

## Seed viability of *Jatropha curcas* in different fruit maturity stages after storage

BAMBANG BUDI SANTOSO<sup>1,\*</sup>, ARIS BUDIANTO<sup>2</sup>, I.G.P. MULIARTA ARYANA<sup>1</sup>

<sup>1</sup>Energy Crops Centre, Faculty of Agriculture, University of Mataram, Jl. Majapahit No. 62 Mataram 83125, West Nusa Tenggara, Indonesia. Tel. +62-0370 621435, Fax. +62-0370 640189, \*email: bbs\_jatropha@yahoo.com

<sup>2</sup>Faculty of Agriculture, University of Mataram, Mataram 83125, West Nusa Tenggara, Indonesia.

Manuscript received: 8 October 2012. Revision accepted: 10 November 2012.

**Abstract.** Santoso BB, Budianto A, Aryana IGPM. 2012. Seed viability of *Jatropha curcas* in different fruit maturity stages after storage. *Nusantara Bioscience* 4: 113-117. The effect of fruit maturity stages and seed storage period to seed viability was investigated. Seed samples of West Lombok, West Nusa Tenggara genotype of *Jatropha curcas* were collected from a stand of two-year-old trees at an experimental field. The seed samples obtained were in four different stages of fruit maturity involving early maturity (green fruit), physiological maturity (yellow fruit), over maturity (brownies fruit), and senescence (black-dry fruit). The results showed that fruit maturity and storage period had an influence on the seed viability of *J. curcas*. The best fruit maturity stage for seed viability including seed oil content was found in yellow fruit and brownies fruit. For germination to be preserved, seeds could be stored in the ambient room storage for at least five months. For oil extraction, seed should preferably be stored not more than four months under ambient room conditions.

**Key words:** germination rate, *Jatropha curcas*, room condition, seed oil content, seed quality

**Abstrak.** Santoso BB, Budianto A, Aryana IGPM. 2012. Viabilitas biji *Jatropha curcas* pada tahapan kematangan berbeda setelah penyimpanan. *Nusantara Bioscience* 4: 113-117. Pengaruh tahapan kematangan dan periode penyimpanan terhadap viabilitas biji telah diteliti. Sampel biji *Jatropha curcas* genotip Lombok barat diambil dari tegakan pohon berusia dua tahun di lapangan percobaan. Sampel biji yang diperoleh memiliki empat tahapan kematangan, yaitu awal kematangan (buah hijau), kematangan fisiologis (buah kuning), kematangan berlebih (buah kecoklatan) dan tua (buah kering hitam). Hasilnya menunjukkan bahwa kematangan dan periode penyimpanan memiliki pengaruh pada viabilitas biji *J. curcas*. Tingkat kematangan terbaik untuk biabilitas biji ditemukan pada buah kuning dan kecoklatan. Untuk mempertahankan perkecambah, biji harus disimpan paling tidak lima bulan di kondisi ruangan penyimpanan. Untuk ekstraksi minyak, biji sebaiknya disimpan tidak lebih dari empat bulan di kondisi ruangan penyimpanan.

**Kata kunci:** laju perkecambahan, *Jatropha curcas*, kondisi ruangan, kadar minyak biji, kualitas biji

### INTRODUCTION

*Jatropha curcas* L. is a multipurpose plant with many attributes and considerable potential. It is a tropical plant that can be grown in low to high rainfall areas and can be planted in reclaimed land as a fence or commercial crop. The seed of this plant produces oil. Because *Jatropha* oil can be used in place of kerosene and diesel fuel, it has been promoted to make rural areas self-sufficient in fuel for cooking, lighting, and motive power (Openshaw 2000). Then, *J. curcas* is expected to be a high potential energy crop in Indonesia (Nazir and Setyaningsih 2010).

Despite these numerous benefits and potential, the production and development program of *J. curcas* in Indonesia has been faced with a number of challenges. One of the constraints is the lack of good seeds in quality and quantity owing to some problems in seed multiplication. Little information is available on quality seed production and post-harvest handling.

Seed quality is often interpreted in terms of genetic traits, germination capacity, purity, and storage potential

(ISTA 1999). Simic et al. (2007) also viewed seed quality as multiple criteria that encompasses several important seed attributes such genetic and chemical composition, germination and vigor, seed water content, and also the presence of seed-borne pathogen. Moreover, poor germination can result from the use of immature seeds (Batin 2011) and storage duration and condition (Dharmaputra et al. 2009; Akowuah et al. 2012). Crop productivity can be increased by increasing the germination rate which is possible by optimizing important parameters which are crucial for germination (Cheema et al. 2010). In the same manner, successful plantation activities for *J. curcas* provide viable seeds for the production of quality seedlings. Seed germination and seedling establishment are the most critical stages for survival during the life cycle of the individual *J. curcas* plant. To date, the potential of this plant is still constrained by the lack of technical information particularly in selecting the best fruit maturity color that could give the most excellent seed germination and seedling growth performance.

As Cheema et al. (2010) state, that seed should be produced in proper condition as seed from harsh environment is not good for further crop production. So, proper selection of fruit maturity of *J. curcas* based on color must be ascertained in order to produce quality planting stocks to meet the increasing demand for *J. curcas* as rehabilitation species and source of oil. Hence, the documentation of fruit color indicating maturation stages of *J. curcas* is necessary to address issues on poor germination and growth. This article described the effect of fruit maturity stages at different time of seed storage period on seed viability.

## MATERIALS AND METHODS

### Plant materials

Seed samples of West Lombok, West Nusa Tenggara, Indonesia genotype of *J. curcas* were collected from a stand of two-year-old trees at an experimental field. The seed samples were obtained in four different stages of maturity involving early maturity (green fruit), physiological maturity (yellow fruit), over maturity (brown fruit), and senescence (black and dry fruit) as shown in Fig.1. The harvested was done in February-March 2010 and the seed storage was done during April-September 2010.

### Procedures

#### *Collection, packaging, and storing of seeds*

Seeds of *J. curcas* were sun-dried for two days and the moisture content was determined using standard hot air oven method at  $105 \pm 1^\circ\text{C}$  for 24 hours (Pradhan et al. 2009). Then two kg of sun-dried *J. curcas* seed was placed in a sac of polypropylene (PP) plastic and then stored in the room condition for duration of 6 months. Three replicates were used for each seed maturity stages. The ambient temperature and relative humidity of the storage room were recorded using a thermohyrometer.

### *Samples handling*

Each sample (100-150 g of seeds) derived from each seed sack (replication) was taken monthly for the determinations of seed water content, seed oil content, seed weight, and seed viability (number of germinating seed and germination rate).

### *Determination of seed viability*

Viability (percentage of germination and germination rate) of seeds from each sample was determined by growing 100 seeds in plastic container containing sand media under greenhouse conditions. Daily germination counts were taken and recorded up to 15 days (time after which no seed was observed to germinate). The results were calculated as percentage of normal seedlings.

### *Determination of seed water content*

Water contents of seeds (based on wet basis) were determined every month based on oven method (gravimetry method). Two samples were used for each replicate (sack).

### *Determination of seed oil (lipid) content*

Seed (kernel) oil (lipid) contents were determined based on Soxhlet extraction method (AOAC 1999) with hexane as the solvent. The extracted lipid was obtained by filtrating the solvent using a rotary evaporator apparatus at  $40^\circ\text{C}$  followed by heating in an oven at  $105^\circ\text{C}$  for three hours to evaporate any remaining solvent and water.

### Statistical analysis

The data were statistically analyzed using mean and standard deviation. Analysis of Variance was applied to test the variation between different stages of fruit maturity through seed viability, seed moisture content, seed oil content, and other characteristics. Least significant difference (LSD at 5% level) was also subjected to significant findings.



**Figure 1.** Maturity stages of *J. curcas* fruit studied. A. early maturity (green fruit), B. physiological maturity (yellow fruit), C. over maturity (brown fruit), and D. senescence (black and dry fruit).

## RESULTS AND DISCUSSION

### Results

The range of ambient temperature and relative humidity of the storage room is presented in Table 1. The temperature ranged between 26.1 and 29.6 °C, whereas the humidity ranged between 71.4 and 83.4 %. The condition of storage room was relatively steady state during the storage period.

**Table 1.** The range of temperature and relative humidity of storage room during storage.

Duration of storage (month)	Temperature (°C)	Relative humidity (%)
0 - 1	26.5 - 28.3	72.2 - 80.5
1 - 2	26.1 - 28.9	75.1 - 83.4
2 - 3	26.3 - 29.4	75.5 - 81.9
3 - 4	26.7 - 29.5	73.4 - 80.8
4 - 5	26.4 - 29.6	72.6 - 80.8
5 - 6	26.3 - 29.4	71.4 - 80.7

Variations and significant difference of fresh fruit and seed characteristic among fruit maturity are given in Table 2. Fruit weight was found maximum (14.7±1.37 g) in green fruit and minimum (5.8±0.78 g) in black-dry fruit of fruit maturity. Not only weight of fruit but also seed moisture content, weight of fruit shell, and weight of seed were found maximum in green fruit and minimum in black-dry fruit.

**Table 2.** Characteristics of fresh fruit and seed after harvest at different fruit maturity stages

Fruit maturity (fruit color)	Seed moisture content (%)	Weight of fruit (g)	Weight of fruit shell (g)	Weight of seed (g)
Green fruit	40.6±1.64	14.7±1.37	11.8±1.02	2.7±0.33
Yellow fruit	32.7±1.42	12.4±1.02	9.4±0.87	2.5±0.21
Brown fruit	29.8±1.11	9.7±0.82	7.9±0.83	2.1±0.11
Black-dry fruit	21.9±0.95	5.8±0.78	4.1±0.82	1.8±0.06
LSD 5%	6.6	3.8	4.5	0.7

Note: ±: value of standard deviation. Means differ significantly at  $P < 0.05$ .

Percentage of seed germination and germination rate of *J. curcas* seeds in this study differ significantly among fruits maturity stages in the storage period of six months (Table 3 and Table 4). Seed taken from yellow fruit and brown fruit had the highest percentage of seed germination during six months of storage period. For germination rate, it was seed taken from yellow fruit, brown fruit, and black-dry fruit had higher rate that of green fruit.

Weight of 100 of *J. curcas* seeds in this study differs significantly among fruit maturity stages in the storage period until six months (Table 5). Decrease of seed weight during storage as consequence of decrease in their moisture content (Table 6). It was observed that there was significant difference in the moisture content of the seeds at room

condition. However, there was no significant difference at the 3 to 5 month of stored seed (Table 6.). Higher and lower seed water content was recorded at green fruit and yellow to black-dry fruit respectively. It could be said that there was a marginal decrease in seed water content of the seeds related to fruit maturity. Seed from green fruit had high water content at the beginning of storage (12.4%), while the water content of seed from yellow to black-dry fruit ranged from 6.7-8.2%. Then, seed water content of *J. curcas* seeds decreased during seed storage.

Oil analysis of seeds was carried out after six months of storage. Seed oil content was influenced by storage period and maturity stage of fruit (Table 7.). The oil content of seeds varied from minimum of 32.3% (green fruit) to maximum which ranged between 35.8 to 36.9% (yellow, brownies, and blk-dry fruit) at the beginning of storage period. After six months of storage, oil content of seeds varied from minimum 8.9% (green fruit) to maximum 23.3% (yellow fruit). Therefore, seed oil content of *J. curcas* seeds harvested at yellow maturity was not different from that at brownies to black-dry maturity fruit. Those phenomena existed during first three months of storage period. At the period of three to six month of storage the seed oil content harvested at yellow fruit was no different from that of brownies fruit.

### Discussion

Seed germination is affected by two factors, i.e. internal and external factors. Internal factor consists of the level of seed maturity, seed size, dormancy, and germination inhibitor. In this study, fruit maturity stages had significant effect on germination of seed, germination rate, and also on the seed weight, water content, and oil content of storage *J. curcas* seeds (one to six months of storage). In present study, the highest percentage of seed germination was observed in seeds taken from yellow fruit and brownies fruit from the beginning of storage until six months of storage.

Seed taken from young fruit (green fruit) produced immature seed, therefore, resulting in low and delayed germination. This proved the claim of Basra (2006) and Batin (2011) that seed viability includes *J. curcas* seed, is higher at the mature stage and decreases at early or late harvest or maturity. Harvesting *J. curcas* fruit too early (green fruit) results in more immature seeds with lower germination of seed and germination rate.

The fresh weight of fruits, shells, and seeds changed during maturation, ripening, and senescence. Fruits, shells, and seeds fresh weight increased significantly when the fruits were ripe (fully yellow) but reduced when they started to senesce. Biomass of *J. curcas* fruits was significantly different according to their maturity stage due to high water content at physiological maturity stage and low water content at senescence stage (Gunaseelan 2009). Germination capacity as seed viability increases during seed maturation. In this *J. curcas*, maximum seed viability coincided with the attainment of maximum seed dry weight or physiological maturity (yellow to brownies fruit color) and decline thereafter. According to Adikadarsih and

Hastono (2007), the lowest percentage of *J. curcas* seed germination (7%) was found in seeds derived from green fruits while the highest percentage (91.6%) was found in seeds derived from yellow fruit. Moreover, Wellbaum and Bradford (1990) found that germination capacity generally increased progressively and coordinately during seed maturation. Probert and Hay (2000) state that maximum seed qualities, normally harvested as dry seeds, were attained at or close to physiological maturity even though for others continue to increase well into the post-abscission phase.

Hard seed coat prevents oxygen and moisture entering the seed and prevents auto-oxidation of linoleic and linolenic acid which is responsible for degradation of cellular organelles (Cantliffe 1998). Therefore, as the time of seed storage increases, so does cellular damage. Gingwal et al. (2004) also reported that germination of *J. curcas* seed fell below 50% within 15 months of storage. In the same manner, Ellis et al. (1990) and Ghasemnezhad and Honemeier (2009) say that melon and sunflower seeds are difficult to store because germination and vigor deteriorate quickly in storage due to the high oil content in the seed. This study also was in agreement with that of Kumari et al. (2011), that the Indias *J. curcas* seeds loses its viability upon storage. In addition, Cheema et al. (2010) state, that there was a decrease in rate of germination of castor seeds due to low availability of moisture (seed water content). Therefore, the seeds of *J. curcas* for seedling should be derived from the yellow and brown fruits with storage period not more than four-month within room condition.

As storage period increased in present study, oil content of seed decreased. Then this study is in agreement with that of Akowuah et al. (2012) which showed that percentage of seed oil content of *J. curcas* gradually decreased with increasing storage time. According to Ahmadhan and Shahidi (2000) and Morello et al. (2004), it was due to the development of rancidity or deterioration of lipids in vegetable oil during storage. As Akowuah et al. (2012) state that aging process naturally affects the quality of seeds during storage at various conditions, especially oil content which is sensitive to deterioration as result of the reaction between unsaturated fatty acid and oxygen. This might be a reason

**Table 3.** Seed germination at different fruit maturity stages after storage

Fruit maturity (fruit color)	Duration of storage (month)						
	0	1	2	3	4	5	6
Green fruit	90.3 a	86.6 a	57.3 a	41.5 a	29.6 a	11.9 a	8.6 a
Yellow fruit	97.5 bc	95.4 b	81.6 b	79.9 c	68.7 c	59.3 c	56.9 c
Brown fruit	98.7 c	96.1 b	79.9 b	70.1 c	61.3 c	53.4 c	48.4 c
Black-dry fruit	92.1 ab	87.1 a	60.1 a	52.2 b	45.8 b	32.9 b	25.5 b
LSD 5%	6.2	5.7	5.8	10.4	9.8	8.5	8.9

Note: numbers in the column with the same letter did not differ significantly at  $P < 0.05$ .

**Table 4.** Rate of seed germination at different fruit maturity stages after storage

Fruit maturity (fruit color)	Duration of storage (month)						
	0	1	2	3	4	5	6
Green fruit	9.3 b	7.8	10.6 b	14.2 b	20.5 b	26.1 b	33.2 b
Yellow fruit	6.6 a	6.9	7.4 a	7.6 a	7.9 a	9.6 a	9.9 a
Brown fruit	6.7 a	6.8	7.4 a	7.5 a	7.8 a	9.8 a	10.2 a
Black-dry fruit	5.8 a	6.5	6.8 a	7.3 a	7.6 a	8.7 a	10.8 a
LSD 5%	2.5	ns	2.6	3.1	4.5	4.7	5.2

Note: numbers in the column with the same letter did not differ significantly at  $P < 0.05$ , ns: not significant

**Table 5.** Weight of 100 seeds at different fruit maturity stages after storage

Fruit maturity (fruit color)	Duration of storage (month)						
	0	1	2	3	4	5	6
Green fruit	108.3 c	98.7 c	68.2 b	63.4 a	61.9 a	52.1 a	47.7 a
Yellow fruit	90.7 b	87.8 b	85.6 b	84.3 b	83.1 b	82.7 c	80.3 c
Brown fruit	81.6 b	78.4 b	76.7 b	75.2 b	74.8 b	74.4 c	74.1 c
Black-dry fruit	69.8 a	67.2 a	66.5 a	65.1 a	64.7 a	63.2 b	62.8 b
LSD 5%	11.5	10.2	9.8	9.4	10.3	10.1	12.2

Note: numbers in the column with the same letter did not differ significantly at  $P < 0.05$ .

**Table 6.** Seed water content at different fruit maturity stages after storage

Fruit maturity (fruit color)	Duration of storage (month)						
	0	1	2	3	4	5	6
Green fruit	12.4 b	10.2 b	8.9 b	7.8	7.2	5.6	4.4 a
Yellow fruit	8.2 a	7.9 a	7.5 ab	7.3	7.1	6.8	6.4 b
Brown fruit	7.4 a	7.2 a	7.1 ab	7.1	6.7	6.6	6.3 b
Black-dry fruit	6.7 a	6.5 a	6.4 a	6.1	5.8	5.2	4.9 a
LSD 5%	2.1	1.9	2.2	ns	ns	ns	1.7

Note: numbers in the column with the same letter did not differ significantly at  $P < 0.05$ , ns: not significant

**Table 7.** Seed oil content at different fruit maturity stages after storage

Fruit maturity (fruit color)	Duration of storage (month)						
	0	1	2	3	4	5	6
Green fruit	32.3 a	31.9 a	26.6 a	23.7 a	18.3 a	14.2 a	8.9 a
Yellow fruit	36.9 b	37.2 b	36.5 b	34.6 c	31.1 b	29.7 c	23.3 c
Brown fruit	36.2 b	36.8 b	36.1 b	34.2 bc	30.8 b	29.1 bc	22.8 bc
Black-dry fruit	35.8 b	36.1 b	35.3 b	32.1 b	29.7 b	27.5 b	20.1 b
LSD 5%	3.3	3.6	4.2	2.1	2.6	2.0	2.7

Note: numbers in the column with the same letter did not differ significantly at  $P < 0.05$ .

why the percentage of oil of stored *J. curcas* seeds tends to reduce during storage. In addition Taiz and Zeiger (2002) and Basra (2006) state that, the metabolism of seed during storage to provide energy for its physiological activities could be another reason for seed oil decrease during storage.

### CONCLUSION

Fruit maturity and storage period had an influence on the seed viability of *J. curcas*. The best fruit maturity stage for good seed viability including seed oil content was found in yellow fruit and brown fruit. Germination percentage and germination rate were statistically the same when the fruits were harvested at yellow and brown ripe. For germination to be or preserved, seeds could be stored in the ambient room storage for at least five months. In addition, for the purpose of oil extraction, seed should preferably be stored not more than four months under ambient room conditions.

### REFERENCES

- Adikadarsih S, Hastono J. 2007. The effect of fruit maturity on the quality of physic nut seeds in Indonesian. Proceeding of Workshop II: Technology State of *Jatropha curcas* plant. Bogor, Indonesia, 29 Nov. 2006.
- Ahmadhan M, Shahidi F. 2000. Oxidative stability of stripped and no stripped borage and evening primrose oils and their emulsions in water. *J Amer Oil Chem Soc* 77: 963-968.
- Akowuah JO, Addo A, Kemausuor F. 2012. Influence of storage duration of *Jatropha curcas* seed on oil yield and free fatty acid content. *ARPN J Agric Biol Sci* 7: 41-45.
- AOAC [Association of Official Analytical Chemists]. 1999. Official methods of the Association of Agricultural Analytical Chemist. Association of Analytical Chemist, Inc., Arlington
- Basra AS. 2006. Handbook of seed science and technology. Haworth Press, New York.
- Batin CB. 2011. Seed germination and seedling performance of *Jatropha curcas* L. fruit based on color at two different seasons in northern Philippines. International Conference on Environment and BioScience IPCBEE 21: 94-100.
- Cantliffe JD. 1998. Seed germination for transplants. *Hort Technol* 8: 4-14.
- Cheema NM, Malik MA, Qadir G, Rafique MZ, Nawaz N. 2010. Influence of temperature and osmotic stress on germination induction of different castor bean cultivars. *Pakistan J Bot* 42: 4035-4041.
- Dharmaputra OS, Worang RL, Syarif R, Miftahudin. 2009. The quality of physic nut (*Jatropha curcas*) seeds affected by water activity and duration of storage. *Microbiol Indon* 3: 139-145.
- Ellis RH, Hong TD, Robert EH. 1990. Effect of moisture content and method of rehydration on the susceptibility of *C. melo* seeds to imbibitional damage. *Seed Sci Technol* 18: 131-137.
- Ghasemnezhad A, Honemeier B. 2009. Influence of storage conditions on quality and viability of high and low oleic sunflower seeds. *Intl J Plant Prod* 3: 39-48.
- Gingwal SH, Phartyal SS, Rawat PS, Srivastava RL. 2004. Seed source variation in morphology, germination and seedling growth of (*Jatropha curcas*) Linn. in Central India. *Silvae Genetica* 54: 76-80.
- Gunaseelan NV. 2009. Biomass estimates, characteristics, biochemical methane potential, kinetics and energy flow *Jatropha curcas* on dry lands. *Biomass Bioeng* 33: 589-596.
- ISTA [International Seed Testing Association]. 1999. International rules for seed testing. Seed Science and Technology. International Seed Testing Association, Bassersdorf, Switzerland
- Kumari A, Joshi PK, Arya MC, Ahmed Z. 2011. Enhancing seed germination of *Jatropha curcas* L. under Central-Western Himalayas of Uttarakhand, India. *Plant Arch* 11: 871-874.
- Morello JR, Motilva MJ, Tovar MJ, Romero MP. 2004. Changes in commercial virgin olive oil (CV Arbequina) during storage with special emphasis on the phenolic fraction. *J Food Chem* 85: 357-364.
- Nazir N, Setyaningsih D. 2010. Life cycle assessment of biodiesel production from palm oil and jatropha oil in Indonesia. 7<sup>th</sup> Biomass Asia Workshop. Jakarta, Indonesia.
- Openshaw K. 2000. A review of *Jatropha curcas*: An oil plant of unfulfilled promise. *Biomass Bioeng* 19: 1-15.
- Pradhan RC, Naik SN, Bhatnagar N, Vijay VK. 2009. Moisture-dependent physical properties of jatropha fruit. *Ind Crop Prod* 29: 341-347.
- Probert RJ, Hay FR. 2000. Keeping seeds alive. In: Black M, Bewley JD (eds). Seed technology and its biological basis. Sheffield Academic Press, Sheffield, UK.
- Simic B, Popovic R, Sudaric A, Rozman V, Kalinovic I, Cosic J. 2007. Influence of storage condition on seed oil content of maize, soybean and sunflower. *CCS Agriculturae Conspectus Scienticus* 72: 211-213.
- Taiz L, E Zeiger. 2002. Plant physiology. 3<sup>rd</sup> ed. Sinauer, Sunderland, MA.
- Wellbaum GE, Bradford KJ. 1990. Water relation of seed development and germination in muskmelon (*C. melo* L.). V. Water relation of imbibition and germination. *Plant Physiol* 92: 1046-1052.