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Genetic diversity in Indian bean (*Lablab purpureus*) germplasm based on morphological traits and RAPD markers

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ABSTRACT

The genetic diversity among 48 Indian bean (*Lablab purpureus* L. Sweet) genotypes, collected from various parts of India, was analyzed based on morphological traits and randomly amplified polymorphic DNA (RAPD) markers. All the genotypes, grown in randomized block design with three replications over two consecutive years, were used to record the 15 morphological traits. Twentyfive RAPD primers amplified 6–12 banding patterns for a total of 215 scorable and 178 polymorphic bands. In cluster analysis based on morphological traits and molecular markers, the genotypes were grouped in three and two clusters, respectively. Result of cluster analysis based on RAPD data showed positive correlation with morphological characters based on Mantel's test (r = 0.1417). The wide genetic variation observed for *L. purpureus* in India indicates that India has a rich genetic diversity for this species and that there is an ample scope for its genetic improvement.

Key words: Cluster analysis, Genetic diversity, *Lablab purpureus*, Morphological traits, RAPD markers

Indian bean (*Lablab purpureus* L. Sweet) (2n=24) is an important legume species, cultivated throughout the tropics and subtropics mostly for human consumption and animal forage (Murphy and Colucci 1999). It is believed to be a native of India, Southeast Asia or Africa (Shivashankar *et al.* 1993). Despite its uses and wide distribution in the tropics, *L. purpureus* is considered as neglected crop with underused potential. *L. purpureus* has been neglected in research and development and consequently our understanding of its genetic diversity is limited (Maass *et al.* 2005). However, its capacity to be used as a vegetable pulse and/or forage crop in tropical region with humid to semi-arid climate has resulted in increased interest in re-assessing its potential in tropical farming system. In India, it is a field crop mostly confined to the peninsular region and cultivated to a large extent in Karnataka, Tamil Nadu, Andhra Pradesh and Maharashtra. In India, Karnataka accounts for nearly 90% of both area and production (18 000 tonnes from 85 000 ha) of Dolichos bean- *Lablab purpureus* (http://www.lablablab.org).

Indo-China region being a centre of diversity is endowed with great variability in terms of morphological characters, especially growth habit, maturity including shape, size, colour of fruit and seed (Rai *et al.* 2006). Morphological and phenological variations and diversity studies in *L. purpureus* germplasm have been done by various workers (Ewansiha *et al.* 2007). Molecular methods are complementary to classical methods such as morphological and physiological traits in determining the genetic similarity of inter- and intra-species and the relationship between the populations.

Previous studies of diversity within *L. purpureus* and subspecies using RAPD (Liu 1996) and AFLP (Maass *et al.* 2005) suggest that there is considerable molecular variation among accessions that comprise lines from Africa, Asia and Europe; and these lines could provide useful and disease traits if incorporated into breeding strategies. In addition to the molecular diversity, *L. purpureus* also has diverse phenotypic characters (Maass 2006) reflected in their different growth habits that have served to provide cultivars fitted to specific environments. Studies in *L. purpureus* were either done through morphological traits or using molecular markers and the present investigation for the first time tries to assess the diversity in 48 genotypes of *L. purpureus* using fifteen morphological traits and assess the grouping so obtained vis-à-vis RAPD markers.

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MATERIALS AND METHODS

The plant material for this study consisted of 48 genotypes, collected from Uttar Pradesh, Bihar, Jharkhand, Chattishgargh, Delhi, Rajasthan, Gujarat, Asom, Himachal Pradesh, Kerala, Meghalaya and Tripura belonging to different agro-climatic zones of India (Table 1). All the plants were grown at research farm of Indian Institute of Vegetable Research, Varanasi during 2006–07 and 2007–08. The experiment was laid out in a complete randomized block design in three replications with 20 plants/genotypes during both years. Seeds were sown on the raised bed in the second week of July with spacing of 2 m between rows and 1 m between the plants. The recommended fertilizers doses and cultural practices along with plant protection measures were followed to raise a good crop. Data for 15 quantitative and qualitative traits were recorded for five well grown individual plants of each genotype in each replication (Table 1).

Fifteen yield and yield-associated morphological traits were assessed across 48 genotypes. These traits were chosen according to *Lablab bean* descriptors. These morphological traits were: (*i*) days to first flowering (DFF), (*ii*) days to first picking (DFP), (*iii*) no. of pods/plants (No. PPP), (iv) per cent fruit set/cluster (PFSPC), (*v*) pod length (PL), (*vi*) pod width (PW), (*vii*) number of seeds/pod (NSPP), (*viii*) seed length (SL), (*ix*) seed width (SW) (*x*) 100 seeds weight (SWt), (*xi*) yield/plant (YPP), (*xii*) flower colour (FC), (*xiii*) pod colour (PC), (*xiv*) seed colour (SC), and (*xv*) seed shape (SS).

The data of means were used for statistical analysis. Univariate statiatical analysis including coefficient of variations, means, maximum and minimum were analyzed. For cluster analysis, the effects of different scales of measurement for different morphological traits were minimized by standardizing the data for each trait separately. Standardization was done by dividing the deviation of mean fraction from the mean for the 48 *Lablab* genotypes with the standard deviation for the given trait; the STAND module of NTSYSpc version 2.1 (Rohlf 1998) software was used to achieve the same. These data were used for generating the distance matrix. The average taxonomic distance was estimated using SIMINT module of NTSYSpc software. Pair wise distance matrix was used as an input for analysis of cluster. UPGMA-based clustering was done using SAHN module of NTSYSpc.

Young leaves, collected from five individual plants of each genotype were grinded in liquid nitrogen to a fine powder, and genomic DNA was extracted using CTAB method with minor modification (Doyle and Doyle 1987). The DNA content was determined by spectrophotometer (Gene Space-III, Hitachi, Japan) with optimal A_{260}/A_{280} ratio ranging from 1.8 to 1.95 and DNA quality was checked on 0.8% agarose gel.

The PCR was accomplished by following the protocol of Williams *et al.* (1990) with minor modifications. A total of 240 random decamer primers, synthesized by Operon Technologies (USA), were evaluated using four most morphologically diverse genotypes for their capability to amplify clear, reproducible and polymorphic DNA bands. The amplification were carried out in 25µl of reaction mixture containing 15 ng genomic DNA, 1.5 mM PCR buffer (MBI Fermentas, USA), 400 µM dNTPs (MBI Fermentas), 1.5 U Taq DNA polymerase (MBI Fermentas) and 0.4 µM primer using a thermal cycler (T1, Biometra, Germany). Thermal cycler program for RAPD involved an initial denaturation at 94°C for 5 min., followed by 38 cycles each of 1 min. denaturation at 94°C, 1 min. primer annealing at 34°C, 1 min primer extension at 72°C and followed by one final extension cycle of 10 min at 72°C. The amplified PCR products and 1 Kb DNA marker ladder (MBI Fermentas) were size fractioned through electrophoresis in 1.4% agarose gel prepared in 1x TAE buffer [40.0 mM Tris-base, 16.65 M acetic acid, 0.5 M EDTA (*p*H 8.0)]. After electrophoresis, gels were stained with ethidium bromide (0.5 µg/ml) and visualized in a gel documentation system (Alfa Imager 2200, Alfa Innotech Corporation, California). The amplification was repeated 2–3 times to ensure that the amplification obtained with the primers is reproducible and consistent. Later, 25 primers, which were found to produce polymorphic DNA amplification products, were used for RAPD analysis of all the 48 genotypes.

The RAPD bands were scored manually for the presence (1) or absence (0) across all the genotypes if visible, regardless of the relative intensity. Only reproducible and clearly distinguished bands were taken into consideration. This data were analyzed using software NTSYSpc. Pair-wise combinations of genotypes were employed to calculate Jaccard's similarity coefficient (GS) = a/(n - d), where a is the number of positive matches, n is the total sample size, and d is the number of negative matches. Genetic distance (GD) between pair of lines was estimated as GD = 1 - GS. This matrix was subjected to cluster analysis by the unweighted pair-group method and the dendrogram was constructed using the SAHN module of NTSYSpc software package. The MXCOMP module of NTSYSpc was used to compare the Jaccard's similarity values estimated from RAPD patterns and the average taxonomic distance values derived from 15 morphological traits.

RESULTS AND DISCUSSION

Morphological evaluation

A total of 15 morphological and yield related traits were recorded for assessment of relationship among *L. purpureus* genotypes. The results showed that the genotypes of *L. purpureus* were significantly different in 15 quantitative and qualitative traits (Table 1), suggesting that selection for relevant characters could be possible. The high coefficient of variation (CV) was observed for the traits per cent fruit set/cluster, number of seeds/pod, seed width, seed weight and pod width.

Table 1 List of 48 *Lablab purpureus* genotypes, morphological characteristics and their mean over two years (2006-2007 and 2007-2008)

Genotypes, source	[a]DFF	DFP	No.PPP	PFSPC	PL	PW	NSPP	SL	SW	SWt	YPP	FC	PC	SC	SS
VRS 501, Asom	45	121.33	231.67	43.67	13.7	2.68	4.7	1.23	0.76	33.33	2.4	1.00	2.00	1.00	1.00
VRS 43, Asom	94.67	145	234.67	44.67	8.6	3.81	4.7	1.85	0.73	35.33	2.37	2.00	1.00	4.00	1.00
VRS 42, Asom	108	141.67	164	55	13.5	1.45	4.3	1.36	0.9	41.33	1.93	1.00	5.00	9.00	4.00
VRS 782, Bihar	117	139.33	126.67	53.33	8.84	1.23	4.7	1.24	0.65	25.67	0.57	2.00	7.00	9.00	3.00
VRS 788, Bihar	91	135.67	155.67	42.33	10.13	1.54	5.0	1.06	0.72	25.67	1.29	1.00	2.00	7.00	1.00
VRS 809, Bihar	76	136	165.33	52.67	10.2	1.13	4.0	1.25	0.78	26.67	0.68	1.00	2.00	2.00	1.00
VRS 772, Bihar	119	136	175	35.33	11.4	0.55	4.0	1.2	0.84	25	1.74	2.00	2.00	9.00	4.00
VRS 791, Bihar	121	151.33	200.67	66.33	8.57	2.33	4.7	1.26	0.87	32	1.41	2.00	3.00	7.00	4.00
VRS 797, Bihar	85	132.33	507.33	46	10.73	1.8	4.0	1.25	0.83	34	3.29	1.00	2.00	9.00	1.00
VRS 45, Chattishgargh	135	165	119.33	44	9.5	3.2	5.0	1.15	0.85	33.33	0.92	1.00	9.00	7.00	1.00
VRS 802, Bihar	107	137	453.33	52.67	8.43	2.74	4.0	1.19	0.82	33.67	2.73	2.00	10.00	5.00	1.00
VRS 117, Chattishgargh	122	152.33	58.33	54.33	11.2	2.04	5.7	1.13	0.74	25	0.47	2.00	7.00	7.00	2.00
VRS 67, Chattishgargh	106	132.67	391.33	45	9.5	2.26	4.3	1.05	0.74	32	1.93	2.00	3.00	2.00	4.00
VRS 107, Chattishgargh	121	136	174.67	46.67	10.95	2.53	6.0	1.15	0.76	32	1.08	1.00	4.00	2.00	4.00
VRS 100, Chattishgargh	118	129	54.33	54	13.11	3.9	6.0	1.23	0.87	37	0.76	2.00	2.00	7.00	1.00
VRS 145, Chattishgargh	101	142	131	51	11.2	2.32	5.0	1.23	0.82	35	1.43	2.00	7.00	9.00	1.00
VRS 115, Chattishgargh	83	130	208	66	10.66	1.59	5.0	1.08	0.81	35	1.86	3.00	11.00	7.00	4.00
VRS 109, Chattishgargh	110	139	344.33	52.33	7.18	1.33	3.7	0.84	0.92	37.33	1.69	1.00	2.00	7.00	4.00
VRS 125, Madhya Pradesh	90	109.33	277.67	52	7.47	1.34	4.0	0.91	0.75	32.67	1.43	1.00	1.00	9.00	1.00
VRS 90, Chattishgargh	105.33	133.33	187.33	43.67	12.57	1.3	4.3	1.12	0.8	32.33	1.3	1.00	1.00	7.00	2.00
VRS 904, Delhi	99	121	343	45	8.5	1.25	3.7	1.16	0.83	35.33	1.75	2.00	6.00	7.00	4.00
VRS 905, Delhi	102	132	183	24	6.71	1.25	4.0	0.83	0.74	36	0.73	2.00	6.00	7.00	4.00
VRS 878, Gujarat	110	141.67	487.33	57.33	10.43	0.95	3.7	1.36	0.7	35.33	2.25	2.00	3.00	7.00	4.00
VRS 820, Gujarat	109.67	144	679.33	47.67	9.37	1.33	4.0	1.14	0.81	31	1.67	1.00	2.00	10.00	1.00
VRS 825, Gujarat	110	134	291	45	11.25	3.59	5.0	1.19	0.84	37	3.5	4.00	12.00	8.00	4.00
VRS 836, Gujarat	107.33	143.67	185.33	39.33	8.63	0.81	3.7	1.05	0.68	29	0.69	4.00	6.00	7.00	1.00
VRS 843, Gujarat	120.33	141.33	114.67	58.67	9.8	2.67	5.3	1.07	0.74	34.33	1.15	4.00	8.00	8.00	4.00
VRS 910, Himanchal Pradesh	55.33	125.33	628.33	34.33	8.75	2.23	4.0	1.1	0.82	39.33	3.08	5.00	3.00	7.00	4.00
HADB 3, Jharkhand	55	95	503.33	52.33	9.43	2.33	4.0	1.15	0.84	42	2.5	5.00	3.00	2.00	4.00
HADB 4, Jharkhand	67	121	240.33	42.33	9.37	2.04	4.0	1.18	0.85	35.33	0.94	2.00	6.00	2.00	5.00
Swarn Utkrisht, Jharkhand	115	149.33	835.33	45	10.75	1.93	3.7	1.23	0.94	32.67	1.06	1.00	2.00	6.00	4.00
KDB 415, Kerala	102	132	145	47	13.89	2.25	5.0	1.2	0.81	45	2.45	1.00	4.00	2.00	4.00
KDB 413, Kerala	104	130	295	38	12.81	2.1	5.0	1.26	0.89	40	2.36	2.00	6.00	7.00	2.00
VRS 301, Meghalaya	104	134	130	50	13.08	3.02	4.0	1.26	0.91	40	2.24	2.00	6.00	7.00	4.00
VRS 101, Meghalaya	111	136	168	48	14.5	3.2	5.0	1.25	0.89	45	2.32	1.00	2.00	1.00	2.00
VRS 201, Meghalaya	107	135	97.33	48	9.75	1.75	5.0	1.1	0.76	25	0.77	2.00	3.00	9.00	4.00
VRS 601, Rajasthan	91	132	212.67	51.33	5.65	2.24	4.0	1.55	0.74	17.67	1.3	2.00	8.00	7.00	4.00
VRS 401, Rajasthan	105	139.33	222.33	61	6.34	2.74	4.0	1.06	0.74	15.33	1.13	1.00	7.00	1.00	1.00
VRS 8, Tripura	104.67	137	154	52.33	6.57	2.23	4.0	1.17	0.86	15	0.85	2.00	6.00	3.00	4.00
VRS 11, Tripura	106	138	406	46.33	8.49	3.12	4.7	1.16	0.85	36.33	3.1	5.00	7.00	7.00	4.00
VRS 186, Tripura	99.67	121.67	225	63	14.57	1.31	5.0	1.41	1.07	33.33	1.85	2.00	2.00	2.00	2.00
VRS 12, Tripura	77	135	186.33	65	12.5	2.27	4.0	1.24	0.85	28.67	1.12	1.00	1.00	6.00	6.00
VRS 6, Tripura	63	115	70	45	13.37	2.53	4.3	1.23	0.85	34.33	0.35	6.00	2.00	8.00	4.00
VRS 804, Uttar Pradesh	101	140	205.33	61	14.25	1.83	5.0	0.84	0.91	32	1.86	1.00	2.00	7.00	4.00
VRS 953, Uttar Pradesh	107	141	257.33	50.33	12.15	1.65	6.0	1.25	0.81	32.33	1.57	2.00	7.00	8.00	1.00
VRS 937, Uttar Pradesh	106	138	169.67	51.67	11.73	1.29	5.3	1.2	0.84	31.67	0.86	1.00	5.00	2.00	4.00
VRS 752, Uttar Pradesh	108	130	380.33	45.33	12.46	2.33	5.3	1.16	0.74	23	2.86	1.00	2.00	7.00	4.00
VRS 720, Uttar Pradesh	98	140	175.67	44.33	7.5	2.37	4.3	1.08	0.76	45	0.85	4.00	2.00	9.00	1.00
Mean	99.99	135.28	253.53	49.17	10.28	2.07	4.56	1.18	0.85	32.59	1.63				
Range	135-45	165-95	835.33-70	66.33-24	14.57-5.65	3.9-0.55	6-3.70	0.83-1.85	1.07-0.65	45-15	3.5-0.35				
CV	1.80	1.83	2.2	10.10	7.5	10.24	10.88	7.33	68.3	12.6	5.80				

CV, Coefficient of variation

[a]DFF, Days to first flowering; DFP, days to first picking; No.PPP, no. of pods/plant; PFSPC, per cent fruit set/ cluster; PL, pod length; PW, pod width; NSPP, no. of seeds/pod; SL, seed length; SW, seed width; SWt, 100 seed weight; YPP, yield/plant; FC, flower colour (1- white, 2, pink, 3- blue white, 4- red, 5- blue); PC, pod colour (1- white, 2- green, 3- red, 4- yellow, 5- dark green, 6- green purple, 7- light green, 8- yellow purple); SC, seed colour (1- reddish brown, 2- reddish black , 3- reddish black with white spotted, 4- deep brown, 5- deep brown red, 6- brown red, 7- black, 8- redish, 9- brown, 10- cream); SS, seed shape (1- round, 2- round oval, 3- round oval kidney shape, 4- flat, 5- flat oval, 6- flat kidney shape)

The germplasm showed very high degree of variation in morphological traits like number of pods/plant, pod length, pod width and seed length. Correlations between morphological traits of *Lablab* genotypes showed that several seed characteristics were in significant correlation with pod characteristics. The genotypes VRS 501, VRS 8, VRS 186, VRS 11, VRS 6 and KDB 413, showed maximum potential for yield/plant due to their high number of pods/plant, pod length and pod width. Presently, these genotypes are under testing in All India Coordinated Research Programme (AICRP) at various agro-ecological zones of India for high yield and good pod quality with resistance/tolerance to DYMV (Dolichos yellow mosaic virus), a devastating problem for its cultivation in northern parts of India. The genotypes with medium pod length and pod width, viz VRS 101 and VRS 843 accompanied with higher yield could be used for improvement of lablab bean for vegetable purposes.

Cluster analysis based on morphological traits

The standardized morphological data were used to estimate taxonomic distances between 48 genotypes for 15 morphological traits; taxonomic distance matrix was employed for cluster analysis and a dendrogram was generated (Fig 1). Genetic dissimilarity values ranged from 0.02 to 0.69. All the genotypes grouped into three major clusters. Dendrogram generated from morphological data showed that the genotypes from north India and Tripura grouped together showing considerable degree of similarity. This may be due to the exchange of genotypes among the institutions for use in breeding programmes which undermined the level of genetic diversity. VRS 791, along with HADB 4, from Bihar and Jharkhand, respectively separated from rest of the genotypes and formed a group. These are the most diverse genotypes from the rest.

RAPD analysis

The selected 25 random decamer primers (Table 2) generated a total number of 215 amplification products of which 178 (83%) were found to be polymorphic. The number of bands generated per primer varied from 6 to 12 and on an average each primer amplified 8.60 products. Each primer on an average yielded 7.12 polymorphic bands; OPC05 and OPF14 produced maximum number (12) of polymorphic bands. The primers viz, OPA18, OPC05, OPE17, 19, OPF13, 14, produced 100% polymorphic products. The size of amplified products from all the primers varied between 300 bp and 3100 bp. The minimum size of 300 bp amplification product was generated from primer OPC04 and OPG13 while the maximum size of 3100 bp was generated with primer OPG13. The primers OPA12, OPA18, OPC04, OPC05, OPD03, OPF14 and OPG02 were found to be most informative based on the level of polymorphism detected by them.

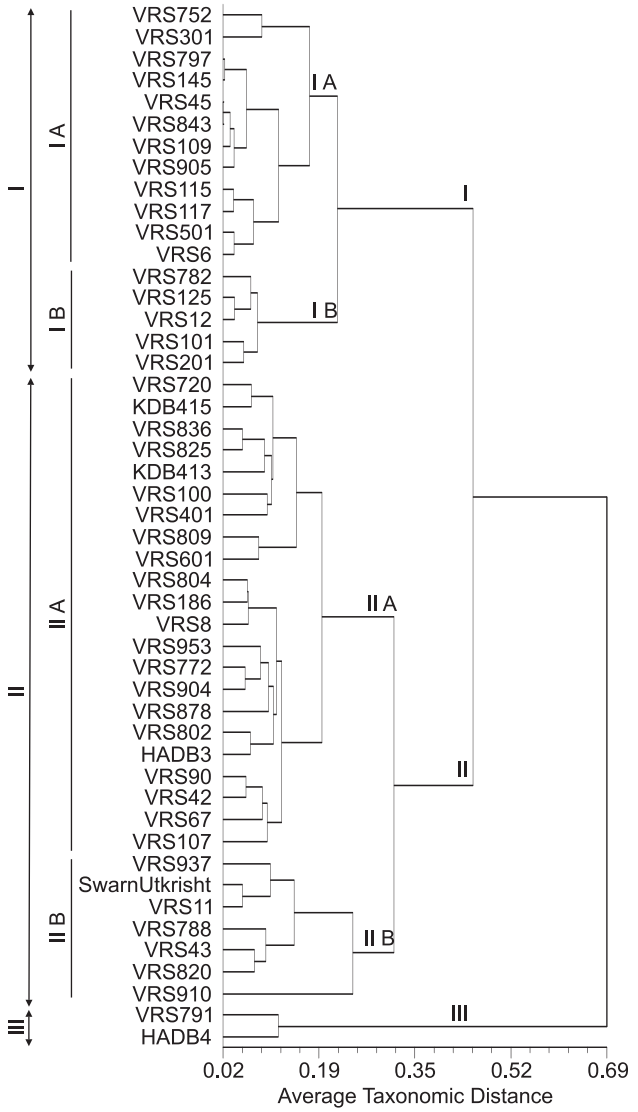

Fig 1 Genetic relationship among 48 genotypes of *Lablab purpureus* based on 15 quantitative and qualitative traits using UPGMA cluster analysis of the distance matrix

Cluster analysis based on RAPD

The relationship between the 48 genotypes based on their genetic relatedness was clustered in a dendrogram through analysis of data on the 215 RAPD markers. Jaccard's similarity matrix was used to generate a dendrogram to obtain the clustering of genotypes (Fig 2). The dissimilarity coefficient ranged from 0.05 to 0.61. The highest similarity coefficient (0.95) was observed between genotypes VRS 720 and VRS 802. The dendrogram showed that all the genotypes were grouped into two major clusters. The bigger cluster, cluster I consisted of 42 genotypes, however, the cluster II consisted of only six genotypes. The wide range of dissimilarity values suggests that the germplasm collection represents a genetically diverse population. The results show that a rich diversity exists between the germplasm collections

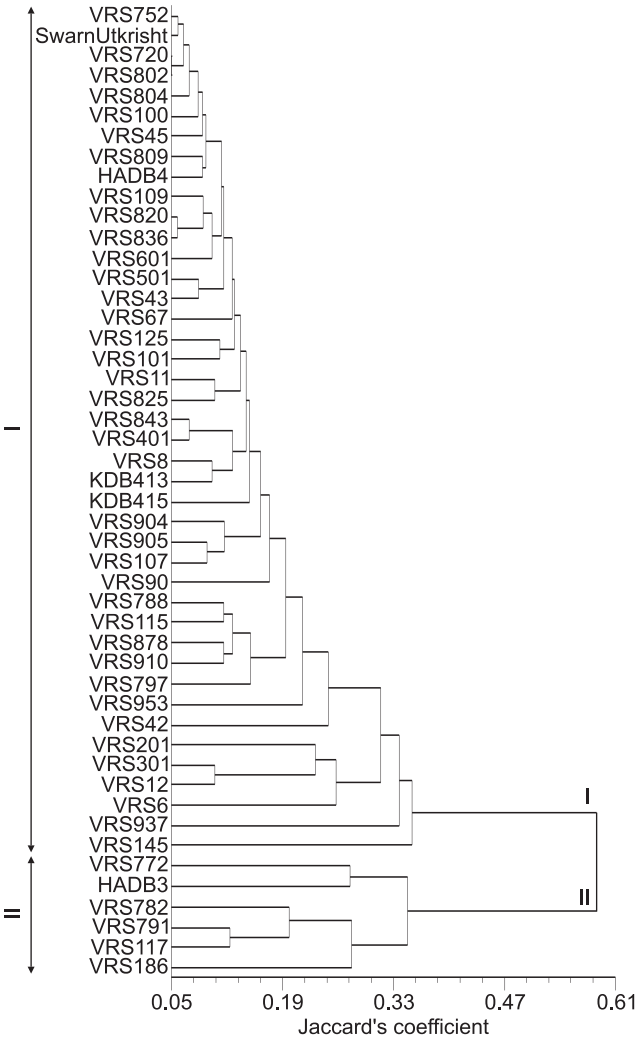

Fig 2 UPGMA dendrogram of 48 genotypes of *Lablab purpureus* based on the 25 random decamer primers

from different geographical regions of the country.

The levels of polymorphism in *L. purpureus* detected by RAPD markers have been estimated to be very low. Dendrogram generated from RAPD analysis showed the clustering of 48 *Lablab* genotypes into two clusters. The genotypes which belong to one region grouped together. The number of amplified bands/primer and their degree of polymorphism detected were found to be low. This is in agreement with Liu (1996) who studied genetic variation in *Lablab* accessions from Asia and Africa with some wild species and found low level of RAPD polymorphism among *L. purpurious* accessions from one region. The level of polymorphism observed in the present study was comparable with that observed utilizing AFLPs (Maass *et al.* 2005) and RAPDs (Liu 1996) depending on the germplasm studied.

In the present study, both of the matrices was in the form of dissimilarity, the correlation obtained between the two matrices (r = 0.1417) indicated little relationship between the two dendrograms. The possible reason behind this may

Table 2 RAPD primers used in present study-their sequence, number of polymorphic products and per cent of polymorphic bands produced by each primer

Primername	Sequence (5'-3')	Total no. of bands amplified	No. of poly-morphic band	Poly-morphic band (%)
OPA 08	GTGACGTAGG	10	7	70
OPA 12	TCGGCGATAG	6	5	83
OPA 18	AGGTGACCGT	7	7	100
OPC 04	CCGCATCTAC	6	4	66.67
OPC 05	GATGACCGCC	12	12	100
OPC 06	GAACGGACTC	9	7	78
OPD 03	GTCGCCGTCA	12	9	75
OPD 09	CTCTGGAGAC	6	4	67
OPE 07	AGATGCAGCC	7	5	71.42
OPE 17	CTACTGCCGT	9	9	100
OPE 19	ACGGCGTATG	9	9	100
OPF 03	CCTGATCACC	8	7	87.5
OPF 13	GGCTGCAGAA	7	7	100
OPF 14	TGCTGCAGGT	12	12	100
OPF 19	CCTCTAGACC	7	6	86
OPG 02	GGCACTGAGG	9	5	55.5
OPG 03	GAGCCCTCCA	10	8	80
OPG 05	CTGAGACGGA	10	8	80
OPG 08	TCACGTCCAC	8	7	87.5
OPG 10	AGGGCCGTCT	8	6	75
OPG 13	CTCTCCGCCA	6	4	67
OPH 02	TCGGACGTGA	7	6	86
OPH 03	AGACGTCCAC	11	9	81.82
OPH 05	AGTCGTCCCC	10	8	80
OPH 18	GAATCGGCCA	9	7	78

be as the morphological traits are controlled by a subset of the genomic regions, while most molecular markers sample random genomic regions (Dahlberg 2000) most of which may not be expressed at the phenotypic level. As a result, markers like RAPDs may accurately assay the degree of genetic change distinguishing two genomes, but they may not necessarily reflect the divergence in terms of changes in morphological traits, which are subjected to selective modifications.

The present investigation concluded that genetic diversity in 48 genotypes of *L. purpureus* based on morphological traits and RAPD markers, both were effective in discriminating the different genotypes and found useful for better management of germplasm resources. However, there is much environmental influence accounting for the morphological variability observed. For a more efficient determination of genetic diversity in *L. perpureus* germplasm, more number of RAPD primers showing maximum number of polymorphic bands could be utilized. Diverse types with peculiar characteristics, identified in the present study may be used as parents in the improvement breeding of lablab bean. Furthermore, for conservation purpose, morphological

traits and RAPD markers would have to be used hand in hand rather than in isolation for identification of duplicates and diverse genotypes to develop a core collection of *L. perpureus* germplasm.

REFERENCES

Dahlberg J A. 2000. Classification and characterization of sorghum. (*in*) *Sorghum: Origin, History, Technology and Production,* pp 99–130. Smith C W, Frederikson R A (Eds), John Wiley and Sons, Inc, New York.

Dolichus Bean - *Lablab purpureus* (L.) Sweet. 2011. http://www.lablablab.org

Doyle J J and Doyle J L. 1987. A rapid DNA isolation procedure from small quantity of fresh leaf material. *Phytochemical Bulletin* **119**: 11–5.

Ewansiha S U, Chiezey U F, Tarawali S A and Iwuafor E N O. 2007. Morpho-phenological variation in *Lablab purpureus*. *Tropical Grassland* **41**: 277–84.

Liu C J. 1996. Genetic diversity and relationships among *Lablab purpureus* genotypes evaluated using RAPD as markers. *Euphytica* **90**: 115–19.

Maass B L, Jamnadass R H, Hanson J and Pengelly B C. 2005. Determining sources of diversity in cultivated and wild *Lablab purpureus* related to provenance of germplasm by using amplified fragment length polymorphism. *Genetic Resources and Crop Evolution* **52**: 683–95.

Maass B L. 2006. Changes in seed morphology, dormancy and germination from wild to cultivated hyacinth bean germplasm (*Lablab purpureus*: Papilionoideae). *Genetic Resources and Crop Evolution* **53**: 1127–35.

Murphy A M and Colucci P E. 1999. A tropical forage solution to poor quality ruminant diets: A review of *Lablab purpureus*. *Livestock Research for Rural Development* **11**: 2.

Rai N, Asati B S, Singh A K and Yadav D S. 2006. Genetic variability, character association correlation and path coefficient analysis in between seed morphology and seeding growth in French bean. *Journal of Assam Science Society* **42**: 40–43.

Rohlf F J. 1998. NTSYS-pc Numerical Taxonomy and Multivariate Analysis System, version 2.01 Exeter Software, Setauket, New York, USA.

Shivashankar G, Kulkarni R S, Shashidhar H E and Mahishi D M. 1993. Improvement of field bean. (*in*) *Advances in Horticulture,* Vol **5**, pp 77–286. Chadha K L and Kallo G (Eds), New Delhi.

Williams J G K, Kubelik A R, Livak K J, Rafalski J A and Tingey S V. 1990. DNA polymorphism amplified by arbitrary primers are useful as genetic markers. *Nucleic Acids Research* **18**: 6531–5.