Vegetative propagation of *Dryobalanops sumatrensis* and *Dryobalanops oblongifolia* subsp. *oblongifolia* by shoot cuttings

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**Abstract.** The existence of *Dryobalanops sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* in Indonesia’s natural forest is increasingly threatened due to land use changes and illegal logging leading to the urgent need of conservation actions in the field. Vegetative propagation by shoot cuttings has been proposed as an alternative method, yet there have been still lacks of information regarding the suitable rooting media and the use of shoots from saplings as cutting materials. This study was to evaluate the survival rate, rooting and shoot sprouting abilities of *D. sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* on two different media (mineral soil and peat). Saplings of the *D. sumatrensis* aged older than 7 years old and *D. oblongifolia* subsp. *oblongifolia* aged less than 2 years old were used as cutting materials. The results showed that the survival rate of *D. sumatrensis* in peat (43.75%) and mineral soil media (43.75%) was lower than *D. oblongifolia* subsp. *oblongifolia* in for peat media (54.55%) and minerals soil media (71.88%). The rooting and shoot sprouting percentage of *D. sumatrensis* in peat and mineral soil media was also lower than *D. oblongifolia* subsp *oblongifolia*. This pattern revealed that the media treatment (peat and minerals) did not affect the survival rate and rooting ability of *D. sumatrensis* shoot cuttings, but has an effect on *D. oblongifolia* subsp. *oblongifolia*.

1. Introduction

The potential for high-value uses of *Dryobalanops sumatrensis* (J.F. Gmelin) A.J.G.H. Kostermans (syn. *Dryobalanops aromatica* C.F.Gaertn.) and *D. oblongifolia* subsp. *oblongifolia* results in a large number of exploitation and illegal logging occurring [1, 2, 3]. As a result of massive logging, the amount found in nature of each of these species decreases. IUCN has granted vulnerable A2cd status to *D. sumatrensis* since 2018 [4] and Endangered A1cd status to *D. oblongifolia* subsp. *Oblongifolia* since 1998 [5]. Therefore, in-situ and ex-situ conservation efforts need to be made to maintain the existence of these two species. However, planting stocks production is still problematic due to irregular flowering and fruiting [6].
Procurement of seedling or planting materials from vegetative parts is the only way to propagate materials as an alternative to the propagation from seeds [4]. Vegetative propagation by shoot cuttings has been proposed as an alternative method because the method of propagation is simple and could be periodically executed or not dependent on flowering and fruiting season [7], yet there have been still lacks of information regarding the suitable rooting media and the use of shoots from saplings as cutting materials.

Two studies on shoot cuttings of *Dryobalanops aromatica* which had been done previously reported that the media used were sand, rice husk media, combination of both media [8], and combination of coco dust and rice husk with ration 2:1 [6, 9]. There are no studies that have tried to use other media such as mineral soil and peat media. Therefore, the aim of this study was to evaluate the survival rate, rooting and shoot sprouting abilities of *D. sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* on two different media (mineral soil and peat).

2. Materials and Methods

2.1 Preparation of cutting media

There were two type of cutting media used for the shoot cuttings of *Dryobalanops sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* in the study i.e peat media and mineral soil media. The peat media were collected from Sungai Beras Village, Tanjung Jabung Timur District, Jambi Province, Sumatera, Indonesia, whilst mineral soil was obtained from Forest Research and Development Center (FRDC), Forestry and Environmental Research Development and Innovation Agency (FOERDIA), Ministry of Environment and Forestry (MoEF) in Bogor, West Java, Indonesia. Each media was crushed and screened in order to get fine powder form of media and dried in open area before being distributed into a pot-tray.

2.2 Preparation of cutting materials

Cutting materials were obtained from orthotropic branches of saplings collections aged older than 7 years old for *Dryobalanops sumatrensis* and less than 2 years old for *D. oblongifolia* subsp. *oblongifolia* at KOFCO (Komatsu-FORDA Conservation) nursery, Forest Research and Development Center (FRDC), Forestry and Environmental Research Development and Innovation Agency (FOERDIA), Ministry of Environment and Forestry (MoEF) in Bogor, West Java, Indonesia. The orthotropic branches of each species were cut between the second and the third node of the shoot (about 5-7 cm) and half of leave area was reduced from its original size to reduce transpiration. The cuttings were placed in the bucket filled with half level of water. The tip of the cuttings was exposed to rooting hormone (Rootone-F) without any difference in the dose of each species and media. The cuttings were planted in pot-tray containing each medium and placed in propagation boxes according to the treatment. The number of *D. sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* shoot cuttings planted for each peat media and mineral soil media was 32 shoot cuttings. In total, 128 shoot cuttings were used in this study. The propagation boxes were stored in a greenhouse. Watering was done twice daily, once in the morning before 10.00 a.m. and once in the late afternoon after 4.00 p.m to ensure the seedlings received enough water during the initial growth stage.

2.3 Data collection

Study on propagation of *D. sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* was carried out nine months from August 2018 to April 2019 at KOFFCO (Komatsu-FORDA Fog Cooling System) greenhouse and nursery, FRDC, FOERDIA, MoEF, Bogor, West Java, Indonesia. The parameters observed in the shoot cuttings were survival percentage, rooting percentage and percentage of shoot sprouting of *D. sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* which were planted on peat media and mineral soil media.
Physical characteristics assessment of cutting media (minerals and peat) samples was carried out at the Forest Influence Laboratory, Department of Silviculture, Faculty of Forestry, IPB University (Bogor Agricultural University), Bogor, West Java, Indonesia. The parameters examined for mineral soil analysis were soil pH, soil texture and soil water content, while parameters examined for peat media were soil pH, soil texture, and peat maturity. Soil pH testing was conducted with pH test paper. Soil texture testing was carried out by using the pipette method [10], soil water content measurement was performed using Gravimetric method [11], whereas peat maturity testing was performed by using the handheld method [12].

2.4 Data analysis

The data of shoot cutting performance such as survival percentage, rooting percentage and percentage of shoot sprouting were analysed using a variance test to figure out whether there were significant differences in both media treatment. Data were analysed for normality by using the Shapiro Wilk test [13] to determine the type of variance test to be carried out. Normally-distributed data were tested using repeated ANOVA tests [14], whereas the abnormally-distributed data were tested using Friedman test [15].

3. Result and discussion

3.1. Cutting media characteristics

Cutting media is one of key factor for root formation [9] and one of the important media characteristics that should be considered is physical characteristic such as media porosity, which can be determined from the water content. Ideal media porosity for cutting material is able to catch water and has sufficient aeration [9].

In this study, the results of physical characteristic of peat and minerals media showed that both media possessed acidic pH of 4.0 for peat media and 5.0 for mineral soil media (Table 1). The largest water content was found in peat media (135.32%), meanwhile the water content in minerals media was only 32.09% (Table 1) indicated that mineral soil materials contain considerably less water than peat media because peat media was able to keep water content better than minerals. Peat media are organic rich materials, usually containing ≥20 mass % [16], which have a unique combination of physical properties of peat, including low bulk density, high total porosity, and the ability to swell and shrink upon wetting and drying [16].

| Media       | pH   | Water content (%) | Texture         |
|-------------|------|-------------------|-----------------|
| Peat        | 4.0  | 135.32            | Fibric          |
| Mineral     | 5.0  | 32.09             | Clay Platters   |

3.2. Shoot cuttings performance

3.2.1 Survival rate percentage.

The survival rate of *D. sumatrensis* shoot cuttings that planted on peat and minerals soil media in the last month of observation were the same (43.75% or 14 shoot cuttings of each media), while the survival rate of *D. oblongifolia* subsp. *oblongifolia* planted on peat media (54.55% or 18 shoot cuttings) was lower than in mineral soil media (71.88% or 19 shoot cuttings) (Figure 1). The result of Friedman test for the survival rate of *D. sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* shoot cuttings planted both on peat media and mineral soil media showed a significant difference with the Chi-square value of 18.11 and a significance value of 0.00. These results indicated that the survival rate of *D. sumatrensis* shoot cuttings was lower than *D. oblongifolia* subsp. *Oblongifolia*, suggesting
that the effectiveness of cutting propagation varied between Dryobalanops species and different media.

The success of cuttings is influenced by the interaction of genetic and environmental factors [17]. Genetic factors mainly include the content of food reserves in the cuttings tissue, water availability, age of planting stock (mother plant), endogenous hormones in cuttings tissue, and species. While, environmental factors include rooting media, humidity, temperature, light intensity and the cuttings technique [18]. In the present study, the low level of survival rate of D. sumatrensis could be caused by the age of planting stock as a source of cuttings material which were quite old, i.e. more than 7 years old. Several studies of Dryobalanops shoot cuttings have reported that the high survival percentage of D. sumatrensis (50% - 80%) [8], and D. lanceolata (80%) [19] was probably caused by the juvenility of cutting source materials.

Figure 1. Survival rate of D. sumatrensis and D. oblongifolia subsp. Oblongifolia shoot cuttings for 9 months of observation (August 2018 - April 2019) in peat and minerals soil media.

3.2.2 Rooting and shoot sprouting ability.

The rooted ability of the two species was observed in shoot cuttings that were still survived. The results of rooting percentage in D. sumatrensis in the last month of observation (April 2019) showed that no roots emerged (0%) in shoot cuttings either in peat or mineral soil media. While in D. oblongifolia subsp. oblongifolia, rooting percentage was high in mineral soil media (15.63%), and was low in peat media (4.55%) (Table 2). This pattern indicated that shoot cuttings of D. oblongifolia subsp. oblongifolia have rooted abilities faster than D. sumatrensis for both media, which could be caused by the juvenility of planting stock of D. oblongifolia subsp. oblongifolia as cutting materials. Juvenile cuttings would increase the chance for roots to grow more optimally, so it can produce a better root formation than cutting material from oldest plant [9,18]. This is because the C/N ratio content in juvenile plants was higher than the older plants’ [18]. The juvenility of the stock plant can also be an overriding factor in root formation especially for plants which are difficult to root [8,20]. The effect of juvenility on rooting ability may be related with low levels of rooting inhibitors as well as high levels of photosynthates, but as the plant grows older, the inhibitor levels increase [21]. In addition, lower rooting ability of D. sumatrensis shoot cuttings may be caused by terpenoid compounds present in cutting materials [8]. However, the potential for D. sumatrensis shoot cuttings to produce roots may still be possible if observation time is increased (more than 9 months).

In the present study, shoot sprouting percentage in D. sumatrensis was higher in mineral soil media (6.25%) than in peat media (3.13%) (Table 2). This result corresponds to the pattern found in D. oblongifolia subsp. oblongifolia, which shoot sprouting percentage was higher in mineral soil media (15.63%) than in peat media (4.55%) (Table 2). Moreover, the results also showed that shoot sprouting
percentage in *D. oblongifolia* subsp. *oblongifolia* was higher than *D. sumatrensis* for both media (Table 2). This pattern can also be influenced by the old age of cutting materials in *D. sumatrensis*. Cutting materials derived from old planting stock usually has a low level of C/N ratio, so shoots tend to sprout first followed by the appearance of roots afterwards [18].

In the present study, the treatment of media differences did not affect the success of *D. sumatrensis* shoot cuttings due to the old age of planting stock (mother plant) used as a source of cutting materials, which was the main cause of the low of survival rate, rooting percentage and shoot sprouting percentage. However, the treatment of media differences on *D. oblongifolia* subsp. *oblongifolia* revealed that the survival rate, rooting percentage and shoot sprouting percentage in mineral soil media were higher than in peat media. This result may be caused by the pH of the peat which was more acidic (low nutrient) than mineral soil so that the cutting conditions become tense. In addition, the success of *D. oblongifolia* subsp. *oblongifolia* shoot cuttings in mineral soil media rather than in peat media may also be due to the type of habitat of this species which grows on mineral soils in natural forests [22], so that shoot cuttings may also prefer to grow on minerals rather than on peat. This media treatment can be a main factor that causes the success of shoot cuttings because the cutting material derived from young saplings.

**Table 2.** Rooting percentage and shoot sprouting percentage of *D. sumatrensis* and *D. oblongifolia* subsp. *oblongifolia* during 9 months of observation

| Species        | Media   | Aug | Sept | Oct | Nov | Dec | Jan | Feb | Mar | Apr |
|---------------|---------|-----|------|-----|-----|-----|-----|-----|-----|-----|
| Root *D. sumatrensis* | Peat    | 0.00| 0.00 | 0.00| 0.00| 0.00| 0.00| 0.00| 0.00| 0.00| 0.00|
|               | Mineral | 0.00| 0.00 | 0.00| 0.00| 0.00| 0.00| 0.00| 0.00| 0.00| 0.00|
| *D. oblongifolia* | Peat    | 0.00| 0.00 | 0.00| 0.00| 0.00| 0.00| 0.00| 0.00| 4.55| 0.00|
|               | Mineral | 0.00| 0.00 | 0.00| 0.00| 0.00| 0.00| 0.00| 6.25| 6.25| 15.63|
| Shoot Sprouting *D. sumatrensis* | Peat    | 0.00| 0.00 | 0.00| 0.00| 0.00| 0.00| 0.00| 0.00| 3.13| 0.00|
|               | Mineral | 0.00| 0.00 | 0.00| 0.00| 0.00| 0.00| 3.13| 6.25| 6.25| 6.25|
| *D. oblongifolia* | Peat    | 0.00| 0.00 | 0.00| 0.00| 0.00| 0.00| 0.00| 0.00| 4.55| 0.00|
|               | Mineral | 0.00| 0.00 | 0.00| 0.00| 0.00| 0.00| 6.25| 6.25| 12.50| 15.63|

4. Conclusion
The present study revealed that the media treatment (peat and minerals) did not affect the survival rate and rooting percentage of *D. sumatrensis* shoot cuttings due to the old age of cutting materials. However, the media treatment (peat and minerals) have an effect on *D. oblongifolia* subsp. *oblongifolia* shoot cuttings, which showed by the higher level of survival rate, rooting percentage and shoot sprouting percentage in mineral soil media than one in peat media. This result may be due to the fact that peat media is more acidic than minerals and habitat preferences of this species is on mineral soils. The unclear patterns of *D. sumatrensis* shoot cutting results in the present study, hence further study using young cutting material of *D. sumatrensis* are needed as a comparison of present study.

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