NUSANTARA BIOSCIENCE
Vol. 3, No. 3, pp. 130-135
November 2011

ISSN: 2087-3948
E-ISSN: 2087-3956
DOI: 10.13057/nusbiosci/n030305

Variation in oil contents and seed and seedling characteristics of *Jatropha curcas* of West Nusa Tenggara selected genotypes and their first improved population

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Manuscript received: 20 July 2011. Revision accepted: 11 September 2011.

Abstract. *Santoso BS. 2011. Variation in oil contents and seed and seedling characteristics of* Jatropha curcas *of West Nusa Tenggara selected genotypes and their first improved population. Nusantara Bioscience 3: 130-135.* This study describes variation in seed and seedling characters of *Jatropha curcas* Linn. of West Nusa Tenggara selected genotypes. Exploration was conducted in several areas where large population of this species grown as fences were found. Five selected genotypes were then grown in experimental fields to let mass selection to obtain the first improved population for each genotype. Seeds of wild population (P0) and those of selected trees as the first improved population (IP-1) were subjected to this study. Seed and seedling characteristics were measured. The result indicated that considerable genetic variability existed among the five *J. curcas* of West Nusa Tenggara selected genotypes and within each genotype population for seed and seedling characteristics. Genotypes of West Lombok, Sumbawa, and Bima performed exceedingly better than those of Central Lombok and East Lombok. Therefore, this study has suggestions for identifying potential seed sources of *J. curcas* and these existing genetic variability provides breeders with materials in crop improvement program.

Keywords: genetic variability, seeds, seedling, selection.

Abstrak. *Santoso BS. 2011. Keragaman kandungan minyak, serta karakteristik biji dan bibit genotipe terpilih* Jatropha curcas *Nusa Tenggara Barat dan populasi pertamanya yang diperkaya. Nusantara Bioscience 3: 130-135.* Kajian ini menjelaskan keragaman karakteristik benih dan bibit *Jatropha curcas* Linn. Nusa Tenggara Barat dari genotipe terpilih. Eksplorasi dilakukan di beberapa daerah dimana populasi-populasi besar jenis ini ditemukan sebagai tanaman pagar. Lima genotipe yang terpilih kemudian ditanam di lahan percobaan untuk memulai seleksi massal untuk mendapatkan populasi yang diperkaya pertama pada setiap kenotipe. Benih dari populasi liar (P0) dan pohon-pohon yang dipilih sebagai populasi yang diperkaya pertama (IP-1) menjadi subyek penelitian ini. Karakteristik benih dan bibit diukur. Hasilnya menunjukkan adanya variabilitas genetik yang cukup tinggi di antara lima genotipe terpilih *J. curcas* Nusa Tenggara Barat dan karakteristik benih dan bibit dari setiap populasi genotipe. Genotipe dari Lombok Barat, Sumbawa, dan Bima memiliki penampilan yang jauh lebih baik dari pada genotipe dari Lombok Tengah dan Lombok Timur. Hasil penelitian ini dapat menjadi acuan dalam mengidentifikasi potensi sumber benih *J. curcas* dan menunjukkan adanya variabilitas genetik yang diperlukan penangkar sebagai bahan untuk program pemuliaan tanaman.

Kata kunci: variabilitas genetik, benih, pembibitan, seleksi.

INTRODUCTION

Physic nut (*Jatropha curcas* L.) is presently grown throughout arid and semi-arid tropical and sub-tropical regions including Indonesia. In Indonesia, it is found in semi-wild condition as fences in the villages and well adapted to various kinds of critical soil conditions and commonly called *jarak pagar* (Santoso 2008; Hasnam 2006). *Jatropha curcas* has gained interest all over the world in comparison to other tree-borne oil seed crops because of its better adaptation to a wide range of environmental conditions, low cost of oilseed production, high oil content, small gestation period and smaller plant size that makes the seeds harvested easier (Sujatha 2006; Sujatha et al. 2008). To reduce the dependence on crude oil and to achieve energy independence, *J. curcas* has been promoted to be developed as an alternative energy source. *J. curcas* has attracted considerable attention as a source of seed oil (Openshaw 2000; Jongschaap 2008; Kumar and Sharma 2008). However, growth and management of this crop have been poorly documented and little results of field experiments have been shared amongst farmers. Until now this crop has not been fully domesticated. Success in establishment and management of this crop plantation is largely determined by factors of plant varieties used and the sources of seed within species.

Jatropha curcas has a high degree of reproductivity and naturally pollinated outcrossing mating system, that ensures large amount of heterozygosity and considerable genetic variability (Ginwal et al. 2004; Das et al. 2010; Parthiban et al. 2011; Zhang et al. 2011). No report on genetic improvement aspect of this species has been published so far in Indonesia, but restricted to few publications at the global level. Studies on genetic basis of *J. curcas* came mostly from India and China researchers. However, early exploration in several locations in Indonesia found that there was variation owing to differences in location creating certain ecotypes such as colors of stem, leaf, and shoot, forms of capsule, and number of seeds per capsule (Hasnam 2006). Makkar et al. (1997) reported that from 18 provenances in West Africa, North and Central America, and Asia there was variation in seed weight, kernel weight, crude protein, and oil content. Heller (1996) also found that from 11 provenances of Sinegal there was variation in weight of capsule, weight of seeds per trees, and weight of 100 seeds. Therefore, those variations can be used as the basis for selection and development of high yielding genotypes.

For West Nusa Tenggara region that has a wide range of dry land and large variations in wild genotype of *J. curcas*, regional or local crop improvement programs will be successful only after assessing local native genetic strength and possible options toward yield improvement. Furthermore, screening of existing populations for oil yield is needed to select the best producing genotypes. It can be used for profitable production before systematic crop improvement program can yield good cultivation and it can also serve as a source for crop improvement material.

Because seed is an important material for plant propagation, and seed containing oil is very important for economic aspect for this crop development, it is necessary to know more about seed and seedling characteristics. Kumar and Sharma (2008) stated that genetic variation in seed morphology and oil content of *J. curcas* is a great potential in tree improvement programs. Callaham (1999) suggested that several other characters should be taken into consideration for provenance description and breeding propose. This article describes variation in seed, oil content, and seedling characteristics of *J. curcas* of West Nusa Tenggara selected genotypes and their first improved population.

MATERIALS AND METHODS

Plant materials

Exploration of plant material was done from May until June 2006 in West Nusa Tenggara (NTB), Indonesia where large population of *Jatropha curcas* grown as garden fences were found. Seeds from each genotype were obtained from tree stand showing good growth and development and representative for each region.

Seeds of *J. curcas* were collected at least from 25 parental plants with a minimum of 10 capsules per cluster (inflorescence) were chosen from each population of each genotype. Those parental plants were grown as fence 100 m in length, and seeds from those plants were collected and labeled; the seeds were then prepared for seed analysis, nursery, field experiment, and storage in seed storage room. Seed sources and their climate condition are given in Table 1.

Cultivation area

Study in cultivation areas was conducted at Amor-Amor Village, Subdistrict Khayangan, North Lombok, West Nusa Tenggara, during 2006-2007. The area has a semi-arid climate with mean annual rainfall of 600-1,000 mm, minimum temperature of 25^{O}C, maximum temperature of 25^{O}C, relative air moisture of 90%, and altitude of 25 m above sea level. Climatic conditions at cultivation site during experiment are given in Table 2.

A uniform pre-treatment was given to the seeds prior to sowing by soaking them in warm water (50^{O}C) for two hours, let it cool and kept soaking for 24 hours. Seed was sown directly in black polythene bags containing media mixture of soil, sand, and manure with a ratio of 1:1:1 (by volume). Seedlings were grown into 2.5-month old saplings.

Two and half month –old saplings of each genotype were planted in the field experiment using Randomized Complete Block design at 2x2 m^{2} spacing. Three replicates of genotypes, each consisting of 24 were applied.

Crop maintenance

Saplings received fertilizers as follows: manure as much as 2 kg.tree^{-1} and urea 25 kg ha^{-1} (10 g.tree^{-1}), SP36 150 kg.ha^{-1} (60 g.tree^{-1}), and KCl 30 kg.ha^{-1} (12 g.tree^{-1}) at the time of planting. The second urea was applied 2 months after planting with a dose of 25 kg.ha^{-1} (10 g.tree^{-1}) (Mahmud et al. 2006). Irrigation was from rainfall only. At the second year of cultivation, the trees were fertilized with the same dose as that of the first year, except for manure fertilization.

Table 1. Region of seeds' sources and their climatic condition.

Seed sources (genotype, district)	Region (subdistrict)	Altitude (m)	Rain fall (mm)	Temperature (^{O}C)		Air moisture (%)
				Max.	Min.	
West Lombok	Khayangan	50-75	600-1,000	31	25	90
Central Lombok	South Praya	30-55	900-1,300	31	24	90
East Lombok	Masbagik	75-100	1,000-1,500	31	25	85
Sumbawa	Alas	50-75	550-1,000	32	25	85
Bima	Sanggar	50-100	600-750	32	24	85

Table 2. Climate condition of experimental site during 2006-2007

Climate component	2006	2007
Rainfall (mm)	965	716
Rainy month (months)	5	5
Rainy day (days)	56	59
Air temperature (^{O}C) min.	24.7	25
Air temperature (^{O}C) max.	31	32
Relative humidity (%)	90	91

Mass selection to meet the first Improved Population (IP-1)

Selection for improved population in this study is to change the population composition with individuals that have higher average value. Selection was based on yield characteristics such as number of capsules per inflorescence and number of capsules per tree identified during generative phase of growth. Intensity of selection was 25% of trees with higher number of capsules per inflorescence (about 15-20 capsules) during the first production cycle. Seeds collected from selected tree as IP-1 were prepared for studying the seedling characteristics.

Parameters observed

Physical characters of seed and seed viabilities were measured and seed oil content was analyzed. Seed length and seed width were measured using a caliper. Seed weight and weight of 100 seeds were measured using an electronic balance. Separate weights of seed coat and kernels, after the seed coat was removed, were measured to calculate kernel weight percentage. Analysis of seed oil content was carried out using extraction the method of Folch et al. (1957) modified by Sudarmadji et al. (1997). Seed viabilities were measured by daily germination observation until 21 days in three replications, each having 100 seeds. Then, seedling growth parameters were measured for two months..

Table 3. Physical characters of *Jatropha curcas* seeds of West Nusa Tenggara genotypes.

Genotypes	Seed size component					
	Length (cm)		Wide (cm)		Weight (g)	
	P0	IP-1	P0	IP-1	P0*)	IP-1
West Lombok	1.82±0.09	1.85±0.01	0.83±0.05	0.85±0.02	0.74 a	0.78
Central Lombok	1.81±0.08	1.83±0.02	0.85±0.06	0.86±0.03	0.64 ab	0.73
East Lombok	1.82±0.10	1.83±0.02	0.85±0.08	0.85±0.05	0.60 b	0.74
Sumbawa	1.79±0.09	1.81±0.03	0.86±0.06	0.86±0.04	0.70 a	0.78
Bima	1.78±0.24	1.80±0.05	0.80±0.06	0.83±0.01	0.68 ab	0.77
LSD 5%	-	-	-	-	0.09	ns

Note for Table 3 until 6: P0: seed from wild plant population, IP-1: seed from the first improved population, *): numbers in the column with the same letter did not differ significantly at P<0.05, ns: not significant ±: value of standard deviation

Table 4. Percentage of kernel weight to total seed weight and 100 seed weight of *Jatropha curcas* seeds of West Nusa Tenggara genotypes

Genotypes	Percentage of kernel weight (%)		100 seeds weight (g)	
	P0*)	IP-1*)	P0*)	IP-1*)
West Lombok	67.27 ab	71.18 a	69.1 a	75.7 a
Central Lombok	58.45 b	65.66 b	66.4 ab	70.1 ab
East Lombok	60.01 ab	68.36 ab	60.6 b	65.7 ab
Sumbawa	64.29 ab	69.08 ab	65.2 ab	69.5 abc
Bima	67.12 ab	69.96 ab	67.8 a	71.0 ab
LSD 5%	10.05	5.05	6.45	9.33

Tabel 5. Kernel oil content of *Jatropha curcas* of West Nusa Tenggara genotypes

Genotypes	Kernel oil content (% b/b)		
	P0 *)	IP-1	
		Rainy season	Dry season
West Lombok	41.7 ab	43.7	44.6
Central Lombok	42.3 a	42.6	43.1
East Lombok	38.8 b	41.9	42.9
Sumbawa	42.9 a	43.6	44.3
Bima	41.1 ab	43.4	44.3
LSD 5%	3.41	ns	ns

Data analyses

Data were subjected to analysis of mean and standard deviation, analysis of variance and Least Significant Difference test using a Minitab-14 computer program.

RESULTS AND DISCUSSION

Jatropha curcas' seed and seedling characteristics varied among five genotypes of Jatropha from West Nusa Tenggara. Variations were also observed among regions where those genotypes were collected.

Seed characters

Seed length

Mean performance of five *J. curcas* of West Nusa Tenggara selected genotypes with respect to seed length is presented in Table 3. Seeds collected from the five regions of West Nusa Tenggara province varied in their seed length. A higher variation was also found within each wild genotypes population (P0) than that in the first Improved Population (IP-1). The standard deviation of seed length in P0 was higher than in IP-1.

Seed width

Variations among genotypes and within each wild genotype population (P0) as that observed in seed length also occurred in seed width (Table 3). Variation decreased after the first mass selection (IP-1) of wild population (P0).

Seed weight

Seed weight ranged from 0.60g to 0.74 g in different genotypes within the wild population (P0). West Lombok and Sumbawa genotypes had the maximum seed weight; in contrast, East Lombok had the minimum. However, there was no significant difference in seed weight of IP-1 (Table 3).

Table 6. Seed viabilities of *Jatropha curcas* seeds of West Nusa Tenggara genotypes

Genotypes	Germination (%)		Germination rate (day)		Seed vigor (%)		Dry weight of 3 weeks old sapling (g)	
	P0	IP-1	P0	IP-1	P0	IP-1	P0*)	IP-1
West Lombok	84.3	88.9	11.40	10.07	89.93	90.06	0.58 a	0.59
Central Lombok	79.3	86.7	12.97	11.03	82.50	85.76	0.45 b	0.52
East Lombok	82.0	85.9	12.49	11.56	89.37	90.35	0.48 ab	0.53
Sumbawa	81.7	87.4	12.67	10.22	89.70	90.54	0.55 ab	0.56
Bima	80.9	88.2	12.40	11.01	86.83	89.88	0.53 b	0.54
LSD 5%	ns	ns	ns	ns	ns	ns	0.12	ns

Table 7. Seedling height, number of leaves, and collar diameter of *Jatropha curcas* of West Nusa Tenggara Genotypes of wild population (P0) and Improved Population- 1 (IP-1)

Genotypes	Seedling height (cm)		Number of leaves		Collar diameter (cm)	
	1 month old	2 month old	1 month old	2 month old	1 month old	2 month old
P0						
West Lombok	15.7±1.078	22.9±0.893	4.6±0.925	8.9±0.908	1.1±1.142	1.3±0.877
Central Lombok	18.3±1.694	21.2±1.132	4.7±1.106	8.2±1.062	0.9±1.333	1.0±0.992
East Lombok	15.2±1.426	20.1±1.426	5.1±1.077	7.8±1.016	0.8±1.434	1.1±0.902
Sumbawa	19.1±1.148	22.2±0.926	5.4±0.945	9.1±0.921	1.0±1.024	1.3±0.743
Bima	17.5±0,992	21.8±0.901	3.8±0.982	7.2±0.933	1.1±1.872	1.4±0.967
IP-1						
West Lombok	17.4±0.789	23.7±0.656	5.2±0.819	9.5±0.445	1.1±0.763	1.4±0.428
Central Lombok	18.9±0.992	22.4±0.793	5.6±0.925	8.9±0.545	1.0±0.784	1.1±0.457
East Lombok	17.3±0.905	21.2±0.885	5.7±0.961	8.7±0.607	0.9±0.825	1.1±0.724
Sumbawa	18.5±0.902	23.6±0.776	5.6±0.786	9.6±0.416	1.0±0.641	1.3±0.409
Bima	17.8±0.814	22.4±0.664	5.1±0.807	8.9±0.378	1.0±0.706	1.3±0.513

Note for Table 7 and 8: P0: seed from wild plant population, IP-1: seed from the first improved population, ±: value of standard deviation

Table 8. Dry weight of seedling shoot and seedling root of *J. curcas* of West Nusa Tenggara Genotypes of wild population (P0) and Improved Population-1 (IP-1)

Genotypes	Seedling shoot (g)		Seedling root (g)	
	1 month old	2 month old	1 month old	2 month old
P0				
West Lombok	4.95±1.798	7.33±1.233	0.94±1.595	1.74±1.013
Central Lombok	3.55±1.882	6.88±1.372	0.63±1.551	1.35±1.154
East Lombok	3.92±1.908	6.91±1.501	0.66±1.462	1.43±1.096
Sumbawa	4.71±1.636	7.16±1.362	0.79±1.357	1.58±1.005
Bima	4.32±1.872	7.43±1.239	0.88±1.623	1.66±1.102
IP-1				
West Lombok	5.79±1.124	9.01±0.863	1.23±1.026	1.95±0.902
Central Lombok	4.95±1.132	8.28±0.994	0.96±1.147	1.69±1.006
East Lombok	5.19±1.164	8.76±0.907	1.12±1.115	1.71±1.021
Sumbawa	5.46±1.087	9.12±0.821	1.29±1.076	1.98±0.942
Bima	5.24±1.139	8.95±0.879	1.22±1.083	2.02±0.879

Kernel weight percentage

Analysis of variance indicated that different genotype had a significant effect ($P<0.05$) on the percentage of kernel weight to total seed weight (Table 4). The maximum kernel weight was found in West Lombok, East Lombok, Sumbawa, and Bima, while the minimum was in Central Lombok. However, after mass selection, it was found that West Lombok had the maximum kernel weight, and Central Lombok possessed the minimum.

100 seed weight (seed index)

The same phenomena as that found in kernel weight percentage were observed in 100 seed weight. The maximum 100 seed weight was found in West Lombok, East Lombok, Sumbawa, and Bima, while the minimum was observed in Central Lombok. After mass selection, it was found that West Lombok had the maximum 100 seed weight with Central Lombok having the minimum (Table 4).

Kernel oil content

Selected genotypes within West Nusa Tenggara province had a significant effect on kernel oil content. However, after mass selection, the five *J. curcas* West Nusa Tenggara selected genotypes showed no variation or no significant difference in the kernel oil content (Table 5).

Seed viabilities

Seed viability component of *J. curcas* seed of West Nusa Tenggara genotypes are given in Table 6. There were no significant effects of genotypes on seed germination, germination rate, and vigor of seed. However, seed viabilities were better after mass selection (IP-1) than the wild genotypes.

Seedling characters

Seedling height

Seedling height ranged from 15.2 cm to 19.1 cm for one-month-old seedlings and from 20.1 cm to 22.9 cm for two-month-old seedlings in different genotype within their wild population (P0). Within their first improved population (IP-1), seedling height also varied among different genotypes, even though there was a decrease in variation within each genotype. The highest (22.9 cm for P0 and 23.7 cm for IP-1) seedling height was found in West Lombok genotype and the lowest (20.1 cm for P0 and 21.2 cm for IP-1) was found in East Lombok genotype.

Number of leaves

Number of leaves varied within five genotypes and it ranged from 7.8 to 9.1 in the wild population (P0) and from

8.7 to 9.6 in the first improved population (IP-1) (Table 7). There was improvement of homogenosity or decreased variation due to mass selection.

Collar diameter

Table 7 showed that genotype had different effect on collar diameter. It ranged from 1.0 cm to 1.4 cm within the wild population (P0), and 1.1 cm to 1.4 cm within their first improved population (IP-1). The variation within each genotype decreased after mass selection of the wild population.

Discussion

This study showed that seed and seedling characteristics of *J. curcas* varied among different selected genotypes of West Nusa Tenggara province. Genotype had genotypic characters according to their location of seed sources. Seed sources varied in their growing habitat with respect to altitude, temperature, and rainfall. The sources used in this study had mean annual rainfall (550-1.500 mm), temperature (24-32^{O}C), and altitude (30-100 m). Sumbawa, Bima, and West Lombok are drier than Central Lombok and East Lombok (Table 1). Therefore, variation in sources with respect to seed and seedling characteristics are mainly due to the fact that those genotypes grow over a wide range of climatic conditions in West Nusa Tenggara.

When the genotypes were grown in experimental field at Amor-Amor, Subdistrict Kayangan, West Lombok they were influenced by local climatic condition affecting seed and seedling performance such as reduction in their variability, especially within IP-1 population). Ginwal et al. (2004), Ginwal et al. (2005) and Kaushik et al. (2007) reported significant variations in seed morphology and seedling growth variables like seedling height, collar diameter, leaves, seed weight, and 100 seed weight in 10 accessions of *J. curcas*.

Since there was no different phenomenon of the influence of genotype within P0 and IP-1, it means that various climatic factors affected the vegetation collectively, but not individually. Considering this fact, genotypes may possess different climatic features that caused genotype variations. The present results are similar to the finding of Zhang et al. (2011), that genetic variation among genotype or provenance may be due to geographical separation.

West Lombok's genotypes followed by Sumbawa's, and Bima's were the best genotypes compared to Central Lombok and East Lombok with respect to their seed and seedling characteristics. There is correlation between good seed characteristic and good seedling characteristic. As Isik (1986) stated, that seed size and seed weight were two important characteristics for improving seedling productivity, hence, it was clear that seeds with greater seed weight produced seedlings with greater shoot and root growth. This may be due to greater nutrient reserves in larger seeds (Bhat and Chauhan 2002; Gonzales 1993).

The purpose of genotype testing is to measure the value of genetic variation and to aid further selection of better adapted and highly productive seed sources or genotypes. Variation in seed and seedling characteristics of *J. curcas* of West Nusa Tenggara genotypes is an indicator of the possibility of selecting the best performance or the highest seed yield of the tree for further crop improvement programs. Due to the fact that no different phenomenon of genetic variability of five *J. curcas* of West Nusa Tenggara selected genotypes both in wild population (P0) and first improved population (IP-1), it can be said that those variability caused by genetic factor with minor effect of environment. Therefore, as Gohil and Pandya (2009) state, that if variability is largely due to genetic cause with least environment effect, probability of isolating superior genotype is a precondition for obtaining higher yield.

The variability in seed, oil content, and seedling characteristic along with variability in early growth performance indicates that economic benefits may be obtained. Although, Parthiban et al. (2011) reported that in India, few native Jatropha species were utilized in their improvement program with limited success; the results of this study will be valuable for strategies for conservation of genetic variation, prospects of improvement and assessment of the potential of locally adapted seed sources. As Boe (2003) mentions, that since seed is the main product of trees selection for increasing seed weight and their content (seed-oil concentration), it may become important selection criterion for new cultivar development.

CONCLUSION

Jatropha curcas improvement program will be successful only after assessing our native genetic strength and the possible option toward yield improvement. Result of the present study revealed that considerable genetic variability existed among the five *J. curcas* West Nusa Tenggara selected genotypes and within each genotype population for seed and seedling characteristics. Genotypes of West Lombok, Sumbawa, and Bima performed exceedingly better than that of Central Lombok and East Lombok in terms of seed and seedling characteristics. These variations could occur from genetic diversity that needs to be studied in detail for their performance on seed production potential and for further *J. curcas* improvement program.

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