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## DIVERSITY ANALYSIS AND ESTABLISHMENT OF CORE SUBSETS OF HYACINTH BEAN COLLECTION OF BANGLADESH

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**ABSTRACT:-** Plant Genetic Resource Centre, Bangladesh Agricultural Research Institute, Gazipur, Bangladesh, maintained 484 hyacinth bean (*Lablab purpureus* L.) accessions. Distribution of vine colors, flower and pod colors, pod curvature and beak position on pod noticed the presence of substantial variation in the collection. Shannon Diversity Index also revealed high phenotypic diversity in vine and pod characters. High diversities were estimated in quantitative characters of inflorescence and pod characters. Considering a core collection strategy, the accessions were stratified into two groups based on vine colors: green and purple. Maximum genotypes of the collection were of green vine. Each of two subgroups were divided into three groups according to colors of pod i.e., green, white and red. Each group again divided itself into two according to beak position as central and marginal of pod. High diversity was also found regarding pod curvature and beak position on it. Strategically, a core subset consisting of 36 accessions out of 484 accessions was determined. Diversity Indices of different characters of the core accessions were of equal or higher magnitudes to the respective characters of base collection indicating the better representation of core to the original collection.

*Key Words: Lablab purpureus; Core Subset; Diversity Index; Agronomic Characters; Bangladesh*

### INTRODUCTION

Hyacinth bean (*Lablab purpureus*) under the family Fabaceae is an important winter vegetable in Bangladesh. The crop has a high nutritive value and is popular among people. Scanty of suitable varieties as well as incidence of pest and disease are the major yield limiting factors of hyacinth bean in Bangladesh. Moreover, many biotic and abiotic stresses are affecting the crop continuously due to ongoing changes in temperatures and

precipitation. Screening and identification of resistant germplasm is fundamental and continuous process for sustainable production (Tanksley and Mc Couch, 1997). However, considerable variations noticed in hyacinth bean genotypes of Bangladesh need to accumulate in the gene bank for characterization and utilization to alleviate all kinds of stresses. To improve hyacinth bean, Plant Genetic Resource Centre (PGRC), Bangladesh Agricultural Research Institute (BARI) collected germplasm of 484

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country bean from home and abroad (Islam et al., 2002 and 2004). Redundant and duplicate accessions within the collection cause difficulty in management and utilization of genetic materials. Use of subset, termed as core collection, increases the efficiency of describing and utilization of collection (Frankel and Brown, 1984; Brown, 1989; 1995). Therefore, the study was undertaken to characterize the bean germplasm and forming a core subset for effective utilization and conservation.

### MATERIALS AND METHOD

The experiment was conducted at the Regional Agricultural Research Station, Jessore during *rabi* season, 2006-07. Seed sample were supplied by the Plant Genetic Resource Centre (PGRC) of the Bangladesh Agricultural Research Institute (BARI), Gazipur. Four hundred eighty four (484) accessions were used as base collection in the experiment. Two plants of each accession were planted in continuous 60m long rows in 8 plots without any replication. Plant to plant distance within accession was 1m and between the accessions was 2m. The distance between two rows was 3m. Irrigation, fertilization and other cultural management were done as and when required. Data on different qualitative and quantitative characters were recorded as stated by IBPGR, 1982) and [www.lablalab.org/attachments/Dolichos%20 Report.pdf](http://www.lablalab.org/attachments/Dolichos%20Report.pdf) (2007). The collected data were summarized and phenotypic diversity for both qualitative and quantitative traits was determined by using Shannon-Weaver Diversity Index (H'). H' ranges from 0 to 1, where 1 indicates

the maximum diversity. H' is determined as:  $H' = -\sum P_i \log P_i$ , where  $P_i$  is the proportion of the total number of genotypes belonging to the  $i^{\text{th}}$  class.

The exact descriptor states, defined the classes for the qualitative characters while the classes for quantitative characters are defined according to the procedure suggested by Yu Li et al. (1996) where each H' value was standardized. The overall genotypes mean ( $\bar{X}$ ) and standard deviation ( $\delta$ ) were used to subdivide the population values (xi) into 10 frequency classes ranging from class (1 - 2) to class 10 ( $\bar{X} + 2\sigma$ ), the class interval being 0.50. The relative frequencies for the different classes were used to calculate the diversity index. The H' for each character was calculated using MS Excel.

The entire collection was stratified based on stem color. Each group was then classified into subgroups according to pod colors. Each subgroup was again split into smaller groups according to pod curvature, which were further divided according to orientation of pod beak. Considering the core collection strategy multivariate analysis of each smallest group was carried out that partitioned accessions in various groups (Hu et al., 2000; van Hintum et al., 1995). Accessions of each group were chosen for core candidate, using a predetermined sampling strategy (Diwan et al., 1995). Thus, a core subset of 36 accessions was built up based on stratification of qualitative characters and multivariate analysis of quantitative traits. In the core subset, 19 genotypes came from accessions of green vine and 16 from accessions of purple or mixed colored vines (Table 1). Multivariate analysis,

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**Table 1. Core candidates of genotypes**

Stem color	Pod color	Pod curvature with beak direction	Intra-cluster distances	Total genotypes	Nature of diversity within individual	Core candidate
Green	Green	C3B1	47.01	146	Uniform	BD-51, BD-1772, BD-8012, BD-8862, BD-62
			40.17			
			43.00			
		C3B2	21.02	34	Moderate	BD-11, BD-8037, BD-8787
			21.04			
			12.28			
		C5B1	23.20	17	Uniform	BD-34, BD2901, BD-8033
			23.39			
			20.49			
			20.81			
		C5B2	27.57	9	Uniform	BD-103
			-			
		C7B1	39.55	63	Diverged	BD 2881, BD-2915, BD-8876
			54.80			
26.68						
C7B2	0	16	Uniform	BD-7991		
	6.6					
Sub-total			9.78	285		16
	White	C3B1	0	9	Uniform	BD-05
			C7B1	0	2	Uniform
Sub-total				11		2
	Red	C3B1	0	2	Uniform	BD-1797
Sub-total				2		1
<b>Total</b>				<b>298</b>		<b>19</b>
Purple	Green	C3B1	37.74	92	Uniform	BD-47, BD8006, BD-8778, BD-8870
			32.88			
			36.52			
			38.12			
		C3B2	0	8		BD-61
		C5B1	47.12	32	Uniform	BD-8833
			48.99			
		C5B2	0	8		BD-150
		C7B1	34.68	11	Moderate	BD-2918
			31.30			
42.25						
				6		BD-8039
				14		BD-71

(Contd.)

Stem color	Pod color	Pod curvature with beak direction	Intra-cluster distances	Total genotypes	Nature of diversity within individual	Core candidate
Purple		C7B2	0	3		BD-8052
Sub-total				174		
	White	C3B1	0	4	Uniform	BD-8868
		C5B1	0	2	Uniform	BD-1786
		C7B1	0	2	Uniform	BD-2913
Sub-total				8		
	Red	C3B1	0	3		BD-1829, BD-8839
		C7B1		1		BD-2882
Sub-total				4		
<b>Total</b>				<b>186</b>		<b>17.00</b>
<b>Grand total</b>				<b>484</b>		<b>36.00</b>
Core percent= [% Core candidates = (484 x 36)/100]						10.12

C3 = Straight pod; C5 = Slight curved pod; C7 = Curved pod; B1 = Central Beak; B2 = Marginal Beak

including principal component analysis (PCA) and clustering were done to assess the pattern of morphological variation within the core subset. Data analysis was carried out using the software SPSS 11.0. Descriptive statistics,  $H'$  and  $\chi^2$  test were used to analyze to test homogeneity of qualitative and quantitative characters of core subset and base collection.

## RESULTS AND DISCUSSION

Green and mixed-purple colored vines were observed in the collection. The distribution of vine colors was statistically different from their expected number of an equal distribution (242). Genotype with green vine were dominant over purple genotypes (Table 2). Sultana (2001) similarly reported domination of green vine genotypes over purple or mixed colored genotypes among 107 hya-

cinth beans germplasm. Islam et al. (2002) also reported two types of vine colors in a collection of 44 hyacinth bean collection where green dominated over purple.

Four distinct flower colors, deep violet, pale violet, violet and white were recorded in the original collection, which distributed independently. Violet and whitish flowered genotypes distributed uniformly but dominated strongly over deep violet and pale violet flowers. Similar pattern of distribution of flower colors was retained on the core subset (Table 2). Lack of dominant/recessive relationship would be argued of independent distribution of flower colors in collection.

Green, whitish and reddish color pods were recorded in the collection. Maximum genotypes produced green pod which strongly dominated over whitish and reddish pod. Dominating

**Table 2. Distribution of qualitative characters of base collection and core sub-set**

Trait	Observed phenotype	Accessions					
		Base collection			Core sub-set		
		Observed number	Expected number	$\chi^2$ value	Observed number	Expected number	$\chi^2$ value
Stem color	Green	298	242	12**	19	18	0.19 <sup>ns</sup>
	Purple	186	242		17	18	
Flower color	Deep violet	19	121	321**	2	9	11.00**
	Pale violet	32	121		4	9	
	Violet	221	121		14	9	
	Whitish	212	121		16	9	
Pod color	Green	459	161	824**	22	12	14.00**
	Whitish	19	161		4	12	
	Reddish	6	161		10	12	
Pod curvature	Straight	70	159	167**	8	12	6.17*
	Slightly curved	287	159		19	12	
	Curved	119	159		9	12	

\*&\*\* = Significant at 5%, and 1% level of probability, respectively; ns = Non significant

of green pod genotypes also found in core subset (Table 2). Sultana (2001) and Islam et al. (2010) also reported the dominance of green pod of hyacinth bean in their studies.

Pigmentation in plant includes wide varieties of defense related compounds to protect foliage damaging from ultraviolet rays as well as pathogens and insect (Freeman and Beattie, 2008). Colorful flower is an important attributes of plants that attract pollinators and facilitates cross-pollination. Cross pollination is a natural source of evolutionary process and enables plants to adapt under changing environment. Pigmentation also

enables plant to absorb light of various wavelengths which pass through leaves and used for maximizing photosynthesis (Alejar et al., 1999). Beans need more energy for accumulating proteins in the seed and pigmentation thus assisting in adaptation process under ongoing changes of climate. However, the pod in bean is economically important part and it has undergone intensive selection by man. Domination of green color might be due result of such selection process.

Three types of pod curvature e.g., straight, slightly curved and curved were recorded in the collection. Genotypes producing slightly curved pod

were maximum and dominating over other types. Similar distribution pattern was retained in the core subset (Table 2). This result is in concurrence of the report of Islam et al. (2010). Richness of particular characters of pod might be due to the needs of farmer, market demand and the influence of environment where the crops grow.

### Diversity Indices

#### Qualitative Characters

Quantification of variability of stem color, flower color and pod curvature using Shannon Weaver Diversity Indices showed high variation in the original collection which were retain almost in core subset too.

**Table 3. Shannon Diversity Indices of qualitative characters of base collection and core subset of hyacinth bean**

Trait	Phenotype	Shannon Weaver Diversity Indices (SWDI)	
		Base collection	Core collection
Stem color	Green	99 (H)	61 (M)
	Reddish		
Flower color	Deep violet	90 (H)	97 (H)
	Pale violet		
	Violet		
	White		
Pod color	Green	21 (L)	81 (H)
	White		
	Red		
Pod curvature	Straight	85 (H)	85 (H)
	Slightly		
	Curved		

H= High, M= Moderate, L=Low

However, diversity indices of stem color was found to slip down in core subset, but sharply increased in pod color (Table 3).

#### Quantitative Characters

Among the 16 characters studied, estimated Diversity Indices  $H'$  in the original collection were moderate to high. In the core subset  $H'$  values of 6 characters were found to improve from moderate to high (Table 4). Increasing trend of  $H'$  values of core subset are indicating the presence of duplicate and similarity of genotypes in original collection. Core subset is quite sufficient to represent the entire genetic spectrum of original collection.

#### Principal Component Analysis

In base collection, first six components accounted for 64.26% of total variation while the first six component of core subset contribution was 73.74%. This relationship is an indication of more linear relationship in genotypes of core subset (Table 5). Linear relationship occurs when the genotypes are diverged to each other. Elimination of redundant and duplicate accession in core has been observed by Mosjidis and Klingler (2006). Mc Khann et al. (2004) also reported maximizing of genetic diversity of core collection in *Arabidopsis thaliana*. Heterosis can be exploited by using the genetically diverged parents of subset (Arunachalam et al., 1984 and Rammanujam et al., 1974).

#### Clustering of Core Subset

Seven variables of core subset were selected based on principal component of variation subjected to Un-weighted Paired Group Method



**Table 4. Diversity Indices of quantitative traits of base collection of hyacinth bean**

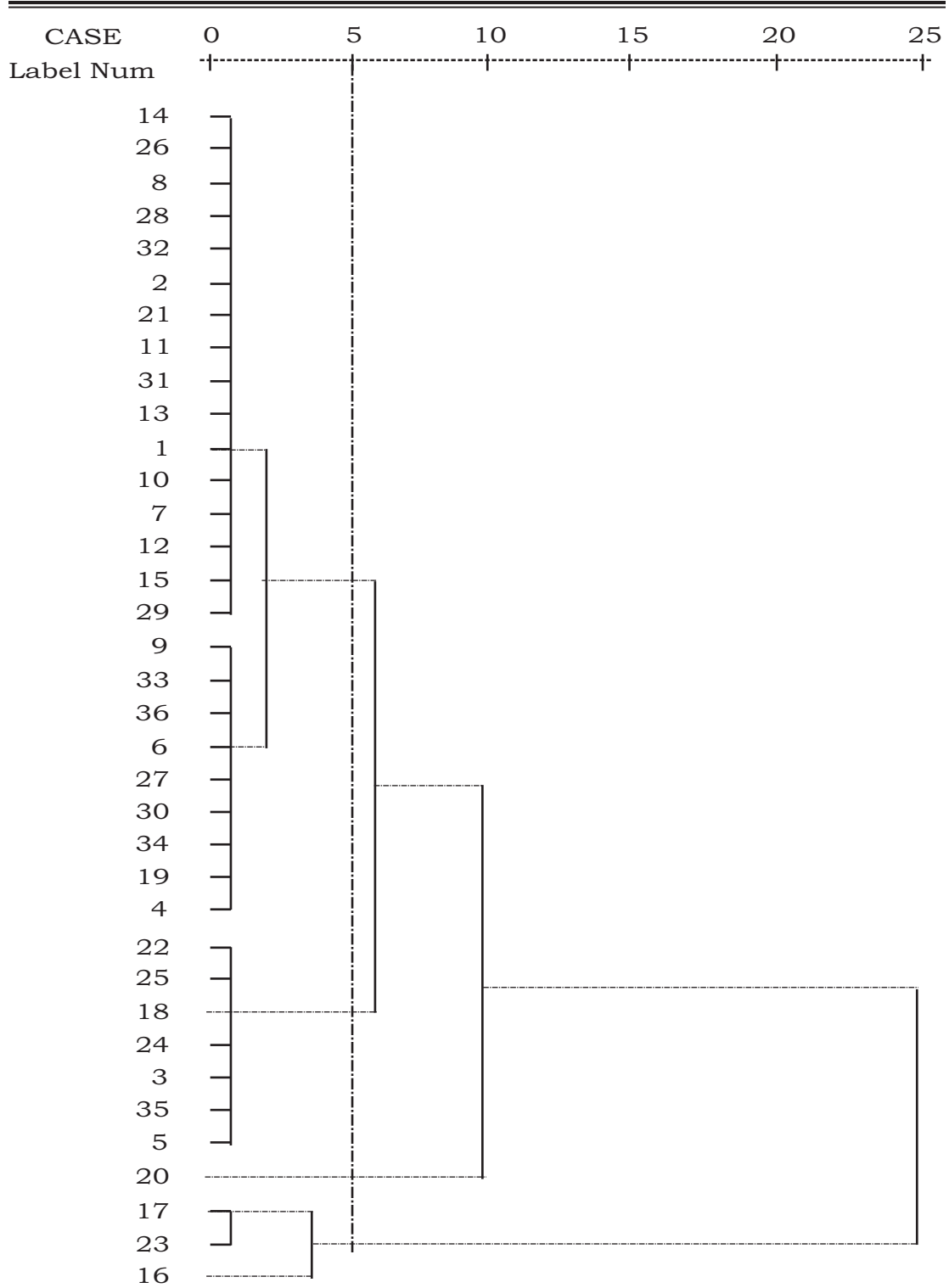
Trait	Shannon Weaver Diversity Indices (SWDI)	
	Base collection	Core collection
Days to germination	67 (M)	67 (M)
Days to flowering	59 (M)	86 (H)
Days to fruit set	55 (M)	83 (H)
Plant height (cm)	53 (M)	86 (H)
Green leave internodes <sup>-1</sup>	88 (H)	78 (H)
Inflorescence plant <sup>-1</sup>	49 (M)	77 (H)
Length of inflorescence (cm)	83 (H)	83 (H)
No. of flower inflorescence <sup>-1</sup>	57 (M)	62 (M)
No. of pod plant <sup>-1</sup>	71 (M)	81 (H)
Length of pod (cm)	84 (H)	84 (H)
Width of pod (cm)	76 (H)	76 (H)
Weight pod <sup>-1</sup> (g)	83 (H)	88 (H)
Weight of green pod without seed (g)	83 (H)	88 (H)
Weight of seed pod <sup>-1</sup> (g)	86 (H)	75 (H)
Yield plant <sup>-1</sup> (g)	71 (M)	75 (H)
Weight of 1000 seed (g)	88 (H)	75 (H)

H= High, M= Moderate

Arithmetic Average (UPGMA) cluster analysis using SPSS-10 (Table 6). From the resulting dendrogram in 1-5 scale measurement, the core subset was grouped into 4 clusters (Figure 1). The largest group was cluster IV, which included 19 genotypes and cluster II and III were the second largest gro-ups. Cluster I included only 1 geno-type. Cluster III was highly hetero-zygous while members of cluster II and IV were moderately diverged (Table 6). The amount of core acce-ssion was worked out at 10.12% of the entire collection. The result can

be validated with the findings of Franco et al. (2005) who demonstrated that core subset at 10% of entire collection was sufficient to represent diversity of entire collection.

It is therefore, concluded that despite the impressive agro-morphological variations, it appears that land races of hyacinth bean in Bangladesh are less diverged. Addition of germ-plasm from home and abroad is therefore, essential to improve genetic diversity for an effective improvement programme. Influence of environment on the observed morpho-



**Figure 1. Dendrogram using Average Linkage (between Groups)**



**Table 5. Eigenvalues of the Covariance Matrix of Principal Components for quantitative characters of base collection and core subset**

Component	Total Variance Explained					
	Base collection			Core subset		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	3.212	18.891	18.891	3.557	20.923	20.923
2	2.783	16.368	35.259	3.086	18.152	39.075
3	1.439	8.462	43.721	1.883	11.076	50.151
4	1.247	7.336	51.057	1.534	9.022	59.173
5	1.174	6.908	57.965	1.359	7.994	67.167
6	1.070	6.294	64.259	1.116	6.565	73.733
7	1.016	5.975	70.234	0.934	5.496	79.229
8	0.928	5.459	75.693	0.848	4.986	84.215
9	0.864	5.081	80.773	0.681	4.007	88.222
10	0.774	4.554	85.327	0.593	3.488	91.710
11	0.691	4.066	89.393	0.435	2.559	94.269
12	0.665	3.914	93.307	0.415	2.440	96.709
13	0.618	3.634	96.941	0.319	1.874	98.583
14	0.420	2.471	99.412	0.207	1.220	99.803
15	0.083	0.486	99.898	0.027	0.158	99.961
16	0.016	0.095	99.993	0.007	0.039	100.000
17	0.001	0.007	100.000	-1.096E-16	-6.445E-16	100.000

**Table 6. Eigenvector in the first six Principal Components of core collection**

	Components					
	1	2	3	4	5	6
Days to germination	0.413	-0.446	0.398	0.198	0.250	0.090
Days to flowering	0.066	0.144	-0.585	0.214	0.471	0.372
Days to fruit set	-0.149	-0.329	0.555	-0.128	-0.392	0.261
Flower/inflorescence	-0.366	-0.038	0.605	0.167	0.598	0.105
Plant height (cm)	-0.069	0.120	-0.066	0.704	-0.322	0.055
Leaf/node	-0.220	-0.366	-0.042	0.532	-0.014	-0.412
Pod length (cm)	0.557	0.364	0.104	0.222	0.002	0.085
Pod weight (g)	0.542	-0.002	0.319	-0.078	0.124	0.298
Inflorescence length (cm)	-0.051	0.393	0.533	-0.211	-0.199	0.063
Inflorescence number	-0.583	0.665	-0.208	0.045	-0.236	-0.032
Pod/plant	-0.703	0.630	0.182	0.135	0.157	0.081
Pod weight (g)	0.789	0.498	0.049	0.012	0.123	-0.240
Yield plant <sup>-1</sup> (g)	-0.440	0.793	0.238	0.167	0.208	0.037
Muscle weight (g)	0.805	0.369	-0.039	0.133	0.163	-0.037
Seed weight (g)	0.334	0.514	0.217	-0.259	-0.031	-0.554
Seed/pod	0.362	0.451	-0.141	-0.039	-0.381	0.492
Thousand seed weight (g)	0.162	-0.029	0.320	-0.626	-0.305	-0.024

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logical and physiological variability might have increased the base collection size without knowing real genetic variability. However, molecular characteristics would have to be used to identify duplicates and diverse genotypes to develop a core subset.

**Table 7. Cluster grouping of 36 core accession of hyacinth bean genotypes**

Cluster	Number of accessions	Intra-cluster distance	Cluster member	
			Green stem	Purple stem
I	1	0	BD-150	
II	8	23.45	BD-34, BD-2881, BD-2915	BD-2913, BD-2918, BD-8833, BD-8839, BD-8870
III	8	49.43	BD-71, BD-8862, BD-111, BD-8033, BD-8037, BD-8039, BD-8052, BD-8876	
IV	19	31.64	BD-51, BD-1829, BD-8012, BD-8787, BD-8868, BD-05, BD-42, BD-47, BD-61, BD-62, BD-103, BD-7991, BD-1772, BD-1786, BD-1797, BD-2882, BD-2901, BD-8006, BD8778	

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