Seed handling of specific forest tree species: Recalcitrant and intermediate seed

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Abstract. Indonesia is a tropical country with an area of rain forests around 128 million ha. Forests have economic, social and environmental functions, therefore those functions must be sustained. Seeds are a very important part for maintaining the sustainability of forest functions, by providing good quality of seeds and seedlings. Nearly 70% of the seeds of forest plants in Indonesia are recalcitrant and intermediate, dominated by species of Dipterocarpaceae, Myrtaceae, Araucariaceae, and Verbenaceae. In general, the characteristics of the seeds can be divided into 3 groups based on their sensitivity of drying and storability, namely orthodox, recalcitrant and intermediates. The viability of orthodox seeds can be maintained for long storage periods in low temperatures, whereas recalcitrant seeds are difficult to store for long periods and lose its viability in a short time. Intermediate seeds are in between of the two characters, that the viability can be kept for a longer time (generally less than 1 year) than recalcitrant seeds. The main problems for recalcitrant and intermediate seeds are their short life time and for some species, the flowering and fruiting periods do not occur every year, especially for the species of Dipterocarpaceae. These problems makedifficult to get large quantities of high-quality seeds. Therefore, seed handling techniques of recalcitrant and intermediates seeds are urgently needed. A proper seed handling is the key to maintain the physical, physiological and genetic quality of seeds which should be carried out by involving several activities including techniques of seed production, seed testing, seed storage, and nursery technique.

1. Introduction

The need of raw materials for the wood industry is increasing, but there are several obstacles in terms of the supply of wood from natural and plantations forests. In 2013, the wood industry needs 70 million m³ of raw material per year while the wood production just reaches 60.4 million m³ per year [1]. According to [2], the coverage of tropical rain forests in the world is decreasing, including in Indonesia due to deforestation and natural disasters such as forest fires. The government's efforts to increase the area of forest cover are conducted by reforestation and rehabilitation through planting activities. Until 2018, the rehabilitation of forests and lands has reached an area of nearly 600,000 ha with a reduction in deforestation rate of 0.5 million ha/year. The deforestation rate decreased compared to the period of 2016-2017 (0.48 million hectares). This condition is a real challenge for establishing productive and sustainable forests. This certainly requires the development of adequate forestry science and technology (IPTEK), one of which is seed science and technology to support the success of various programs. Seed science and technology is needed to maintain the physical, physiological and genetic quality of seeds.
Forest in Indonesia is mostly dominated by tropical rain forests that affect seed characters. One characteristic of the seeds from tropical rain forest plants is a high moisture content, caused by the conditions of rainfall and high humidity in the tropics [3]. In general, the characteristics of the seeds are divided into 3 groups based on their sensitivity to drying and storability, namely orthodox, recalcitrant and intermediates. Orthodox seeds are dry seeds with long storage periods of time, whereas recalcitrant seeds are difficult to store for long periods and lose their viability in short time. Meanwhile, character of intermediate seeds is between the two characters above, that is the moisture content of intermediate seed can be lowered and stored for a longer time (generally less than 1 year). The characters of the seed certainly influence the technique of seed handling [4].

Seed procurement activities are often constrained by the lack of the information of proper seed handling technique, especially for seeds that are rapidly lost their viability and damaged (recalcitrant and intermediates). To date, the development of forest plantation with orthodox seeds is more desirable because of they are easy to handle and less risk of damage. On the contrary, recalcitrant and intermediate seeds dominating tropical forest are rarely chosen for establishing the forest plantations because of the difficulty in obtaining large quantities of high-quality seeds. Even though a lot of seeds available, most of them have died; indicating that the characteristics of recalcitrant or intermediates seeds are still a problem in the seed procurement, especially in an effort to store in long period of time [5]. It is a challenge to consider that nearly 70% seed of forest plants in Indonesia's tropical rainforests is recalcitrant and intermediate, which is dominated by species from the Dipterocarpaceae, Myrtaceae, Araucariaceae and Verbenaceae.

Another problem of some species of recalcitrant and intermediates seeds is that the flowering and fruiting seasons do not occur every year, especially for Dipterocarpaceae. Flowering and fruiting patterns of Dipterocarpaceae is 5-8 years; and the record on ramin (Gonystylus bancanus (Miq.) Kurz.) is 4-5 years or even more [6]. Therefore, there will be abundant fruit in the good year of flowering season, but there are still not enough seeds available for the next planting season. The limitation of seed storability and the irregular flowering and fruiting season are a challenge in providing good quality seed of recalcitrant and intermediates. Therefore, seed handling techniques of recalcitrant and intermediate seeds are urgently needed through a proper seed handling, in order to maintain the physical, physiological and genetic quality of seeds which should be carried out by involving several activities including techniques of seed production, seed testing, seed storage, and nursery techniques.

2. Classification and characteristics of seeds
There are several classifications of the characters of the seeds. According to [7], there are 2 categories of seeds (orthodox and recalcitrant) based on the differences in physiological responses during storage, that are moisture content and temperature. Orthodox seeds can be dried to low moisture content (2% - 5%) without damage. The seed can be stored in long periods, in line with decreasing of moisture content and temperature of storage room. Conversely, recalcitrant seeds cannot be dried until the optimum moisture content for survival varied from 12% to 31%. The characteristics of seed are classified into 4 groups, based on storage condition and their adaptability to the natural habitats [8]:

1. “True orthodox”, seed can be stored for long periods in low temperature and seed moisture content lower than 10% (< 10%), e.g. Tectona grandis, Araucaria cunninghamii, Pinus spp, etc.
2. “Suborthodox”, seed can be stored at the same condition for true orthodox seed, but only for short periods. Ex. Gmelina arborea
3. “Temperate-recalcitrant”, seed can not be dried without damage, but seeds can be stored for several years at low temperature, close to freezing. Ex. Quercus spp
4. “Tropical-recalcitrant”, seeds that are sensitive to damage from drying and low temperatures (10-15°C or lower). Example: Hopea spp. and Shorea spp.
[9] proposed a third category, namely intermediate seeds, because this group was identified as having storage behaviour between orthodox and recalcitrant [10]. Intermediate seeds are resistant to slow drying (dried at room temperature) with a moisture content range from 10% to 20%, and seeds will be damaged at moisture levels below those ranges. These additional categories began recognized and accepted to complement the categories presented by [7]. The three main categories of seed storage behaviour that have been recognized are orthodox, intermediate, and recalcitrant [9].

Physically, orthodox seed can be distinguished from non-orthodox seeds. It can be distinguished from size, weight, thickness of fruit or testa, colour, seed moisture content, germination type, dormancy etc. The size of orthodox seeds relatively smaller and thicker than others. A distinctive characteristic to distinguish orthodox seeds from others is moisture content, and it is often used as a limitation of understanding between orthodox and non-orthodox seeds. Orthodox seeds have a low moisture content which ranges from 5% - 10% [11]. Several species of orthodox seeds include Falcataria moluccana, Adenanthera microsperma, Acacia spp., Albizia procera, Hibiscus macrophyllus, Intsia bijuga and others.

According to [7], recalcitrant seeds cannot be dried without causing damage and also unable to maintain their viability during storage, while [12] define recalcitrant seeds as having high-moisture content that are sensitive to drying, active metabolism with high respiration rates and high levels of intracellular differentiation. This condition causes the seeds to germinate quickly if the water content is maintained in high condition, so that storage in humid conditions can only be done in a short time. Examples of several species of recalcitrant seeds are: Shorea selanica, Peronema canescens, Azadirachta indica, Macaranga triloba, Macaranga hypoleuca, Shorea balangeran, Pongamia piñata, and Podocarpus sp.

Intermediate seeds have characteristics between orthodox and recalcitrant. Although the initial (fresh seed) moisture content is relatively high, intermediate seeds can be dried to a certain moisture content and stored for several months (generally <1 year). Other consideration of intermediate seeds is they are sensitive of direct drying in the sunlight. Loss of viability after drying or during storage depends on the species, degree of maturity and method of extraction or handling. In general, seeds that are physiologically mature more tolerant to drying and can be stored longer in conditions with relative humidity of 40-50% and the moisture content of the seeds is around 10% [4]. Examples of several species of intermediate seeds are Maesopsis eminii, Swietenia macrophylla, Santalum album, Duabanga moluccana, Samanea saman, Sterculia foetida, Toona sureni, Litsea cubeba, Gmelina arborea, and Styrax benzoin.

The characteristic of recalcitrant, intermediate and orthodox seeds in Table 1 [3].

| Natural condition | Recalcitrant | Intermediate | Orthodox |
|-------------------|-------------|--------------|----------|
| Tropic, sub tropic and several species found in dryland areas | Tropic, sub tropic and several species found in dryland areas | Dominantly found in arid and semi arid areas, sub tropical, tropical highland and a pioneer in wet climate areas |
| Family/Genus | Artocarpus, Dipterocarpaceae, Meliaceae, Triplochiton, Durio, Rhizophoraceae, Araucaria, Syzigium, Quercus, Agathis | Meliaceae, Pinaceae, Myrtaceae, Casuarinaceae. | Leguminosae (Fabaceae), Myrtaceae, Casuarinaceae, Pinaceae. |
### Seed moisture content and storage room temperature

| Intolerant of drying and low temperature storage, except for several species in sub-tropic. Tolerance level is influenced by species, generally seed moisture content for storage around 20-35%, with the temperature of room storage varies 12-15°C, and for several species temperature could beat 15-20°C. |
| Seed viability of some species can not be maintained at below of minimum seed moisture content. Seeds can be dried to 9-10% of moisture content level |
| Seeds are tolerant to drying and can be stored at low temperature (0-20°C, with moisture content varies at 5-7%). The seed has to be prepared at the moisture content of 2-4% and stored at -15 to -20°C for cryopreservation |

### Potential storage periods

| True recalcitrant seed can be stored for several days, and several months. Some of Dipterocarpus sp. can be stored for years at low moisture content (10%-12%) and temperature below freezing (-20°C to -30°C) |
| The seed can be stored for several months to 1 year. |
| The seed can be stored for years or decades at optimum condition, |

### Seed characteristic

| Generally, seed size is medium – large, with initial seed moisture content more than 30% |
| Seed size varies from small, medium and large, with initial moisture content more than 20% |
| Seed size varies from small to medium and often have thicker testa |

### Maturity characteristic

| Fruit/seed dry weight is increasing until fully mature or fall down. Initial seed moisture content varies from 30% to 70% with large variation among individual seeds. |
| Fruit/seed dry weight is increasing until fully mature or fall down. Initial seed moisture content varies at the minimum of 20% with moderate variation among individual seeds. |
| Fruit/seed dry weight is increasing until fully mature or fall down. Seed moisture content can be dried at 6%-10% with small variation among individual seeds |

### Dormancy

| No dormancy or weak dormancy. The maturity and germination occur in the same time |
| No dormancy or weak dormancy |
| Dormancy is often occurs |

Sources: [10]; [3] in [4]

Orthodox seeds do not have problems in terms of storage, because they cannot maintain their viability even though they are stored for a long time, with optimal storage conditions. However, maintaining the viability of recalcitrant and intermediate seeds after storage is still a problem. In general, storage periods less than 6 months still possible for intermediate seeds, but not for recalcitrant seeds, such as Shorea leprosula that loses its viability in several days [5].

The viability of recalcitrant seeds can be maintained at moisture content levels less than full imbibition, i.e. at equilibrium conditions at 98-99% relative humidity and optimum temperature varies 7°C-17°C. If germination occurs during storage, then the seed undergoes active metabolism which is the reason why recalcitrant seeds are sensitive to drying [13]; [12].
3. Seed collection

Obtaining good quality seed depends on the proper time of seed collection. The physiological seed maturity is the best indicator for harvest because at that stage the level of seed maturity is optimum. The fruit maturity can be seen from the change color of the fruit skin, and it is often used as a basis of time for seed collection. Several examples of fruit maturity by colour change are as follows: meranti fruits are harvested when the color of wings and calyx are brown, fruit color of neem is yellowish to yellow, the cone of agathis is marked with a dark green and accompanied by brown spots, and ebony is characterized by dark green yellowish with brown spots and soft hairy, and dramin should be picked when the fruit is reddish [14].

Seed collection or harvesting can be done in various ways depending on the species. Several ways in seed collection are: conducted on the forest floor (naturally), threshing, climbing or picking. Seed collection on the forest floor is carried out for medium to large fleshy fruit, such as Dipterocarpaceae (Shorea sp., Dryobalanops sp., Hopea sp., Dipterocarpus sp., Anisoptera sp., and Vatica sp.), Intsia bijuga, Gmelina arborea and Tectona grandis.

The low capability in seed collection causes slow of seed production. In addition, understanding of reproductive biology also influences the level of seed production. Therefore, efforts to increase seed production need to be accompanied by recognizing and understanding reproductive biology which includes flowering phenology. The phenology and rate of development of a plant are influenced by various climate factors such as temperature, day length and water supply. Phenology in the tropics has a number of distinctive characteristics compared to temperate. Understanding the phenology of flowering and fruiting will improve the quality and quantity of seeds through predicting the accuracy of harvest time and seed production each year [15].

The time period of flowering and fruiting of several plants growing in the tropics is shorter (less than 1 year) compared to those growing in the temperate [16]. However, the beginning of the reproductive cycle in plants in the tropics is less consistent than temperate plants [17]. This can be understood related to the climate that occurs in the tropics which has a slight difference in seasons (dry and rainy), so that the flowering process until fruiting is not disturbed or stopped due to the influence of four seasons of climate. One group of flowering periodicity in tropical plants is seasonal flowering category where flowering occurs in response to dry season, for example, kemenyan (Styrax benzoin) [15].

Phenology observation of several forest tree species having recalcitrant and intermediate seeds have been done, including neem (Azadirachta indica A. Juss), suren (Toona sureni Merr), kemenyan (Styrax benzoin) and bambang lanang (Magnolia champaca). Flowering and fruiting period was indicated by the appearance of shoots to mature fruit; and it varies with species. Kemenyan needs 8-9 months [15], suren needs 6 months [18] while Bambang lanang only takes 3 months to develop flowers into mature fruit [19].

4. Seed handling

The seed procurement of high physical and physiological qualities needs the knowledge of the accuracy of seed handling. Seed handling includes activities from post-harvest seed processing, seed testing quality (physical and physiological) and storage.

4.1. Post-harvest seed processing

Seed handling activities after fruit collection include temporary fruit packaging, temporary storage, fruit transportation, fruit/seeds treatment in processing and post-harvest processing [20]. Fruit packaging after fruit collection can be done using porous containers and plastic bags. The use of porous
containers to avoid fungal growth and increase the temperature and humidity which can reduce seed viability. Plastic bags can be used with the upper part opened.

The transportation of recalcitrant seeds from the location of fruit collection must be immediately carried out using special vehicles with temperature conditions 20°± 5ºC and 80%-95% of relative humidity. Recalcitrant and intermediate seeds must be avoided from drying rapidly [20].

Seed processing includes extraction by separating seeds from other parts of fruit (skin, flesh, stalks, wings), seed cleaning, selection, and sortation. Seed extraction consists of 2 methods, namely dry and wet extractions. Extraction with dry method is carried out on fruits that do not have flesh, usually for pods, follicles, capsules, and cones/scales. Especially for recalcitrant and intermediate, drying is prohibited to avoid the rapid decrease of the moisture content.

Extraction of winged fruit (Dipterocarpaceae) is done by removing the wing manually and carefully to avoid physical damage of seeds. In several species, wing discharge during extraction can reduce the viability of seeds. For example, for the seeds of *Hopea macrophylla*, if the wings are not removed, their germination is high (89%), but when the wings are removed from the fruit, the germination percentage decreases to 76.5% [5].

Wet extraction is done on fleshy fruit such as neem (*Azadirachta indica*), where fruit is soaked in water for 24 hours and the soaking is stopped when the flesh is soft and seeds can easily be removed from the fruit. For fruit with hard shell, the flesh must be peeled, and seeds can be cleaned by scraping or using a modification of coffee peeler or blender. Seed can be cleaned from their flesh by using fine sand or other materials in running water [5].

4.2. Seed Quality Testing

The quality of recalcitrant and intermediate seeds is based on parameters of physical and physiological characteristics. Physical quality of intermediate seeds is seen from the initial moisture content (12-30%) and the purity requirement which is 100% for large-sized seeds and at least 70% for small-sized seeds [21]. The physical quality of recalcitrant seeds which generally have medium-large seed size is determined from the initial moisture content (30%-70%) and 100% of purity. Meanwhile, the physiological quality requirements of recalcitrant and intermediate seeds are characterized by the level of germination that should reach 60%-100% [3].

Seed testing is needed to find out the physical and physiological quality of seeds. It includes several stages, starting with seed sampling, determination of seed moisture content, analysis of seed purity, determination of seed weight, and germination test. The data obtained from the results of seed quality testing is very useful for calculating seed requirements for seedling procurement in the nursery.

4.3. Seed Storage

Seed storage is carried out after seed testing process has been done, because based on the initial seed testing, it can be seen the character of a seed lot. Seed storage is only carried out on seeds with an initial germination of more than 50%, because under this value indicates that the seeds are damaged. The main purpose of seed storage is to guarantee the supply of quality seeds for a planting program. Storage techniques for recalcitrant seeds are different from those for intermediate seeds. Therefore, it is necessary to understand the character of the seed, because this affects the storage technique.

- Recalcitrant seed storage
  The recalcitrant seeds have a low storage capability, hence their viability decreases rapidly in various conditions of the storage room. The recalcitrant seeds require sufficiently moist and cool
storage conditions, combined with aeration (air exchange) and are attempted not to overheat due to seed moisture and respiration. Recalcitrant seed must maintain high moisture content during storage, which is around >30%-50%, and 60%-70% for some species of true recalcitrant [3]. To maintain the high moisture content, the seed should be mixed with several materials, such as sawdust, coconut fiber, or cocopeat, perlite, and other materials that have been moisturized [22]. The container used for storage must be porous or allows air and water exchange. This means the container must be able to maintain humidity, such as cotton cloth, paper bags, or gunny sacks. According to [23], recalcitrant seeds can be kept in the storage with room temperature (temperature 27°-30°C and relative humidity 70%-80%) and air conditioned (AC) room (temperature 18°-20°C, relative humidity of 50%-60%).

- Intermediate seed storage,

Seeds of some intermediate species can be dried up to 12%-17% and can be stored for several months [3]. For intermediate seed storage, containers such as gunny sack, cotton, or thin plastic bags allow oxygen exchange. Plastics with a thickness of 0.1-0.25 mm can prevent loss of moisture, but still provide adequate ventilation. Control of insects, fungi, and other diseases must be done because the seeds are stored at a temperature and water level that allows the fungus to grow and develop. The development of the disease can be prevented by giving CO₂ fumigation or short immersion in cold or warm water. Fungicides can be given in wet conditions by soaking seeds in a solution or by dry treatment. The age of intermediate seed storage can be extended by hydration and dehydration treatment. This treatment activates the self-healing mechanism during the hydration stage [3]. According to [24], storage conditions should be aimed at avoiding drying, reducing microbial contamination, preventing germination, and maintaining oxygen availability.

4.4. Germination and nursery

The purpose of the germination test is to get information on seed viability that will reflect the growth of seed lots in the field. Germination tests can be carried out in the laboratory using germinators and in greenhouses. Testing in the laboratory is using a paper test and testing in a greenhouse using a mixture of sterilized soil and sand (1:1). The best germination test method for seeds in the greenhouse is by sowing the standing seed (wings above) with 2/3 parts of the seeds immersed in soil media [5].

The results of the study [25] show that germination testing of several species of meranti (Shorea spp.) was carried out in greenhouses with conditions of temperature fluctuated at 30°-34°C, relative humidity 47%-78% and light intensity of 5180-19400 lux. The ideal germination condition for Shorea roxburghii, Shorea robusta, and Shorea almon was at temperatures of 26°-31°C [26]. The same germination conditions were stated by [27] that reported the best germination was achieved at relatively high temperatures (25°-30°C) and temperature below 15°C would decrease the ability of S. roxburghii seeds to germinate, and was not possible for seeds of Symphonia globulifera and Simarouba amara, although Hopea odorata seeds could still germinate at 5°C.

Seed germination test of nyawai (Ficus variegata) in the laboratory with germinator can be done using paper test methods (UDK) with filter paper. Germination in greenhouses can be done with sand+soil media (1:1 v/v) and or rice husk charcoal + sand (1:1 v/v), and germination bed/box covered with transparent plastic [28]. Species of agarwood (Aquilaria malaccensis) as one of the forest species with recalcitrant seed character achieved the best germination of 92% in a greenhouse with a cover of two layers of shading net [29].

5. Seed handling for several recalcitrant and intermediate seed

Research and development of seed handling of several species with recalcitrant and intermediate seed have been done by BP2TPTH. Almost 36 species of tropical forest categorized as recalcitrant and intermediate have been compiled by [4], and arranged as a basis data of information for stakeholder
about these two characters of seed. The information includes: Taxonomy, seed characteristic, seed collection, seed transportation, seed processing, lowest-safe moisture content, seed storage and seed germination. All of the information of 36 species can be seen at The Book entitled “Characteristics and Seed Handling Principles of Forest Tree Species: Intermediate and Recalcitrant”, published by Center of Research and Development Forestry Tree Seed Technology, Bogor.

6. Seedling production in supporting tree improvement program

Handling of recalcitrant and intermediate seeds in tree improvement program is still a problem. Seeds produced from breeding activity must be handled properly, for maintaining the quality of seed. Storage activities will be important when fruiting season occurred and seeds are produced from the seed orchard, but the problem arises when the seeds that have been harvested could not be stored for long period, especially for dipterocarps species. Therefore, strategies of seed and seedling procurement of superior recalcitrant and intermediate seed which comes from the breeding activities are necessary. Several strategies that could be done to solve the problem of seed and seedling procurement for recalcitrant seed are as follows:

- **Storage in form of Seedlings**
  Seeds that have been harvested from seed source must be sown immediately, to anticipate the decreasing of its viability, and seedling must be kept to maintain their slow growth. Storage in form of seedling will be a solution in seedling procurement of recalcitrant seed. The retained growth of seedlings makes the transportation of seedling easier. Storage in form of seedling has been conducted for several species of recalcitrants such as meranti, bakau, cempaka, gaharu, agathis, kayu bawang and mimba [30]; [31]; [32]; [33]; [34]; [35]. The treatment applied to hold seedling growth rate is by using several retardance substances (paclobutrazol or saline solution) which could keep the growth of seedling slow, especially for high growth. The objectives of this treatment is to control the size of seedling until times to planting, and the augmentation of the substances must be stopped when the seedling is ready to plant. Others treatment that could be applied to suppress seedling growth are controlling the environment, such as percentage of shading level or seedling media. Several studies showed that there was no significant difference in growth at planting site between treated seedling and untreated seedling. It means that the residue of the inhibitor is not permanently stored in the seedlings, so the plants can grow to normal performance after being planted.

- **Hedge Garden**
  Testing and selection have been done in tree improvement program, resulting in superior clones which can produce superior seed and seedling. Applying vegetative propagation to multiply the clones is the best alternative in seed procurement, especially for recalcitrant seed. The development of hedge garden came from selected superior clones and it will be a source for shoot cutting. The shoot cutting methods for several dipterocarps have been known. This information is very important to supporting procurement of good quality of recalcitrant seedlings.

- **Wildings (Stump) method**
  Wildings or stump is another method to support procurement of either recalcitrant or intermediate seed. This method has been used in relation with seedling production for reforestation, especially in natural forest. Stump is made from wildings on the forest floor, where most of characteristics of dipterocarps fruits have wings, and fruits can fly around their mother trees. Stump is made from wildings on forest floor and germinate immediately in optimum environment (temperature, humidity and light). Seedling seed orchard is built from selected superior mothers tree which means the seed or seedling produced from this area inherits the characters of their parents. The abundance of wildings on the forest floor could be utilized for stump production. Stump is made from wildings on forest floor where parts of leaves and roots of wildings are cut off to minimize the transpiration and to make transportation easier. Most of seedlings could be produced by this method.
7. Closing remarks
The main challenge in handling of recalcitrant seed is the long period of storage without reducing its viability. Conventional recalcitrant seed storage has been widely understood, but is still limited in a relatively short period of time (6-8 weeks). The intermediate seeds are still able to be stored in several months or less than one year in specific conditions. Several alternatives that can be selected, such as storage in form of seedling, shoot cuttings from hedge gardens and stumps from wildings have been done to support the production of seedlings from breeding activities especially for recalcitrant seeds.

8. References
[1] DBUK (Dirjen Bina Usaha Kehutanan) 2014 Statistik kehutanan tahun 2013 (Jakarta: Kementerian Kehutanan Republik Indonesia)
[2] The Ministry of Environment and Forestry 2018 Status Hutan dan Kehutanan Indonesia 2018 (Jakarta: Kementerian Lingkungan Hidup dan Kehutanan Republik Indonesia)
[3] Schmidt L 2000 Pedoman Penanganan Benih Tanaman Hutan Tropis dan Subtropis. Terjemahan (Jakarta: PT. Gramedia)
[4] Sudrajat, DJ 2017 Klasiﬁkasi dan karakteristik benih rekalsitran dan intermediate tropis Bunga Rampai: Karakteristik dan Prinsip Penanganan Benih Tanaman Hutan Berwatak Intermediet dan Rekalsitraned Siregar IZ and Mindawati N (Bogor: IPB Press)
[5] Nurhasybi, Sudrajat, D J, Pramono A Aand Budiman B 2007 Review Status Iptek Perbenihan Tanaman Hutan Publikasi Khusus Balai Penelitian Teknologi Perbenihan 6(6)
[6] Sumbayak ESS, Komar TE, Pratiwi, Nurhasybi, Triwilaida, Pradjadinata S, Rosita DT and Ramdhani N 2014 Pedoman Teknik Pembuatan Stek Pacuk Ramin (Gonystylus bancanus (Miq.) Kurz.) (Bogor: Forda Press)
[7] Robert EH 1973 Predicting of the storage life of seeds Seed Science of Tech. 1499-514
[8] Bonner FT 1990 Storage of seeds: Potential and limitations for germplasm conservation Forest Ecology and Management 35 35-43
[9] Ellis RH, Hong TD and Robert EH 1990 An intermediet category of seed storage behavior Coffee Jurnal of Exp. Botany 41 1167-1174
[10] Hong TD and Ellis RH 1996 A Protocol to Determine Seed Storage Behaviour (Rome: International Plant Genetic Resources Institute)
[11] Bonner FT 2015 Storage of Seeds In Bonner FT and KamfaltRP (eds).Woody Plant Seed Manual Part 1. (USDA: Forest Series) p 155
[12] Pammenter NW and Berjak P 2013 Development of the understanding of seed recalcitrant and implications for ex situ conservation BiotecnologíaVegetal. 13(3) 131-144
[13] Berjak P and Pammenter NW 2008 From Avicennia to Zizania: Seed recalcitrance in perspective. Annals of Botany 101 213-228
[14] SudrajatatDJ2017 Database penanganan benih rekalsitran dan intermediate beberapa jenis tanaman hutan tropisBunga Rampai: Karakteristik dan Prinsip Penanganan Benih Tanaman Hutan Berwatak Intermediet dan Rekalsitraned Siregar IZ and Mindawati N (Bogor: IPB Press)
[15] Syamsuwida D, Aminah A, Nurochman N, Sumarni EB and Ginting J 2014 Flowering and Fruiting Development Cycle and Fruit Set of Kemenyan (Styrax benzoin) at Aek Nauli J.P.H.T. 11(2) 89-98
[16] Tenorio GC and Manriquez GI 2007 Plant Reproductive Phenology in a Temperate Forest of The Monarch Butterfly Biosphere (Mexico: Reserve) pp 445-452
[17] Owen-Smith N and Ogutu JO 2013 Controls over reproductive phenology among ungulates: allometry and tropical-temperate contrasts. Ecography 36(3) 256-263
[18] Aminah A and Syamsuwida D 2010 Tahapan perkembangan pembentukan bunga dan buah suren (Toona sureni Merr) J.P.H.T. 7(3) 113-115
[19] Rustam E, Pramono AA and Syamsuwida D 2014 The Development of Flowering and Fruiting of Bambang Lanang (Michelia champaca) J.P.T.H 2(2) 67-76
[20] Marzialina M and Krishnapillay B 2002 Seed Procurement and Handling In Krishnapillay, B. (ed). Malayan Forest Records No. 45. (Kuala Lumpur: FRIM)
[21] Standar Nasional Indonesia [SNI] 7627 2011 Mutu Benih Tanaman Hutan (Jakarta: Badan Standarisasi Nasional)
[22] Yuniarti N and Zanzibar M 2017 Prinsip dan strategi penyimpanan benih berwatak rekalsitran dan intermediate Bunga Rampai: Karakteristik dan Prinsip Penanganan Benih Tanaman Hutan Berwatak Intermediet dan Rekalsitran Siregar IZ and Mindawati N (Bogor : IPB Press)
[23] Sadjad S 1980 Panduan Pembinaan Mutu Benih Tanaman Kehutanan Indonesia (Jakarta: Ditjen Reboisasi dan Rehabilitasi Lahan Departemen Kehutanan dengan Institut Pertanian Bogor)
[24] King MW and Roberts EH 1979 The Storage of Recalcitrant Seeds – Achievements and Possible Approaches (Rome: IBPGR)
[25] Nurhasybi, Danu, Sudrajat DJ and Dharmawati 2003 Kajian Komprehensif Benih Tanaman Hutan Jenis-jenis Dipterocarpaceae (Bogor; Balai Penelitian Teknologi Perbenihan Hutan) Publikasi Khusus 3(4)
[26] Tompset PB 1998 Seed physiology A Review of Dipterocarps: Taxonomy, ecology and silviculture Appanah S and Turnbull JM (Bogor: CIFOR)
[27] Corbineau F and Come D 1989 Germination and storage of recalcitrant seeds of some tropical forest tree species Annals Science of For. 46 89-91
[28] Widyani N, Nurhasybi, Ismiati E, Abay and Sanusi M 2012 Standardisasi Metode Pengujian Mutu Benih Tanaman Hutan Penghasil Kayu Jenis Tembesu (Fagraea fragrans) dan Nyawai (Ficus variegata) Laporan hasil penelitian 12 (Bogor:BPTPTH)
[29] Tabin T and Shristava K 2014 Factors effecting seed germination and establishment of critically endangered Aquilaria malaccensis (Thymelaeaceae) Asian J. of Plant Science and Research 4(16) 4-46
[30] Syamsuwida S, Aminah A and Hidayat A 2008 Growth of Gaharu (Aquilaria malaccensis) Seedling Following The Application of Paclobutrazol During Storage J.P.H.T. 5(1) 21-31
[31] Syamsuwida D and Aminah A 2010 Metode penyimpanan semai bawang (Rhizopora apiculata) dengan berbagai kondisi tempat dan media simpan serta bahan penghambat pertumbuhan. J.P.T.H.4(3) 125–136
[32] Irawan A, Halawane J E and Hidayah H N 2018 Teknik penyimpanan semai cempaka wasian (Magnolia tsiampaca (Miq.) Dandy) menggunakan zat penghambat tumbuh dan perlakuan media tanam J.P.H.T. 15(2) 67-145
[33] Satjapradja O, Setyaningsih L, Syamsuwida D and Rahmat A 2006 The Study of paclobutrazol on the growth of Agathis loranthifoliasedlings J.M.H.T.XII (1) 63-73
[34] Kusmana C, Kalingga N and Syamsuwida D 2011 Pengaruh media simpan, ruang simpan, dan lama penyimpanan terhadap viabilitas benih Rhizophora stylosa Griff. J.Silv.Trop.3(1) 82–87
[35] Syamsuwida D and Aminah A 2011 Teknik penyimpanan semai kayu bawang (Dysoxylum moliscimum) melalui pemberian zat penghambat tumbuh dan pengaturan naungan J.P.T.H.8(3) 147–153

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