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IN-SITU ASSESSMENT OF PHENOTYPIC DIVERSITY IN *CAJANUS SCARABAEOIDES* (L.)THOUARS

NILAMANI DIKSHIT[1]*, VISHNU K GITE[2] AND NATARAJAN SIVARAJ[3]
[1]ICAR-National Bureau of Plant Genetic Resources,
Regional Station, Dr. P.D.K.V. Campus, Akola - 444 104 Maharashtra, INDIA
[2]Agricultural Research Station, Badnapur, Jalna - 431 202, Maharashtra, INDIA
[3]ICAR-National Bureau of Plant Genetic Resources, Regional Station, Hyderabad - 500 030,Telangana, INDIA
e-mail: dikshitn@gmail.com

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***Corresponding author**

ABSTRACT
In-situ assessment of phenotypic diversity in 13 wild populations of *Cajanus scarabaeoides* (L.) Thouars were carried out in five districts of Vidharbha region of Maharashtra state, India. Significant variability in qualitative traits ranging from stem pigmentation (light green, green to purple and some accessions turned even red during maturity), flower pattern (sparse to medium streaks on yellow flower) and seed coat colour (brown, mottled to black) were observed. Plant height ranged from 80.0-190.0 cm, number of branches per plant (3.0-10.0), pod length (1.7-3.0 cm), pod width (0.7-1.0 cm), number of seeds per pod (4.0-6.0), number of pods per plant (5.7-30.0) and 100 seed weight (1.47-3.02g). Promising lines were identified for select traits which can be utilized in pigeonpea crop improvement programme. For example, D/G-9, a collection from Gujgawan, Chandrapur district was recorded promising for plant height (190.0 cm), number of seeds per pod (6.0) and number of pods per plant (30.0). DIVA-GIS grid maps were generated to delineate the diversity rich pockets for select quantitative traits. GIS analysis indicated that Northern Vidarbha region was the rich pocket of *C. scarabaeoides* diversity.

INTRODUCTION

Pigeonpea is a multi-use pulse crop and a source of protein in the vegetarian diet. It is widely grown by the resource poor farmers of semi-arid and rainfed tropics and the crop perform well in poor soils and regions where moisture availability is unreliable or inadequate. India has the largest area (3.38 million ha) and accounts for over 70% of the World's production and the per capita availability of protein is 28 g/day which is much lower than the FAO recommended level of 80g/day (Nagy *et al.*, 2013, Prasad *et al.*, 2013, Saroj *et al.*, 2013). It is followed by Myanmar (580,000 ha), China (150,000 ha), and Nepal (21,360 ha) (FAO, 2007). Though, considerable progress in pigeonpea crop improvement has been made, the crop still suffer from several problems such as biotic and abiotic stresses, low pod setting, etc.

The crop is attacked by a host of diseases, insect pests and prone to water logging, salinity etc. Broadening the genetic base and introduction of resistance genes for various biotic and abiotic factors will help in increasing the productivity of the crop. Exploitation of desirable traits available in the wild relatives of cultivated types is useful for dynamic crop improvement programme and therefore conservation and evaluation of secondary and tertiary gene pools assume great importance.

Wild *Cajanus* species have contributed desirable agronomic traits such as cytoplasmic male sterility (CMS) (Mallikarjuna and Saxena 2005), dwarf growth habit (Saxena and Sharma (1995) and high protein content (Saxena *et al.*, 2002). *Cajanus scarabaeoides* (L.) Thouars- an Indian origin wild relative of pigeonpea belongs to the secondary gene pool, possess genes for earliness and high pod setting in addition to multiple disease and pest resistance. The scope of utilization of wild relatives has been increasing gradually with our increasing capacity to study the genes responsible for various traits and their exploitation (Arora and Pandey, 1996). Stable cytoplasmic male sterile (CMS) and high seed protein content lines were derived from crossing *Cajanus cajan* with *C. scarabaeoides*. Morpho-agronomic diversity has been reported earlier in a world collection *of* 102 accessions *of Cajanus scarabaeoides* germplasm (Upadhyaya and Gowda 2009, Upadhyaya *et al.*, 2011). However, the *Cajanus scarabaeoides* collections of Vidarbha region has not been studied in detail so far. Increasing threats to natural habitats due to climate change, human settlement, overgrazing, irrigation project etc. make it imperative to explore diverse collection of *C. scarabaeoides* before valuable material is lost forever. Hence, the objective of the programme was to collect and conserve wild germplasm from Vidarbha region of Maharashtra state and assess the phenotypic diversity of *C. scarabaeoides in-situ*.

MATERIALS AND METHODS

In order to collect the gene pool of wild species of *Cajanus scarabaeoides,* an exploration and collection programme was undertaken in selected five districts of Vidarbha region of

Maharashtra during 7th Nov. -16th November, 2013. The districts explored include Akola, Yavatmal, Chandrapur, Bhandara and Gondia. The planning and sampling procedures were followed as per the guidelines suggested by Engels *et al.*, (1995) and Brown and Marshall (1995). For *in-situ* assessment and to characterize the wild species of *C. scarabaeoides*, 13 diverse sites with good plant population were identified and at each site five random plants were selected. In-situ assessment was carried out by analysing the eight quantitative traits *viz.* plant height, number of branches per plant, number of pods per plant, pod length, pod width, number of seeds per plant, 100 seed weight and trachoma density and six qualitative traits *viz.* stem pigmentation, leaflet shape, flowering pattern, flower streak pattern, seed coat colour pattern and seed shape. The measurements were recorded as per the methodology suggested (Remanandan *et al.,* 1988). Hand held geographical positioning system (GPS-Garmin 12) was used to record the geographical coordinates. Digimatic calliper (Mitutoyo Corporation, Japan) was used to record some of the pod traits. DIVA-GIS software version 7.5.0 (Hijmans *et al.*, 2012) was used for diversity analysis of *C. scarabaeoides* (www.diva-gis.org). Statistical Analysis Software, SAS Enterprise Guide 4.2 was used for statistical analysis.

RESULTS AND DISCUSSION

Cajanus scarabaeoides is an Indian origin wild relative of pigeon pea having source for stable CMS, resistant to pigeon pea pod borer (*Helicoverpa armigera*), pod fly and has multiple disease resistance. In Vidarbha region, *Cajanus scarabaeoides* occur in open grasslands, dry scrub vegetation on hill slopes and ridges between cultivated fields. It was also located along the road side and water canals where reasonable amount of sunlight was available. However, its population was low in the deep inland bushes and dark forests. The source of *in-situ* assessment and germplasm collection sites are provided in Table 1. DIVA-GIS mapping of collection and *in-situ* assessment sites of *C. scarabaeoides* is given in Fig.1. Significant variations were observed for quantitative and qualitative traits among the 13 accessions collected from diverse sites of Vidarbha region. Morphological characteristics and variability observed in *Cajanus scarabaeoides in-situ* from Vidarbha region of Maharashtra are provided in Table 2.

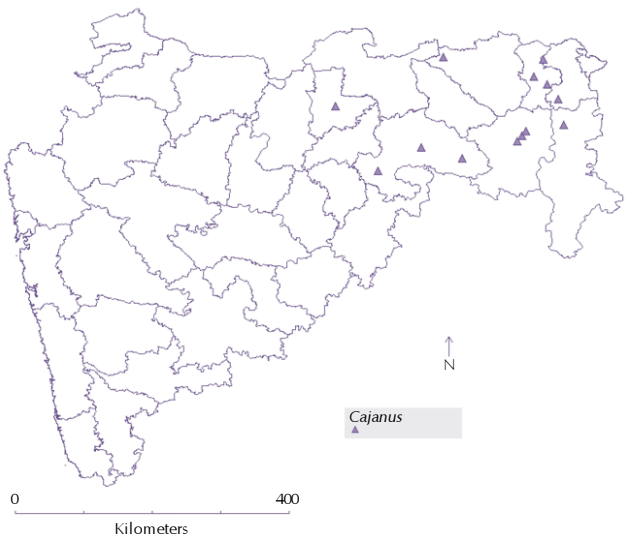

Figure 1: Map showing the collection sites of *Cajanus scarabaeoides* in parts of Vidarbha region of Maharashtra state, India

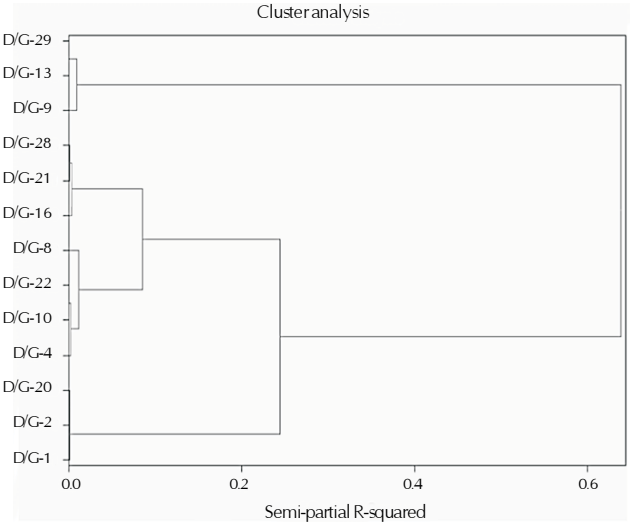

Figure 2: Ward's minimum variance dendogram generated for *Cajanus scarabaeoides* germplasm assessed *in-situ*

Table 1: Source particulars of *Cajanus scarabaeoides* for *in-situ* assessment in Vidarbha region of Maharashtra state, India

Coll. No.	Place of collection/ District	Description of the site	Associated species	Latitude	Longitude
D/G-1	Akola/ Akola	Road side, lateritic soil	Grass species	20°43′ 701	77°04′ 507
D/G-2	Kasola/Yavatmal	Field bund, lateritic soil	*Hyptis suaveolens*	19°51′ 500	77°38′ 882
D/G-4	Parbha/Yavatmal	Fencing, road side	-	20°10′ 363	78°12′ 040
D/G-8	Moharli/Chandrapur	Road side forest, periphery of Tadoba forest	Ber, forest plant	20°19′ 083	79°33′ 559
D/G-9	Gujgawan/Chandrapur	Road side	*Hyptis suaveolens*, Ber, *Barleria*	20°15′ 83	79°29′ 998
D/G-10	Sindewahi/Chandrapur	Farm fencing	Ber	20°28′ 803	79°66′ 104
D/G-13	Chandapur/Chandrapur	Road side	*Hyptis suaveolens*	20°01′ 773	78°45′ 085
D/G-16	Sawaragaon/Chandrapur	Road side (social forestry)	*Hyptis suaveolens*	20°23′ 183	79°36′ 904
D/G-20	Palladi/Bhandara	Hillock, stone cutting site	*Hyptis suaveolens*	21°07′ 825	79°42′ 247
D/G-21	Sawarbandh/Bhandara	Canal side	*Hyptis suaveolens*	21°01′ 905	79°53′ 128
D/G-22	Arjuni Moregaon/Gondia	Road side	*Hyptis suaveolens*	20°49′ 902	80°02′ 228
D/G-28	Mundikota/Gondia	Road side	*Hyptis suaveolens*	21°21′ 675	79°50′ 423
D/G-29	Hiware/Bhandara	Road side	*Hyptis suaveolens*	21°23′ 562	78°30′ 572

Table 2: Morphological characteristics and variability observed in *Cajanus scarabaeoides* from Vidarbha region of Maharashtra

Coll. No.	Plant height (cm)	No. of branches	Pod length (cm)	Pod width(cm)	No. of seeds /pod	No. of pods /plant	Seed length (mm)	Seed width (mm)	100 seed weight (g)
D/G-1	80.0	7.2	2.2	0.75	5.5	12.0	4.48	3.6	2.56
D/G-2	81.4	7.3	2.7	0.86	4.75	05.7	4.42	3.18	2.16
D/G-4	112.2	7.6	3.0	1.0	6.0	15.0	4.08	3.06	3.02
D/G-8	100.6	8.2	1.7	0.8	4.0	15.2	4.0	2.92	1.47
D/G-9	190.0	10.0	3.0	0.9	6.0	30.0	4.08	3.0	2.0
D/G-10	120.4	6.5	1.9	0.6	4.6	13.0	4.08	3.0	1.75
D/G-13	180.2	8.2	2.8	0.9	6.0	20.0	4.0	3.0	1.67
D/G-16	150.3	7.2	2.4	0.8	5.8	17.0	4.04	3.04	1.73
D/G-20	80.2	3.0	1.8	0.7	5.2	06.0	4.04	2.98	1.84
D/G-21	140.4	6.2	2.0	0.8	5.8	18.0	4.08	3.0	1.85
D/G-22	120.2	5.4	2.2	0.7	5.4	15.0	3.9	2.96	1.59
D/G-28	140.0	4.4	2.0	0.7	5.4	12.0	4.18	2.92	1.80
D/G-29	180.2	8.2	2.8	0.8	5.8	16.0	4.0	3.12	1.87
Descriptive Statistical Analysis									
Minimum	80.0	3.0	1.7	0.6	4.0	5.7	3.9	2.92	1.47
Maximum	190.0	10.0	3.0	1.0	6.0	30	4.48	3.6	3.02
Mean	128.9	6.9	2.3	0.8	5.4	15.0	4.1	3.06	1.94
Standard Error Mean	10.72	0.5	0.13	0.03	0.17	1.7	0.04	0.05	0.12
CV%	30.0	26.5	19.81	13.39	11.49	41.0	4.04	5.81	21.7

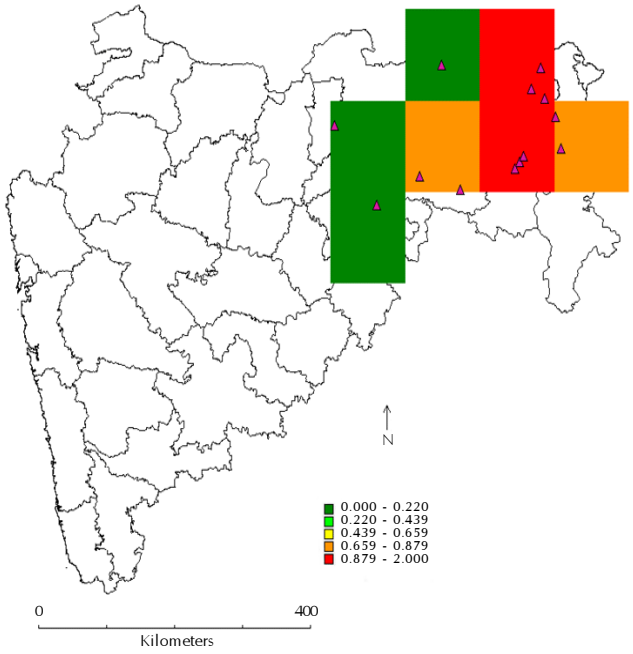

Figure 3: DIVA-GIS grid map generated for diversity analysis of trait pods per plant in *Cajanus scarabaeoides*

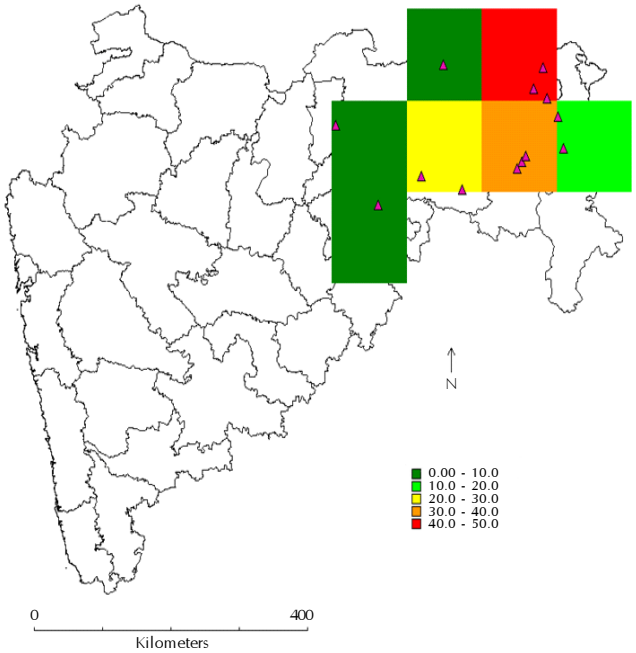

Figure 4: DIVA-GIS grid map generated for statistical analysis (Co-efficient of Variation %) of trait pods per plant in *Cajanus scarabaeoides*

All the accessions of *C. scarabaeoides* expressed the creeper-climber growth habit. It is mostly supported by the plant species *Hyptis suaveolens* in majority of the explored sites. In the forest area of Chandrapur district, it is also associated with *Ber* (*Ziziphus* sp.) and desert forest plants. Stem pigmentation trait varied from light green, green to purple and some accessions turned even red during maturity. Leaves are pinnately trifoliate, lower surface densely pubescent and leaflet varied from lanceolate to ovate shape in most of the accession. Flowering habit is indeterminate and sporadic and its duration ranged from August to late November. Flower pattern varied from sparse to medium streaks on yellow flower. Pods are oblong, purple or dark purple colour, densely covered with golden brown long and short hairs. The observed trichome density on pods varied from sparse to medium dense and dense pubescent. Seeds are rectangular, rounded, brown, mottled to black colour. Upadhyaya *et al.* (2011) observed the similar type of variation in *C. scarabaeoides* accession for stem pigmentation, leaflet shape, flower pattern, seed coat colour and trichome density.

A habitat of *C. scarabaeoides*

Collection from field bund

Flowering branch

Twining nature of *C. scarabaeoides*

***C. scarabaeoides* plant with pods**

Close view of pods

Twining red stem with a pod

***C. scarabaeoides* on a fence**

Figure 5: *Cajanus scarabaeoides*, a wild relative of pigeonpea

In-situ phenotypic diversity assessment revealed that the maximum variation was observed for plant height which was ranged from 80.0 cm in D/G-1 accession to 190.0 cm in D/G-9 accession (Table 2). The branches are straight or winding, quite woody at base. Number of branches per plant ranged from 3.0 in D/G-20 accession to 10.0 in D/G-9 accession. Pod length varied from 1.7-3.0 cm and pod width varied from 0.7-1.0 cm. The maximum number of pods per plant was observed in D/G-9 accession (30.0) while minimum was observed in D/G-2 accession (5.7). Number of seeds varied from 4.0-6.0 per pod and 100 seed weight ranged from 1.47g in D/G-8 accession and 3.02g in D/G-4 accession. Ward's minimum variance dendrogram generated for the in-situ phenotypic assessment of quantitative characters is provided in Fig. 2. The germplasm accessions of *C. scarabaeoides* were grouped into three clusters at the semi-partial R squared distance of 0.6.

DIVA-GIS grid maps generated for diversity and statistical analyses (CV %) for the trait pods per plant in *Cajanus scarabaeoides* are provided in figures 3 and 4 respectively. This study indicated that diversity rich pockets for *C. scarabaeoides* distributed in Northern Vidarbha region of Maharashtra. DIVA-GIS have been successfully used for *in-situ* assessment of *Jatropha curcas* (Sunil *et al.*, 2008). Further, it has been successfully used in assessing biodiversity and in identifying areas of high diversity in *Phaseolus* bean (Jones *et al.*, 1997); wild potatoes (Hijmans, and Spooner 2001); medicinal plants and agrobiodiversity (Varaprasad *et al.*, 2007, 2008), *Jatropha curcas* (Sunil *et al.*, 2009), linseed (Sivaraj *et al.*, 2012) and Proso millet (Dikshit and Sivaraj 2013). As many crop wild relatives worldwide including *Cajanus scarabaeoides,* are currently threatened with loss of diversity and/or extinction (Stolton *et al.*, 2006), the *in-situ* assessment study will be useful in assessing diversity rich pockets for planning *in-situ* conservation strategies in the future.

C. scarabaeoides is a useful genetic resource for Pigeonpea breeders. Besides having earliness, high pod setting, high protein content, pest and disease resistance, it is useful for development of stable CMS lines (Tikka *et al.*, 1997). Development of *C. scarabaeoides* based CMS system has shown promise in breaking the yield barriers in Pigeonpea. Keeping this in view, D/G-9 a collection from Gujgawan, Chandrapur district was recorded promising for plant height (190.0 cm), number of seeds per pod (6.0) and number of

pods per plant (30.0). Hence, this accession may be utilized in further breeding programme. Thus, the wild relatives of crop plants are important resources of good agronomic traits variability in terms of tolerance/resistance to biotic and abiotic stresses. The collected germplasm has been conserved under Medium Term Storage module of the station for further distribution and utilization by breeders.

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