

GENETIC DIVERSITY ANALYSIS IN MUNGBEAN
(*Vigna radiata* (L.) WILCZEK)

BY

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CERTIFICATE

*This is to certify that thesis entitled, "Genetic Diversity Analysis in Mungbean (*Vigna radiata* (L.) Wilczek)" submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of **MASTER OF SCIENCE** in **GENETICS AND PLANT BREEDING**, embodies the result of a piece of bona fide research work carried out by **MST. SUFARA AKHTER BANU**, Registration No. 07-2610 under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.*

I further certify that such help or source of information, as has been availed of during the course of this investigation has duly been acknowledged.

*Dated: December, 2008
Place: Dhaka, Bangladesh*

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**Dedicated to
My
Beloved Parents**



SOME COMMONLY USED ABBREVIATIONS

| Abbreviation | Full Word |
|----------------|--|
| AEZ | Agro-Ecological Zone |
| BARI | Bangladesh Agricultural Research Institute |
| SAU | Sher-e-Bangla Agricultural University |
| HI | Harvest Index |
| % | Percent |
| g | Gram(s) |
| kg | Kilogram(s) |
| cv. | Cultivar(s) |
| t/ha | Tonnes per hectare |
| hr | Hour(s) |
| ppm | Parts per million |
| °C | Degree Celsius |
| m ² | Meter square |
| NS | Non significant |
| cm | Centi-meter |
| No. | Number |
| var. | Variety |
| <i>et al.</i> | And others |
| etc. | etcetra |
| RCBD | Randomized Complete Block Design |
| m | Meter |
| G | Genotype |
| GN. | Genotype Number |
| BD | Bangladesh |
| MOA | Ministry of Agriculture |
| <i>Univ.</i> | University |
| <i>J.</i> | Journal |
| <i>Sci.</i> | Science |
| <i>Agric.</i> | Agriculture |
| <i>Agron.</i> | Agronomy |
| <i>Agril.</i> | Agricultural |
| <i>Res.</i> | Research |
| CVA | Canonical Variate Analysis |
| CLU | Cluster Analysis |
| PCO | Principal Coordinate Analysis |
| σ^2 | Variance |
| σ | Standard deviation |
| RFLP | Restriction fragment length polymorphism |
| RAPD | Rapid amplified polymorphic DNA |



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GENETIC DIVERSITY ANALYSIS IN MUNGBEAN (*Vigna radiata* (L.) WILCZEK)

By

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ABSTRACT

Forty mungbean genotypes was evaluated during February, 2008 to May, 2008 at Sher-e-Bangla Agricultural University farm to identify genotypes to be utilized in mungbean breeding for the improvement of economic traits. High variance was observed for days to maturity, branches per plant, pods per plant, seeds per pod, seed yield per plant, biological yield per plant and harvest index. Seed yield per plant showed significantly positive correlation with branches per plant, pods per plant, biological yield per plant and harvest index. Negative association of biological yield with harvest index showed physiological inefficiency for appropriate partitioning of total dry matter towards economic yield, consequently the accessions with low grain yield attained low harvest index. The germplasm accessions were grouped into six clusters on average linkage basis. First three principal components (PCs) with eigen values > 1 contributed 71.47% of the variability amongst the accessions. The populations with greater PC₁ values were high yielding, late maturing and were characterized by high number of branches with more pods. The second component was strongly associated with earliness, more number of seeds and high harvest index. Green seed coat color and shiny seed luster proved their importance in selection for improving most of the yield contributing traits.



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Chapter I

Introduction



CHAPTER I INTRODUCTION

The genus *Vigna* is pan tropical and now has been broadened to include about 170 species, 120 from Africa, 22 from Indo-Pak sub-continent and Southeast Asia, and a few from other parts of the world (Ghafoor, *et. al.*, 2001). Only seven species of *Vigna* are cultivated as pulse crops mostly in Asia, Africa and some parts of Latin America (Anishetty and Moss, 1988). It is generally considered that two of these cultivated species are of African origin (subgenus *Vigna*) and five are Asiatic origin (subgenus *Ceratotropis*). The Asiatic group consists, mungbean/greengram (*Vigna radiata* L. Wilczek), blackgram (*Vigna mungo* L. Hepper), mothbean (*Vigna aconitifolia* Jack. Marechal), adzukibean (*Vigna angularis* Willd, Ohwi and Ohashi) and ricebean (*Vigna umbellata* Thunb, Ohwi and Ohashi). The subgenus *Ceratotropis* of the genus *Vigna* includes five important Asian pulses; mungbean, blackgram, ricebean, mothbean and adzukibean. Mungbean and blackgram have been the major pulses in Asia since ancient times (Paroda and Thomas, 1988). At present, mungbean cultivation spreads worldwide because it is easily digested as compared to blackgram (Smartt, 1990). The subgenus *Ceratotropis* is considered to have originated in Asia and is called Asian *Vigna*. It forms a discrete group of about seventeen species largely confined to Asia and the Pacific. *Ceratotropis* can be subdivided into two groups by the seedling characteristics, i.e., (1) "mungbean

group" showing an epigeal germination and (2) "adzuki bean group" showing a hypogeal germination. *Vigna radiata* var. *sublobata* belongs to the subgenus *Ceratotropis* in the genus *Vigna*. This species was formerly treated by many authors as *Phaseolus sublobatus* Roxb, which was considered as the common ancestor of both *V. radiata* and *V. mungo* (Verdcourt, 1970). After further studies, it was revealed that the taxon contained two different forms, one related to *V. radiata* and the other to *V. mungo* (Arora, et. al., 1973; Singh, et. al., 1974; Jain and Mehra, 1980). Lukoki, et. al., (1980) accepted the specific distinction between the two forms and their relations as wild ancestors to the cultivated species. They treated two forms as *V. radiata* var. *sublobata* (Roxb.) Verdcourt and *V. mungo* var. *silvestris* Lukoki. This was confirmed by Chandel, et. al. (1984) and Miyazaki, et. al. (1984), based on the detailed morphological and biochemical studies.

Mungbean (*Vigna radiata* L. Wilczek) is an annual food legume. It is one of the important crops well suited to dry areas, mainly under irrigated conditions. It is cultivated traditionally by small landholders throughout tropical, subtropical and temperate zones of Asia including Bangladesh, Pakistan, India, Srilanka, Nepal, Thailand, China, Korea and Japan. Since it has a short maturity span (60-75 days) mungbean is grown under various cropping systems, hence contributing to the increase of the small landholders' income as well as to the improvement of the soil conditions (Fernandez and Shanmugasundaram, 1988). Mungbean is used to make daal, which is the most common dish in the South Asia. It is also used to make various kinds of sweet, bean jam, sweetened bean soup, vermicelli, and bean sprout.



In Bangladesh it is grown under a wide range of agro-ecological zones of both rainfed and irrigated nature mainly in the Barisal and Potuakhali district. During 2006-2007, it was cultivated over an area of 65 thousand ha with 40 thousand tones production, and average yield of 0.62 tones/ha (MOA of Bangladesh, 2007). The average yield is much low than its potential, and the yield obtained in many other countries. One of the reasons of low yield is unavailability of high yielding cultivars with better adaptability. World agriculture has been successful in the past century in meeting the demand for cereals that are more researched as compared to legumes. To increase the agricultural productivity, it is necessary to use a broader range of the plant genetic diversity, particularly of legumes and minor crops genetic resources. The importance of genetic diversity in plant breeding is obvious from the results obtained in different crops (Rabbani, *et. al.*, 1998; Ghafoor, *et. al.*, 2001; Upadhyaya, *et. al.*, 2002; Upadhyaya, 2003). The knowledge of genetic diversity is useful tool in gene-bank management and planning experiments because it facilitates efficient sampling and utilization of germplasm either by identifying and/or eliminating duplicates in the gene stock ultimately resulting in the development of core collection philosophy. Smith and Smith (1989) considered morphological characterization as an important step in description and classification of crop germplasm because a breeding programme mainly depends on the magnitude of genetic variability (Smith, *et. al.*, 1991). The multivariate analysis, and in particular the principal component and cluster analyses (Mardia, *et. al.*, 1979) have been utilized for the evaluation of germplasm when studying various traits and a large number of accessions.

Variances of relatively highly heritable, quantitative genetic markers provide an estimate of genetic diversity. Sokal (1965) advocated calculating generalized variances the determinant of the variance-covariance matrix derived from morphological characters as indices of intrapopulation diversity. For example, Goodman (1968) estimated the comparative intra-accession variability of several maize and cotton genotypes. Subdividing the variance into its components assists the genetic resources conservation and their utilization. It enables planning for use of appropriate gene pool in crop improvement for specific plant attributes (Pecetti, *et. al.*, 1996). Various numerical taxonomic techniques (Brown and Weir, 1983; Nei, 1987; Weir, 1990) have been successfully used to classify and measure the pattern of phenotypic diversity in the relationship of germplasm collections in a variety of crops by many scientists as in mungbean (Ramana and Singh, 1987; Singh, 1980; Ghafoor, *et. al.*, 2000), blackgram (Shanmugam and Rangasamy, 1982; Dasgupta and Das, 1984 and 1985; Ghafoor, *et. al.*, 2001).

The availability of transgressive segregant in any breeding program depends upon the diversity of the parents. The quantification of genetic diversity through biometrical procedures such as Mahalanobis's D^2 - statistic and Canonical Variate Analysis (CVA) has made it possible to choose genetically diverged parents. The Mahalanobis generalized distance (D^2 - statistic) may be an efficient tool in the quantitative estimation of genetic diversity. Inclusion of more diverse parents (within a limit) in hybridization is supposed to increase the chance of obtaining maximum heterosis and give broad spectrum of variability in segregating generations. Selection



of parents based on Genetic divergence has been successfully utilized in different crop species (Das, *et. al.*, 1993). Germplasm resources are of little value unless they are used by the breeders for crop improvement. Keeping in view the importance of mungbean as a pulse crop of Bangladesh, a wide range of lines were collected from BARI and evaluated under field conditions for various qualitative and quantitative traits with the following objectives:

- 1) To study the genetic variability for different quantitative and qualitative characters.
- 2) To determine the nature of relationship between yield and yield contributing characters and relative contribution of each character towards seed yield in mungbean through the correlation co-efficient analysis.
- 3) To identify the genetically diverse parents for utilization in future breeding program.



Chapter II

Review of literature

CHAPTER II REVIEW OF LITERATURE

Estimates of genetic diversity and relationship between germplasm are very important for facilitating efficient germplasm collection, evaluation and utilization. Many tools are now available for identifying desirable variation in the germplasm including total seed protein, isozymes and various types of molecular markers. However, morphological characterization is the first step in the description and classification of germplasm (Singh and Tripathi, 1985; Smith and Smith, 1989). Bekele (1985) thoroughly discussed the importance of a hierarchical approach to quantitatively define the variance in the centre of genetic diversity over a range of micro environments. Subdividing the variance into its components may assist in genetic resources conservation and utilization by determining the relative contribution of the different levels of variability to the total diversity available in any one area. This would enable planning of future germplasm sampling, establishment of *in-situ* gene conservation, or use of appropriate gene pools in crop improvement for specific plant attributes (Bekele, 1984; Pecetti, *et. al.*, 1992). Germplasm evaluation must be considered as the first step in plant breeding programme and it is commonly based on a simultaneous examination of a large number of populations for several characters of both agronomic and physiological interest (Pezzoti, *et. al.*, 1994).

In mungbean, Tomar, *et. al.*, (1973) and Khalid, *et. al.*, (1984) observed positive correlation of yield with yield components, whereas, Malik, *et. al.*,



(1987) reported negative correlation of yield with maturity, pod length and seed weight. Malik, *et. al.*, (1983) investigated maximum relative selection efficiency for branches per plant in mungbean. Malhorta, *et. al.*, (1974) observed positive association of yield with days to maturity, plant height, pods and pod length, whereas, negative with seed weight. Lal (1967), Singh (1977), Singh, *et. al.*, (1980), Patel and Shah, (1982), Malik, *et. al.*, (1981, 1986), Khan and Malik (1989) and Ghafoor, *et. al.*, (1993b) gave emphasis for the selection of legume genotypes on the basis of high harvest index. Positive correlation among yield and its components has been reported by Rani and Rao (1981) in blackgram.

Virmani, *et. al.*, (1983) categorized mungbean germplasm in various groups for different traits. The genetic diversity between *V. radiata* and *V. mungo* was reported by Chen, *et. al.*, (1983) and Egawa (1988), Singh and Srivastava (1985) categorized pea germplasm into various groups. Ghafoor, *et. al.*, (1989) classified blackgram germplasm and selected eleven pure-lines for further exploitation. Bakhsh, *et. al.*, (1992) categorized lentil germplasm on the basis of quantitative traits and suggested the utilization of short stature lentil genotypes for crop improvement. In a study on mungbean, Ghafoor, *et. al.*, (1992) selected twenty eight genotypes on the basis of high yield potential and resistance to diseases.

Lin and Cheng (1988) evaluated mungbean germplasm collected from 20 provinces of China. They reported three classes of growth habit (erect, semi-trailing and trailing), two of seed coat colour (green and yellow) and two of seed surface luster (shiny and dull). The seed colour in mungbean exhibits a wide variation from

yellow, greenish yellow, light green, dark green, dull green, black, brown and mottled with black (Paroda and Thomas, 1988). Sandhu, *et. al.*, (1988) screened 2028 mungbean accessions against Mungbean Yellow Mosaic Virus (MYMV) and Urdbean Leaf Crinkle Virus (ULCV) and reported less than 10% lines as resistant against both diseases. Ghafoor, *et. al.*, (1998) evaluated 285 blackgram accessions; they observed 25 accessions resistant to both MYMV and ULCV. They also classified blackgram germplasm on the basis of growth habit and seed coat colour.

Results reported by Falcinelli, *et. al.*, (1988) and Veronesi and Falcinelli (1988) showed multivariate analyses to be a valid system to deal with germplasm collection. Nevertheless, the qualitative traits must be often used for separating varieties when a limited range of quantitative traits is found in certain groups (Sneddon, 1970). Broschat (1979) considered principal component analysis (PCA), a useful data reduction technique which worked by removing inter-relationship among variables.

Dasgupta and Das (1984) conducted multivariate analysis in blackgram and considered it a method of choosing parents for hybridization using D^2 analysis. Data on 12 characters on forty strains of blackgram collected from India and Nepal were used. The genotypes were grouped into seventeen different clusters and no clear association was observed between clusters and geographical origin. Similarly genetic divergence was conducted in 38 genotypes of blackgram by Dasgupta and Das (1985) using D^2 statistics. No relationship was observed between geographic distribution and genetic divergence of the varieties. Flowering time and seed size



exhibited maximum contribution to the total divergence. Environmental conditions exerted considerable impact on the number and composition of clusters. Suggestion has been made for selecting suitable stable diverse parents so as to initiate a crossing programme for increased grain yield.

Seventy two landraces of pea (*Pisum sativum*) evaluated for 19 morphological characters exhibited broad genetic diversity as reported by Amurrio *et. al.*, (1993). Seven landraces were selected for special attention for having promising breeding value. Amurrio, *et. al.*, (1995) reported a wide genetic diversity in 105 pea landraces at the intraspecific level based on 19 quantitative characters. Taxonomically useful results were provided and six groups were established but the grouping pattern of these landraces did not reflect any association with geographic origin. Smith, *et. al.*, (1991) studied principal components and average cluster analyses in alfalfa and established six geographically distinct groups. Significant regional variation was observed within the germplasm evaluated but ecotypes from neighboring countries were generally closely associated. All elite germplasm accessions fell in one group and this revealed that only a small portion of genetic diversity has been used in formal breeding. Multivariate analyses have been used successfully to classify and order variation observed in both qualitative and quantitative traits in collection of crop germplasm (Singh, 1988; Peeters and Martinelli, 1989; Caradus, *et. al.*, 1989). Rumbaugh, *et. al.*, (1988) used discriminate analysis of morphological and agronomic characters to place 146 accessions of alfalfa from Morocco into five geographical groupings that were defined initially based on the area of collection.

Smith *et. al.*, (1991) conducted average linkage cluster and PCA, and reported the utility of these results in preservation and utilization of germplasm. The landraces of tetraploid wheat from two provinces (Shewa and Tigray) of Ethiopia were found to be distinctly different (Pecetti, *et. al.*, 1996). This divergence was attributed to the differences in environmental conditions between them. Wide differentiation among landraces within each province was also present. The proportion of total variance due to differences among agrotypes within landraces was by far the greatest found in this study. Various reasons were advocated for the occurrence of a great diversity in wheat, such as isolation from other wheat germplasm, primitive farming systems, heterogeneous environments, field mixture, and natural cross fertilization due to field mixtures, knowledge on the pattern of variation for important morpho-agronomic traits is needed for a proper improvement and better exploitation of gene pool (Jain, *et. al.*, 1975).

Perry and McIntolsh (1991) characterized soybean germplasm from 78 countries for seventeen traits and determined variation within and among all regions for most characters. Canonical discriminate analyses and clustering of the canonical means delineated four regional clusters: i) India and Africa ii) China, Europe, New World and Southeast Asia iii) Korea and Japan and iv) Southwest Central Asia. The cluster containing the Korean and Chinese accessions was the most diverse. Based on the diversity and number of accessions, Africa, India and Southeast Asia seemed under-represented in the collection. One approach for building gene pool is to collect material from diverse geographical origins with a concentration of accessions



from proposed centre of diversity. This should capture inherent and unexploited diversity in the individual samples. Representative samples from the complete geographical range of the crop species can help to ensure that co-adapted gene complexes (or correlated adaptations) are conserved (Frankel and Soule, 1981; Frankel, 1984). Brown (1978) advocated that the maximum genetic conservation would be achieved by sampling populations from as many distinct environments as possible.

Malhotra and Singh (1971) while working on genetic divergence in blackgram reported narrow range of variability for 100-seed weight and pod length whereas, Shanmugam and Rangaswamy (1982) while analyzing 45 genotypes of blackgram reported that yield per plant contributed most to the genetic diversity. Malik, *et al.*, (1985) studied genetic divergence in 12 indigenous varieties of mungbean for six quantitative characters. The study indicated the presence of ample genetic variation among the cultivars irrespective of their origin. They suggested that plant height, days to flowering and grain yield should be considered for selecting genetically divergent lines in mungbean.

Tawar, *et al.*, (1988) conducted genetic divergence using D^2 analyses in 34 genotypes of mungbean and these were grouped in five clusters. Variability observed in the parents was related to genetic diversity of the parents selected under study. First canonical root contributed 88% of the total variation. Inclusion of such genotypes from distinct clusters and their implication in mungbean breeding programme was suggested. Singh, *et al.*, (1991) examined the organization of diversity for

morphological and agronomic characters in 306 landraces of cultivated common bean (*Phaseolus vulgaris* L.) by analyzing data for multivariate statistical analyses and observed genetic variance within and between groups. Kumar and Arora (1992) presented observation of 40 genotypes of chickpea collected from various geographical regions for 18 characters including seed yield. Multivariate analyses revealed 10 clusters. No definite relationship was established between genetic diversity and geographical distribution. Maximum hybrid vigor was observed among most diverse genotypes.

Mishra and Rao (1990) reported thirteen clusters in a comparative study of D^2 and meteroglyph analyses of 117 chickpea genotypes. Cluster I had the maximum number of genotypes. Meteroglyph analysis did not show similar type of clustering as observed in D^2 analysis, but canonical analysis showed similar type of clustering. Gupta, *et. al.*, (1991) and Dias, *et. al.*, (1993) reported no association between morphological characters and geographical origin in cabbage and kale landraces whereas Revilla and Tracy (1995) observed a low level of morphological variability amongst widely used open-pollinated sweet corn cultivars.

Clements and Cowling (1994) investigated the pattern of morphological diversity in relation to geographical origins of 157 accessions of wild *Lupinus angustifolius* using multivariate technique. Genetic diversity was extremely large for most of the morphological traits, with significant variation detected among localities in Greece and within and between collection sites for same trait. Thirteen

groups were identified by hierarchical clusters analysis. Accessions from northern Greece grouped together as late flowering, shorter, and smaller seed size, but some accessions from southern Greek Islands were grouped with the northern mainland types. Multivariate analyses provide a good evaluation of landraces by identifying those that should be further evaluated at the genetic level (Rouamba, *et. al.*, (1996). Laghetti, *et. al.*, (1998) suggested collecting expedition to the areas where genetic erosion takes place in cowpea along with the areas where genetic existing diversity has not yet gathered (Padulosi, 1993).

Ahmad, *et. al.*, (1997) observed that cluster analyses on the basis of quantitative characters were phenotypically more distinct and exhibited more breeding value. Though cluster analyses grouped together accessions with greater morphological similarity, the cluster did not necessarily include all the accessions/genotypes from the same or nearby sites. Maqbool, *et. al.*, (1997) reported phylogenetic relationship of 15 genotypes of the genus *Lens* and seven of their interspecific hybrids were determined by morphological (quantitative and qualitative) characters.

Rabbani, *et. al.*, (1998) determined the extent of diversity and relationship among *Brassica juncea* germplasm from Pakistan for 35 morphological characters in 52 accessions using cluster and principal component analyses. The germplasm was categorized into six groups. Landrace group was primarily associated with morphological differences among the accessions and secondarily with the breeding objectives and horticultural uses. The germplasm showed a comparatively



low level of phenotypic variation which revealed that the evaluated germplasm appears to have a narrow genetic base and undergoes a high level of genetic erosion. Though cluster analyses grouped together accessions with greater morphological similarity, the cluster did not necessarily include all the accessions from the same or nearby sites.

Simply inherited characters are important for plant description (Kurlovich, 1998) and are mainly affected by the consumers' preference, socio-economic scenario and natural selection. Nakayama, *et. al.*, (1998) reported that foxtail millet landraces with low amylase allele were distributed only in Southeast Asia mainly because of preference followed selection.

Ghafoor, *et. al.*, (2000) conducted cluster analysis in mungbean for nine quantitative traits. They observed significant negative correlation of days to maturity with all the characters except branches per plant and suggested that short to medium maturity mungbean cultivars were to be selected for high yield. They identified 44 pure-lines on the basis of important agronomic traits that were recommended for testing under wide range of agro-ecological condition in pursuit of best mungbean cultivars.

Ghafoor, *et. al.*, (2001) studied genetic diversity in 484 blackgram germplasm accessions. Quantitative traits were analyzed for cluster and principal component analyses. The first four PCs with eigenvalues >1 contributed 79.5% of the total variability amongst accessions. The germplasm was categorized in five clusters based on average linkage. The first two principal components were

plotted to observe relationship between the clusters. Clusters II, III and IV showed more clear separation than clusters I and V.

Elizabeth, *et. al.*, (2001) investigated nineteen *Sesbania* accessions to characterize them on morphological and agronomic data using multivariate methods. Principal component analysis indicated that variance accumulated by the first two components for morphological and agronomic data was 74.4% and 77.0% respectively. The cluster analysis performed with the eight selected characters classified the accessions into five groups. Upadhyaya, *et. al.*, (2002) studied phenotypic diversity for morphological and agronomic characteristics in 1956 accessions of chickpea core collection, comprising desi, Kabuli and intermediate types. The Kabuli and intermediate types were not significantly different for growth habit and seed colour, while they differed significantly from desi types for both traits. Principal component analysis showed that days to 50% flowering, flowering duration, apical secondary branches, tertiary branches, 100-seed weight, seed colour and seed testa texture were important traits in explaining multivariate polymorphism. Manivannan (2002) analyzed 33 mungbean genotypes derived from ten crosses to determine genetic diversity using multivariate analysis. The genotypes were grouped into seven clusters. Among the characters studied, 100-seed weight and powdery mildew reaction contributed the most towards the total divergence. Based on the performance, they suggested seven genotypes to be included in the hybridization programme.


Thirty seven diverse genotypes of blackgram and three of mungbean resembling to blackgram, were studied by Ghafoor, *et. al.*, (2002) to determine the extent of genetic variation based on morphological characters. High variance was observed for plant height, days to maturity, branches per plant, pods per plant, pod length, seeds per pod, biological yield per plant, grain yield per plant and harvest index (%). First four components of PCA with eigenvalue > 1 contributed 78.7% and 79.1% of the total variance amongst 40 genotypes during two consecutive years.

Upadhyaya (2003) evaluated 1704 accessions of groundnut for various morphological and agronomic characteristics to estimate phenotypic diversity and determine importance of different descriptor traits. Principal coordinate and principal component analyses showed that 12 morphological descriptors and 15 agronomic traits, respectively, were important in explaining multivariate polymorphism. The results of their study indicated that there was a significant variation for morphological and agronomic traits in the groundnut core collection. The phenotypic correlations depended upon the subspecies group. The mean pod length, 100-seed weight and yield per plant was higher in the *hypogaea* group than in the *fastigiata* group while it was opposite for plant height. Thirty genotypes of chickpea were evaluated by Ghafoor *et al.*, (2003a) for 10 quantitative traits using multivariate techniques. The first three PCs with eigenvalues >1 contributed 83.38% of the variability amongst genotypes. Populations with high PC₁ values were characterized by high yield potential, 100-seed weight and harvest index. The populations with high PC₂ values were early in



maturity, high in biological yield and pods but low in seed weight and harvest index. The scattered diagram on the basis of first three factors gave separation of two groups.

Twenty two blackgram genotypes representing a broad based germplasm were analyzed by Ghafoor, *et. al.*, (2003b) using multivariate analyses for two consecutive years. High genetic variance was observed for plant height, maturity, pods, seed weight, biomass, grain yield and harvest index. First four PCs contributed 80.0% of the variation during 1998, and 80.9% during 1999. Five yield contributing traits, i.e. branches, pods, pod length, biomass and grain yield were observed important for first component during both the years. PC₂ was more related to maturity rather than reproductive traits. First two PCs which exhibited about 60% of the variance were plotted to observe the relationship between the cultivars. Five genotypes were separated from others during both the years.



Chapter III
Materials and Methods

CHAPTER III MATERIALS AND METHODS

Agricultural research uses a large number of procedures and techniques for successful conduction of field experiment. The techniques to be adopted depend on the nature of the research trial and its objectives. Success of field experiment largely depends on the appropriateness of establishment. This means how precisely different aspects of field plot techniques are considered and adopted to maximize non-treatment variations or errors.

3.1 Site of experiment:

The experiment was conducted at the field laboratory of the Department of Genetics and Plant Breeding, Sher-e-Bangla Agricultural University, Dhaka during the period from February 2008 to May 2008 (Fig.1). The experimental site was at 90022" E longitude and 23041" N latitude at an altitude of 8.6 meters above the sea level. The physical and chemical characteristics of the soil have been presented in Appendix I.

3.2 Materials:

A total of forty genotypes (40) of mungbean originated from different places of Bangladesh were used in this experiment (Table 1). The materials were collected from Genetic Resources Centre of BARI, Gazipur.



Table 1. List of mungbean genotypes with their sources

| Genotype No. | Name/Acc No. (BD) | Source |
|---------------------|--------------------------|---------------|
| 1 | BD-6901 | BARI |
| 2 | BD-6902 | BARI |
| 3 | BD-6903 | BARI |
| 4 | BD-6904 | BARI |
| 5 | BD-6905 | BARI |
| 6 | BD-6907 | BARI |
| 7 | BD-6909 | BARI |
| 8 | BD-6911 | BARI |
| 9 | BD-6912 | BARI |
| 10 | BD-6913 | BARI |
| 11 | BD-6914 | BARI |
| 12 | BD-6918 | BARI |
| 13 | BD-6924 | BARI |
| 14 | BD-6925 | BARI |
| 15 | BD-6926 | BARI |
| 16 | BD-6927 | BARI |
| 17 | BD-6932 | BARI |
| 18 | BD-6933 | BARI |
| 19 | BD-6934 | BARI |
| 20 | BD-6936 | BARI |
| 21 | BD-6875 | BARI |
| 22 | BD-6876 | BARI |
| 23 | BD-6877 | BARI |
| 24 | BD-6879 | BARI |
| 25 | BD-6880 | BARI |
| 26 | BD-6881 | BARI |
| 27 | BD-6882 | BARI |
| 28 | BD-6884 | BARI |
| 29 | BD-6885 | BARI |
| 30 | BD-6886 | BARI |
| 31 | BD-6888 | BARI |
| 32 | BD-6889 | BARI |
| 33 | BD-6890 | BARI |
| 34 | BD-6891 | BARI |
| 35 | BD-6893 | BARI |
| 36 | BD-6894 | BARI |
| 37 | BD-6895 | BARI |
| 38 | BD-6897 | BARI |
| 39 | BD-6898 | BARI |
| 40 | BD-6900 | BARI |

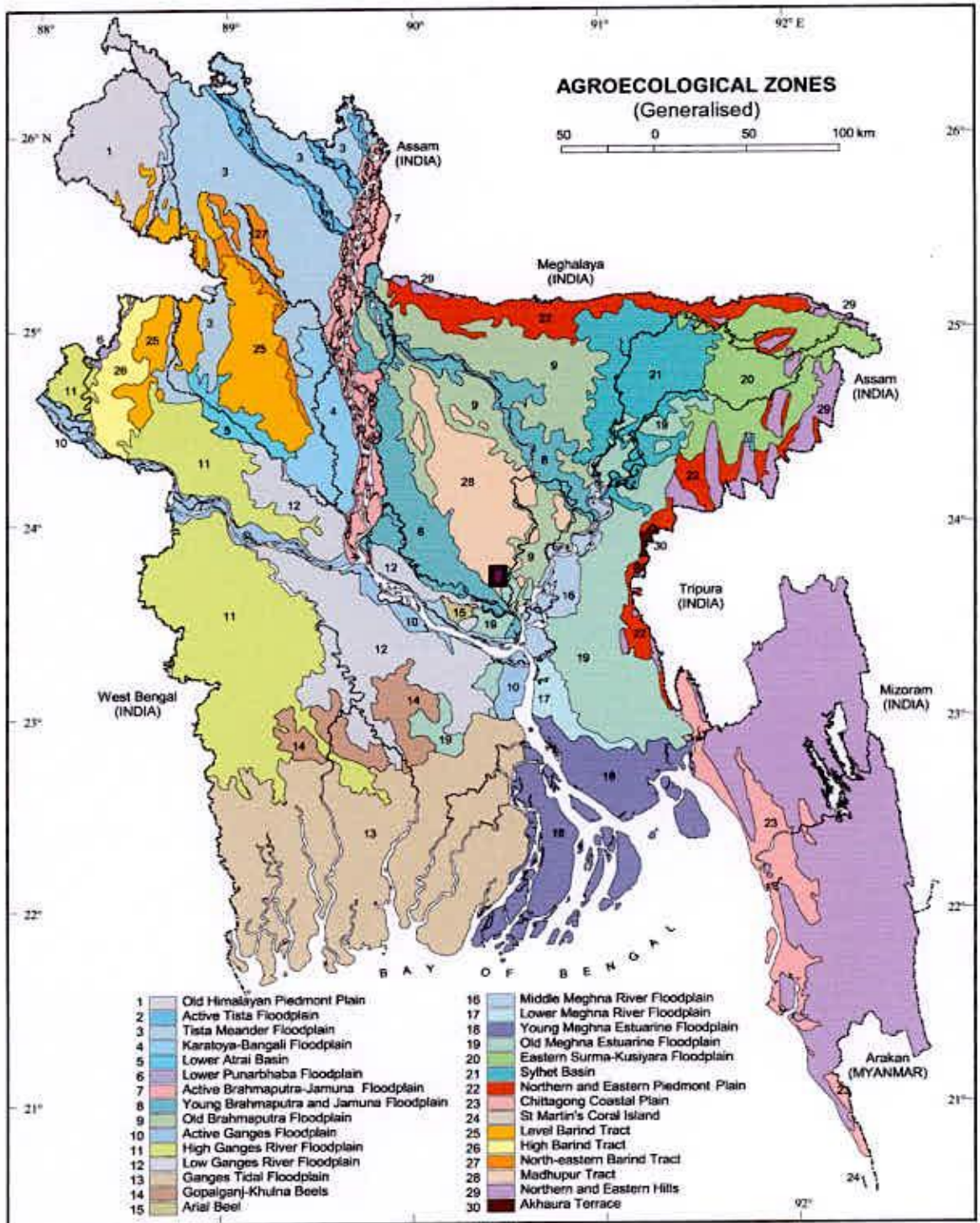


Fig. 1. Location of experimental field



3.3 Soil and climate:

The land belongs to Agro-ecological region of 'Madhupur Tract' (AEZ 28) of Nodda soil series. The soil was sandy loam in texture having pH 5.47- 5.63. The mean temperature of the growing period was 24.36° C with average maximum and minimum being 30.00 C and 18.67° C respectively. The monthly total rainfalls, average sunshine hour, temperature during the study period are shown in Appendix II.

3.4 Experimental design and layout:

The study was laid out in Randomized Complete Block Design (RCBD) with three (3) replications. The plant to plant distance was 10 cm and line to line distance was 30 cm. The total land size was 126.75 m². The block to block distance was 2.5 m. The genotypes were randomly distributed to each row within each block (plate 1 and plate 2).

3.5 Land preparation:

The experimental plot was prepared by ploughing with tractor followed by harrowing and laddering by cows. Weeds and stubbles were removed. Recommended doses of manures and fertilizers were applied before the final land preparation. Irrigation channels were made around each plot. The final land preparation was done on 24 February 2008.

3.6 Manure and fertilizer:

Due to the ability of nitrogen fixation from the atmosphere mungbean require less nitrogen application. But for initial establishment of plant up to the stage of nodule formation a starter dose of 20kg-40kg-20kg NPK respectively was applied.



Plate 1: The overall view of the experimental field



Plate 2: A single mungbean plant in the experimental field

In this study fertilizer was applied as per the recommendation of Bangladesh Agricultural Research Institute (BARI). The following doses of fertilizers and manures were applied to the plot for Mungbean cultivation.

| Fertilizers/ Manures | Dose (kg) | |
|-------------------------|------------------------|-------------|
| | Applied in the plot | Quantity/ha |
| Urea | 1.71 | 45 |
| TSP | 3.23 | 85 |
| MP | 1.33 | 35 |
| Cow dung | Applied earlier | 1.5 ton |

Urea, TSP, MP and Gypsum were applied at the time of final land preparation. Cow dung was applied two weeks before sowing during the land preparation.

3.7 Sowing of seeds and intercultural operation:

The seeds of 40 mungbean genotypes were sown in the field on 25 February 2008. Intercultural practices were done uniformly for all the genotypes. Thinning was done 25 days after sowing and weeding was done twice-the first during thinning and the second after about two months of sowing.

3.8 Harvesting:

Different genotypes matured at different times. The harvesting was completed by 14 May 2008. Ten plants from each plot were randomly selected to collect data and these were harvested by uprooting. Border plants were discarded to avoid border effect.



3. 9 Recording of Experimental Data:

Data on the following characters were recorded on individual plant basis from 10 randomly selected plants from each genotypes in each replicate (plate 3). For the quantitative characters, out of 9 characters, days to maturity were recorded in the field condition and the data on the other characters were recorded in the field Laboratory after harvest.

3.9.1 Days to maturity: Data on days to maturity was recorded from date of sowing to date of pod maturity.

3.9.2 Branches per plant (primary): The total number of primary branches and secondary branches including the main stem was counted.

3.9.3 Pods per plant: The total number of pods in individual plants was recorded.

3.9.4 Pod length: length of pod was measured in cm.

3.9.5 Seeds per pod: Total number of seed in each pod within the individual plants was counted.

3.9.6 Weight of 100 seed: One hundred clean sun dried seeds were randomly taken from each line and weighed in gram (g).

3.9.7 Grain yield per plant : Yield per plant was measured in gram by using electrical balance.

3.9.8 Biological yield per plant: Sun dried plants were weighted by electrical balance, randomly counting ten plants from each of the line.



Plate 3: The author recording data in her experimental field

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12.3.15

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3.9.9 Harvest index: This was measured as the ratio of grain yield to the biomass or biological yield expressed as percentage.

For qualitative characters growth habit (erect, semi erect and spreading), seed coat color (green and black) and seed luster (shiny and dull) were also studied.

3.10 Analysis of data:

In hybridization programme selection of parents based on Mahalanobis's D^2 statistic is more reliable as requisite knowledge of parents in respect of a mass of characteristics is available prior to crossing. Statistical analysis such as Mahalanobis D^2 and Canonical Variate Analysis (CVA), which quantify the differences among several quantitative traits are efficient method of evaluating genetic diversity. Mean data of each quantitative character were subjected to both univariate and multivariate analysis. For univariate analysis of variance, analysis was done individually and least of significance was done by F- Test. Mean, range, standard deviation and correlation was estimated using MSTAT computer programme. Genetic diversity was estimated following Mahalanobis's (1936) generalized distance, Multivariate analysis viz., Principal Component Analysis (PCA), and Cluster Analysis (CLU) were done by using GENSTAT programme.

3.10.1 Principal Component Analysis (PCA):

Principal Component Analysis, one of the multivariate techniques, is used to examine the inter-relationships among several characters. It can be done from the sum of squares and products matrix for the characters. Thus PCA finds linear combinations of a set variety that maximize the variation contained within them; they

are expressed by displaying most of the original variability in a smaller number of dimensions. Therefore, principal components were computed from the correlation matrix and genotype scores obtained for the first components (which has the property of accounting for maximum variance) and succeeding components with latent roots greater than unity (Jeger *et. al.*, 1983).

3.10.2 Clustering:

To divide the genotypes of a data set into some number of mutually exclusive groups clustering was done using non- hierarchical classification. In GENSTAT, algorithm was used to search for optimal values of chosen criteria.

3.10.3 Computation of average intra-cluster distances:

When the clusters are formed, the average intra-cluster distance for each cluster was calculated by taking possible D^2 values within the members of a cluster obtained from the principal Coordinate Analysis (PCO). The formula used was D^2/n , where D^2 is the sum of distances between all possible combinations (n) of the genotypes included in a cluster. The square root of the average D^2 values, represent the distance (D) within cluster.



Chapter IV

Results and Discussion

CHAPTER IV RESULTS AND DISCUSSION

The knowledge of genotypic variation within genotypes in relation to morphology, phenology and yield would help to screen better genotypes. Therefore, to generate in the degree of diversity forty lines of mungbean were raised in the growing season of 2008 at the field of Sher-e-Bangla Agricultural University, Dhaka. The data in respect of days to maturity, branches per plant, pods per plant, pod length, seeds per pod, 100 seed weight, grain yield per plant, biological yield per plant and harvest index for quantitative characters and for qualitative characters like growth habit (erect, semi erect and spreading) seed coat color (green and black) and seed luster (shiny and dull) were recorded, analysed and presented in this chapter.

The availability of transgressive segregants in breeding program depends upon the divergence of the parents. So, the accurate information on the nature and degree of diversity of the parents is the pre-requisite of an effective breeding program. Performance of 40 mungbean genotypes was investigated and the findings of present study have been discussed under different morphological characters. The result of the study showed marked variation in different characters and the variation of different characters are presented in following tables, figures and plates.

The data pertaining to nine characters were computed and statistically analyzed and the results obtained are described below:



4.1 Quantitative Characters

Basic statistics for measured quantitative traits, viz., days to maturity, branches per plant, pods per plant, pod length (cm), seeds per pod, 100 seed weight (g), grain yield per plant (g), biological yield per plant (g) and harvest index (%) is presented in Table 2. High variance was observed for all the characters except pod length, seeds per pod and 100 seed weight. Low genetic variability of these three characters seemed to restrict the scope of selection for these traits in the present germplasm collection. The genes for these important economic traits should be investigated and exploited from other sources i.e., inter-specific hybridization or mutation. Large scale testing of broad base germplasm needs to be built up by making extensive local collection and obtaining germplasm from abroad to develop a sound breeding programme (Jain, *et. al.*, 1975 and Ghafoor, *et.al.*, 1992). Brown (1978) and Laghetti, *et. al.*, (1998) advocated that maximum genetic conservation would be achieved by sampling populations from as many environments as possible.

The maturity period ranged from 62-95 days after planting, branches per plant from 2.8-50.0, pods per plant from 2.33-90.00, pod length from 4.20-11.72 cm, seeds per pod from 2.0-14.8, 100 seed weight from 1.88-7.75 g, grain yield per plant from 0.5-32.5 g, biological yield per plant from 6.5-145.3 g and harvest index from 4.71-60.52 (%). The frequency distributions for various quantitative traits are presented in the graphic form (Fig. 2-10).

4.1.1 Days to maturity

For days to maturity, maximum accessions 15 which were 37.5 % of the population, matured within the range of 71-75 days after planting (Fig. 2). It was followed by nine and eight accessions which matured in 76-80 and 66-70 days, respectively. Ten accessions (G1, G2, G7, G8, G11, G18, G24, G25, G26, and G32) which matured in less than 68 days, were selected on the basis of early maturity as presented in Table 3.

4.1.2 Branches per plant

For branches per plant, frequency distribution is depicted in Fig. 3 and it was observed that maximum 15 accessions which were 37.5 % of the total germplasm, had 5-10 branches per plant. Seven accessions had up to 5 branches per plant. Six accessions were observed to be bushy and produced more than 25 branches per plant. Eight accessions (G1, G2, G3, G5, G19, G24, G32 and G34) which produced over 20 branches per plant, were selected on the basis of branches per plant as presented in Table 3.

4.1.3 Pods per plant

Pods per plant ranged from 2.33 to 90.00 and on the basis of class interval, it was observed that 10 accessions produced 20.1 to 30 pods per plant which was followed by the group from 30.1 to 40 pods per plant (8 accessions) and this group can be considered medium pod bearing which were about 20 % of the population (Fig. 4).



Table 2: Range, mean, variance and coefficient of variation for nine morphological characters in 40 mungbean genotype

| Traits | Range | Mean±S.E | σ^2 | σ |
|--------------------------------|-------------|--------------|------------|----------|
| Days to maturity | 62.00-95.00 | 74.06 ± 0.32 | 27.25 | 5.22 |
| Branches per plant | 2.80-50.00 | 8.27 ± 0.38 | 36.98 | 6.08 |
| Pods per plant | 2.33-90.00 | 37.03 ± 1.14 | 341.75 | 18.49 |
| Pod length (cm) | 4.20-11.72 | 7.60 ± 0.06 | 0.93 | 0.96 |
| Seeds per pod | 2.00-14.80 | 12.41 ± 0.10 | 2.62 | 1.62 |
| 100 seed weight (g) | 1.88-7.75 | 3.46 ± 0.05 | 0.70 | 0.84 |
| Grain yield per plant (g) | 0.50-32.50 | 11.05 ± 0.37 | 35.01 | 5.92 |
| Biological yield per plant (g) | 6.50-145.33 | 36.85 ± 1.54 | 618.75 | 24.87 |
| Harvest index (%) | 4.71-60.52 | 32.37 ± 0.59 | 92.51 | 9.62 |

σ^2 = Variance and

σ = Standard deviation



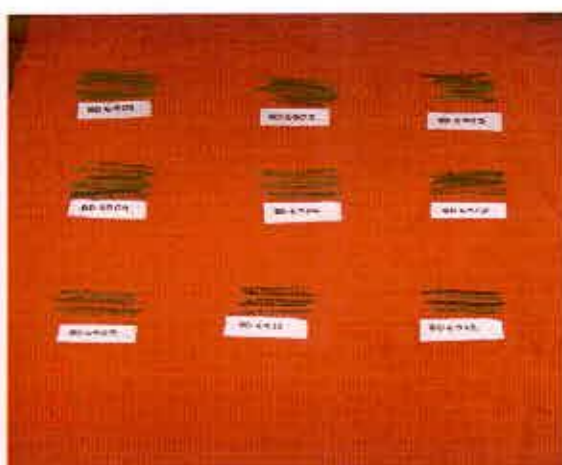


Plate 4: Photograph showing the different type of pods of mungbean genotypes



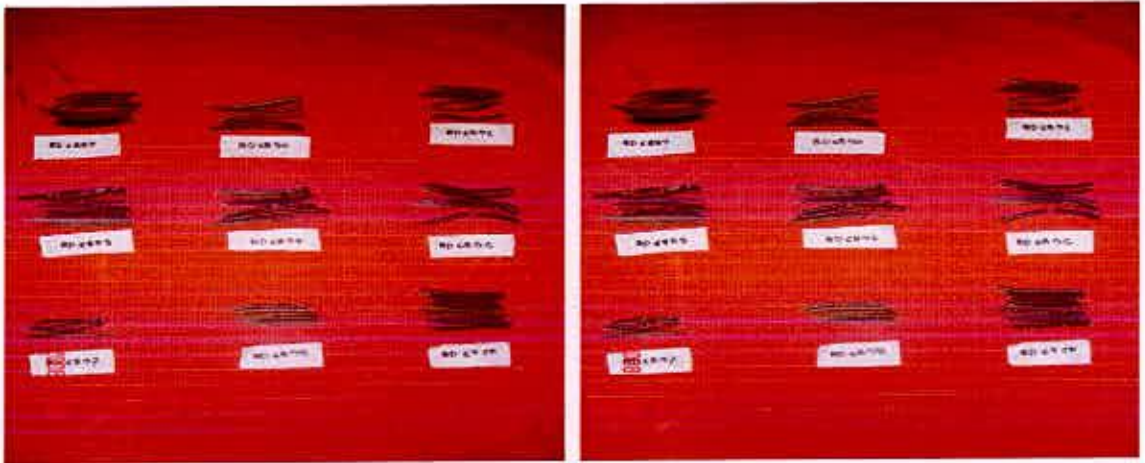
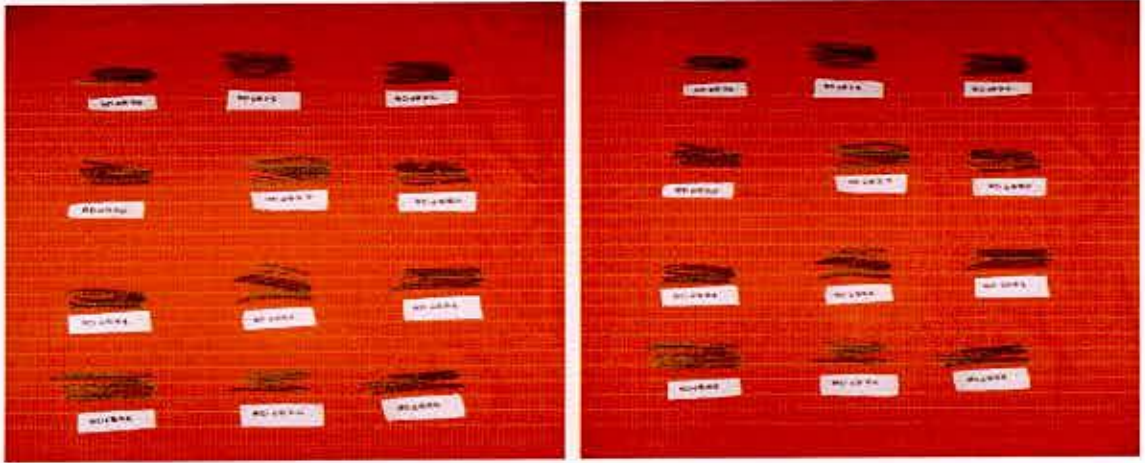


Plate 4: Continued

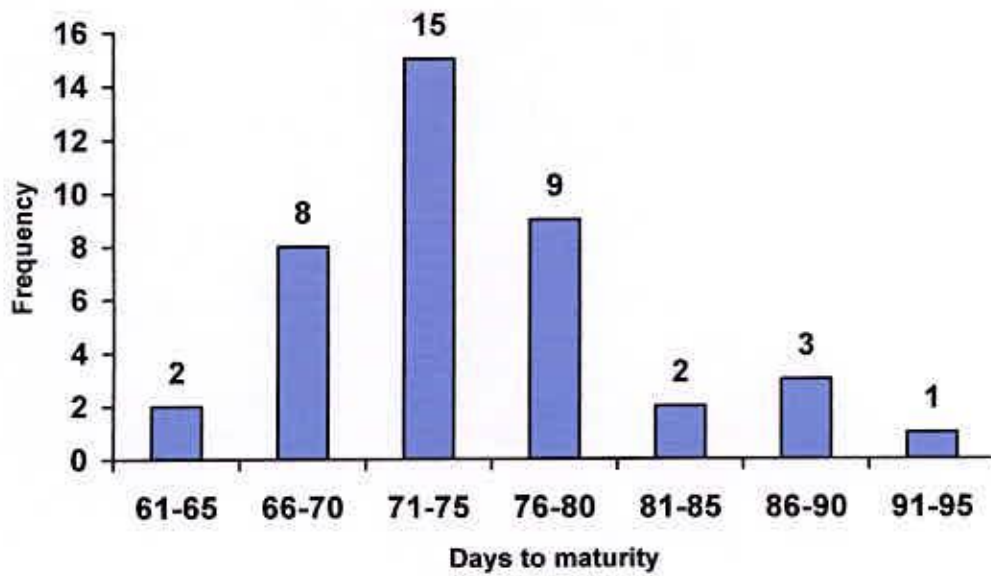


Fig. 2 : Frequency distribution for days to maturity in 40 mungbean genotype

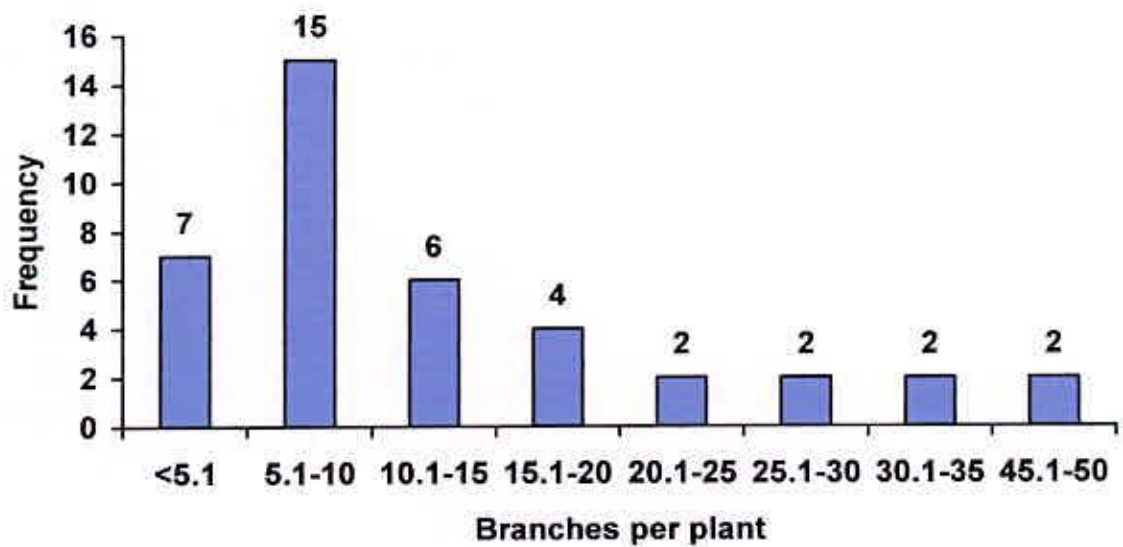


Fig. 3: Frequency distribution for branches per plant in 40 mungbean genotype

4.1.4 Pod length

Pod length ranged from 4.20-11.72 cm and the frequency distribution is presented in Fig. 5. Maximum accessions (15), which were 37.5% of the population, had 7.1 -8.0 cm pod length, while only two accessions produced long pods (>10 cm).

4.1.5 Seeds per pod

Twelve accessions which were 30 % of the population produced 12.1-14 seeds per pod and it was followed by 10 accessions with 10.1-12 seeds per pod (Fig. 6). Four accessions (10 %) were observed with maximum (14.1-16) seeds per pod.

4.1.6 100 seed weight

The frequency distribution regarding 100 seed weight as depicted in the Fig. 7 revealed that 12 accessions were having 3.1-3.5 g seed weight, followed by 7 and 5 accessions which produced 2.6-3.0 g and 3.6-4.0 g seed weight, respectively. 100-seed weight ranged from 1.88-7.75 g and only 6 accessions produced more than 5 g seed weight.

4.1.7 Grain yield per plant

Grain yield per plant ranged from 0.5-32.5 g. Frequency distribution revealed that maximum number of accessions (15) which were 37.5 % of the population, produced 5.1 to 10 g grain yield per plant (Fig 8). It was followed by the range 10.1-15 g where 7 accessions were observed. Seven accessions (17.5 %) produced more than 20 g grain yield per plant and hence were selected for higher grain yield production. These accessions are G1, G11, G14, G18, G19, G25 and G26, therefore could be utilized in improving yield potential of mungbean.



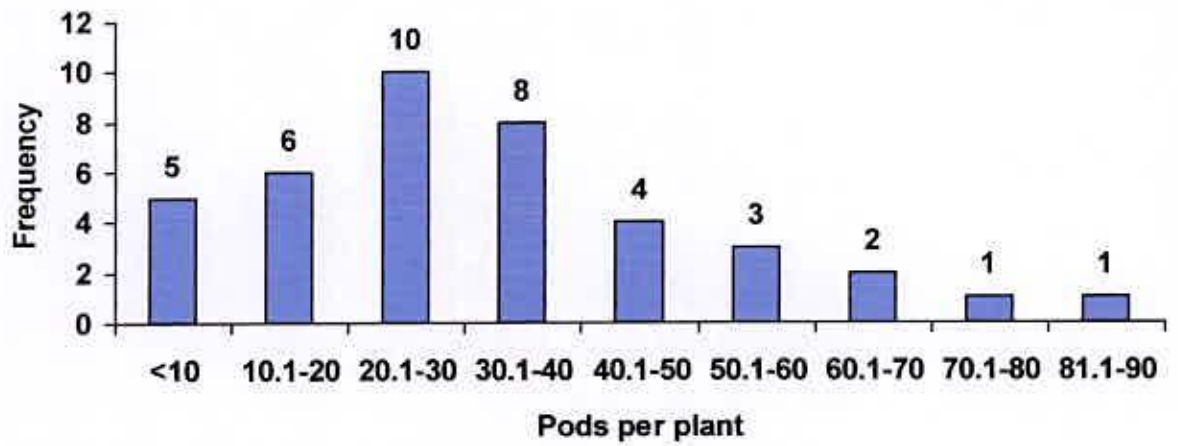


Fig. 4: Frequency distribution for pods per plant in 40 mungbean genotype

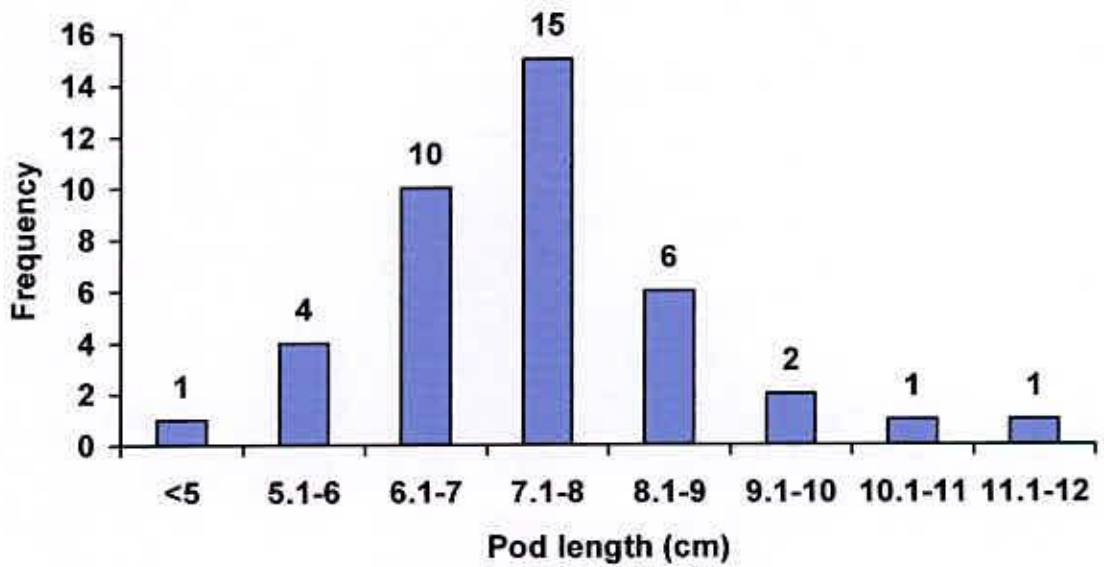


Fig. 5: Frequency distribution for pods length in 40 mungbean genotype

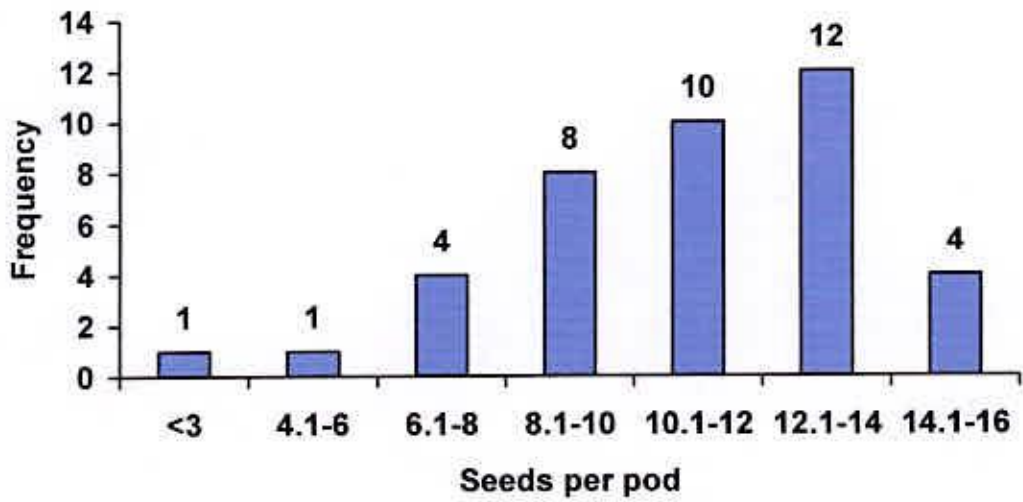


Fig. 6: Frequency distribution for seeds per pod in 40 mungbean genotype

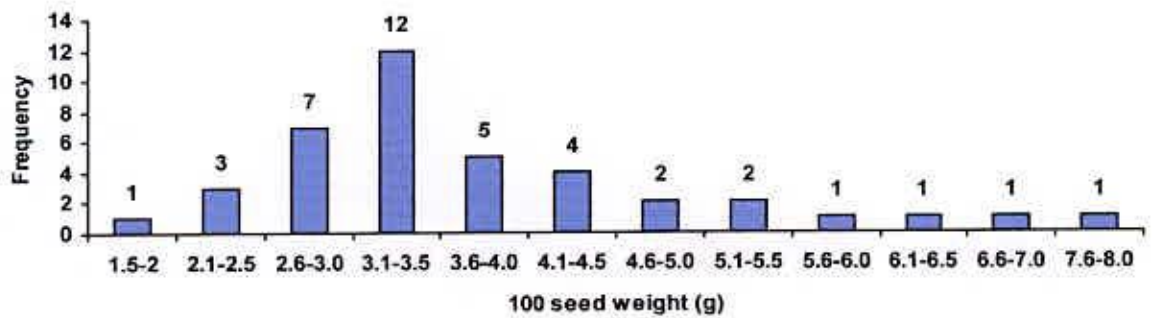


Fig. 7: Frequency distribution for 100 seed weight in 40 mungbean genotype



4.1.8 Biological yield per plant

On the basis of biological yield per plant, the germplasm ranged from 6.5145.3 g. Frequency distribution presented in Fig. 9 revealed that the maximum number of accessions (10) which were 25 % of the population produced 20.1- 40.0 g biological yield per plant. It was followed by 8 and 6 accessions with 40.1-60.0 g and 60.1-80.0 g biological yield per plant, respectively. Seven accessions produced maximum biological yield per plant (>100 g).

4.1.9 Harvest index

The frequency distribution regarding harvest index in the present mungbean germplasm (Fig. 10) revealed that maximum number of accessions (12) which were 30% of the total population, produced harvest index ranging from 30.1-40%. It was followed by 8 accessions producing 20.1-30% harvest index and this group can be considered in the medium range. Nine accessions (Table 3) gave >45% harvest index in the present study.

The important yield traits, i.e., days to maturity, branches per plant, pods per plant, seeds per pod, grain yield per plant, biological yield per plant and harvest index exhibited high range alongwith high variation which, in general revealed that the selection for these economic traits is effective in developing high yielding varieties of mungbean. Subdividing the variance into its components assists the genetic resources conservation and utilization and it enables planning for use of appropriate gene pools in crop improvement for specific plant attributes (Bekele, 1984, 1985; Pecetti, *et al.*, 1992, 1996).

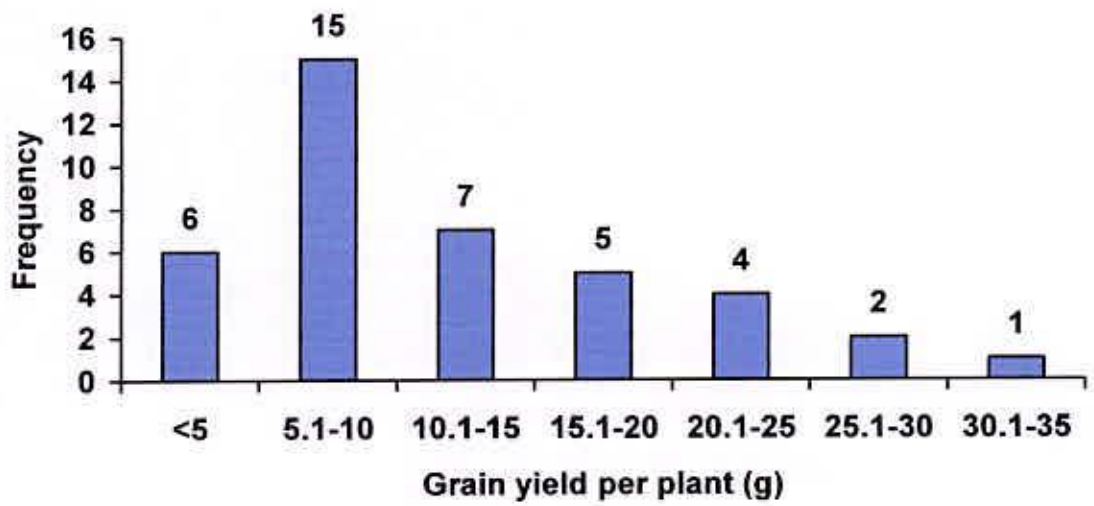


Fig. 8: Frequency distribution for grain yield per plant in 40 mungbean genotype

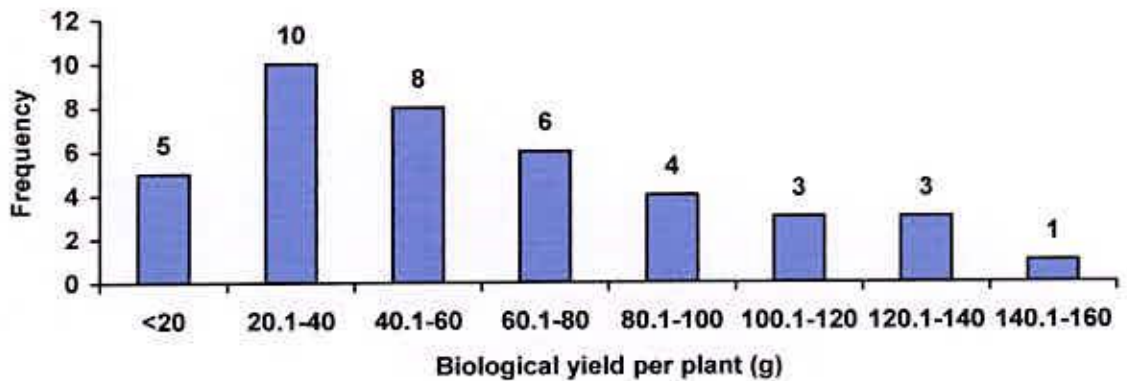


Fig. 9: Frequency distribution for Biological yield per plant in 40 mungbean genotype

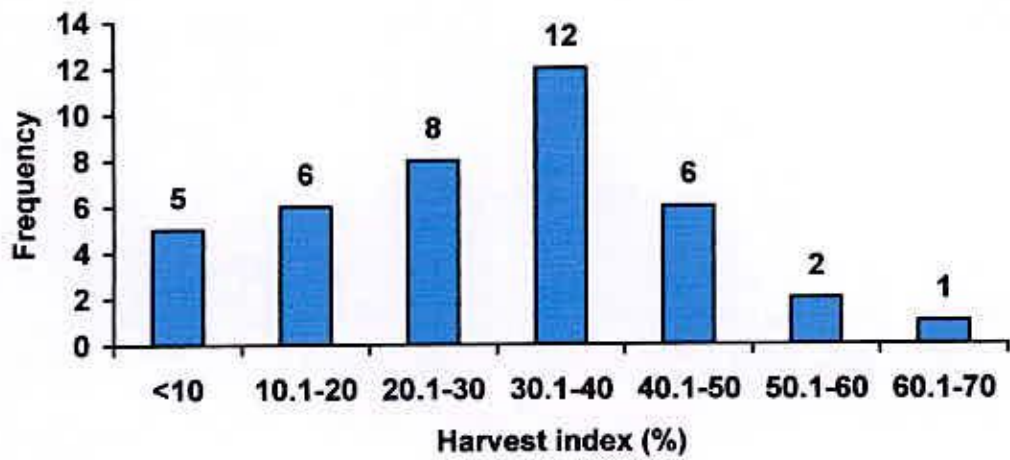


Fig. 10: Frequency distribution for harvest index in 40 mungbean genotype

The classification of present mungbean germplasm gave rise to some elite lines for specific characters and the accessions for days to maturity (G1, G2, G7, G8, G11, G18, G24, G25, G26 and G32), branches per plant (G1, G2, G3, G5, G19, G24, G32 and G34), pods per plant (G2 and G32), pod length (G3 and G24), seeds per pod (G1, G5, G14 and G19), 100-seed weight (G1, G5, G11, G18, G25 and G34), grain yield per plant (G1, G11, G14, G18, G19, G25 and G26), biological yield per plant (G3, G5, G7, 18, G24, G32 and G34) and harvest index (G2, G5, G11, G18, G19, G24, G25, G26 and G32) have been selected for exploitation in breeding programme (Table 3). It was observed that some of the accessions possessed desirable genes for more than one character and hence these could be utilized directly or included in hybrid program already been suggested by many researchers like Donald (1962), Lal (1967), Singh (1988 for varietal development. The selection on the basis of best performance has 77), Singh, *et. al.*, (1980), Khan and Malik (1989) and Ghafoor, *et. al.*, (2000).

Table 3: The selected mungbean accessions on the basis of best performance for specific characters

| Traits | Range | Name of Genotype | No. of Genotype |
|--------------------------------|--------------|--|------------------------|
| Days to maturity | <68 | G1, G2, G7, G8, G11, G18, G24, G25, G26, G32 | 10 |
| Branches per plant | >20 | G1, G2, G3, G5, G19, G24, G32, G34 | 8 |
| Pods per plant | >75 | G2, G32 | 2 |
| Pod length (cm) | >10 cm | G3, G24 | 2 |
| Seeds per pod | >14 seeds | G1, G5, G14, G19 | 4 |
| 100 seed weight (g) | >5 g | G1, G5, G11, G18, G25, G34 | 6 |
| Grain yield per plant (g) | >20 g | G1, G11, G14, G18, G19, G25, G26 | 7 |
| Biological yield per plant (g) | >100 g | G3, G5, G7, 18, G24, G32, G34 | 7 |
| Harvest index (%) | >45 % | G2, G5, G11, G18, G19, G24, G25, G26, G32 | 9 |

4. 2 Qualitative Characters

Distinct classes of three plant traits, viz., growth habit, seed coat colour and seed lustre were recorded on line basis and the tabulated results are presented in Table 4. In mungbean, Lin and Cheng (1988) reported three classes of growth habit (erect, semitrailing and trailing), two of seed coat colour (green and yellow) and two of seed surface lustre (shiny and dull). The seed colour in mungbean exhibits a wide variation for yellow, greenish yellow, light green, dark green, dull green, black, brown and mottled with black (Paroda and Thomas, 1988).

4.2.1 Growth habit

In the present study, growth habit was recorded as erect, semi-erect and spreading types. Twenty (50%) accessions were erect, twelve (30%) were semi-erect and eight (20%) were spreading types. In a study on blackgram, 59 accessions were observed as erect, 202 as semi-erect and 24 were spreading types (Ghafoor, *et. al.*, 1998)

4.2.2 Seed coat colour

Thirty four genotypes which were 85% of the population with green seed coat and only six genotypes (15%) were with black seed coat colour. The seed lustre was also observed and it was noted that twenty eight (70%) genotypes were having shiny seed lustre, twelve (30%) were dull. In mungbean germplasm, collected from 20 provinces of China, Lin and Cheng (1988) reported over 95% accessions with green seed coat. They also reported shiny and dull seed lustre, but majority of the accessions with shiny surface.

Qualitative characters are important for plant description (Kurlovich, 1998) and are mainly affected by the consumers' preference, socio-economic scenario and natural selection. Nakayama, *et. al.*, (1998) reported that foxtail millet landraces with low amylose allele were distributed only in Southeast Asia mainly because of preference followed selection. Some of these traits may be influenced by biotic or abiotic stresses. Prostrate plant type is preferred for planting under rainfed conditions as it facilitates in moisture conservation. In irrigated conditions, erect mungbean genotypes are preferred to obtain high yields by increasing number of plants per unit area. From consumer's point of view, green seed coat and shiny seed lustre is preferred over black seedcoat colour having dull seed lustre.



Table 4: Frequency distribution of qualitative traits in 40 mungbean genotypes

| Plant trait | Frequency | Percentage |
|------------------------|------------------|-------------------|
| Growth habit | | |
| Erect | 20 | 50 |
| Semi-erect | 12 | 30 |
| Spreading | 8 | 20 |
| Seed coat color | | |
| Green | 34 | 85 |
| Black | 6 | 15 |
| Seed luster | | |
| Shiny | 28 | 70 |
| Dull | 12 | 30 |

4.3 Correlation analysis

Knowledge of the relationship among plant characters is useful for selecting traits to combine for yield improvement. The correlation coefficients were computed among all the measured quantitative traits (Table 5). The results revealed that number days to maturity was significantly and positively correlated with pods per plant, whereas it had significant and negative correlation with pod length and 100-seed weight. Branches per plant had significant and positive correlation with pods per plant, grain yield per plant and biological yield per plant, while it had significant and negative association with pod length, seeds per pod and harvest index. Significant and positive association of pods per plant was observed with grain yield per plant, biological yield per plant and harvest index. Pods per plant were significant and negatively correlated with pod length and 100 seed weight. Pod length showed significantly positive correlation with seeds per pod and 100-seed weight. Seeds per pod had significantly positive association with harvest index. It revealed significantly negative association with 100 seed weight and biological yield per plant. Grain yield per plant showed significantly positive association with biological yield per plant and harvest index. Biological yield per plant, the important character in determining the yield in mungbean, exhibited significant positive correlation with branches per plant, pods per plant and grain yield per plant, whereas significant negative association with seeds per pod and harvest index.

Table 5: Correlation coefficients among nine characters in 40 mungbean genotypes

| Traits | BPP | PPP | PL | SPP | SW | GYPP | BYPP | HI |
|--------|-------|--------|---------|---------|---------|--------|---------|---------|
| DM | -0.08 | 0.15* | -0.18** | 0.03 | -0.13* | 0.07 | 0.06 | -0.11 |
| BPP | | 0.46** | -0.13* | -0.17** | 0.07 | 0.62** | 0.75** | -0.28** |
| PPP | | | -0.29** | 0.08 | -0.25** | 0.79** | 0.59** | 0.21** |
| PL | | | | 0.23** | 0.53** | -0.10 | -0.09 | 0.09 |
| SPP | | | | | -0.19** | 0.03 | -0.21** | 0.39** |
| SW | | | | | | -0.01 | 0.06 | -0.08 |
| GYPP | | | | | | | 0.78** | 0.18** |
| BYPP | | | | | | | | -0.37 |

** and * = Significant at 1% and 5% probability level respectively

DM = Days to maturity, BPP = Branches per plant, PPP = Pods per plant, PL = Pod length, SPP = Seeds per pods, SW = 100 seed weight, GYPP = Grain yield per plant, BYPP = Biological yield per plant and HI = Harvest index

The correlation is a measure of the degree to which variables vary together or a measure of intensity of association (Steell and Torrie, 1980). Generally, a high magnitude of correlation with positive signs was observed between different traits.

In mungbean, Tomar, *et. al.*, (1973) and Khalid, *et. al.*, (1984) observed positive correlation of yield with yield components, whereas, Malik, *et. al.*, (1987) reported negative correlation of yield with maturity, pod length and seed weight. Significant positive correlation of grain yield with other yield contributing characters has been reported by Rani and Rao, (1981) in blackgram. Malik, *et. al.*, (1983) investigated maximum relative selection efficiency for branches per plant in mungbean, and Malhorta, *et. al.*, (1974) observed positive association of yield with days to maturity, plant height, pods and pod length, whereas, negative with seed weight. In the present material, significant positive correlation of grain yield with branches per plant, pods per plant, biological yield and harvest index revealed that these characters are really yield contributing characters. Malik, *et. al.*, (1987) and Ghafoor, *et. al.*, (1993b) reported positive association of grain yield with biological yield. Negative association of biological yield with harvest index showed physiological inefficiency for appropriate partitioning of total dry matter towards economic yield, consequently the accessions with low grain yield attained low harvest index.



Plate 5: Photograph showing the different type of seeds of mungbean genotypes



Plate 5: Continued

4. 4 Cluster analysis

Forty mungbean genotypes were grouped into six clusters. Members of each cluster are given in Table 6. The distribution pattern indicated that the maximum number of genotypes were included in cluster IV (13), while cluster II, V and VI included same number of genotypes (4) and cluster I and III included 8 and 7 genotypes respectively.

Genetic diversity is generally associated with geographical diversity but the former is not necessarily directly related with geographical distribution. The genotypes with in the same cluster although formed specific clusters but were originated from different geographic regions of the world. This indicated that the geographical and genetic distribution did not follow the same trend. Non corresponding genetic diversity and geographic distribution were also reported earlier by Shunmugam, *et. al.*, (1982).

Euclidean dissimilarity coefficients among six clusters are presented in Table 7. Maximum euclidean distance (39.62) was observed between clusters IV and V followed by the distance (38.64) between clusters V and VI. Minimum distances were observed for clusters III and VI (6.53), and for clusters IV and VI (6.60). These indicate that the genotypes in cluster IV were far diversified from those of cluster V and those in cluster III were somewhat close to cluster VI. The genotypes belonging to the distant cluster could be used in hybridization program for obtaining a wide spectrum of variation among the segregant.



Table 6: Distribution of 40 genotypes of mungbean in six clusters

| Cluster no | No. of genotypes | Genotypes |
|------------|------------------|--|
| I | 8 | G3, G6, G24, G29, G31, G32, G34 and G36 |
| II | 4 | G1, G14, G15, and G19 |
| III | 7 | G12, G13, G23, G28, G35, G39 and G40 |
| IV | 13 | G4, G7, G8, G11, G16, G17, G25, G26, G27, G30, G33, G37, and G38 |
| V | 4 | G2, G5, G9 and G10 |
| VI | 4 | G18, G20, G21 and G22 |

Table 7: Euclidean distances among 6 clusters based on nine characters among 40 genotypes of mungbean

| | Cluster II | Cluster III | Cluster IV | Cluster IV | Cluster VI |
|-------------|------------|-------------|------------|------------|------------|
| Cluster I | 14.35 | 10.14 | 20.17 | 28.63 | 16.08 |
| Cluster II | | 16.21 | 22.95 | 17.03 | 21.82 |
| Cluster III | | | 10.22 | 32.99 | 6.53 |
| Cluster IV | | | | 39.62 | 6.60 |
| Cluster V | | | | | 38.64 |



Mean, standard error and variance computed for 6 clusters are presented in Table 8. In the cluster I, the accessions gave the mean value for days to maturity (76.05 ± 1.02), pods per plant (66.27 ± 2.09), grain yield per plant (17.38 ± 0.59), biological yield per plant (45.21 ± 1.50) and harvest index (38.97 ± 1.02), therefore classified as late maturing with medium grain yield. Due to high variance, there is a scope of selection for maturity in this group. Cluster II is categorized as medium maturing (73.91 ± 1.43) with medium grain yield (18.13 ± 1.31). Cluster III comprised of late maturing group (75.21 ± 0.59) with an average grain yield (11.1 ± 0.26). In the cluster IV, in general the accessions were medium maturing (73.89 ± 1.02) with low grain yield per plant (4.29 ± 4.32), therefore, were classified as medium maturing with lower grain yield. The accessions in cluster IV had the highest mean value for 100 seed weight (3.78 ± 0.25), hence can be exploited in breeding bold seeded mungbean varieties. Cluster V exhibited highest average for branches per plant (22.68 ± 4.16), grain yield per plant (24.15 ± 1.94), biological yield per plant (127.15 ± 3.66) and a high number of pods (62.66 ± 3.51), while it showed lowest mean for days to maturity (72.22 ± 2.36). This cluster is therefore classified as early maturing with high grain yield potential. The accessions in cluster VI were also early maturing (72.63 ± 10.34), but due to low grain yield average (7.79 ± 0.18), were considered as low yielding with early maturity. The grouping of accessions by multivariate methods in this study is of practical value to mungbean breeders. Representative accessions may be chosen from particular groups for hybrid programme with other approved varieties. Several potentially important agronomic

types have been identified and these may be exploited for genetic potential to transfer the desirable genes and this facilitates the assembly of a core collection of accessions from the large genetic resources collection (Tolbert, *et. al.*, 1979; Frankel, 1984; Singh, 1988; Clements and Cowling, 1994). Tawar, *et. al.* (1988) conducted genetic divergence in 34 diverse genotypes of mungbean which were grouped in five clusters, and they observed that variability in the parents was related to genetic diversity. Inclusion of such genotypes from distinct clusters and their implication in mungbean breeding programme was suggested.

Table 8: Mean, standard error and variance for six clusters based on nine characters of 40 mungbean genotypes

| | Cluster I | | Cluster II | | Cluster III | | Cluster IV | | Cluster V | | Cluster VI | |
|-----------------------------------|------------|----------|------------|----------|-------------|----------|------------|----------|-------------|----------|------------|----------|
| | Mean | Variance | Mean | Variance | Mean | Variance | Mean | Variance | Mean | Variance | Mean | Variance |
| Days to maturity | 76.05±1.02 | 40.58 | 73.91±1.43 | 46.81 | 75.21±0.59 | 23.22 | 73.89±1.02 | 36.22 | 72.22±2.36 | 49.94 | 72.63±0.34 | 10.59 |
| Branches per plant | 8.97±0.49 | 9.26 | 18.86±1.91 | 84.06 | 7.39±0.32 | 6.67 | 5.72±0.28 | 2.83 | 22.68±4.16 | 155.76 | 5.45±0.16 | 2.40 |
| Pods per plant | 66.27±2.09 | 170.29 | 47.52±3.07 | 216.33 | 39.25±0.97 | 62.17 | 15.75±1.03 | 36.8 | 62.66±3.51 | 110.78 | 25.76±0.55 | 27.00 |
| Pod length | 7.20±0.10 | 0.41 | 7.44±0.16 | 0.59 | 7.55±0.11 | 0.84 | 7.58±0.23 | 1.79 | 7.39±0.15 | 0.20 | 7.87±0.10 | 0.93 |
| Seeds per pod | 12.83±0.26 | 2.54 | 11.31±0.40 | 3.76 | 13.02±0.11 | 0.85 | 11.45±0.34 | 4.13 | 11.07±0.79 | 5.63 | 12.56±0.14 | 1.75 |
| 100 seed weight | 3.04±0.09 | 0.31 | 3.63±0.11 | 0.26 | 3.26±0.08 | 0.41 | 3.78±0.25 | 2.25 | 3.76±0.13 | 0.14 | 3.59±0.07 | 0.50 |
| Grain yield per plant | 17.38±0.59 | 13.67 | 18.13±1.31 | 39.45 | 11.10±0.26 | 4.32 | 4.29±0.32 | 3.68 | 24.15±1.94 | 34.03 | 7.79±0.18 | 2.87 |
| Biological yield per plant | 45.21±1.5 | 87.24 | 79.09±2.31 | 122.46 | 34.26±0.80 | 42.12 | 21.14±1.22 | 52.28 | 127.15±3.66 | 120.45 | 21.41±0.54 | 26.06 |
| Harvest index (%) | 38.97±1.02 | 40.61 | 23.17±1.58 | 57.22 | 33.28±0.90 | 53.15 | 20.51±1.26 | 55.52 | 19.07±1.53 | 21.18 | 37.13±0.67 | 40.22 |

4.5 Principal component analysis

Variance was further studied by principal component analysis (PCA). Eigenvalues of nine principal components have been shown in the screw plot (Fig. 11). A principal component matrix is given in Table 9 which revealed that the first three components with eigenvalues >1 contributed 71.47% of the variability amongst 40 accessions of mungbean evaluated for nine quantitative traits. In 30 genotypes of chickpea, first three PCs with eigenvalue >1 contributed 83.3% variability (Ghafoor, *et. al.*, 2003a). Ghafoor, *et. al.* (2003b) analysed 22 blackgram genotypes for two consecutive years using multivariate analyses. They reported that first four PCs with eigenvalue > 1 contributed 80.0% variation during 1998 and 80.9% during 1999.

Principal component 1 possessed 34.36%, PC₂ 19.78% and PC₃ had 17.34% of the total variation. The characters that contributed more positively to PC₁, were branches per plant (0.809), pods per plant (0.811), grain yield per plant (0.888) and biological yield per plant (0.905), whereas days to maturity, pod length, seeds per pod, 100 seed weight and harvest index contributed negatively to first component. This means that the populations with high PC₁ values are high yielding and formed by late maturing plants characterized by high number of branches with more pods, but having smaller pod length, lesser seeds per pod and low seed weight. The populations in this component are also associated negatively with harvest index which revealed that the accessions in the PC₁ failed in appropriate partitioning of economic yield which ultimately reduced harvest index. Days to maturity (0.521), seeds per pod (0.594) and harvest index (0.670)

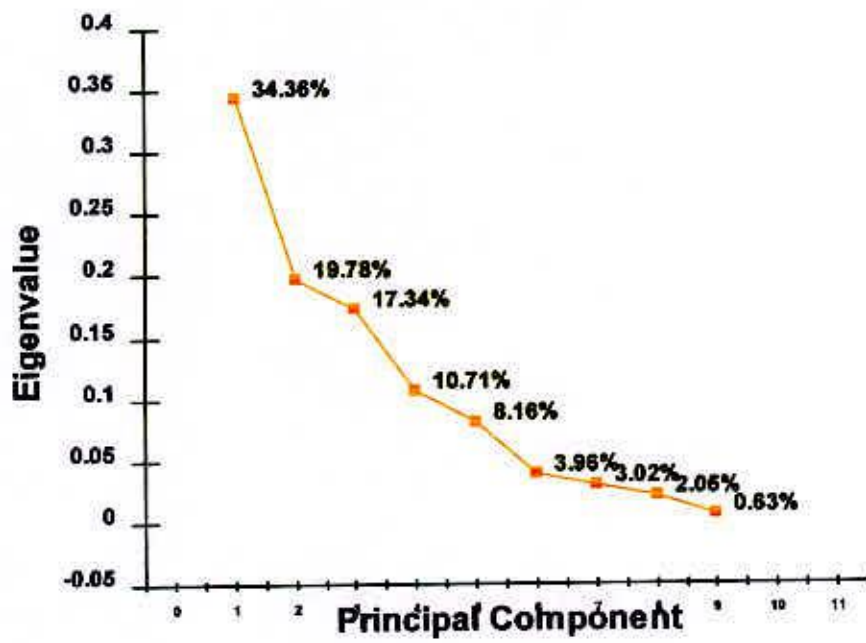


Fig. 11: Eigenvalues alongwith nine principal component

contributed maximum genetic variance to PC₂. The second component was strongly associated with earliness, more number of seeds and high harvest index and moderately with pods per plant. Eight characters contributed positively for PC₃, whereas three characters (pod length, 100 seed weight and harvest index) were observed with highest values for this component. Seeds per pod had a moderate value for third component. It is evident that all the quantitative characters under study contributed more positively to first three principal components and hence these could be given considerable importance for the genetic material under investigation. Results reported by various researchers [Holcomb, *et. al.*, (1977), Ramana and Singh, (1987), Falcinelli, *et. al.*, (1988), Veronesi and Falcinelli, (1988); Brown, (1991) and Humphreys, (1991), Elizabeth, *et. al.*, (2001); Ghafoor, *et. al.*, (2002) and Upadhyaya, (2003)] showed multivariate analyses to be a valid system to deal with germplasm collection. Ghafoor, *et. al.*, (2001) categorized five clusters based on average linkage for 484 blackgram accessions and observed more clear separation of clusters II, III and IV than clusters I and V after first two PCs plotted in a scattered diagram.



Table 9: Principle components (PCs) for nine characters in 40 mungbean genotypes

| | | PC₁ | PC₂ | PC₃ |
|-----------------------------------|----------------------|-----------------------|-----------------------|-----------------------|
| Eigen value | | 3.092 | 1.780 | 1.561 |
| Proportion | | 34.356 | 19.776 | 17.339 |
| Cumulative | | 34.356 | 54.133 | 71.472 |
| | Communalities | | Eigen vectors | |
| Days to maturity | 0.498 | -0.009 | 0.521 | -0.367 |
| Branches per plant | 0.744 | 0.809 | -0.291 | 0.067 |
| Pods per plant | 0.844 | 0.811 | 0.414 | 0.118 |
| Pod length | 0.767 | -0.308 | -0.348 | 0.742 |
| Seeds per pod | 7.588 | -0.160 | 0.594 | 0.457 |
| 100 seed weight | 0.730 | -0.112 | -0.668 | 0.520 |
| Grain yield per plant | 0.920 | 0.888 | 0.160 | 0.324 |
| Biological yield per plant | 0.898 | 0.905 | -0.276 | 0.051 |
| Harvest index (%) | 0.745 | -0.156 | 0.670 | 0.518 |



Plate 6: Genotype bearing moderate number of pods



Plate 7: Genotype bearing minimum number of pods



Plate 8: Genotype bearing maximum number of pods



Chapter V

Summary and Conclusion

CHAPTER V SUMMARY AND CONCLUSION

Research reported in this manuscript was conducted on mungbean at Sher-e-Bangla Agricultural University (SAU), Dhaka-1207 to estimate (a) genetic diversity based on morphological traits analysis (b) to study the nature of correlation of grain yield and its components. The ultimate aim was to identify genotypes to be utilized in mungbean breeding for the improvement of economic traits.

Forty mungbean genotypes were evaluated for various agro-morphological traits under field condition at SAU farm. Data were recorded on nine quantitative traits (days to maturity, branches per plant, pods per plant, pod length, seeds per pod, 100 seed weight, grain yield per plant, biological yield per plant and harvest index) and three qualitative traits (growth habit, seed coat colour and seed lustre).

High variance was observed for days to maturity, branches per plant, pods per plant, seeds per pod, grain yield per plant, biological yield per plant and harvest index. Results of association analysis revealed that grain yield per plant was significantly and positively correlated with branches per plant, pods per plant, biological yield per plant and harvest index. Negative correlation of biological yield with harvest index showed physiological inefficiency for appropriate partitioning of total dry matter towards economic yield, consequently the accessions with low grain yield attained low harvest index.

All the mungbean genotypes under study were grouped into six clusters on

average linkage basis. As the number of accessions from various sources differed considerably, it was difficult to establish any relationship between origin and clustering pattern. Variance was further studied by principal component analysis (PCA). First three principal components (PCs) with eigenvalues >1 contributed 71.47% of the variability amongst the genotypes. The populations with high PC₁ values were high yielding, late maturing and were characterized by high number of branches with more pods, but having smaller pod length, lesser seeds per pod and lower seed weight. The second component was strongly associated with earliness, more number of seeds and higher harvest index, and moderately with pods per plant. Eight characters contributed positively for PC₃, whereas three characters (pod length, 100 seed weight and harvest index) were observed with highest values for this component. It is evident that all the quantitative characters under study contributed more positively to first three principal components and hence these could be given considerable importance for the genetic material under investigation.

Seed coat colour and seed lustre were observed into two distinct classes. It was observed that in general the genotypes having green seed coat colour facilitated early maturity, while those having black seed coat were late in maturity. All other quantitative characters exhibited increasing effect with the presence of genes responsible for green seed coat which revealed that these genes had association in increasing yield and yield components. Similarly, shiny lustre genes had increasing effect for most of the yield contributing characters. In the present study, hence the green seed coat and shiny seed lustre characters, provide their worth in selection for improving most of the yield contributing traits.



The germplasm under investigation displayed a wide range of diversity for most of the traits. Some accessions possessed unique characters which could help to identify landraces with suitable traits to be used in hybridization programme to broaden the genetic base.

The genotypes for days to maturity (10), branches per plant (8), pods per plant (2), pod length (6), seeds per pod (4), 100-seed weight (6), grain yield per plant (7), biological yield per plant (7) and harvest index (9) were earmarked for use in the breeding programme. It was observed that some of these selected accessions possessed desirable genes for more than one character and hence these could be utilized directly or included in hybridization programme for mungbean varietal development.



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Appendices



Appendix I. Morphological, Physical and chemical characteristics of initial soil (0-15 cm depth)

A. Physical composition of soil

| Soil separates | % | Method employed |
|----------------|-----------|----------------------------------|
| Sand | 36.90 | Hydrometer method (Day, 1915) |
| Silt | 26.40 | Do |
| Clay | 36.66 | Do |
| Texture class | Clay loam | Do |

B. Chemical Composition of the soil

| Sl. No. | Soil characteristics | Analytical data | Methods employed |
|---------|--------------------------|-----------------|----------------------------|
| 1 | Organic carbon (%) | 0.82 | Walkley and Black, 1947 |
| 2 | Total N (kg/ha) | 1790.00 | Bremner and Mulvancy, 1965 |
| 3 | Total S (ppm) | 225.00 | Bardsle and Lanester, 1965 |
| 4 | Total P (ppm) | 840.00 | Olsen and Sommers, 1982 |
| 5 | Available N (kg/ha) | 54.00 | Bremner, 1965 |
| 6 | Available P (kg/ha) | 69.00 | Olsen and Dean, 1965 |
| 7 | Exchangeable K (kg/ha) | 89.00 | Pratt, 1965 |
| 8 | Available S (kg/ha) | 16.00 | Hunter, 1984 |
| 9 | Ph (1:2.5 soil to water) | 5.55 | Jackson, 1958 |
| 10 | CEC | 11.23 | Chapman, 1965 |

Appendix II. Monthly record of air temperature, relative humidity and rainfall of experimental site during the period from November 2007 to May 2008

| Month | Year | *Air temperature (°c) | | Relative Humidity (%) at 12 p.m. | ** Rainfall (mm) |
|----------|------|-----------------------|---------|-------------------------------------|------------------|
| | | Maximum | Minimum | | |
| November | 2007 | 29.07 | 18.80 | 65.13 | 0 |
| December | 2007 | 27.07 | 15.65 | 63.80 | 3 |
| January | 2008 | 24.76 | 13.46 | 69.53 | 0 |
| February | 2008 | 31.26 | 19.42 | 51.27 | 0 |
| March | 2008 | 33.20 | 22.00 | 46.13 | 0 |
| April | 2008 | 33.74 | 23.81 | 61.40 | 185 |
| May | 2008 | 33.66 | 24.95 | 46.27 | 180 |

* Monthly average

** Monthly total

Source: Bangladesh Meteorological Department (Climate Division) Agragoan, Dhaka-1212.

গণপ্রজাতন্ত্রী বাংলাদেশ সরকার
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 ১০/১২/১৫

