



B.B. Bandyopadhyay et al./ Elixir Agriculture 105 (2017) 46169-46174 Available online at www.elixirpublishers.com (Elixir International Journal)

Agriculture



Elixir Agriculture 105 (2017) 46169-46174

Genetic Diversity Analysis in Jatropha (Jatropha curcas L.) genotypes

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ARTICLE INFO

Article history: Received: 14 February 2017; Received in revised form: 28 March 2017; Accepted: 18 April 2017;

Keywords

Jatropha curcas, Morphological traits, Genetic parameters, Genetic divergence, Selection.

ABSTRACT

Steady decrease in crude petroleum oil reserve from earth and increasing demand of petroleum oil as source of generating energy for automobile and other sectors of industry, has led to adopt an alternative strategies for the production of bio-fuel from plants genetic resources to avert the risk of impaired supply of fuels in future from resource countries. Jatropha curcas is known as bio-fuel production crop. The oil present in its seeds used for extraction of bio-fuel. Genetic diversity analysis was performed during 2015-16 on 10 years old plants of 20 Jatropha curcas genotypes that were collected from different regions of India. The seeds were planted in randomized block design with three replications during 2005. Observations were recorded on 14 morphological characters. Field data were used for computation of ANOVA, estimation of genetic parameters and Mahalanobis D^2 statistics. Significant differences were registered for all traits. Days to flowering, number of fruit clusters per plant and 100 seed weight showed additive genetic effect. Twenty genotypes were grouped into 3 clusters comprised of three, sixteen and one genotypes respectively and suggested that geographical diversity not related to the genetic diversity. Seed yield per plant, fruits per cluster, post floral period and secondary branches per plant contributed maximum towards genetic divergence. Three lines were found promising for the development of superior genotype.

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Introduction

India possesses precious wealth of plant genetic materials. Jatropha curcas is one of such an important plant genetic material that could be used as bio-fuel production crop (Umamaheswari et.al., 2010) as an alternative resource of crude petroleum oil used for generating energy for the growth of developed and developing countries in the world. Geopolitical features, increase in hike of crude petroleum oil price (Asif and Munee, 2007) and growing demand of fuel among these countries seem to cast a shadow over the supply of crude petroleum oil in many countries including India. The government of India has adopted alternative strategies for the production of cheap and renewable liquid fuel based on vegetable oil (Shukla, 2005) among plants in order to avert the risk of impaired supply of fuels from resource countries. The seeds of Jatropha contain 46 to 58% of oil on kernel weight and 30 to 40% on seed weight (Subramanyam et.al., 2010). The oil/fat contains about 21% saturated fatty acids and 79% unsaturated fatty-acid (Raja et.al., 2011). Jatropha is reported to introduce in Asia and Africa by Portuguese as an oil yielding plant. In India the plant grows in the state of Chhattisgarh, Maharashtra, Gujarat, Tamil Nadu and Rajasthan and thrives well to semi-arid condition, alkalinity /salinity of soil and marginal land (Rao et.al., 2008). It also find in natural habitation in Uttarakhand state. Jatropha species are essentially cross pollinated, which offer a high degree of variation and provide ample scope for screening and selection of superior genotypes with desired traits and high seed oil content (Ginwal et.al., 2004; Kumar et.al., 2014). However, little is known about the genetics, breeding, physiology and agronomic aspects of the species.

In this present investigation emphasis is given to study "Genetic divergence analysis in *Jatropha curcas* L. based on morphological characters".

Materials and Methods

A set of Jatropha germplasm were collected during July, 2005 from all over India. Jatropha curcas is perennial crop. The original seeds of 20 different genotypes were planted on 31st July, 2005 in Randomized Block Design (RBD) with three replications at Medicinal Plant Research and Development Centre (M.R.D.C.) of G.B. Pant University of Agriculture & Technology, Pantnagar (29^oN latitude, 79.3^oE) at an altitude of 243.84 m above mean sea level. The present study was undertaken on 10 years old plants of 20 Jatropha genotypes during 2015-16. Observations were recorded on five randomly selected plants from each replication for fourteen morphological characters viz. plant height (m), collar diameter (cm; estimated over the values recorded for circumferences of respective plant), number of primary branches per plant, number of secondary branches per plant, days to flowering (days required for flowering from 31st July in 2015 i.e., same date of planting seeds as on 2005), post floral period (days to commencement of flowering to days to final harvest; the seeds of all Jatropha genotypes were harvested on same day in the month of December), number of fruits per cluster, number of fruit-clusters per plant, leaf area (cm²), leaf index (1:b ratio), fresh weight of 100 fruits(Kg), dry weight of 100 fruits(Kg), 100 seed weight (g), seed yield per plant(Kg). Field data were subjected to statistical analysis. Analysis of variance (Panse and Sukhatme, 1967) was performed. The partitioning of variances into genotypic, phenotypic and error variances were worked out.

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The genotypic coefficient of variation, phenotypic coefficient of variation (Burton and Devane, 1953), broad sense heritability (Johnson et.al., 1955), genetic advance (Allard, 1960) and genetic gain over percent of mean (Johnson *et al.*, 1955) were calculated. Mahalanobis D² statistics (1936) of generalized distance (Rao, 1952) was estimated to measure the genetic divergence present among 20 genotypes of Jatropha and D² values (Murty and Arunachalam, 1966) were estimated for group constellation Jatropha genotypes.

Results and Discussion

The variability studies are the prerequisite and paramount importance for developing tree improvement strategies (Gairola et.al., 2011). Germplam serve as a most valuable reservoir of variability on which selection can be made directly for evolving superior genotypes. Effective selection thus depends on the existence of sufficient genetic variability in the available breeding material. In the present study ANOVA (Table 1) exhibited significant differences for all characters among Jatropha genotypes indicating that naturalized stand of Jatropha curcas gneotypes, that grows under varied climatic condition in India, shows high degree of variability and offer ample scope for selecting superior genotypes from germplasm for desire traits. Significant variability for yield and related traits (Singh et.al., 2013; Saadaoui et.al., 2015), collar diameter, number of branches, fruit yield, seed yield per plant and 100 seed weight (Tripathi et.al., 2013), number of primary branches per plant, secondary branches per plant, seed yield per plant and 100 seed weight (Kumar et.al., 2015) were reported by other researchers, which are in agreement to our study. Contrarily, Das et al. (2010) reported non-significance variation for primary branches per plant and fruits per plant which are not in conformity with results of present study. The superior genotypes over checks (PJ 03004 and PJ 03031), on the basis of overall mean performance for different traits can be exploited directly. Mean performance (Table 2) of TNMC-3, TNMC-2 and IGAU Surguja exhibited superior performance in seed yield per plant over checks. TNMC-3 registered higher number of fruit per cluster while TNMC-2 established superiority in leaf area, post floral period, number of fruit clusters per plant and days to flowering over best checks. Similarly IGAU Surguja showed evidence of superiority for 100 seed weight and secondary branches per plant. A perusal on genetic parameter (Table 3) revealed higher values of phenotypic variance and genotypic variance for number of fruit clusters per plant (2222.703, 22.278), leaf area (158.913, 140.991) and days to flowering (31.388, 29.979) while relatively low phenotypic and genotypic variance registered in dry weight of hundred fruits (0.00068, 0.00065).

In contrast Kaushik et al. (2015) reported high phenotypic variance and genotypic variance for 100 seed weight but Shabanimofrad et al. (2013) observed higher values in plant height, seed yield per plant and 100 seed weight. The estimates of phenotypic coefficient variation (PCV) were higher than genotypic coefficient of variation (GCV) for all the characters studied, however, the differences between PCV and GCV appeared less. This indicated that selection would be effective in improving the genetic constitution of cultivar. Seed yield per plant showed the highest phenotypic coefficient of variation (56.79 %) followed by number of fruit clusters per plant (35.258%), number of secondary branches per plant (16.172%), number of fruits per cluster (15.525%) and number of primary branches per plant (15.358%). The result of Das et al. (2010) and Shabanimofrad et al., (2013) bear close affinity to our findings with high PCV and GCV values except for plant height, 100 seed weight. On the contrary, Kaushik et al. (2015) observed high PCV and GCV for 100 seed weight which is not in confirmation with the present study. Heritability measures the genetic relationship between the parents and their offspring. Effective selection depends on the existence of the genetic variability in heritability of a trait. High heritability revealed that the expression of a trait largely depend on genetic components while the influence of environment was found less. Experimental analysis found that very high heritability (>85%) for characters. The results on heritability described by Shabanimofrada et al. (2013) accorded to our findings for primary branches and 100-seed weight except collar diameter although Das et al., (2010) reported moderate to high heritability for plant height and 100 seed weight, collar diameter, secondary number of branches per plant and seed vield per plant. Heritability and genetic advance are considered as important genetic parameters and will help in deciding the scope for improvement of a trait through selection (Johnson et.al., 1955). In the present investigation the higher values of genetic advance was estimated for fruit clusters per plant (30.683), leaf area (23.04) and days to flowering, post flowering period (11.02). The genetic advance over per cent of mean recorded high for seed yield per plant (116.678%), number of fruit clusters per plant (72.92%), number of secondary branches per plant (30.541%), number of fruits per cluster (30.252%), number of primary branches per plant (27.468%), fresh weight of 100 fruits (26.192%), leaf index (23.034%), and days to flowering (22.320%). Expected genetic advance indicates the expected genetic progress for a particular trait under a selection cycle and measures the extent of its stability under selection pressure.

characters	Source				
	Replication (2)	Genotypes (19)	Error (38)		
Plant height	0.032	0.198**	0.005		
Collar diameter	0.963	17.765**	0.944		
Primary branches /plant	0.213	4.489**	0.219		
Secondary branches / plant	0.079	24.256**	0.713		
Days to flowering	3.738	91.346**	1.409		
Post floral period	3.741	91.340**	1.410		
Fruits/cluster	0.052	12.322**	0.230		
Fruit clusters /plant	0.060	667.258**	0.425		
Leaf area	30.590	440.89**	17.922		
Leaf index (l:b ratio)	0.002	0.053**	0.002		
Fresh weight of 100 fruits	0.0001	0.056**	0.001		
Dry weight of 100 fruits	0.0001	0.002**	0.00003		
100 seed weight	0.894	43.235**	0.640		
Seed yield/plant	0.0001	0.498**	0.0004		

Table 1. Analysis of variance for 14 morphological characters in 20 Jatropha curcas genotypes.

**Significant at 5 per cent level

d.f. represent the figure given under parenthesis

Sl.	Treatments	riormances of 14 morphological characters* in 20 <i>Jatropha curcas</i> genotypes and their SEM, CD, CV and range values. Characters													
No.		1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	IGAU Raipur	3.95	30.59	11.11	23.87	41.75	90.25	8.54	65.4	136.54	1.02	1.02	0.24	60.42	0.80
2	IGAU Bilaspur	4.34	32.26	9.59	20.06	49.34	82.66	9.28	35.09	154.7	1.02	1.06	0.24	53.22	0.34
3	IGAU Surguja	4.52	32.64	9.01	23.37	50.79	81.21	12.72	62.5	145.79	1.05	1.22	0.25	63.64	1.18
4	TNMC-2	4.34	33.45	9.14	18.22	29.55	102.45	16.52	66.52	157.65	1.03	1.14	0.19	59.42	1.58
5	TNMC-3	4.55	34.97	9.14	20.76	54.46	77.54	18.98	65.41	152.73	1.09	1.13	0.24	56.03	1.76
6	TNMC-4	4.7	34.3	9.08	18.96	52.32	79.68	13.01	26.82	134.77	1.18	0.86	0.22	53.89	0.54
7	TNMC-5	4.68	34.08	7.6	17.05	50.28	81.72	13.36	25.48	148.48	1.06	0.97	0.26	48.87	0.33
8	TNMC-7	5.11	36.17	7.67	17.62	49.05	82.95	16.33	24.25	128.5	1.08	1.1	0.28	48.17	0.25
9	TNMC-22	4.76	37.45	8.54	17.67	49.29	82.71	16.05	21.99	130.93	1.12	1.25	0.27	56.04	0.46
10	Sagar (SFRI,Jabalpur)	4.92	31.73	6.13	14.46	50.49	81.51	13.68	26.08	148.06	1.07	1.04	0.28	58.65	0.35
11	Indore (SFRI,Jabalpur)	4.93	31.95	7.94	17.33	44.97	87.03	14.61	27.62	130.03	1.11	1.29	0.27	52.76	0.40
12	TFRI-1	4.79	34.03	7.78	16.59	51.95	80.05	15.16	41.33	156.96	1.17	0.86	0.26	55.23	0.82
13	TFRI-2	4.93	31.89	7.79	15.44	49.76	82.24	12.57	36.2	123.65	1.09	1.11	0.28	53.89	0.63
14	Pant J. sel-1	4.66	35.29	7.97	15.71	50.49	81.51	13.24	45.32	129.11	1.05	0.76	0.28	56.94	0.71
15	Pant J.sel-2	4.68	34.72	7.48	15.44	53.13	78.87	13.33	54.68	127.56	1.63	1.04	0.28	52.96	1.08
16	RJ-117	4.79	37.84	10.64	22.26	49.58	82.42	12.39	40.3	122.92	1.07	0.9	0.26	56.17	0.60
17	PKVJ-MKV-1	4.66	40.06	7.16	19.72	54	78	11.89	36.2	123.43	1.1	0.96	0.29	54.67	0.44
18	PKVJ-DHW-1	4.72	35.26	7.04	14.23	51.43	80.57	13.43	45.67	143.37	0.94	0.97	0.28	51.22	0.69
19	PJ-03004 (LC-I)	4.66	33.36	8.4	15.18	53.37	78.63	12.17	51.55	126.28	1.13	0.96	0.28	51.99	0.66
20	PJ-03031 (LC-II)	4.88	37.63	7.34	17.89	51.72	80.28	13.07	48.12	129.82	1.07	0.95	0.27	52.22	0.72
	Grand mean	4.68	34.48	8.33	18.09	49.39	82.61	13.52	42.33	137.56	1.1	1.03	0.26	54.82	0.72
	SEM±	0.39	0.56	0.27	0.49	0.69	0.69	0.29	0.38	2.49	0.26	0.21	0.33	0.46	0.12
	CD (5%)	0.11	1.61	0.77	1.4	1.96	1.96	0.82	1.08	7.11	0.74	0.6	0.94	1.32	0.35
	CV (%)	1.45	2.82	5.62	4.67	2.4	1.44	3.69	1.54	3.13	4.09	3.52	2.2	1.46	2.92
	Range	3.95-	30.59-	6.13-	14.23-	29.55-	77.54-	8.54-	21.99-	122.92-	0.94-	0.76-	0.19-	48.17-	0.25-
		5.11	40.06	11.11	23.87	54.46	102.45	18.98	66.52	157.65	1.63	1.25	0.29	63.64	1.76

Table 2. Mean performances of 14 morphological characters* in 20 Jatropha curcas genotypes and their SEM, CD, CV and range values.

*1 = Plant height (m); 2 = Collar diameter (cm); 3 = Primary branches/ plant; 4 = Secondary branches/ plant; 5 = Days to flowering (after 31st July); 6 = Post flowering period (days); 7 = Fruits/ cluster; 8 = Fruit clusters/ plant; 9 = Leaf area (cm²); 10 = Leaf index (l:b ratio); 11 = Fresh weight of 100 fruits(Kg); 12 = Dry weight of 100 fruits(Kg); 13 = 100 seed weight (g); 14 = Seed yield/ plant (kg)

			Table 5. E	stimates of g	enetic parameter	5 IOI 14 IIIOI p	nonogicai	character	s III Jau	opna cui cas	genotypes.			
Characters/	Plant	Collar	Primary	Secondary	Days to	Post	Fruits/	Fruit	Leaf	Leaf	Fresh	Dry weight	100 seed	Seed
genetic	height	diameter	branches/	branches/	flowering (after	flowering	cluster	clusters/	area	index (1:b	weight of	of 100	weight	yield/
parameter	(m)	(cm)	plant	plant	31st July)	period		plant	(cm ²)	ratio)	100 fruits	fruits(Kg)	(g)	plant
						(days)				,	(Kg)			(kg)
PCV (%)	5.609	7.422	15.385	16.172	11.344	6.782	15.525	34.994	9.164	12.523	11.73	8.965	7.007	56.79
GCV (%)	5.418	6.867	14.323	15.485	11.087	6.628	15.099	34.96	8.632	11.83	11.678	8.912	6.831	56.571
ECV (%)	0.191	0.555	1.062	0.687	0.257	1.43	0.426	0.034	0.532	0.689	0.049	0.053	0.176	0.2185
h^2	93.291	85.583	86.669	91.674	95.51	95.51	94.591	99.801	88.72	89.288	99.17	98.824	95.023	99.736
G A	0.505	4.513	2.288	5.525	11.023	11.02	4.022	30.386	23.04	0.254	250.1	47.979	7.516	0.838
GA as % of	10.78	13.086	27.468	30.541	22.32	13.343	30.252	71.945	16.75	23.034	23.96	18.251	13.71	116.67
the mean														
V _P	0.069	6.551	1.642	8.561	31.388	22.997	5.927	222.703	158.9	0.019	0.019	0.00068	14.838	0.166
Vg	0.0643	5.607	1.423	7.848	29.979	29.978	5.697	222.278	140.9	0.017	0.018	0.00065	14.198	0.166
Ve	0.005	0.944	0.219	0.13	1.409	1.411	0.23	0.425	17.92	0.002	0.001	0.00003	0.64	0.0004

 Table 3. Estimates of genetic parameters for 14 morphological characters in Jatropha curcas genotypes.

	Table 4. Clustering patterns and average inter and intra-cluster D ² and D* values of 20 <i>Jatropha curcas</i> genotypes.								
Cluster	Genotypes included	Number of	Inter and intra-cluster D^2 and D values						
		genotypes	Ι	II	III				
Ι	IGAU Raipur, IGAU Surguja, TNMC-3	3	10.937 (3.307)	30.963 (5.564)	30.845 (5.54)				
Π	IGAU Bilaspur, TNMC-4, TNMC-5, TNMC-7, TNMC-22, Sagar (SFRI, Jabalpur), Indor (SRFI, Jabalpur), TFRI-1,TFRI-2, Pant J. sel-1, Pant J. sel-2, RJ-117, PKVJ-MKV-1, PKVJ-DHW-1, PJ-03004 (LC-I), PJ-03031 (LC-II)	16		15.651 (3.956)	48.504 (6.964)				
III	TNMC-2	1			0.000 (0.000)				

Diagonals are intra-cluster values for respective cluster

* D values are given under parenthesis

Heritability, genetic advance and genetic advance as per cent of mean are important genetic parameters for the selection of desirable traits. High heritability (broad sense) couple with high genetic gain indicated additive genetic control in the inheritance of these traits suggesting that early generation phenotypic selection would be effective for these traits in *Jatropha curcas*. Additive genetic control was observed for plant height (Saadaoui et.al., 2015; Tripathi et.al., 2013), 100 seed weight, 100 fruit weight (Kaushik et.al, 2007), seed yield per plant, number of fruits per plant and secondary branches per plant (Das et.al., 2010).

In genetic divergence analysis existing variability for all characters among 20 genotypes were grouped into three clusters (Table 4). The cluster I comprised of three genotypes. cluster II had sixteen genotypes, while cluster III was represented by single genotype. It was presumed that Jatropha originating from same geographical region were genetically close to each other, exhibited similar morphological characters and fall in the same cluster. In the present investigation, however, the pattern of group constellation proved that geographical diversity need not necessarily be related to the genetic diversity because those 3 genotypes that constituted cluster I had collected from different regions in India. Similarly sixteen genotypes including two checks (viz., PJ-03004and PJ-03031) of diverse origin were grouped in to cluster II. This suggested that Jatropha originating from same geographical region tend to fall into different cluster and the genotypes included in the same cluster had different geographical region. Similar observations have been reported by Gohil and Pandya (2008) and Singh et al. (2013). Different clustering pattern were also reported recently on Jatropha by some other workers (Gohil and Pandya, 2008; Singh et.al., 2013; Kaushik et.al., 2015). Seed yield per plant contributed maximum towards genetic divergence (Table 5) followed by number of fruits per cluster, post floral period, number of secondary branches per plant, number of primary branches per plant, leaf index, leaf area, days to flowering and fresh weight of 100 fruits. These characters were considered to be the most important for constituting genetic diversity in the population and could be utilized in the effective breeding programme to achieve desire genetic gain.

Maximu intra-cluster average D^2 values found in cluster II (15.651) with sixteen genotypes (Tables 4) followed by cluster I (10.937) with three genotypes. The minimum intra-cluster distance was found for Cluster III (0.000) which was represented by only one genotype. The inter-cluster average D^2 - value and the genetic distance (D) were found maximum between cluster II and III (48.504, 6.964) suggesting the genotypes belong to these two cluster were genetically more diverse in comparison to that observed between cluster I and II (30.963; 5.564) and between cluster I and III (30.845; 5.54). It was therefore assumed that the genotypes belong to clusters I maintained equal distance from both cluster II and cluster III. This may attribute that the genotypes from divergent clusters can be used to inter-cross to obtain high heterotic response and also to recover desirable transgressive- segregants. Gohil and Pandya (Gohil and Pandya, 2009) and Singh et al. (2013) also arrived to the similar conclusion and stating that use of divergent parent exhibited heterotic response through favourable combination of genes from two divergent, contrast but complementary parents in a cross, which help to produce superior plants through transgressive segregation. Seed yield per plant, fruits per cluster, post floral period, and secondary branches per plant contributed maximum towards genetic

divergence (Table 5). A comparative assessment of cluster mean values (Table 5) and individual performance of Jatropha genotype for each trait (Table 2) suggested that IGAU Raipur, TNMC 3 (both from cluster I) and TNMC2 (cluster III) were found promising and could be utilized as parent in a cross to produce new genotypes with superior combination of traits for augmenting seed yield per plant of Jatropha population.

Table 5. Cluster mean values of 14 morphological
characters in Jatropha curcas genotypes and their
contribution towards genetic divergence.

Character		Cluster	Contribution		
Character	Ι	II	III	(percentage)	
Plant height (m)	4.341	4.765	4.343	2.475	
Collar dia. (cm)	32.732	34.876	33.445	3.190	
No. of primary	9.756	8.012	9.144		
branches / plant				6.638	
No of secondary	22.667	17.226	18.217		
branches / plant				7.103	
Days to flowering	49	50.698	29.55		
(days)				5.050	
post flowering	83	81.302	102.45		
period				7.963	
No. of fruits /cluster	13.415	13.347	16.522	15.927	
No. of fruit clusters /	64.435	36.669	66.517		
plant				3.983	
Leaf area (cm ²)	145.02	134.91	157.65	5.474	
Leaf index (l: b	1.053	1.118	1.027		
ratio)				6.012	
Fresh weight of	1.125	1.004	1.136		
100fruits (Kg)				4.397	
Dry weight of 100	0.242	0.267	0.191		
fruits (Kg)				3.130	
100 seed weight (g)	60.03	53.556	59.417	3.019	
Seed yield / plant	1.25	0.564	1.58		
(kg)				25.637	

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