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South African Journal of Botany 79 (2012) 102–105

SOUTH AFRICAN JOURNAL OF BOTANY

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Genetic variability studies for morphological and qualitative attributes among *Jatropha curcas L.* accessions grown under subtropical conditions of North India

T.A. Wani, S. Kitchlu, G. Ram *

Biodiversity & Applied Botany Division, Indian Institute of Integrative Medicine (CSIR) Canal Road Jammu, India

Received 8 June 2011; received in revised form 17 October 2011; accepted 18 October 2011

Abstract

Evaluation of *Jatropha curcas* germplasm comprising seven accessions indicated a wide range of variability in vegetative growth and other qualitative attributes. These characteristics could be harnessed in future improvement programme of *Jatropha curcas*. Seed yield/plant had a positive and significant correlation with number of branches/plant, oil yield, plant spread (r=0.806, 0.802, 0.782), plant spread had a highest correlation with plant height (r=0.840). The seeds analyzed for proximate composition, fatty acid and physiochemical characteristics revealed that fiber and ash content in seed flour were high (16.5% and 4.35%). Oil content varied from 24.5% to 37.9%. The lower value of the viscosity suggests it as diesel oil. Accession JC006 could be an alternative source of linoleic acid (51%) while the accession JC001 could be a source for oleic acid (48%) and linoleic acid 42.4%. Stearic acid was highest in accession JC003 (42.9%).This evaluation has helped to identify cultivar with specific yield and vegetative growth features. Among all the seven accession evaluated accession JC007 is found to be promising which could be taken as productive genotype for commercial exploitation.
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Keywords: Biofuel; Correlation; *Jatropha curcas*; Oil composition; Seed oil content; Variability

1. Introduction

Plant species, which provide a diesel fuel substitutes, have received the interest globally in recent years (Farooqi et al., 2004). In this plant category *Jatropha curcas* L. (Euphorbiaceae) came into prominence. It is a small tree or shrub with smooth gray bark, which exudes a whitish colored, watery, latex when cut, ranges from 3 to 7 m in height. Ratan jyot or Safed-Erand is native of tropical America, but now thrives in many parts of the tropics and sub-tropics in Africa/Asia (Gubitz et al., 1999; Kumar and Sharma, 2008; Martinez-Herrera et al., 2006; Openshaw, 2000).The species serve as an eco-friendly renewable source of biodiesel replacing dependence on imported fossil fuel besides contributing towards economic upliftment of farmers, rejuvenation of wastelands/arid lands, reducing soil erosion (Datta and Pandey, 1993).the wood and fruit can be used for numerous purposes including fuel. The seeds of *Jatropha* contain viscous oil, which can be used for manufacture of candles and soap, in cosmetic industry, as a diesel substitute. These characteristics along with its versatility make it of vital importance to developing countries (Kumar and Sharma, 2008). The use of biodiesel has grown dramatically during the last few years. Feedstock costs account for a large percent of the direct biodiesel costs, including capital costs and returns (Bozbas, 2006). With no competing food uses, this characteristic turns attention to *Jatropha curcus* L., which grows in tropical and sub-tropical climates across the developing world (Openshaw, 2000). In Madagascar, Cape Verde and Benin, *Jatropha* oil was used as mineral diesel substitute during the World War II (Agarwal and Agarwal, 2007). In India, where 70% of the country requirement for diesel fuel is met through imports, developing an eco-friendly substitute domestically has a great relevance. In order to cultivate this

* Corresponding author. Tel.: +91 9419183639.
E-mail address: gandhiram123@rediffmail.com (G. Ram).

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doi:10.1016/j.sajb.2011.10.009

species on larger area it is highly desirable to select out elite clone for the purpose. Therefore, before starting a selection programme, a study of the genetic diversity among populations appears to be useful. As such, the present investigation was undertaken to determine the magnitude of variability for yield and oil composition among *Jatropha curcas* L. accessions collected from seven natural populations' resource base and identify productive accessions for commercial cultivation.

2. Materials and methods

The present investigation consisted of seven natural populations growing under subtropical conditions of Jammu and Kashmir State in Northwest part of India. Sampling of seven accessions (JCOOI-JC007) from seven different sites includes Jammu, Sunderbani, Rajouri, Kathua, Udhampur, Basohli and Vaishno Devi respectively. The minimum distance between locations was 30 km. The populations were grown in randomized block design with 3 replicates during 2007–2008 at Experimental Field Station of Indian Institute of Integrative Medicine(IIIM) Jammu (32°44′ (N) Latitude and 74°55′ (E) longitude and 400 m above the sea level with a temperature range Summer(30–44 °C), Winter (5–20 °C) and total annual rainfall of 506 mm). From each population 30 even aged phenotypically uniform plants were selected. The accessions were evaluated for various attributes viz. plant height, number of branches per plant, plant spread, leaf length, leaf width, seed yield per plant, and 100 seed weight.

Seeds being the part of commerce were harvested and air dried and was analyzed for crude fat, crude protein, crude fiber, ash, moisture, and carbohydrate according to standardized procedures (ISI-1980). Oil from seeds was recovered by solvent extraction, and analyzed for fatty acids by Gas chromatography (GC) equipped with auto-sampler and Flame Ionization Detector. FAME composition was analyzed on Shimadzu 17 A GC using SP 2380, column 30 meters × L.-D.O.25 mm × 0.251 lm film thickness using FID, 170 °C to 260 °C 4 °C/min. Injection and detector temperature were 260 °C and 280 °C, respectively. FAMEs were identified by comparison of retention times with those of standards. Fatty acid content was computed using GC area counts for different FAMEs. Crude fat, crude protein, crude, fiber, ash, moisture and carbohydrate (by difference) were analyzed by method of AOAC (2000). Mean, range of variations, coefficient of variations (CY %) and correlations were computed following Sundarajan et al. (1972).

3. Results and discussion

The results presented in (Table 1) reveals a wide range of variation in different parameters of vegetative growth ranging (CY) from 3.15% to 14.67%, highest being in number of branches/plant followed by seed yield/plant and lowest in oil yield. Seed yield/plant, oil% oil yield/plant, plant spread and 100 seed wt. ranged from 222 to 635 g 22.43% to 35.46%, 77.60–220.3 cc, 1.66 to 3.6 em and 89.9 to 109 g with a mean value of 432.44 g, 32.37, 137.49 cc 2.86 cm and 100.07 g respectively. Accessions *lC007, lC005,* JCOOI had more oil yield than the mean value of 137.49 cc; *lC007* leads among all the accessions. Accessions *lC007, lC005, lC004* and JCOOI had more seed yield/plant (g) than the mean value of 432.44 g. Another important attribute for selection, number of branches/plant and plant spread, the yield contributing characters, variability ranged from 2.5 to 4.6 and 1.66 to 3.6 em respectively. Less plant height and maximum yield are important characters determining the suitability in production system. Majority of the accessions evaluated had a shorter plant height than the mean value. The accession JC007 having higher seed yield, less plant height and more oil yield than their mean value is found to be an elite genotype among all the accessions.

Correlation coefficient for 10 metric traits is presented in (Table 2) Most of the correlations were significant. Seed yield/plant had a positive correlation with all the characters except leaf width, 100 seed weight and oil percent, highest being with number of branches/plant (r = 0.806) followed by oil yield (0.800) and plant spread (r = 0.782). Oil percent had a negative correlation with all characters, significant negative correlation with seed yield/plant, 100 seed weight and plant spread (r = −0.404, -0.404, −0.424) respectively showing thereby that if the oil percentage increases seed yield decreases and vice versa. Number of branches had a positive and significant correlation with plant height, plant spread, stem diameter, seed yield/plant and oil yield/plant (r = 0.647, 0.634, 0.733, 0.806, 0.724) respectively, while as plant spread had a positive significant correlation with stem diameter, leaf length, seed yield/plant, oil yield/plant (r = O.072, 0.536, 0.783, 0.658) respectively.

Table 1
Mean values for morphometric characters and yield attributes in seven accessions of *Jatropha curcas*.

Accession	Plant height (m)	Number of branches / Plant	Plant spread (m)	Stem diameter (cm)	Leaf length (cm)	Leaf width (cm)	Seed yield (g)	Oil %	Oil yield/ plant (cc)	100 seed wt (g)
JC001	4.85 ± 0.164	4.6 ± 0.382	3.57 ± 0.100	13.83 ± 0.51	12.03 ± 0.398	0.25 ± 0.279	635.5 ± 35.79	24.49 ± 1.127	155.60 ± 2.68	96.79 ± 3.68
JC002	3.87 ± 0.127	2.5 ± 0.158	2.22 ± 0.110	10.4 ± 1.067	8.44 ± 0.132	8.44 ± 0.029	222.8 ± 17.89	34.83 ± 1.064	77.60 ± 1.193	10.89 ± 9.63
JC003	5.2 ± 0.121	3.0 ± 0.245	1.66 ± 0.080	9.14 ± 0.248	10.9 ± 0.121	8.08 ± 0.109	333.2 ± 7.33	35.46 ± 1.120	118.13 ± 1.426	95.20 ± 6.92
JC004	4.87 ± 0.132	3.4 ± 0.253	3.39 ± 0.132	11.63 ± 0.233	12.39 ± 0.030	9.16 ± 0.074	433.2 ± 22.13	29.43 ± 1.826	127.46 ± 1.272	109.00 ± 3.64
JC005	3.81 ± 0.104	3.3 ± 0.284	3.0 ± 0.363	11.40 ± 0.240	12.80 ± 0.148	10.70 ± 0.166	478 ± 31.70	31.26 ± 1.146	149.4 ± 0.551	99.09 ± 1.82
JC006	3.46 ± 0.045	2.9 ± 0.300	2.64 ± 0.066	9.72 ± 0.343	10.50 ± 0.024	7.52 ± 0.024	342.2 ± 26.01	33.26 ± 1.554	113.80 ± 0.809	101.58 ± 1.09
JC007	2.93 ± 0.110	4.2 ± 0.369	3.6 ± 0.066	14.25 ± 0.139	12.00 ± 0.183	9.20 ± 0.060	582 ± 48.41	37.86 ± 1.747	220.3 ± 5.804	89.900 ± 2.34
Mean	**4.14 ± 0.104**	**3.40 ± 0.288**	**2.86 ± 0.138**	**11.48 ± 0.327**	**11.717 ± 0.217**	**8.76 ± 0.191**	**432.44 ± 25.77**	**32.37 ± 1.38**	**137.47 ± 2.50**	**100.07 ± 5.79**
CV %	**4.342**	**14.677**	**8.337**	**4.930**	**3.203**	**3.776**	**10.322**	**7.42**	**3.15**	**10.13**

Table 2
Correlation among different yield traits in *Jatropha curcas.*

Characteristics	No. of branches/plant	Plant spread (m)	Stem diameter (cm)	Leaf length (cm)	Leaf width (cm)	Seed yield/plant (g)	100 seed wt. (g)	Oil%	Oil yield/Plant (cc)
Plant height (m)	0.647 *	0.840 *	0.857 *	0.555 *	0.265	0.676 *	−0.059 g	−0.271	0.628 *
No. of branches/plant		0.634 *	0.733 *	0.328	0.171	0.806 *	−0.222	−0.242	0.724 *
Plant spread (m)			0.872 *	0.556 *	0.321	0.782 *	−0.065	−0.424	0.652 *
Stem dia (cm)				0.528 *	0.298	0.809 *	−0.218	−0.303	0.761 *
Leaf length (cm)					0.854 *	0.475 *	−0.023	−0.319	0.416
Leaf width (cm)						0.299	0.009	0.004	0.377
Seed yield/plant (g)							−0.340	−0.404	0.800 *
100 seed weight (g)								−0.404	−0.463 *
Oil %									0.096

* $P \leq 0.05$.

Plant height had a significant correlation with all characters except leaf width, 100 seed weight, and oil percent. 100 seed weight had positive correlation with oil yield ($r=0.800$) and a negative correlation with oil percent (−0.404). This suggests that the number of branches/plant, plant spread and plant height are the important factors to which further improvement can be made. The proximate composition of the seed flour is given in Table 3. The crude fiber and ash content values are important parameters to evaluate the suitability of the seed cake for compounding of animal feed. In the present case the values for crude fiber and ash are (16.5% and 4.35%) respectively which is high so it would be unsuitable for animal feed. The protein content of the oil seeds is slightly higher or comparable to the less known pulse crop oil seeds, Sunflower (19.5%) Peanut (30.3%) (Amubode and Feluga, 1984). Thus the seeds could serve as an alternative source of protein.

The fatty acid composition of seed oil is presented in Table 4. The oil content from the seeds varied from 24.5% to 37.9%. The quantities of palmitic, stearic, oleic and linoleic acid varied among accessions. Linoleic acid which is one of the most important polyunsaturated fatty acid in human food because of its prevention of distinct heart vascular diseases (Boelhouwer, 1983) is the most abundant fatty acid in accession JC006 (51%) followed by JC001, JC002 (42.4 and 41.0%), respectively. Accession JCOOI has predominant oleic acid (48%) which is more than the Indian standard. Except in accession JCOO 1 all other accessions had more stearic acid than the international standard (Anonymous, 1959) highest being in accession JC003 while accession JC005 had predominant palmitic acid (25.1%). All the accessions displayed significant variation among themselves for individual fatty acids. Broader range of variation was recorded for all the four fatty acids. Such variation in fatty acid composition points to an inheritable capacity.

Table 3
Proximate composition of Jatropha seed.

Assay	Percentage
Crude fat	29.9 ± 1.153
Crude protein	30.2 ± 1.332
Crude fiber	16.5 ± 0.701
Ash	4.35 ± 0.17
Moisture	5.1 ± 0.252
Carbohydrate (by difference)	12.5

Values are mean for triplicate determination.

In earlier studies Sukarin et al. (1987) failed to observe any morphological difference between clones collected from different locations in Thailand. The evaluation of five cultivars from Hissar revealed a good degree of variation for plant height, branches per plant and seed yield (Hooda et al., 2004). In another study Heller (1992) reported the existence of genotypes specifically adapted to marginal conditions. In the present investigation, seed yield, 100 seed weight, and crude fat content indicate possibilities of selection of an elite genotype which could serve as a panacea for energy shortage in the future world.

Among all the accessions studied, accession JC007 was found to be promising for majority of its traits. Seed oil of accession JC007 was evaluated for physicochemical analysis as depicted in Table 5. The iodine value is a measure of the unsaturation of fats and oils. Higher iodine value indicated higher unsaturation of fats and oils (Knothe, 2002; Kyriakidis and Katsiloulis, 2000). The iodine values of *Jatropha* oil seed suggest their use in production of alkyl resin, shoe polish, varnishes, etc. (Akintayo, 2004). High saponification value indicated that oils are normal triglycerides and very useful in production of liquid soap and shampoo industries. Viscosity increases with molecular weight but decreases with increasing unsaturation and temperature (Nourredini et al., 1992a, 1992b). Different methods such as preheating, blending, ultrasonically assisted methanol transesterfication and supercritical methanol transesterfication are being used to reduce the viscosity and make them suitable for engine applications (Pramanik, 2003; Banapurmath et al., 2008).

4. Conclusion

The present study reveals that there is considerable variability among the populations of genotypes collected from subtropical conditions of north India. Comparing the accessions by statistical and physico-chemical analysis, JC007 was identified as an elite type which can be exploited for commercial purposes.

Acknowledgements

The authors are grateful to the Director Indian Institute of Integrative Medicine (IIIM) Jammu for providing facilities. Thanks are also due to National Medicinal Plant Board (NMPB) for providing financial support.

Table 4
Fatty-acid composition of oil of seven accessions of *Jatropha curcas.*

Accession no	Location	Total oil %	Palmitic acid %	Stearic acid %	Oleic acid %	Linoleic acid %
JC001	Jammu	24.49	8.2±0.165	5.1±0.165	48.0±0.194	42.4±0.248
JC002	Sunderbani	34.83	17.5±0.261	15.1±0.155	16.8±0.266	41.0±0.155
JC003	Rajouri	35.46	5.1±0.187	42.9±0.119	23.7±0.165	28.1±0.275
JC004	Kathua	29.43	16.3±0.138	26.6±0.222	17.7±0.185	28.6±0.104
JC005	Udhampur	31.26	25.1±0.149	35.5±0.222	30.3±0.229	34.5±0.194
JC006	Basoli	33.26	12.5±0.191	23.4±0.202	19.4±0.23	51.1±0.149
JC007	Vaishno Devi	37.86	12.0±0.287	23.8±0.147	−19.6±0.24	26.2±0.138
Mean		**32.37**	**13.81**	**24.62**	**25.07**	**35.98**
Standard (International)		–	**12-17**	**5-10**	**37-63**	**19-40**
National (Indian)		**30-40**	**13.6**	**9.6**	**40.9**	**32.0**

Values are mean for duplicate determination.

Table 5
Physico-chemical characteristics of selected accession JC007 of *Jatropha curcas* seed oil.

Parameters	Values
Free fatty acid (mg/g)	5.8±0.100
Acid value (as oleic acid)	2.89±0.095
Saponification value (mg k OH/g)	172.5±1.217
Iodine value with (mg/iodine/g)	105.2±0.529
Refractive index (25 °C)	1.468
Specific gravity (25 °C)	0.912
Hydroxyl value	2.15±0.015
Ester value	1.667±0.153
Viscosity (30 °C) cst	16.9
Unsaponificable matter (%)	0.7±0.050

Values are mean of triplicate determination.

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Edited by E Balazs