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GENETIC DIVERSITY OF WILD AND CULTIVATED *Mucuna pruriens* (L.) DC. ACCESSIONS ANALYZED USING THIRTY MORPHO-AGRONOMICAL CHARACTERS

[DIVERSIDAD GENETICA DE VARIEDADES SILVESTRES Y DOMÉSTICADAS DE *Mucuna pruriens* (L.) DC. ANALIZADA MEDIANTE EL EMPLEO DE TREINTA CARACTERES MORFO-AGRONOMICOS]

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SUMMARY

Genetic diversity and relatedness among 31 wild and 9 cultivated *Mucuna pruriens* accessions collected from diverse eco-climatic regions in India were evaluated using 30 morpho-agronomical characters. The experiments were carried by growing the plants in Randomized Complete Block Design (RCBD) in four replications for two seasons. The results of PCA for 22 quantitative and 8 qualitative characters indicated that the first two PCs together explained 65.18% and 67.50% of the total variation respectively, which were drawn across a range of traits. Cluster analysis demonstrated on the whole high morphological diversity (63.05%) amongst wild and cultivated accessions and low diversity (36.95%) within cultivated accessions. Analysis of six yield traits based on $LSD_{5\%}$ values revealed distinct genotypes under each category which are of potential value in breeding programs. Low morphological diversity within cultivated accessions emphasizes the need for expanding its genetic base in India.

Key words: *M. pruriens*; genetic diversity; L-Dopa.

RESUMEN

Se evaluó la diversidad genética y parentesco de 31 variedades silvestres y 9 cultivadas de *Mucuna pruriens* colectadas en diferentes regiones agroclimáticas de la india mediante el empleo de 30 caracteres morfo-agronómicos. Se empleó un diseño de bloques al azar con cuatro repeticiones para dos estaciones. Los resultados del análisis de componentes principales para 22 caracteres cuantitativos y 8 cualitativos indicaron que 2 componentes explicaron 65.2% y 67.5% de la variación total respectivamente. El análisis de conglomerados mostró la existencia de una gran diversidad morfológica (63.05%) en las variedades silvestres y una baja diversidad (36.95%) en las variedades cultivadas. El análisis de seis caracteres productivos (basado en $DMS_{5\%}$) permite identificar los genotipos en cada categoría con potencial para ser empleados en programas de mejoramiento. La baja diversidad morfológica en las variedades cultivadas enfatiza la necesidad de ampliar la base genética del cultivo en la India.

Palabras clave: *M. pruriens*; diversidad genética; L-Dopa.

INTRODUCTION

The genus *Mucuna* belongs to family Fabaceae (Leguminosae) and includes 150 species of annual and perennial legumes of pantropical distribution. Reported to be indigenous to India, China and Malaysia (Burkill, 1966), all the species of this genus are climbing plants with long, slender branches and lanceolate leaves with large flowers and hairy pods.

Mucuna pruriens, an important member of this group is a native of southern Asia (Wilmot-Dear, 1987), and was introduced to USA in the late 1800s and from there re-introduced to tropical and subtropical areas in 1900s (Duke, 1981). Described as "*featured example of green manures contribution to sustainable agricultural systems*" (Buckles, 1995), a growing body of evidence has established its potential to rapidly establish ground cover, produce large above-ground biomass, and accumulate nutrients, particularly nitrogen in various environments (St-Laurent *et al*., 2002). Its impact as green manure cover crop on main crop yield is well documented in number of earlier studies (Carsky and Ndikawa, 1998, Jorge *et al*., 2007).

M. pruriens has gained increased global attention in recent times as promising source of protein diet due to presence of 20-30% of protein content in the seed (Buckles, 1995). However, despite nutritional profile similar to other food legumes (Bressani *et al*., 2003), the plant suffers from poor edible attributes due to presence of high levels of L-Dopa (1-5%) – a non protein amino acid that acts as a precursor of the neurotransmitter drug dopamine used in the treatment of Parkinson's disease (Haq, 1983). When consumed as food, L-Dopa (L-3, 4- dihydroxy phenylalanine) induces serious side effects such as nausea, anorexia and vomiting in human beings (Szabo and Tebbett, 2002) and intestinal disorders in ruminant animals (Pugalenthi *et al.*, 2005). Past experiences have shown this problem as the major bottleneck for popularization of *M. pruriens* cultivation in the farmer's field and thus efforts are needed to breed improved varieties with safe levels of L-Dopa to make its cultivation broad based and acceptable.

The role of genetic resource characterization in plant breeding endeavors is well recognized (Querol, 1987). The possible tradeoff of genetic material from such an effort has often resulted in newer combination of traits with long lasting impact on breeding of relevant crop species. However, despite presence of rich genepool, such options are explored to minimal in *M. pruriens*, probably due to lack of data on germplasm attributes. Therefore, there is a need to undertake detailed characterization of *M. pruriens* genetic resources in India to reinforce the local crop improvement program. In view of this, a detailed analysis on variability among 31 wild and 9 cultivated accessions collected from diverse geographic locations in India have been carried out with an objective to (a) determine the variability for morpho-agronomical characters among these accessions (b) ascertain their relationships, and (c) identify potential germplasm lines which can be used in future breeding programs.

MATERIALS AND METHODS

The study was carried out in an experimental field of Sir M Visvesvaraya Institute of Technology, Bangalore. The site is located in south-eastern part of the state of Karnataka situated in Southern India at 13.15° N and 77.61° E at an altitude of 914 m above the sea level. The area receives mean annual rainfall of 842 mm yr^{-1} with primary rainy season falling between June and September (South-West monsoon) and secondary season falling between November and December (North-East Monsoon). The mean annual temperature is recorded to be 24.3° C with hottest season occurring between March and May. The soil is classified as red-laterite, which is well drained, deep reddish brown with moderately acidic pH.

Forty *M. pruriens* accessions comprising 31 wild (var. *pruriens*; itching bean) and 9 cultivated (var.*utilis*; Velvetbean) genotypes collected from diverse agro-climatic regions in India through regular field visits during 2007-2008 as well as those obtained from repository of National Bureau of Plant Genetic Resources (NBPGR), New Delhi, were used in the present study (Table 1). All the accessions were grown for seed bulking between April and December 2008 before proceeding with main study in Jan. 2009. For seed germination, five to eight seeds were planted in each hole initially and were later thinned to one seedling three-four weeks after planting. The seeds were directly sown in the field at a spacing of 2.0 m x 2.0 m. All the experimental plots were set into natural showers and sunlight supported with manual irrigation once in 7-8 days. Other agronomic practices including weeding and top dressing were conducted uniformly and as required. No external fertilizer was applied at any stage of experiment. The first season experiment was conducted between January and June 2009, followed by the second season experiment between July and December, 2009. The experiment was laid out in a Randomized Complete Block Design (RCBD) with four replications.

Data on 8 qualitative and 22 quantitative characters including leaf, flower, fruit and seed characteristics were recorded adopting descriptor prepared by the present authors (unpublished). Each qualitative descriptor was scored by observing four tagged plants per accession taking one plant from every block (replicate). The data were recorded directly on living plant using 1-9 scale or as binary recording after segregating the variable into discrete or rank ordered factor. Quantitative descriptors were scored as mean value integers of four measurements made on four plants per replicate. Among the quantitative characters, vegetative data was recorded after 45 days; flower and pod data at 50% maturity; and days to maturity when 90% of the pods were about to dry.

Statistical analysis on 22 quantitative characters was carried out using different statistical softwares. First descriptive statistics (mean, standard deviation, and coefficient of variation) were generated using JMP software version 8 (SAS Inst. Inc., 2008) with UNIVARIATE procedure. Principal component analysis (PCA) was later performed using XLSTAT software version 2010.4.01. (Addinsoft Inc., 2010). In this procedure, first similarity matrix was used to generate Eigen values and scores for accessions. The first two principal components which accounted for the highest variation were then used to plot two dimensional scatter plots. For performing cluster analysis, data was organized into matrix and analyzed using NTSYS-pc software version 2.21 (Rohlf, 2009) using SIMQUAL function and Unweighted pair-group

method with arithmetic averages (UPGMA). Statistical uncertainty of the resulting clusters was determined through multi-scale bootstrapping using WINBOOT software (Yap and Nelson, 1996). In addition, eight yield traits such as days to flowering, number of flower buds per cluster, number of pods per cluster, days to maturity, hundred seed weight and fertility index, after normalization, were subjected to frequency distribution analysis to determine the arrangement of accessions in each frequency class which was later used to ascertain the extremes of phenotypes. For all the genotypes showing significant ANOVA, $LSD_{5\%}$ scores were determined to further identify the accessions which are important from the point of breeding these traits.

Table 1. *Mucuna pruriens* accessions used in the study

Sl. No.	Name of the Accession	Accession Number	Place of collection	Latitude and longitude
1.	*M. pruriens* var. *utilis*	500102KA	Karnataka	-
2.	*M. pruriens* var. *utilis*	IC369144	NBPGR, Latehar, Jharkand	23° 74’ N, 84° 50’ E
3.	*M. pruriens* var. *utilis*	IC185926	NBPGR	-
4.	*M. pruriens* var. *utilis*	IC385928	NBPGR	-
5.	*M. pruriens* var. *utilis*	IC385841	NBPGR, Pakud, Jharkand	24° 62’ N, 87° 84’ E
6.	*M. pruriens* var. *utilis*	IC471870	NBPGR, New Delhi	28° 62’ N, 77° 23’ E
7.	*M. pruriens* var. *utilis*	IC385842	NBPGR, Mohanpur, Jarkhand	24 °48’ N, 86 ° 69’ E
8.	*M. pruriens* var. *utilis*	IC385926	NBPGR, Dhangadih, Jharkand	24° 26’ N, 87° 24’ E
9.	*M. pruriens* var. *utilis*	500159PY	Madagadipet, Puducherry	-
10.	*M. pruriens* var. *pruriens*	500109KA	Shimoga, Karnataka	13° 56' N, 75° 38’ E
11.	*M. pruriens* var. *pruriens*	500110KA	Devarayanadurga, Karnataka	13° 33’ N, 77° 10’ E
12.	*M. pruriens* var. *pruriens*	500112KA	Mysore, Karnataka	12° 18' N, 76° 42' E
13.	*M. pruriens* var. *pruriens*	500113MH	Triambakeshwar, Maharashtra	20° 00’ N, 73° 77’ E
14.	*M. pruriens* var. *pruriens*	500115TN	Ayurvedic Vendor, Madurai, TN	-
15.	*M. pruriens* var. *pruriens*	500123KL	Seed company, Kerala	-
16.	*M. pruriens* var. *pruriens*	500130KA	Gokarna, Karnataka	14° 42' N, 74° 04' E
17.	*M. pruriens* var. *pruriens*	IC265577	NBPGR, Kottayam, Kerala	9° 58’ N, 76° 52’ E
18.	*M. pruriens* var. *pruriens*	500138TN	Chittanavasal, Tamil Nadu	10° 38’ N, 78° 82’ E
19.	*M. pruriens* var. *pruriens*	500142KA	Humnabad, Karnataka	17° 43’ N, 77° 12’ E
20.	*M. pruriens* var. *pruriens*	500154AP	Srisailam, Andhra Pradesh	-
21.	*M. pruriens* var. *pruriens*	500151AP	Kadam, Andhra Pradesh	19° 05’ N, 78° 45’ E
22.	*M. pruriens* var. *pruriens*	500152AP	Khanaparti, Andhra Pradesh	18° 48’ N, 79° 09’ E
23.	*M. pruriens* var. *pruriens*	500136TN	Valapar kovil, Tamil Nadu	-
24.	*M. pruriens* var. *pruriens*	500146AP	Ghanapur, Andhra Pradesh	18° 39’ N, 78° 10’ E
25.	*M. pruriens* var. *pruriens*	500147AP	Durzgaon, Andhra Pradesh	18° 39’ N, 78° 10’ E
26.	*M. pruriens* var. *pruriens*	500148AP	Mancheppa, Andhra Pradesh	18° 40’ N, 78° 10’ E
27.	*M. pruriens* var. *pruriens*	500135KA	Chandrapalli, Karnataka	17° 47' N, 77° 43' E
28.	*M. pruriens* var. *pruriens*	500165AP	Mahanandi, Andhra Pradesh	15° 26' N, 79° 06' E
29.	*M. pruriens* var. *pruriens*	500149AP	Pochampad dam, Andhra Pradesh	18° 89’ N, 78° 10’ E
30.	*M. pruriens* var. *pruriens*	500144AP	Pathur, Andhra Pradesh	18° 03’ N, 78° 18’ E
31.	*M. pruriens* var. *pruriens*	500145AP	Dichpalli, Andhra Pradesh	18° 39’ N, 78° 10’ E
32.	*M. pruriens* var. *pruriens*	500133KA	Hudugi, Karnataka	17° 43’ N, 77° 12’ E
33.	*M. pruriens* var. *pruriens*	500134KA	Hospet, Karnataka	15° 16' N, 76° 26' E
34.	*M. pruriens* var. *pruriens*	500129KA	Karwar, Karnataka	14° 48' N, 74° 12' E
35.	*M. pruriens* var. *pruriens*	500131KA	Chittapur, Karnataka	17° 12’ N, 77° 08’ E
36.	*M. pruriens* var. *pruriens*	500122TN	Herbal drug vendor, Madurai	-
37.	*M. pruriens* var. *pruriens*	500120TN	Herbal drug vendor, Madurai	-
38.	*M. pruriens* var. *pruriens*	500162PY	Herbal drug vendor, Puducherry	-
39.	*M. pruriens* var. *pruriens*	500163PY	Herbal drug vendor, Puducherry	-
40.	*M. pruriens* var. *pruriens*	500164PY	Herbal drug vendor, Puducherry	-

RESULTS AND DISCUSSION

PCA for quantitative and qualitative traits among total studied accessions

Principal component analysis, when applied to 22 quantitative characters indicated first two PCs to explain total variation of 66.34% of which 46.51% corresponded to PC 1 alone. According to values of correlation (Table 2), this was drawn across almost all the characters such as: Terminal leaf length (TLL), Terminal leaf width (TLW), Adjacent leaf length (ALL), Adjacent leaf width (ALW), Petiole length (PL), Flower length (FL), Pod length (POL), Pod width (PW), Seed length (SL), Seed width (SW), Seed thickness (ST), Individual seed weight (ISW), and Hundred seed weight (HSW). Other characters such as Days to Flowering (DF), Inflorescence length (IL), Number of buds per inflorescence (NBPI), Days to first mature pods (DFMP), Number of pods per cluster (NPPC) and Number of pods per plant (NPPP) corresponded to PC 2 explaining 19.83% of the variation. PC 3 showed 9.78% variation for characters like Days to emergence (DE) and Fertility index (FI), but, as the overall variation was less than 4%, this was considered less significant.

Representing these values in a graph (Figure 1a and 1b) it was observed that the plants are distributed in all the 4 quadrants of scatter plots indicating good variability for wild and cultivated accessions spread over large number of traits. Similar observation is also reported in *Dioscorea* spp. (Mwirigi *et al*., 2009). Pugalenthi and Vadivel (2007) have observed significant morphological variations among 11 *M. pruriens* var. *utilis* accessions collected from South India.

Table 2. Correlations between variables and PCs based on 22 quantitative characters

Characters	PC 1	PC 2	PC 3	PC 4
Days to emergence	-0.139	0.111	0.542	**0.624**
Terminal leaf length	**0.780***	0.431	0.329	-0.128
Adjacent leaf length	**0.777**	0.451	0.328	-0.150
Terminal leaf width	**0.807**	0.295	0.448	-0.127
Adjacent leaf width	**0.802**	0.360	0.409	-0.097
Petiole length	**0.711**	0.244	0.458	-0.153
Days to flowering	-0.585	**0.678**	0.148	0.237
Inflorescence length	-0.101	**0.822**	-0.413	-0.019
No. of flower buds per inflorescence	-0.137	**0.847**	-0.377	-0.027
Flower length	**0.657**	0.329	-0.338	0.357
Days to first mature pods	-0.627	**0.639**	0.171	0.250
Pod length	**0.932**	0.072	-0.092	0.078
Pod width	**0.657**	-0.022	-0.332	-0.018
No. of pods per cluster	-0.231	**0.562**	-0.273	-0.119
No. of pods per plant	-0.111	**0.797**	0.070	-0.260
Days to maturity	-0.783	0.236	0.347	0.185
Seed length	**0.859**	-0.105	-0.253	0.195
Seed width	**0.849**	-0.080	-0.104	0.162
Seed thickness	**0.893**	-0.032	-0.060	0.114
Individual seed weight	**0.923**	0.017	-0.142	0.187
Hundred seed weight	**0.958**	-0.064	-0.128	0.098
Fertility index	0.014	-0.531	**0.392**	-0.053

*values in bold indicate the most relevant character that contributed most to the variation of the particular component.

Among 9 qualitative characters, the total variation of 67.50% was explained by first two principal components with many characters falling among these two (Table 3). PC 1 explained 45.43% of variation in the data set which was highly correlated to characters such as Leaf texture (LTX), Plant hairiness (PH), Pod itchiness (PI), Seed color (SC) and Seed texture (STX); while 22.07% variation explained by PC 2 was related to characters such as Pod curvature (POC) and Pod trichome color (PTC).

Table 3. Correlations between variables and PCs based on 8 qualitative characters

Characters	PC 1	PC 2	PC 3	PC 4
Leaf texture	**0.762***	0.148	0.348	**0.458**
Plant hairiness	**0.704**	0.516	0.129	-0.246
Pod trichome color	-0.710	**0.389**	**0.401**	-0.315
Pod curvature	0.206	**0.845**	-0.270	-0.099
Pod itchiness	**0.849**	0.197	-0.102	0.027
Seed color	**0.675**	-0.458	-0.379	-0.334
Seed texture	**0.616**	-0.377	0.531	-0.286

*values in bold indicate the most relevant character that contributed most to the variation of the particular component.

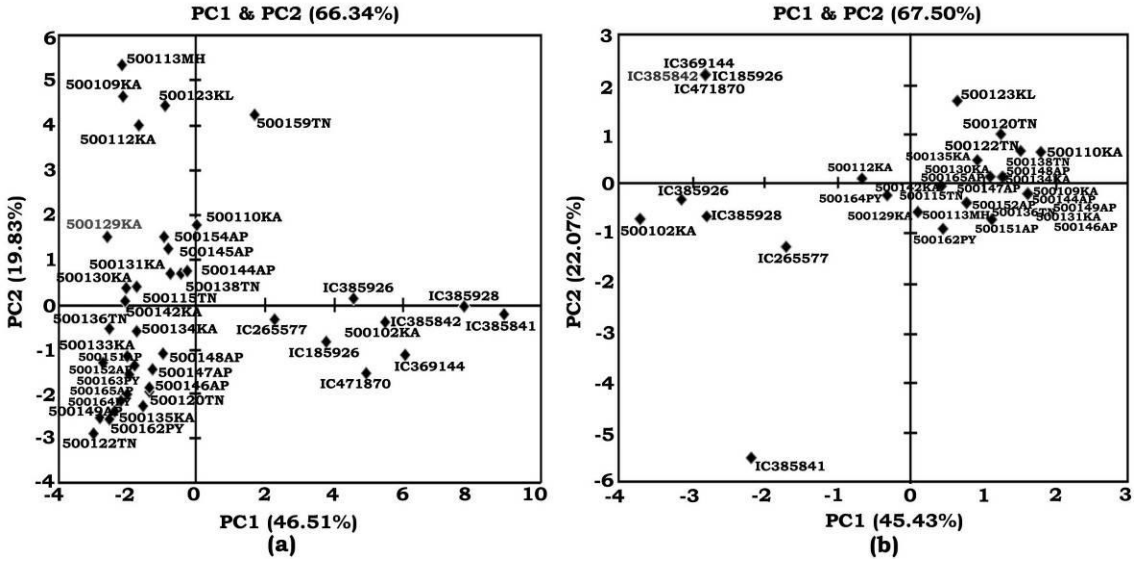

Figure 1. Principal component analysis carried out for 40 germplasm accessions: Scatter plots obtained for (a) 22 quantitative and (b) 9 qualitative characters.

Cluster Analysis

Results of cluster analysis demonstrated good variability (SI-13.7 to 93.3%) among the studied accessions (Fig. 2) with broad delineation of wild and cultivated genotypes in two distinct clusters. All the cultivated Velvetbean accessions grouped in cluster I, while wild forms separated in cluster II. Greater variability, as evidenced by similarity values ranging from 0.38-0.93 with mean at 0.65, was observed within wild genotypes while cultivated accessions showed similarity value ranging from 0.56-0.93 with mean at 0.75. Cluster II also showed formation of many sub-clusters and out-grouped accessions possibly due to representation of diverse genotypes in this group. No geographical affiliation, however, was observed for any of the two groups except for few instances, wherein, accessions from Andhra Pradesh grouped together (500146AP, 500147AP, 500148AP, 500151AP, 500152AP and 500165AP). The correlation coefficient (r) value of 0.96 between the data matrix and the tree matrix determined through Mantel test (Mantel, 1967) indicated that the cluster analysis is well represented by the data matrix based on the phenotypic data. Padmesh *et al*. (2006) using RAPD analysis have also reported separation of *Mucuna pruriens* var. *utilis* (cultivated) and var. *pruriens* (wild) accessions collected from Kerala in two distinct clusters with var. *pruriens* exhibiting greater diversity (SI-53 to 93%) than var. *utilis* (SI-72 to 92%). Our observation is in further conformity with the observations of these authors.

PCA for wild and cultivated accessions

The results of PCA carried out separately for wild and cultivated accessions are given in Fig. 3. Within wild group, the quantitative characters revealed 63.19% of total variation from PC 1 and PC 2 of which, 44.68% contributed by leaf and pod characters was concentrated in PC 1. The PC 2 had maximum contribution from seed characters showing 18.51% of the total variation. For qualitative characters, the PC 1 and PC 2 values presented the variation of 39.66% and

25.05% respectively together explaining 64.71% of the total variation. Within cultivated accessions the percentage variation given by PC 1 (41.75) and PC 2 (18.12) for quantitative traits showed 59.87% of variance which was less in comparison to the wild accessions. However, the latter was found to be more diverse for qualitative traits with PC 1 and PC 2 explaining 66.75% of the total variance which might due to diversity in flowers (white and purple flowers) and pod coat colour (velvety black and green) as against only one flower colour (purple) and pod coat colour (orange bristles) in wild genotypes. The alignment of accessions in PCA scatter plots (Fig. 3 a-d) confirmed good variability for quantitative and qualitative characters among both wild and cultivated germplasm. Previous study by Pugalenthi and Vadivel (2007) on agro-biodiversity of 11 cultivated *M. pruriens* var. *utilis* accessions has demonstrated considerable variability within this group. However, their low variability in the present study might be due to difference in the accessions sampled by us and the approach of comparative evaluation adopted.

Identification of elite germplasm for yield traits

Twenty two quantitative characters evaluated in the present study also included six yield based traits such as: days to flowering, number of flower buds per cluster, number of pods per cluster, days to maturity, hundred seed weight and fertility index which are important from breeding perspective. Hence, a detailed analysis was carried out on data from these traits to understand their diversity among the on-hand accessions and to identify key variants in each group based on $LSD_{5\%}$ score (data not shown). The results are summarized in Table 4; Figures 4 and 5.

The number of days for flowering varied from minimum 50 days to maximum of 190 days dividing 40 accessions into four major frequency classes (Fig. 4a). Nearly 50% of the accessions belonged to early flowering type (50-85 days), while 5% exhibited tendency for late flowering (155-190 days). Cultivated plants exhibited greater prevalence of early flowering (72%) (Figure 4c) when compared to wild accessions, in which, majority of the accessions were in the medium range (85-155 days) of flowering days (Fig. 4b). The least significant difference ($LSD_{5\%}$) scores indicated that the genotypes 500112KA, 500133KA and 500135KA among the wild and 500159TN and IC369144 of the cultivated group are of interest in breeding for early flowering trait. The number of days for flowering is an important trait, which in addition to other advantages, reduces the incidence of pests and diseases, thereby improving the yield and quality of the pods and seeds (Pugalenthi and Vadivel, 2007).

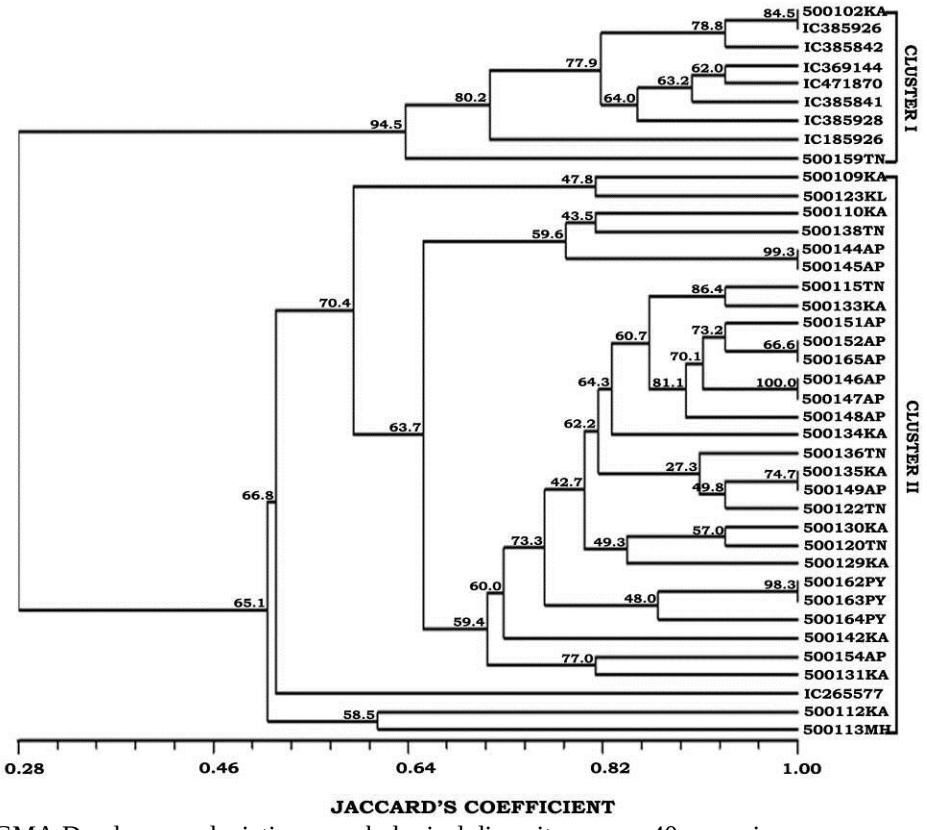

Figure 2. UPGMA Dendrogram depicting morphological diversity among 40 accessions

Earliness, determined by the plants ability to reach flowering and maturity cycle in a short period is another important trait associated with the yield. Reducing crop cycle has, in many cases, helped to avoid unfavorable temperatures and low air humidity during flowering and podding stages (Angelova and Stoilova, 2008). The days to maturity among the studied accessions varied from minimum of 90 days to maximum of 270 days dividing 40 accessions into four major frequency classes (Figure 4d). About 20% of the accessions in our germplasm exhibited tendency for early maturity (90-135 days), while 5% were late maturing; as many as 50% grouped in medium range of maturity period (180-225 days). Among the two groups studied, cultivated plants exhibited greater prevalence of early maturity (72%) (Figure 4f) when compared to wild accessions, in which, majority belonged to medium range of maturity days (180-225 days) (Figure 4e). Based on $LSD_{5\%}$ value, accessions IC471870, IC385928 with 500159TN were noted to be important for breeding maturity period in *M. pruriens*.

The hundred seed weight among different accessions varied from minimum of 13 g to maximum of 205 g dividing 40 accessions into six frequency classes (Figure 4g). Sixty four percent of our germplasm was found to be poor seed yielding type (13-45 g), while 5% exhibited high yielding nature (141-205 g). One of the accessions, IC385841 showed very high yield of 205 g/100 seeds and is of interest in genetic improvement programs. Among the wild and cultivated groups, almost all the wild accessions were in the range of very low to low seed yield (13-77 g) (Figure 4h); while cultivated plants exhibited extensive variation ranging from 45 g to 205 g with 64% of them peaking the medium range (109-141 g) (Figure 4i). Germplasm accessions 500159TN, IC471870 and IC385841 based on $LSD_{5\%}$ value were found to be important for breeding seed yield trait in *M. pruriens*. The fertility index (FI) is an important agronomical character which has direct bearing on the seed yield (Pugalenthi and Vadivel, 2007). In the present study, the FI among the different germplasm accessions varied from minimum of 17 to maximum of 100 dividing 40 accessions into seven frequency groups (Figure 5j). Large variability was observed among both wild and cultivated accessions with almost no preference for any particular range (Figure 5 k-l). The least significant difference ($LSD_{5\%}$) scores indicated that accessions 500159TN, IC385926 and IC369144 are diverse genetic stocks for this trait.

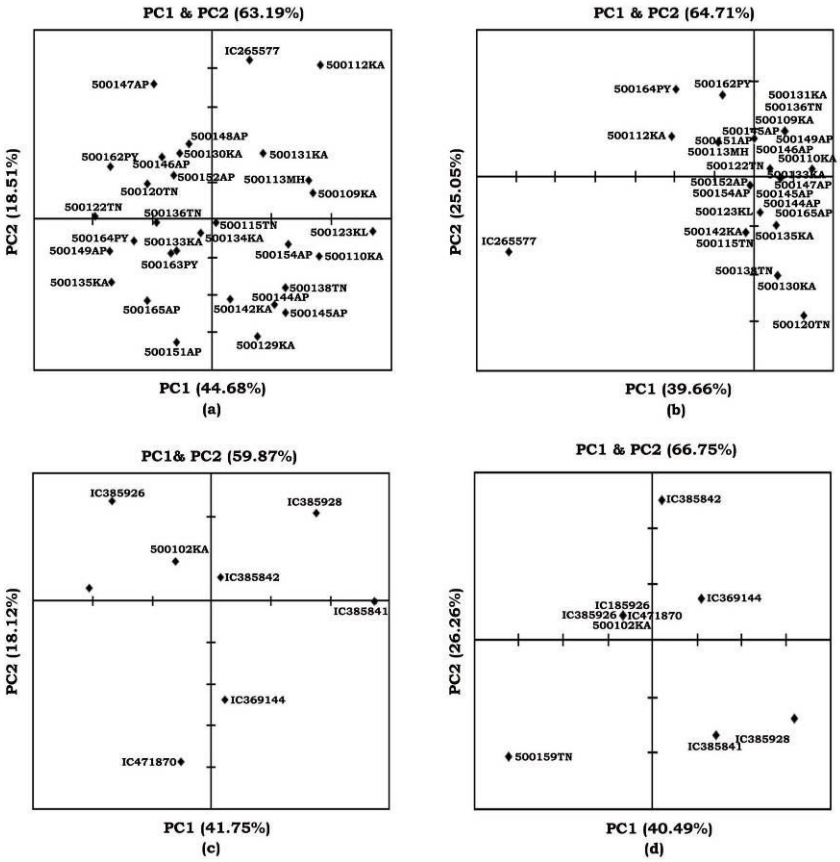

Figure 3. Principal component analysis for wild (a & b) and cultivated (c & d) accessions carried out separately for quantitative (a & c) and qualitative (b & d) characters.

The number of flower buds per cluster ranged from 2 to 78 among different accessions dividing 40 accessions into six frequency classes (Figure 5m) with 57.50% of them placed under 'low' (less than 13); 25% under 'medium' (>13 but less than 30); 12.5% under 'high' (>30 but less than 52) and 5% under 'very high' pod setting (>52) category (Figure 5 m-o). The $LSD_{5\%}$ scores showed that accessions 500113MH, 500149AP, 500165AP and 500109KA among the wild and 500159TN and IC471870 of the cultivated groups are of interest in breeding programs. Similarly, for pod setting ability, determined through number of pods/cluster, of the 6 frequency classes formed (Figure 5p), 60% of the accessions showed poor pod setting ability (less than 9); 33% were in the medium range (>9 but less than 23) and only 6% showed tendency for high pod setting (>29) (Figure 5 q-r). Accessions 500136TN, 500162PY and IC471870 based on $LSD_{5\%}$ value were noted to be important for breeding programs.

Table 4. Variability for Quantitative characters among *M. pruriens* accessions

Character	**Range**	**Mean**	**SD**	**CV%**	**F-ratio**	**F-test**	**$LSD_{5\%}$**
Days to emergence	5-16 days	6.93	1.98	28.49	9.21	*	0.019
Terminal leaf length	4.78-11.73 cm	8.43	2.30	27.59	8.76	*	0.029
Adjacent leaf length	4.83-11.35 cm	8.25	2.22	27.18	8.94	*	0.027
Terminal leaf width	2.98-8.68 cm	5.27	1.69	32.38	11.78	*	0.013
Adjacent leaf width	2.92-8.38 cm	5.25	1.63	31.29	9.20	*	0.014
Petiole length	6.00-16.40 cm	5.98	2.54	42.07	16.86	*	0.031
Days to flowering	52 – 240 days	94.24	34.64	37.04	32.25	**	0.332
Inflorescence length	1.38-93.0 cm	13.76	16.66	121.1	94.99	*	0.186
No. of flower buds / inflorescence	5-84 flowers	17.28	17.87	103.41	28.80	**	0.660
Flower length	4.05-5.05 cm	4.54	0.361	8.00	37.76	NS	0.000
Days to first mature pods	65-280 days	117	37.49	32.45	11.46	*	0.115
Pod length	6.00-16.4 cm	9.64	2.60	27.26	85.11	*	0.005
Pod width	1.20-2.70 cm	2.09	1.34	65.15	1.27	*	0.028
No. of pods / cluster	3-33 pods	17.13	7.60	78.12	17.89	**	0.158
No. of pods/ plant	15-340 pods	130	90.72	69.79	22.43	**	0.143
Days to Maturity	90-360 days	178	50.05	28.12	44.02	**	0.127
Seed length	6.92-18 mm	11.40	3.24	28.59	16.28	*	0.037
Seed width	4.98-15.19 mm	8.78	2.58	29.55	15.53	*	0.024
Seed thickness	3.01-12.02 mm	5.71	2.13	38.08	14.26	*	0.017
Individual seed weight	0.13-2.02 g	0.56	0.476	38.38	32.33	**	0.257
Hundred seed weight	12.80-200.9 g	54.40	44.29	82.78	54.10	**	0.713
Fertility index	17.30-98.50 %	64.80%	22.18	34.66	66.06	**	0.632

*Significant at P< 0.05; ** Highly Significant at P<0.05; NS- non-significant

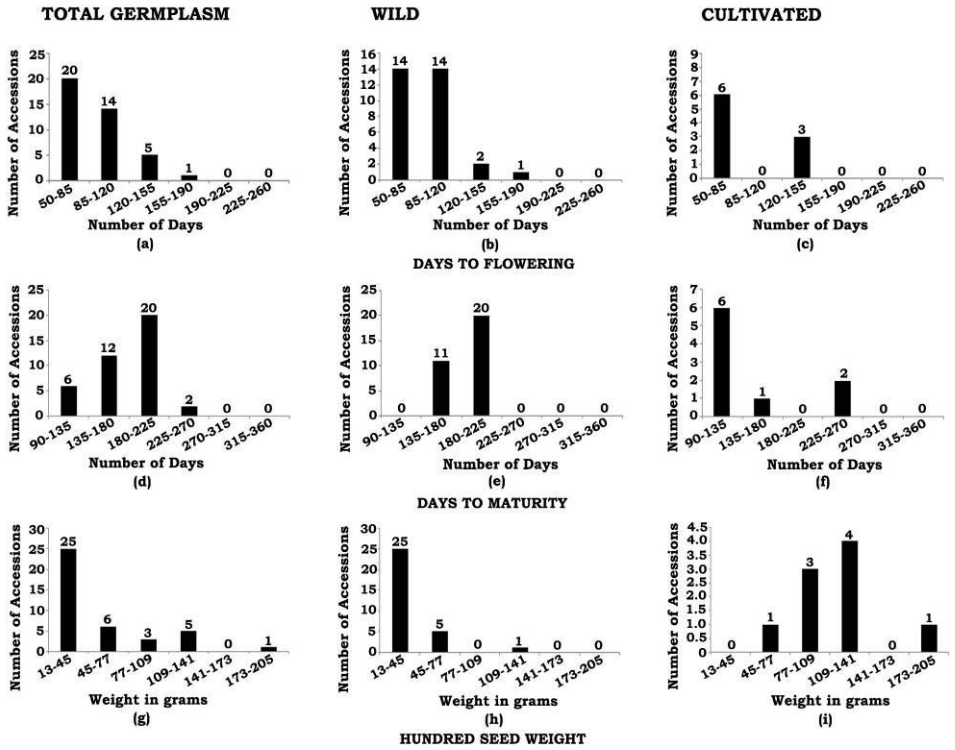

Figure 4. Frequency distribution analysis of trait values carried out for total germplasm as well as wild & cultivated accessions independently in three yield traits *viz.*, days to flowering (a-c); days to maturity (d-f); hundred seed weight (g-i).

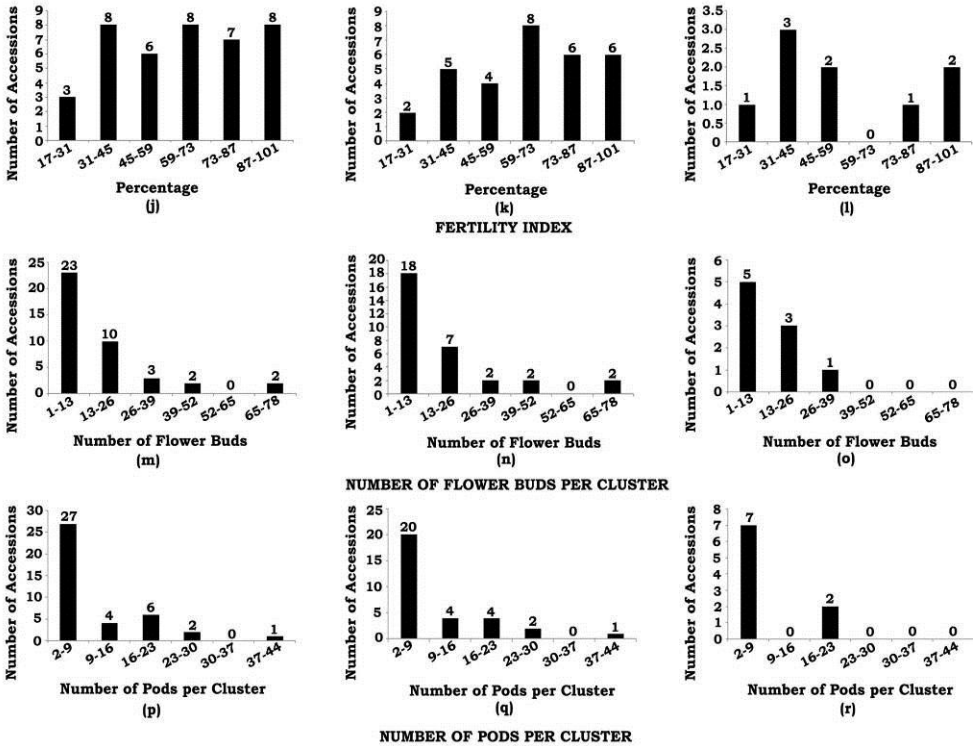

Figure 5. Frequency distribution analysis of trait values carried out for total germplasm as well as wild & cultivated accessions independently in three yield traits *viz.*, fertility index (j-l); number of flower bud per cluster (m-o) and number of pods per cluster (p-r).

CONCLUSIONS

The overall results from the present analyses indicated that 40 on-hand accessions are diverse for several morpho-agronomical characters with potential for exploitation in breeding programs. It has also revealed good scope for introducing wild germplasm in the ongoing breeding programs, as improvement efforts so far have concentrated only around cultivated genotypes. Less variability recorded for cultivated genepool call for need to enhance the genetic base of these plants through mutation breeding, wide-hybridization and other approaches. At the same time, considering that fact isolated use of morphological markers might lead to incomprehensive conclusions due to inherent limitation of this marker (Smith and Smith, 1992, Leela Tatikonda *et al.*, 2009), it is desirable to overlay the results of present findings with those from DNA-based markers to gain realistic insight into diversity pattern represented in the current *M. pruriens* collection before embarking on specific breeding program.

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