season Blackgram [*Vigna Mungo* (L.) Hepper]

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Abstract

Thirty-seven diverse genotypes of *Vigna mungo* (L.) Hepper were studied for 13 quantitative traits. Determinate Maturity Scores were calculated as days taken to maturity from first pod formation to identify determinate genotypes with higher yield potential. Cluster analysis assigned genotypes into groups on the basis of agronomic performance rather than their origin. Grouping of all advanced breeding lines into one cluster indicates that only a portion of available genetic diversity has been exploited for blackgram improvement. Scattered diagram indicates that sixteen genotypes were determinate in nature and took less than 10 days to mature after first pod formation during two consecutive years. Eleven genotypes grouped in upper right box of the scattered diagram showed desirable determinate maturity and were least affected by the environmental fluctuations and, among these, two (Pk-45855 and Pk-45703) were selected with combined effects of yield potential and determinate maturity. The genotypes studied were pure-lines and selected ones could be tested under a wider range of environmental conditions to evaluate for their adaptability and yield potential with determine maturity trait. They could also be incorporated in breeding programmes to develop superior cultivars with a determinate and stable growth potential.

Keywords: Cluster analysis, Mash, Principal Component Analysis, Rice-based farming system.

Introduction

Blackgram or mash [*Vigna mungo* (L.) Hepper] is an important pulse crop of the tropic and sub-tropic areas and has been identified as a potential crop in many countries (Smartt, 1990; Girish et al., 2012). The economic product of blackgram is seed grain, which provides significant dietary protein. The yield of this crop is very low because of the non-availability of high yielding and stable cultivars and the best cultivars of one year may not necessarily repeat the performance in the following years (Anonymous, 2011). The lack of stable and high yielding cultivars is one of the major constraints governing its expansion. Evaluation and exploitation of germplasm for various yield related characters is necessary for the development of improved varieties. High yielding genotypes selected from germplasm prove their superiority in advance testing under various agro-ecological conditions (Ghafoor & Arshad, 2011).

Blackgram has potential to be cultivated as a Spring crop, but one of the major problems is that

available cultivars are mostly in-determinate types and unstable when planted over various environments and locations, especially during spring season. It is also cultivated during the Summer season (July-October) in most of the South Asian countries. If determinate types of blackgram are developed, it could be planted successfully during the Spring season (April to mid July) by incorporating in the rice-wheat systems of the Indo-Gangetic Plains (IGP). The IGP offers 13.5 m ha which is 20 % of the total geographic area of four countries (India, Nepal, Bangladesh and Pakistan) and half of this are irrigated (Ladha et al., 2000). This area could be utilized successfully by incorporating short duration determinate types that complete their life cycle from April to mid July. Incorporation of blackgram alongwith mungbean in IGP is important to enhance the productivity of poor countries to meet the protein requirements (Zahid et al., 1998). Under a rice-wheat farming system, after the harvest of wheat (end April- mid May) and before transplanting the rice (August) the

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land is available for 80 to 90 days and short duration uniform maturing genotypes of blackgram could be incorporated to enhance protein and soil productivity. Although, blackgram has been identified as a potential crop in most of the Asian countries, but due to instability its adaptation is limited. Although, genetic diversity seems low for qualitative traits but this crop is known for low genetic diversity for various plant traits and a little information is available on plant descriptors in blackgram. Low genetic diversity along with instability is the most limiting factors for wider adaptation of this crop.

One of the reasons for less breeding and genetic work is associated with low genetic variability in blackgram as compared to other summer legumes (mungbean, cowpea). The present study was conducted to assess and identify sources of high yielding and determinate germplasm suitable for spring sowing which can provide base material for the crop improvement.

Materials and Methods

In the present study thirty-one black seeded and six green seeded genotypes of *Vigna mungo* from diverse origin obtained from the gene bank of Institute of Agro-Biotechnology and Genetic Resources (IABGR) which were collected from were used to study the extent of genetic variation based on morphological and agronomic traits. The germplasm from Pakistan included in this study was collected from seven agro-ecological zones (Ghafoor et al., 2001). The genotypes used in present study were self pollinated for 2 consecutive years to establish purity. The experiments were conducted with three replications during summer seasons of 1997 and 1998 at the National Agricultural Research Centre, Islamabad, Pakistan (33°40' N and 73°07' E). The genotypes were sown during fourth week of July for summer seasons and second week of April for spring cultivation.

Five rows of 4-meter length for each genotype were made with 10 cm intra-row spacing, whereas inter-row distance was kept 50 cm. The central three rows were used to record data to minimize any border effect. Mash 1 (approved variety) was included as a check in all the experiments. Agronomic and crop protection measures were taken to raise a healthy crop. Data were recorded following the IPGRI descriptors

for *Vigna mungo* and *V. radiata* (IBPGR, 1985). Days to flowering and maturity were recorded at 50% of flowering and 90% maturity and these variables were represented by a single value for each genotype. Plant height (cm), branches plant⁻¹, pods plant⁻¹, grain yield plant⁻¹ (g) and biomass plant⁻¹ (g) were recorded on 10 plants sampled randomly. Pod length (cm) and seeds/pod were recorded on ten pods sampled at random for each genotype. The 100-seed weight was recorded in grams and the harvest index was determined as economic yield expressed in percentage over total biomass. The duration from first pod formation to 90 % maturity was observed in days, and termed the Determinate Maturity Score. A minimum duration was considered to be the best criterion for determinate genotypes. Based on two years' results, the determinant genotypes were selected and further evaluated for days to maturity and grain yield during two seasons (spring and summer) for six years to identify the best genotype/s for future use in the breeding program. The data were analyzed for simple statistics and numerical taxonomic techniques using a cluster analysis procedure (Sneath & Sokal, 1973) with the help of the computer software "Statistica 7.0" and "SPSS 13.0" for Windows 2003. On the basis of performance, eight high yielding and determinate genotypes were selected and further evaluated for spring (mid April to mid July) and summer (end July to end October) seasons for two years and data were recorded for days to maturity, grain yield plant⁻¹ (g) and harvest index (%) for presentation.

Results

On the basis of maturity trait, the difference between the days to first pod matured and days to attain full maturity determines the growth habit, i.e., determinate or in-determinate. The determinate types are very useful for mechanized cultivation and to fit the crop in various cropping patterns, especially in the rice-wheat system of Indo-Gangetic Plains. The cultivars Pak 45921, Pak 45832, Pak 45845, Pak 45851, Pak 45826, Pak 45920, Pak 45854, Pak 45831, Pak 45839, Pak 45838 and Pak 45729 were observed to be determinate and least affected by environmental changes (Fig. 1). Sixteen genotypes in the lower left box were of determinate nature and took less than 10 days to mature after first pod formation. On the basis of cluster analysis, the genotypes were classified into 3 clusters during 1997 and 4

clusters during 1998, whereas 3 clusters were observed on the basis of pooled data (Fig. 2). Twelve genotypes were grouped in cluster I, eleven in cluster II and fourteen in cluster III during 1997, whereas during 1998, cluster I

consisted of 7 genotypes, cluster II six, cluster III nineteen and cluster IV consisted of five genotypes. On the basis of pooled analysis, 12 genotypes were observed in cluster I, seventeen in cluster II and eight in cluster III.

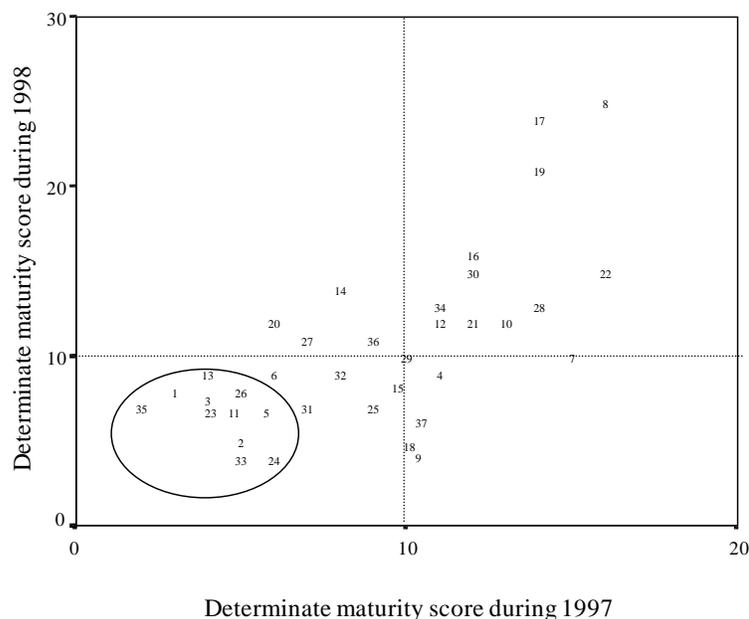


Fig. 1:- Scattered diagram based on determinate maturity scores for 2 years in blackgram. The serial numbers are 1- Pak 45921, 2- Pak 45832, 3- Pak 45845, 4- Pak 45846, 5- Pak 45851, 6- Pak 45834, 7- Mash 1, 8- Pak 45855, 9- Pak 41117, 10- PL-2, 11- Pak 45826, 12- Pak 41096, 13- Pak 45920, 14- Pak 45853, 15- Pak 45852, 16- Pak 45738, 17- Pak 45703, 18- Pak 41065, 19- Pak 45426, 20- Pak 45844, 21- MM 33-40, 22- Pak 45740, 23- Pak 45854, 24- Pak 45831, 25- Pak 41085, 26- Pak 45839, 27- Pak 45849, 28- Pak 45737, 29- Pak 45304, 30- Pak 45736, 31- 9065, 32- Pak 45701, 33- Pak 45838, 34- Pak 45836, 35- Pak 45729, 36- Pak 45841, 37- Pak 41016

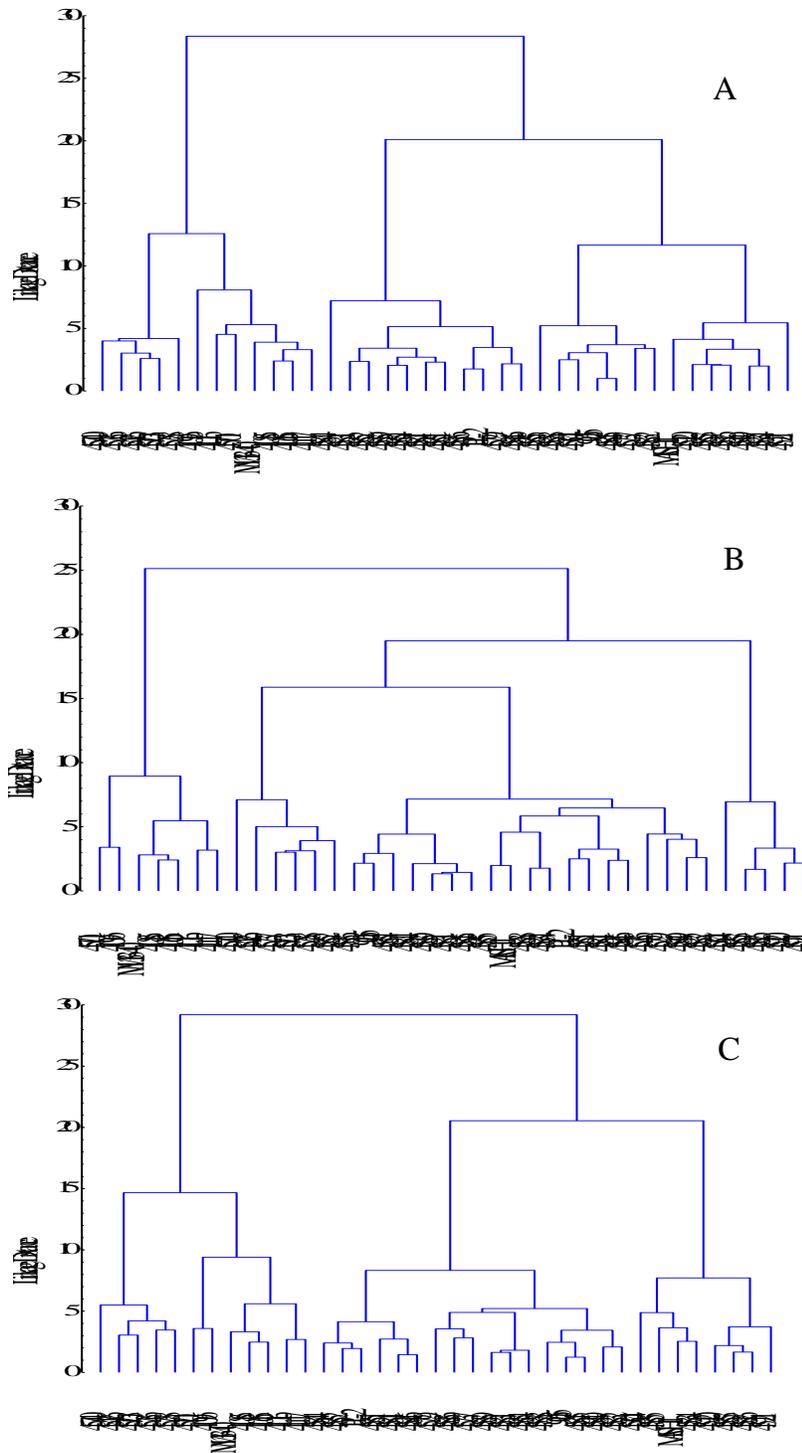


Fig. 2:-Phenogram based on morphological traits for 1998(A), 1999 (B) and average(C)

The clusters were based on agronomic performance rather than origin. All the six green-seeded genotypes (Pak 41117, Pak 41096, Pak 41065, MM 33-40, Pak 41085, Pak 41016) either

local or exotic were grouped together in cluster I in all three sets of analyses. All these genotypes were low yielding and late in maturity which grouped together on the basis of their agronomic performance. The genotypes from varied sources differed considerably; it was difficult to establish any relationship between origin and clustering pattern. Although, these types of results are not uncommon in self-pollinated crops like blackgram but this indicated the validity of multivariate techniques for identification of particular groups of genotypes to be used in future, either evaluation under multilocal testing or including in hybridization programme depending upon the situation. Eight selected genotypes were grouped together in clusters II & III during 1997 and in clusters III and IV during 1998. Mean values along with standard deviation for each cluster for both the years are presented in Table 1. The genotypes in cluster IV were early maturing and high yielding during 1998. The cluster means and SD for different traits provided an interesting picture of nature of the genetic diversity. Genetic diversity based on Nei's genetic distances plotted for two years revealed that, in general, advanced breeding and approved variety lines were more consistent in performance and less influenced by year effect, whereas, genotypes selected from

local collections and exotic origin were more influenced by year effect (Fig. 3).

Among the selected genotypes which were evaluated for two seasons and six years, the genotype Pak 45921 was early maturing and high yielding for both the seasons, although it produced 50 percent less grain yield during the spring season as compared to the summer season (Table 2). In general almost all the genotypes took more days to mature during the spring season. Four genotypes, Pak 45921, Pak 45834, Pak 45826 & Pak 45920, were able to complete their life cycle from April to early July and produced reasonably good grain yield even during the spring season. These could be incorporated successfully in the rice-wheat system of IGP during the spring season when land is available from the end April (after harvest of wheat) to August (before transplanting rice). Fig.4 presents the average performance of eight selected genotypes along with the check variety for two seasons and two years. All the selected genotypes matured in < 80 days except Pak 45701 which was selected from local landraces. Two genotypes in the upper left box were high yielding (> 12 g/plant) and matured in < 70 days and, hence, could be used as base material for development of future cultivars with better adaptability for the spring season.

Table 1. Mean and standard deviation for clusters during 1997 and 1998 based on agronomic characters

Character	1997			1998			
	Cluster I	Cluster II	Cluster III	Cluster I	Cluster II	Cluster III	Cluster IV
Frequency	12	11	14	7	6	20	4
Plant height	50.6±9.33	29.22±6.21	38.9±7.61	44.9±7.57	45.7±15.18	32.7±11.88	38.6±6.37
Days to flowering	53±7.53	43±6.26	40±2.32	57±2.20	46±8.93	39±3.65	35±2.86
Days to first pod matured	88±15.51	65±13.05	61±4.63	100±2.11	71±7.30	61±3.94	61±0.32
Days to maturity	99±11.35	73±12.61	70±4.98	110±2.20	84±8.21	69±5.02	67±2.79
Branches/plant	9.3±4.11	12.4±3.96	17.07±4.22	21.6±7.29	11.2±11.46	17.7±8.55	27.6±8.99
Pods/plant	24.5±14.37	26.7±7.63	55.05±23.2	33.7±22.62	18.6±13.33	39.1±14.30	90.6±15.99
Pods/branch	2.9±1.82	2.3±0.73	3.2±0.87	1.5±0.81	2.1±1.11	2.3±0.69	3.5±0.93
Pod length (cm)	4.4±0.16	4.1±0.21	4.4±0.07	5.03±0.41	3.9±0.19	4.4±0.17	4.9±0.41
Seeds/pod	5.9±0.27	5.5±0.47	6.1±0.36	6.2±0.77	5.1±0.48	6±0.33	6.5±0.85
Seed weight (g)	4.56±0.64	4.21±0.48	4.23±0.43	4.64±0.58	4.71±0.68	4.34±0.31	4.52±0.45
Biomass/plant (g)	23.9±7.86	16.4±4.93	31.8±9.25	29.7±11.82	20.9±13.98	26.26±8.58	54.22±7.20
Grain yield (g)	4.41±2.14	4.72±1.83	10.23±3.99	6.62±4.55	2.27±1.51	6.74±2.72	17.66±1.58
Harvest index (%)	19.12±7.88	28.90±7.33	31.87±6.19	20.75±8.52	11.21±2.35	26.43±6.98	32.81±1.97

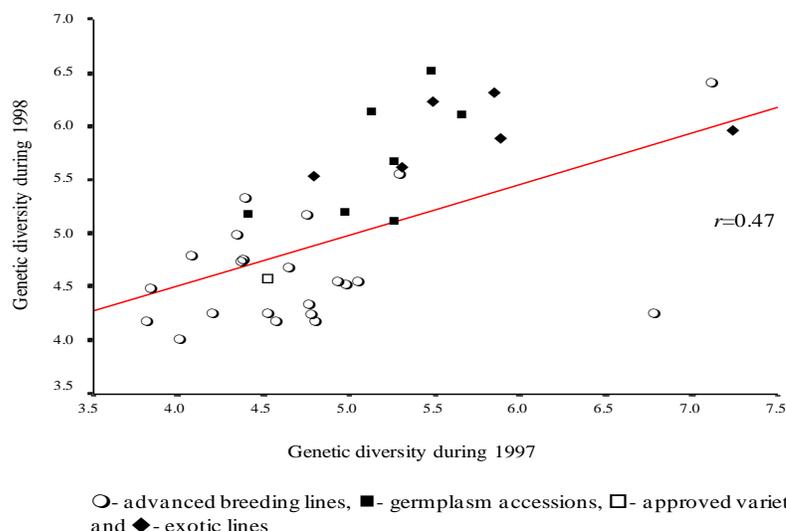


Fig. 3:- Scatter diagram for blackgram evaluated for morphological and agronomic traits during two years

Table 2. Days to maturity and grain yield (g) of nine genotypes of blackgram evaluated during spring and summer seasons during six years.

	1999		2000		2001		2002		2003		2004		SD (DM)	SD (GY)												
	Spring	Summer																								
	DM	GY																								
Pak 45921	72	12.2	68	19.2	76	8.6	68	19.8	70	10.9	67	20.4	75	10.1	71	17.9	74	9.5	66	20.9	77	11.9	66	19.4	4.0	4.9
Pak 45834	76	7.9	65	16.6	74	8.1	68	10.2	77	5.2	66	15.1	71	6.1	74	9.8	79	6.5	66	14.2	80	8.9	69	11.9	5.3	3.7
Mash 1	86	4.1	72	9.45	79	5.4	76	8.8	87	3.2	77	8.2	84	3.2	79	8.9	89	3.9	70	5.8	82	5.8	78	9.0	5.9	2.4
Pak 45826	70	11.8	67	14.0	71	9.5	68	18.0	71	11.1	65	16.1	74	12.2	71	15.2	75	10.1	69	13.9	74	8.5	62	17.2	3.8	3.1
Pak 45920	82	9.4	70	8.61	81	9.1	65	16.0	87	6.2	74	6.5	81	6.8	68	20.1	85	9.8	71	7.9	88	10.0	66	17.2	8.4	4.6
Pak 45853	75	7.6	78	13.8	77	6.8	70	10.9	74	6.7	77	11.4	81	6.1	74	11.1	79	6.5	77	14.0	79	5.9	74	9.9	3.0	3.0
Pak 45304	81	6.9	71	7.05	86	4.1	71	19.2	85	2.1	72	5.2	94	5.2	77	18.4	86	5.4	70	5.6	89	4.8	72	21.0	8.4	6.7
Pak 45701	0	0	108	8.68	115	2.1	108	12.5	119	3.0	109	9.5	119	1.1	110	9.0	0	0	112	9.0	119	2.5	110	11.9	44.1	4.7
Pak 45838	80	5.4	70	6.73	76	5.4	67	10.2	83	3.6	74	4.3	79	3.9	82	11.3	85	4.2	77	4.9	79	6.3	69	9.9	5.8	2.7
SD	26.4	3.8	13.1	4.5	13.2	2.5	13.3	4.3	14.8	3.3	13.3	5.5	14.7	3.4	12.6	4.4	27.7	3.3	14.3	5.4	13.6	2.9	14.3	4.6		

DM- days to maturity, GY- grain yield plant⁻¹ (g). SD is the standard deviation. The genotype, Pak 45701 did not mature during spring season of 1999 and 2003.

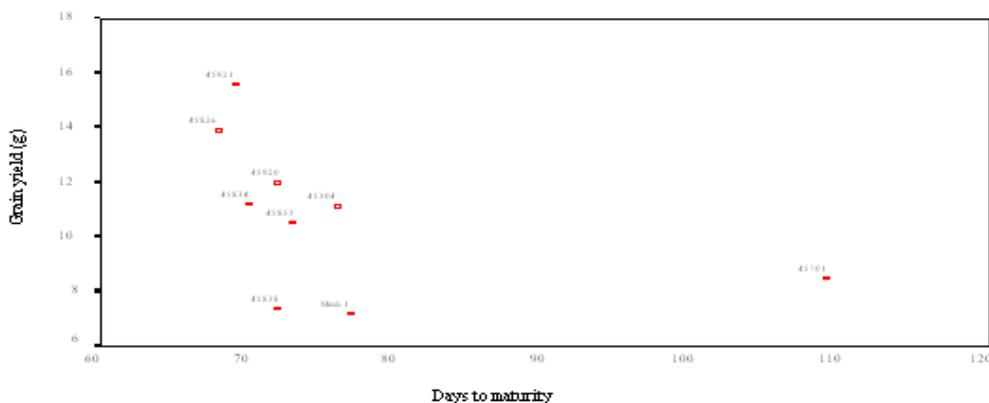


Fig. 4. Scatter diagram based on days to maturity and grain yield/plant of nine genotypes of blackgram evaluated for Spring and Summer seasons during six years.

Discussion

If one of the goals is to bring together cultivars with genetically similar characteristics, quantitative characters may be useful for grouping (Sanni et al., 2012), nevertheless, the qualitative traits must be used often for separating varieties when a limited range of quantitative traits is found (Kurlovich, 1998). In order to develop a determinate plant type, genotypes which took fewer days to mature after first pod formation were selected for testing in 2 distinct seasons for two years and out of these, 2 genotypes (Pak 45921, Pak 45826) gave encouraging results. The genotypes involved in the experiment were pure-lines and the genotypes identified could be tested under a wide range of environment for their adaptability and yield potential. Further, these could also be incorporated in breeding programmes to develop superior cultivars with determinate and stable nature. Cluster analysis was able to isolate the desirable genotypes which could be utilized by the breeders to develop new varieties. Grouping of advanced breeding lines in one cluster revealed that only a portion of the available genetic diversity has been exploited for blackgram improvement and this needs to be broadened by involving diverse parents in a hybridization programme (Shafique et al., 2011).

Although genotypes with greater morphological similarity grouped together, a cluster did not necessarily include genotypes from the same source or origin. Salimi et al., (2012) reported no association between morphological traits and origin or source in soybean. On the basis of grain yield, genotype Pak 45921 was the best, followed by Pak 45826, Pak 45920, Pak 45834 and Pak 45304. Classification of germplasm gave a rise to some elite lines, viz., Pak 45921, Pak 45826, Pak 45920, Pak 45834, Pak 45304 and Pak 45853 which may provide base material for developing dual season blackgram cultivars. It was determined that blackgram germplasm evaluated in the present studies displayed a wide range of genetic diversity for most of the quantitative traits studied alongwith some genotypes with unique characters that enabled us to identify short duration and determinate genotypes without losing yield potential. Two identified genotypes could be exploited to enhance farm productivity in the rice-wheat cropping system of the IGP. Short duration cultivars of blackgram are

unavailable in South Asian countries at present, although mungbean is being cultivated due to the development of short duration mungbean cultivars (Chandna et al., 2012).

Clusters II and III during 1997 and III and IV during 1998 possess desirable genes for days to maturity and yield potential. These could be utilized for future development involving them in crossing programme, because the parents with high performance from various clusters produce better combinations in blackgram (Solomon et al., 2012). During both the years, 23 genotypes were common and out of eight genotypes, seven except Pak 45701 were in these clusters, whereas all the four genotypes identified for spring cultivation were in these clusters that revealed the validity of cluster analysis. At present, mungbean (*Vigna radiata*) is being cultivated under rice-wheat system of IGP due to early maturing genotypes. In our study, the blackgram genotypes with same maturity range (as in mungbean) have been identified and tested under natural ecological zone of blackgram. Due to high yield potential of blackgram as compared to mungbean, we expect high farm production of rice-wheat system and further the areas where mungbean could not be cultivated due to climatic requirements; blackgram could have a chance to fit in the system.

Insignificant differences due to seasons for harvest index, and year-season interaction (days to maturity & harvest index) were attributed to effects of two distinct seasons (spring & summer). Anyhow within one season (either summer or spring), selected genotypes proved their consistency. In the present studies, reproducibility of the experiments over the seasons and years indicates that selection for desirable traits could be effective. Careful selection could give rise to genotypes that may be used for both growing seasons, i.e., spring and summer. Subdividing genotypes into various groups assists in the conservation of genetic resources and their utilization. It enables planning in the use of appropriate gene pools in crop improvement for specific plant attributes (McCouch, et al., 2012; Maxted et al., 2012).

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