The drought stress tolerance of physic nut (Jatropha curcas Linn.) genotypes

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Abstract. Physic nut (Jatropha curcas Linn.) is one of tropical plant that has a potential as an alternative energy source. This study aimed at finding out the drought tolerance level of six genotypes of J. curcas based on the morphology and production of the first harvest time. The plant was treated under a condition of 100 %, 70 % and 40 % field capacity. The tested genotypes were 5-1-14 (SP8 × SP16), 6-2-10 (SP8 × SP38), 7-2-8 (SP33 × HS49), 18-1-14 (SM35 × SP38), IP3A and IP3P. The drought stress of 40 % ground water inhibited the plant vegetative and generative growth (the amount of leaves, male flower, carpel, hermaphrodite flower and the percentage of formed fruit) while at the same time it tended to reduce the amount of fruit and the seeds’ dry weight. The level of drought that reached 70 % resulted in the reduce of seeds’ dry weigh up to 87.76 % and upon 40 % of drought level, the seeds’ dry level was reduced nearly to 97.92 %. At 40 % moisture content, the highest percentage of fruit formation is achieved by genotypes 5-1-14 and 6-2-10.

Keywords: Bio-fuel crop, biofuel tree, breeding, renewable energy.

1. Introduction
The Indonesia’s dependence toward fossil-based fuel especially petroleum in fullfiling the internal consumption reaches 98 % (petroleum 48 %, gas 18 % and coal 30 %) of total national energy consumption so that it requires the development of bio-fuel alternative energy [1]. President Decree of Republic of Indonesia number 5 year 2006 on national energy policy had stipulated that biofuel target user from 0.2 % will increase to more than 5 % of total national energy consumption in 2025 [2]. Indonesia has 16 different plants that have potential as alternative energy source, one of which is Jatropha curcas Linn. [3]. Unfortunately the success rate of J. curcas in Indonesia is still low due to the limitation of qualified seed and plant [4]. The result of the study showed that the high productivity of J. curcas, was obtained on maximum irrigation with water content > 65 %, while the plant without irrigation experienced production decreased of 72.8 % [5].

The area of dry land in Indonesia reaches up to 123.1 ha. There are several things to be done to optimize the sub-optimum land, they are: through improvement of plant genetic capacity both conventionally and biotechnologically, through improvement of production system in sub-optimum area, improvement of agricultural infrastructure and through improvement of technical capacity and
farmer institutional as the main production agent. The appropriate area for *J. curcas* in Indonesia reaches $49.53 \times 10^6$ ha, which is categorized into three criteria of highly appropriate of $14.28 \times 10^6$ ha, moderately appropriate of $5.53 \times 10^6$ ha and marginal compliance of $29.72 \times 10^6$ ha. This land criteria were established based on land’s biophysical, climate and environment conditions [4].

Support for conversion of fuel oil to bio-fuel through alternative of *J. curcas* oil and the usage of dry land in Indonesia must be conducted by inventing drought proof varieties of *J. curcas*. This study aimed at testing the drought tolerance level of six genotypes F1 of physic nut (*J. curcas* Linn.) based on the morphological and yield capacity in the first harvest.

### 2. Materials and methods

The research was conducted by applying split plot design. There were two factors treatment with with the main plot is the soil water content, consisting of three levels of soil water content: 100 % (Control), 70 % and 40 % field capacity. The sub plots are *J. curcas* genotypes (six levels), which were: 5-1-14; 6-2-10; 7-2-8; 18-1-14; IP-3A and IP-3 P, in green house of Faculty of Agriculture and Animal Husbandry, University of Muhammadiyah Malang, East Java.

Plant seed applied in the study were cutting of hybridized *J. curcas* [6] with 20 cm length and 1 cm to 2 cm of diameter. After 30 d, the plant seeds were removed to polybags of (40 × 40) cm which was already filled with 10 kg of planting media. The dosage of fertilizer was 20 g Urea + 50 g SP36 + 10 g KCl [7]. The fertilized was given after the plant reached the age of 1 wk, in the form of half dosage of N fertilized and whole dosage of P and K fertilizers (20 g Urea + 50 g SP-36 + 10 g KCl), while half dosage of nitrogen fertilizer (10 g of Urea) was given when the plant reached the age of 8 wk. The treatment for drought level was given when the land water content reached 100 %, 70 % and 40 % of field capacity [8].

The study was conducted in two levels and the first treatment was drought treatment level which was conducted during the drought level reached 100 %, 70 % and 40 % of capacity field. The plant dessication in accordance to the treatment and water addition was conducted on daily basis during 2 mo period. The second treatment was recovery session after the dessication treatment. The land water content of field capacity was conducted using gypsum moisture meter. The dry moisture content is determined by: i) Weighing the empty bottle with the lid (A g), ii) Filling the growing media into a weighing bottle with half the volume of the bottle then closed and weighed (B g), iii) inserting the bottle into the oven with the lid open on constant temperature for 12 h, iv) After 12 h, the bottle is closed and removed until it reaches room temperature, v) Considering the cold bottle (C g), vi) Calculating the moisture content of dry soil with equation (1) [8]:

$$\text{Moisture content} = \frac{B \cdot C}{C - A} \times 100\%$$  \hspace{1cm} (1)

A = weight of empty bottle and bottle cap (g),
B = weight of bottle after filling with planting media (g),
C = weight of bottle and lid after being cooled (g),
value (CA) = sample weight of absolute dry soil,
value (BC) = the weight of water in the soil sample.

The morphological characteristics which were in the form of the number of formed leaves, fallen leaves, formed flower, the percentage of formed fruit, the number of fruit, total dry weight of seed and the length of root were observed during last session of the study. The obtained data were analyzed using variance analysis and the Duncan test.

### 3. Results and discussions

The genotype treatment affected the number of formed leaves. The biggest amount of formed leaves was achieved by genotype 5-1-14 which was not different with genotype IP-3P and 18-1-14. On 29 d to 56 d after the planting, the number of formed leaves were affected by the drought stress.
drought stress disturb the leaves formation, this was shown by land water content of 40 % as the treatment with the lowest amount of formed leaves, and it gradually increased as the percentage got higher which was on 70 % to 100 % (table 1).

The leaves growth is very sensitive toward water stress [9]. Different respond of plant growth on genotype which is rather tolerant and rather sensitive showed that the physiological responds toward drought stress varied based on the types of genotype. On vegetative phase, the drought stress may disturb the plant height, leaves formation and leaves surface accretion [10]. While on 57 d to 84 d (recovery phase), the number of formed leaves did not affect by land water content, the result of average leave amount on stress treatment of 40 % after recovery showed not much different amount. It was suspected the increase of formed leaved occurred due to the plant’s ability to recover from the drought stress effect.

Table 1. The average amount of formed leaves from variety of *J. curcas* genotypes and land water content during drought phase (1 d to 56 d after planting) and recovery phase (57 d to 84 d after planting).

| Genotype  | The amount of formed leaves (days after planting) |
|-----------|---------------------------------------------------|
|           | (1 to 28) d | (29 to 56) d | (57 to 84) d |
| 5-1-14    | 21.17 c     | 11.67 a      | 15.00 a      |
| 6-2-10    | 16.00 bc    | 7.00 a       | 9.83 a       |
| 7-2-8     | 12.50 ab    | 10.08 a      | 10.33 a      |
| 18-1-14   | 20.67 c     | 11.42 a      | 15.67 a      |
| IP3A      | 9.00 a      | 8.00 a       | 12.00 a      |
| IP3P      | 22.08 c     | 17.92 a      | 22.50 a      |

Soil water content (% field capacity)

| Soil water content | The amount of formed leaves (days after planting) |
|--------------------|---------------------------------------------------|
| 100 %              | 28.75 a | 19.21 c | 9.17 a |
| 70 %               | 18.75 a | 8.50 b  | 14.83 a|
| 40 %               | 3.21 a  | 5.33 a  | 18.67 a|

Note: Numbers which are followed by similar alphabet in the same column are similar during Duncan test of 5%.

The amount of fallen leaves on 1 d to 56 d (drought stress phase) and 57 d to 84 d (recovery phase) were not affected by land water content treatment and genotype (table 2). The higher water stress treatment is, the bigger intensity of leaves damage value resulting in chlorosis in the leaves [11]. This resulted in the decrease of leaves amount, which eventually affected the area of chlorophyll for photosynthesis. Some of morphological responses toward drought stress are to decrease water content by reducing heat adsorption through smaller leaves’ surface, leaf rolling, leaf folding or leaf falling [12].

Table 2. The average of fallen leaves from *J. curcas* genotypes and soil water content on drought phase (1 d to 56 d after planting) and recovery phase (57 d to 84 d after planting).

| Genotype  | The amount of fallen leaves |
|-----------|----------------------------|
|           | Drought phase | Recovery phase |
| 5-1-14    | 10.42 a        | 2.34 a         |
| 6-2-10    | 8.00 a         | 3.67 a         |
| 7-2-8     | 6.25 a         | 1.17 a         |
| 18-1-14   | 8.17 a         | 2.17 a         |
| IP3A      | 7.25 a         | 3.00 a         |
| IP3P      | 11.00 a        | 2.50 a         |

Table 2 continue to the next page.
The highest percentage of stamens decrease occurred on genotype IP-3A while the lowest was on genotype 5.1.14. During recovery phase, the genotype of IP-3A did not produce any stamens while the genotype of 5.1.15 was the one with the highest stamens increase which reached 2.61 % (table 3). During drought stress, the genotype IP-3A was the genotype with the highest stamens decrease due to the decrease of land water content to 40 %. On recovery phase, the genotype 6.2.10 was the genotype with the highest amount of carpels increase (table 4).

**Table 2.** The amount of stamens of various *J. curcas* genotypes during drought and recovery phases.

| Genotype | Drought phase | Change (%) | Recovery phase | Change (%) |
|----------|---------------|------------|----------------|------------|
|          | 100 % | 70 % | 100 % | 70 % | 100 % | 70 % |
| 5.1.14   | 261.00 a | 170.75 a | -48.84 | 191.50 a | 203.00 a | 6.01 |
| 6.2.10   | 260.00 a | 215.25 a | -17.21 | 196.00 a | 283.50 a | 44.64 |
| 7.2.8    | 332.50 a | 241.50 a | -27.37 | 163.00 a | 206.50 a | 26.69 |
| 18.1.14  | 275.75 a | 258.00 a | -6.44 | 377.00 a | 140.00 a | -62.86 |
| IP-3A    | 251.75 a | 80.25 a | -68.12 | 120.00 a | 66.00 a | -45.00 |
| IP-3P    | 111.25 a | 97.25 a | -12.58 | 326.50 a | 195.00 a | -40.28 |

**Table 4.** The amount of carpels of various *J. curcas* genotypes during drought and recovery phases.

| Genotype | Drought phase | Change (%) | Recovery phase | Change (%) |
|----------|---------------|------------|----------------|------------|
|          | 100 % | 70 % | 100 % | 70 % | 100 % | 70 % |
| 5.1.14   | 8.00 a | 3.00 a | -85.71 | 14.00 a | 23.50 a | 67.86 |
| 6.2.10   | 4.50 a | 5.50 a | -62.07 | 6.00 a | 15.00 a | 150.00 |
| 7.2.8    | 4.25 a | 12.50 a | -12.28 | 7.50 a | 9.00 a | 20.00 |
| 18.1.14  | 7.00 a | 13.00 a | -23.53 | 8.50 a | 6.00 a | -29.41 |
| IP-3A    | 0.75 a | 2.50 a | -76.74 | 6.00 a | 6.50 a | 8.33 |
| IP-3P    | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 a | 0.00 |

Note: Numbers which are followed by same alphabet in the same column are similar during Duncan test of 5 %.
produce more fruit [15]. The low rate of production was assumed also as a result of the inhibited meristems, fruit and seed.

The water content might cause the plant to reduce the leaf surface area in order to lower the photosynthesis weight seed compared to the control (100 % of land water content) (table 6). The decrease of leaf water content might cause the plant to reduce the leaf surface area in order to lower the photosynthesis rate which eventually reduce the photosynthesis product result as well as its translocation to the meristems, fruit and seed. J. curcas really needs the support of vegetative characteristics in order to produce more fruit [15]. The low rate of production was assumed also as a result of the inhibited

| Genotype | Drought phase | Change (%) | Recovery phase | Change (%) |
|----------|---------------|------------|----------------|------------|
|          | 100 % 40 %    | 100 % 70 % | 100 % 40 %     | 100 % 70 % |

Note: Numbers which are followed by same alphabet in the same column are similar during Duncan test of α 5 %; (-) indicates decrease value.

Table 5. The amount of hermaphrodite flower of various genotypes of physic nut during drought and recovery phases.

| Genotype | Drought phase | Change (%) | Recovery phase | Change (%) |
|----------|---------------|------------|----------------|------------|
|          | 100 % 40 %    | 100 % 70 % | 100 % 40 %     | 100 % 70 % |

Note: Numbers which are followed by same alphabet in the same column are similar during Duncan test of α 5 %; (-) indicates decrease value.

The percentage decrease of the amount of flower was suspected due to water supply for the photosynthesis was obstructed resulting in the low amount of photosynthesis result which eventually was unable to form its flowers. The growth and productivity of J. curcas becomes distracted during long dry season. During the dry season, J. curcas only blooms once a year due to water limitation, while in the area with irrigation system it can bloom all year long [13]. Based on plant reproduction structure, there are two types of plants namely monoecious (unisexual) and andromonoecious [14]. The monoecious flowers own genotypes of 5-1-14, 6-2-10, 7-2-8, 18-1-14 and IP-3A. While the IP-3P belongs to andromonoecious, resulting in more amount of hermaphrodite flowers than other genotypes.

The land water content treatment and plant genotype did not affect significantly toward the amount of fruit and seed dry weight. The 40 % of land water content decreased the amount of fruit and dry weight seed compared to the control (100 % of land water content) (table 6). The decrease of leaf water content might cause the plant to reduce the leaf surface area in order to lower the photosynthesis rate which eventually reduce the photosynthesis product result as well as its translocation to the meristems, fruit and seed. J. curcas really needs the support of vegetative characteristics in order to produce more fruit [15]. The low rate of production was assumed also as a result of the inhibited
transportation process of photosynthesis result through phloem tissues of plant. The phloem transportation is highly affected by turgor pressure of cell. During drought stress, there will be a decrease of phloem water potential which directly hamper the activity of photosynthesis product [16].

**Table 6.** Average fruit amount during drought phase (1 to 56) d and recovery phase and dry weight of seed per plant.

| Genotype | Amount of fruit (day after planting) | Seed dry weight/plant (g) |
|----------|-------------------------------------|--------------------------|
|          | 1 to 65 | 66 to 84 |          | 1 to 65 | 66 to 84 |          |
| 5-1-14   | 4.50 a   | 8.67 a   | 1.82 a   |          |          |          |
| 6-2-10   | 3.25 a   | 4.67 a   | 1.11 a   |          |          |          |
| 7-2-8    | 2.67 a   | 4.00 a   | 0.91 a   |          |          |          |
| 18-1-14  | 4.25 a   | 3.33 a   | 1.98 a   |          |          |          |
| IP-3A    | 1.83 a   | 3.33 a   | 0.89 a   |          |          |          |
| IP-3P    | 1.75 a   | 9.17 a   | 0.95 a   |          |          |          |

**Land water content**

|          | 100 %   | 70 %    | 40 %    |          |          |          |
|----------|---------|---------|---------|----------|----------|----------|
|          | 6.54 a  | 7.67 a  | 3.35 a  |          |          |          |
|          | 2.00 a  | 5.92 a  | 0.41 a  |          |          |          |
|          | 0.58 a  | 3.00 a  | 0.07 a  |          |          |          |

Note: Numbers which are followed by same alphabet in the same column are similar during Duncan test of α 5 %

**Table 7.** Percentage of fruit formed on 56 d and 84 d after drought stress.

| Land water content (%) | Genotype | Percentage (%) of fruit formed | The percentage difference (%) |
|------------------------|----------|--------------------------------|-------------------------------|
|                        |          | (1 to 56) d (Drought phase)    | (57 to 84) d (Recovery phase) |
| 100                    | 5.1.14   | 56.94 bc                       | 44.64 b                       | -21.60 |
|                        | 6.2.10   | 48.28 bc                       | 16.67 a                       | -65.47 |
|                        | 7.2.8    | 35.09 b                        | 43.33 b                       | 23.48  |
|                        | 18.1.14  | 58.82 bc                       | 14.71 a                       | -74.99 |
|                        | IP-3A    | 46.51 bc                       | 54.17 b                       | 16.47  |
|                        | IP-3P    | 11.27 a                        | 45.71 b                       | 305.59 |
| 70                     | 5.1.14   | 83.33 c                        | 12.77 a                       | -84.68 |
|                        | 6.2.10   | 18.18 a                        | 21.67 a                       | 19.20  |
|                        | 7.2.8    | 24.00 b                        | 13.89 a                       | -42.13 |
|                        | 18.1.14  | 13.46 a                        | 58.33 bc                      | 333.36 |
|                        | IP-3A    | 20.00 a                        | 26.92 a                       | 34.60  |
|                        | IP-3P    | 25.49                          | 55.56 b                       | 117.97 |
| 40                     | 5.1.14   | 60.00 c                        | 18.75 a                       | -68.75 |
|                        | 6.2.10   | 70.00 c                        | 20.37 a                       | -70.90 |
|                        | 7.2.8    | 0.00 a                         | 17.65 a                       | 100.00 |
|                        | 18.1.14  | 22.22 b                        | 5.56 a                        | -74.98 |
|                        | IP-3A    | 0.00 a                         | 0.00 a                        | 0.00   |
|                        | IP-3P    | 0.00 a                         | 50.00 b                       | 100.00 |

Note: Numbers which are followed by same alphabet in the same column are similar during Duncan test of α 5 %; Percentage obtained from the calculation of carpels and hermaphrodite and formed fruit; (-) Indicates a decrease value.
Genotype with the highest percentage of fruit formed by the carpels was 5-1-14 on 70 % water content, while the lowest percentage was genotype 7.2.8, IP-3A and IP-3P on 40 % water content. While during recovery process for 28 d, the genotype 18.1.14 held the highest percentage. The fruit formation during recovery process resulted in various amount, the highest occurred on genotype 7.2.8 and IP-3P. Several genotypes were not able yet to increase the number of formed fruit from its flowers. This was assumed due to the energy recover for flowering did not yet run perfectly. The similar process occurred in Pakuwon, where the flowering and fertilization of physic nut were affected by the environment in contact with the genetic factors of the plant [7]. Even the excellence provenance IP3 was only able to perform at its maximum extent on an optimum area. Upon no irrigation system, the seed production of *J. curcas* will reduce to 70 % [5]. Drought stress and genotype did not have any effect on the root length. The average root length can be viewed on table 8. Agronomic factors play an important role in determining the success of *J. curcas* cultivation [17]. The increase of root growth is one of the plant tolerance mechanisms against the drought stress [18]. This occurrence was possibly due to usage of polybag as the planting media with a limited root catchment area, and also due to the inhibited rate of photosynthesis product. The decrease of root length due to drought stress was reported occurred on Albizia and Populus (*Paraserianthes iophanta* Willd. and *Populus nigra* L.) species [12]. On Soya plant (*Glyxine max* L.), the drought stress with Poly Ethilene Glicol (PEG) 10 % had shorter root than the control [19].

### Table 8. Average root length on 56 d and 84 d after drought stress.

| Genotypes | Root length (cm) | 56     | 84     |
|-----------|------------------|--------|--------|
| 5-1-14    |                  | 56.85 a | 58.47 a|
| 6-2-10    |                  | 53.18 a | 58.53 a|
| 7-2-8     |                  | 47.12 a | 56.73 a|
| 18-1-14   |                  | 55.40 a | 55.13 a|
| IP-3A     |                  | 55.67 a | 54.95 a|
| IP-3P     |                  | 53.83 a | 47.28 a|
| Soil water content |          |        |        |
| 100 %     |                  | 61.67 a | 58.89 a|
| 70 %      |                  | 52.58 a | 59.02 a|
| 40 %      |                  | 46.78 a | 47.64 a|

Note: Numbers which are followed by same alphabet in the same column are similar during Duncan test of α 5 %.

The sensitivity index calculation showed that genotypes 5.1.14 and 6.2.10 included in moderate criteria, while the genotypes 7.2.8, 18.1.14, IP-3A and IP-3P included in rather tolerant criteria. Plant tolerant value toward drought stress was the physical tolerance expression toward the drought stress. Plant with rather tolerant criteria toward drought stress produced better plant growth than the moderate genotypes. The different growth response on rather tolerant and moderate genotypes showed the variety of physiological response toward drought stress beased on the variety of genotypes [20].

Drought stress in plants can be caused by a lack of water supply in the root area and excessive water demand by leaves due to the rate of evapotranspiration that exceeds the rate of water absorption even though the condition of groundwater is sufficiently available. In *J. curcas*, drought stress decreases the relative water of the leaves, and the density of the upper and lower leaf stomata [8]. Drought stress by reducing water to 40 % of field capacity inhibits vegetative growth of *J. curcas*, with a decrease in the size and dry weight of plants by an average of more than 50 % [13].
4. Conclusions
The drought stress to 40% of soil water content inhibits the vegetative and generative growth of plant (the amount of leaves, stamen, carpel, hermaphrodite flowers and the percentage of formed fruits), and tend to decrease the number of fruit and seed dry weight. The drought that reached up to 70% lowered the seed dry weight to 87.76% and when the drought reached 40% it would lower the seed dry weight to 97.92%. At 40% moisture content, the highest percentage of fruit formation is achieved by genotypes 5-1-14 and 6-2-10. In further research activities it is necessary to test the plants tolerance to drought stress in a field scale. Genotypes that show a tolerant response to drought stress, can be used by plant breeders as candidates for superior crosses elders.

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