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Short Communication

First identification of core accessions of *Jatropha curcas* from India based on molecular genetic diversity

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Abstract

We report on identification of core collection from 192 accessions collected from 12 Indian states and five other countries based on 109 polymorphic amplified fragment length polymorphism (AFLP) markers. Pairwise Jaccard's similarity coefficient for accessions varied from 0.25 to 1 with a maximum genetic distance of 0.75 obtained between accessions Jc428 (from Mexico) and J204 (from Madurai, Tamil Nadu). Both UPGMA (Unweighted Pair Group Method of Arithmetic Averages) clustering and principal coordinate analyses showed similar grouping of accessions in three major clusters in which Mexican accessions clustered separately from Indian, Chinese and African accessions. Results obtained from analysis of molecular variance indicated that 59% of the genetic variation was distributed among the populations, while 41% of variation was within the populations. A total of 16 (8.3% of the entire collection) core accessions were identified, which contained the entire allelic diversity of 192 accessions with respect to the sampled AFLP loci. The core accessions would be highly useful for future genetic improvement of *Jatropha*. To the best of our knowledge, this is the first report on identification of core accessions in *Jatropha*.

Keywords: AFLP; biodiesel; core collection; genetic diversity; *Jatropha curcas*

Introduction

Jatropha curcas L. has been recognized as a major biodiesel crop (Fairless, 2007). It has a special feature as a renewable energy source due to its comparatively higher oil content than other non-edible oilseeds (Openshaw, 2000). It is native to Mexico and Central America from where it has been taken to different tropical and subtropical regions of the world (Heller, 1996; Pamidimarri and Reddy, 2014).

Availability of a genetically diverse germplasm is important for genetic improvement programmes. High phenotypic variability with respect to oil traits has been reported in Indian accessions of *J. curcas* (Kaushik *et al.*, 2007). However, most of the genetic diversity studies conducted using DNA-based markers have shown low genetic diversity within Indian accessions (Ranade *et al.*, 2008; Sujatha *et al.*, 2008; Tatikonda *et al.*, 2009; Mastan *et al.*, 2012).

Identification of a core collection is an important step towards management and utilization of genetic resources. The core collection, a subsample of the whole collection, typically comprises approximately 10% of all available accessions and is intended to provide a set of genetically

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diverse material (Brown, 1989; Schoen and Brown, 1993). A number of studies have used amplified fragment length polymorphism (AFLP) markers for identification of core collections due to their high abundance and ease of genotyping (Jansen and van Hintum, 2007; Bamberg and del Rio, 2014).

In this study, AFLP markers were used for characterization of genetic diversity and identification of core accessions from an existing collection of *J. curcas* accessions collected solely based on geographical locations. The accessions of this core collection are conserved in TERI's *Jatropha* germplasm and can be made available for research purpose on request.

Experimental methods

A set of 192 accessions collected from India and five other countries were used (Tables S1 and S2, available online). AFLP genotyping was carried out using the standard AFLP protocol (Vos *et al.*, 1995). The amplification products were labelled with ^{32}P and detected by autoradiography. Various marker attributes such as polymorphism information content (PIC) (Roldan-Ruiz *et al.*, 2000), marker index (MI) and resolving power (RP) (Prevost and Wilkinson, 1999) were calculated. The binary matrix was used to estimate genetic similarity using Jaccard's coefficient. Clustering was performed based on UPGMA (Unweighted Pair Group Method of Arithmetic Averages) using only polymorphic bands. All the above-mentioned analyses were performed using the NTSys-PC software (version 2.20; Exeter software, New York, USA) (Rohlf, 2000). The reliability and robustness of the dendrogram topology were analysed by bootstrap analysis (1000 re-samplings) using the DARwin software (version 5) (Perrier and Jacquemoud-Collet, 2006). Principal coordinate analysis (PCoA) and analysis of molecular variance were carried out using the software GenAlEx version 6.5 (Peakall and Smouse, 2006). Core collections were identified based on AFLP data using PowerCore version 1.0 through heuristic search option (Kim *et al.*, 2007).

Discussion

A total of 319 bands were obtained using five primer combinations, out of which 109 (34.1%) were polymorphic (Table S4, available online). Primer combination E-AA × M-CTA displayed the highest values for PIC (0.16), RP (4.55) and MI (3.32), whereas the primer combination E-AG × M-CTA had the lowest values for these attributes. Pairwise Jaccard's similarity coefficient for accessions ranged from 0.25 (Jc428 (Mexico) *vs.* J204 (Tamil Nadu, India) to 1. A large number of accessions showed 100% similarity to other accessions indicating existence of duplicate accessions. While most of the

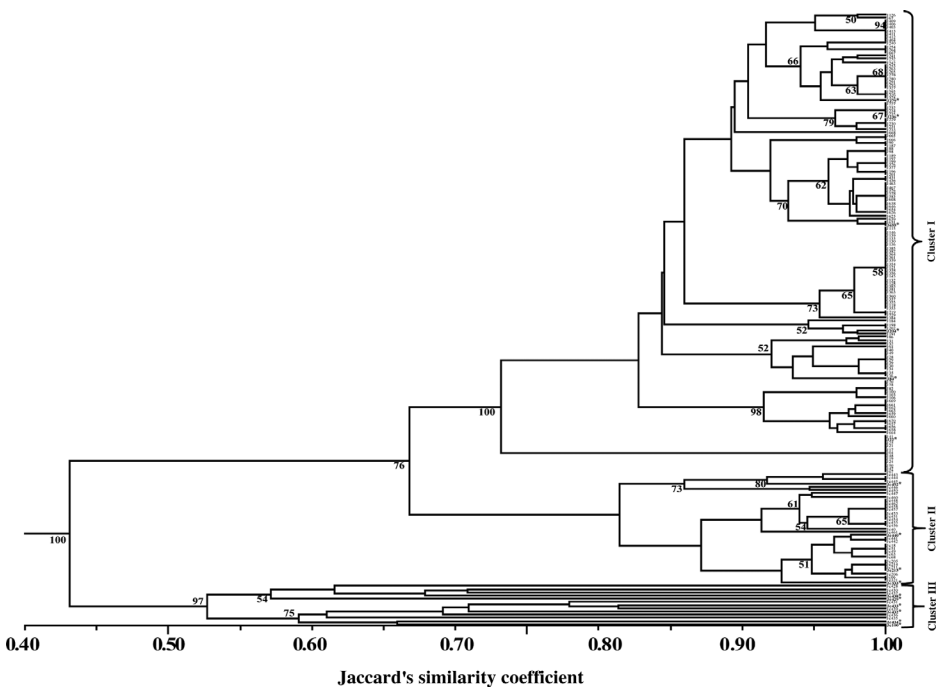

Fig. 1. Dendrogram showing clustering of *Jatropha curcas* accessions using the UPGMA method. Cluster I: Indian accessions; Cluster II: African, Chinese and Indian accessions; Cluster III: Mexican accessions. The asterisk marks (*) against the accession IDs denote core accessions. Bootstrap values greater than 50 were considered significant and are shown in the figure.

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Table 1. *Jatropha curcas* core accessions identified using AFLP data

S. No.	Core accessions	Country/State	Germplasm Institute	Cluster
1	J236	Haryana	HAU, Hisar	I
2	J84	Kerala	KAU, Thrissur	I
3	J22	Maharashtra	PDKV, Akola	I
4	J204	Tamil Nadu	MKU, Madurai	I
5	J655	Tamil Nadu	MSSRF, Chennai	I
6	J279	Uttarakhand	HAPPRC, Srinagar	I
7	Jc215	Congo	TERI, New Delhi	II
8	Jc452	Ethiopia	TERI, New Delhi	II
9	Jc333	Karnataka	TERI, New Delhi	II
10	Jc440	Rajasthan	TERI, New Delhi	II
11	Jc100	Mexico	TERI, New Delhi	III
12	Jc429	Mexico	TERI, New Delhi	III
13	Jc432	Mexico	TERI, New Delhi	III
14	Jc433	Mexico	TERI, New Delhi	III
15	Jc434	Mexico	TERI, New Delhi	III
16	Jc428	Mexico	TERI, New Delhi	III

duplicate accessions were found within and across the Indian states, several duplicate accessions across different countries were also found (Table S3, available online).

High genetic similarity has been reported in accessions from India, China, Singapore, Africa and Southern America (Ranade *et al*., 2008; Sujatha *et al*., 2008; Sun *et al*., 2008; Ambrosi *et al*., 2010; Wang *et al*., 2011). Low genetic diversity in *J. curcas* germplasm seems to be a common problem in all countries where it has been introduced. This may be a result of founder effect, which occurs due to introduction of a population of highly related individuals (Wright, 1931; Provine, 2004).

Out of the three major clusters obtained, Cluster I included 144 accessions all of which were from India. Cluster II contained 35 accessions from Congo, Ethiopia, Nigeria, China and India, which may be indicative of germplasm flow between these countries. Cluster III included 13 Mexican accessions revealing their high divergence from other accessions. This is in agreement with a prevalent view of Mexico and Central America being the centre of origin of *Jatropha* (Ovando-Medina *et al*., 2011; Pamidimarri and Reddy, 2014). PCoA also revealed similar grouping (Fig. 1 and Fig. S1, available online). Fifty-nine percent of the genetic variation was distributed among the clusters, while 41% of variation was within the clusters.

A total of 16 (8.3% of the entire collection) core accessions were identified using the PowerCore program (Table 1). The core accessions harboured the entire allelic diversity of the germplasm collection. Six accessions were selected each from clusters I (India) and III (Mexico), whereas four accessions were identified from cluster II (Congo, Ethiopia and India). Among the eight Indian accessions, four accessions were from southern India, two from northern India and one accession each from west and southwest India. The Indian core accessions thus represented seven states covering almost 90% of latitudinal span of the country (Table S1, available online). One of these accessions, Jc440, collected from northwest India (Rajasthan) was highly distinct with triangular fruits and extremely short internodes leading to a dwarf phenotype, which is highly desirable for high-density plantation and manual harvesting. Introduction of dwarfing genes such as *Rht* in wheat and *sd-1* in rice has a well-known history of success in increasing productivity of these crops during 1960s leading to green revolution (Spielmeyer *et al*., 2002; Borojevic and Borojevic, 2005). The current genetic and phenotypic information would enhance the optimal utilization of these accessions in future breeding programmes. It would be logical to pyramid together the various desirable traits such as dwarf plant height, large fruit size, etc., to develop varieties with increased seed yield. The core accessions could also be used as a reference for germplasm acquisition/introduction initiatives in India. Overall, the core accessions would be highly useful for future genetic improvement of *Jatropha*.

Supplementary material

To view supplementary material for this article, please visit http://dx.doi.org/10.1017/S1479262115000088

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References

Ambrosi DG, Galla G, Purelli M, Barbi T, Fabbri A, Lucretti S, Sharbel TF and Barcaccia G (2010) DNA markers and FCSS analyses shed light on the genetic diversity and reproductive strategy of *Jatropha curcas* L. *Diversity* 2: 810–836.

Bamberg J and del Rio A (2014) Selection and validation of an AFLP marker core collection for the wild potato *Solanum microdontum*. *American Journal of Potato Research* 91: 368–375.

Borojevic K and Borojevic K (2005) The transfer and history of "reduced height genes" (*Rht*) in wheat from Japan to Europe. *Journal of Heredity* 96: 455–459.

Brown AHD (1989) Core collections: a practical approach to genetic resources management. *Genome* 31: 818–824.

Fairless D (2007) Biofuel: the little shrub that could-maybe. *Nature* 449: 652–655.

Heller J (1996) *Physic Nut* Jatropha curcas *L. Promoting the Conservation and Use of Underutilized and Neglected Crops*. Gatersleben/Rome: Institute of Plant Genetics and Crop Plant Research/International Plant Genetic Resources Institute.

Jansen J and van Hintum T (2007) Genetic distance sampling: a novel sampling method for obtaining core collections using genetic distances with an application to cultivated lettuce. *Theoretical and Applied Genetics* 114: 421–428.

Kaushik N, Kumar K, Kumar S, Kaushik N and Roy S (2007) Genetic variability and divergence studies in seed traits and oil content of *Jatropha* (*Jatropha curcas* L.) accessions. *Biomass and Bioenergy* 31: 497–502.

Kim K-W, Chung H-K, Cho G-T, Ma K-H, Chandrabalan D, Gwag J-G, Kim T-S, Cho E-G and Park Y-J (2007) PowerCore: a program applying the advanced M strategy with a heuristic search for establishing core sets. *Bioinformatics* 23: 2155–2162.

Mastan SG, Pamidimarri DVNS, Rahman H, Ghosh A, Rathore MS, Ravi Prakash C and Chikara J (2012) Molecular characterization of intra-population variability of *Jatropha curcas* L. using DNA based molecular markers. *Molecular Biology Reports* 39: 4383–4390.

Openshaw K (2000) A review of *Jatropha curcas* L: an oil plant of unfulfilled promise. *Biomass and Bioenergy* 19: 1–15.

Ovando-Medina I, Sanchez-Gutierrez A, Adriano-Anaya L, Espinosa-Garcia F, Nunez-Farfan J and Salvador-Figueroa M (2011) Genetic diversity in *Jatropha curcas* populations in the State of Chiapas, Mexico. *Diversity* 3: 641–659.

Pamidimarri DVNS and Reddy MP (2014) Phylogeography and molecular diversity analysis of *Jatropha curcas* L. and the dispersal route revealed by RAPD, AFLP and nrDNA-ITS analysis. *Molecular Biology Reports* 41: 3225–3234.

Peakall R and Smouse PE (2006) GENALEX 6: genetic analysis in Excel. Population genetic software for teaching and research. *Molecular Ecology Notes* 6: 288–295.

Perrier X and Jacquemoud-Collet JP (2006) DARwin software, version 5.0.158. Available at http://darwin.cirad.fr/darwin

Prevost A and Wilkinson MJ (1999) A new system of comparing PCR primers applied to ISSR fingerprinting of potato cultivars. *Theoretical and Applied Genetics* 98: 107–112.

Provine WB (2004) Ernst Mayr: genetics and speciation. *Genetics* 167: 1041–1046.

Ranade SA, Srivastava AP, Rana TS, Srivastava J and Tuli R (2008) Easy assessment of diversity in *Jatropha curcas* L. plants using two single-primer amplification reaction (SPAR) methods. *Biomass and Bioenergy* 32: 533–540.

Rohlf FJ (2000) *NTSYS-pc: Numerical Taxonomy and Multivariate Analysis System, Version 2.2*. Setauket, NY: Exeter Software.

Roldan-Ruiz I, Dendauw J, VanBockstaele E, Depicker A and Loose MD (2000) AFLP markers reveal high polymorphic rates in ryegrasses (*Lolium* spp.). *Molecular Breeding* 6: 125–134.

Schoen DJ and Brown AHD (1993) Conservation of allelic richness in wild crop relatives is aided by assessment of genetic markers. *Proceedings of the National Academy of Sciences USA* 90: 10623–10627.

Spielmeyer W, Ellis MH and Chandler PM (2002) Semidwarf (*sd-1*), "green revolution" rice, contains a defective gibberellin 20-oxidase gene. *Proceedings of the National Academy of Sciences USA* 99: 9043–9048.

Sujatha M, Reddy TP and Mahasi MJ (2008) Role of biotechnological interventions in the improvement of castor (*Ricinus communis* L.) and *Jatropha curcas* L. *Biotechnology Advances* 26: 424–435.

Sun Q-B, Li L-F, Li Y, Wu G-J and Ge X-J (2008) SSR and AFLP markers reveal low genetic diversity in the biofuel plant *Jatropha curcas* in China. *Crop Science* 48: 1865–1871.

Tatikonda L, Wani SP, Kannan S, Beerelli N, Sreedevi TK, Hoisington DA, Devi P and Varshney RK (2009) AFLP-based molecular characterization of an elite germplasm collection of *Jatropha curcas* L., a biofuel plant. *Plant Science* 176: 505–513.

Vos P, Hogers R, Bleeker M, Reijans M, van de Lee T, Hornes M, Friters A, Pot J, Paleman J, Kuiper M and Zabeau M (1995) AFLP: a new technique for DNA fingerprinting. *Nucleic Acids Research* 23: 4407–4414.

Wang CM, Liu P, Yi C, Gu K, Sun F, Li L, Lo LC, Liu X, Feng F, Lin G, Cao S, Hong Y, Yin Z and Yue GH (2011) A first generation microsatellite- and SNP-based linkage map of *Jatropha*. *PLoS ONE* 6: e23632.

Wright S (1931) Evolution in Mendelian populations. *Genetics* 16: 97–159.