UNIVERSITI MALAYSIA SARAWAK

UNIMAS

Faculty of Resource Science and Technology

EFFECT OF PGR ON ROOTING OF CUTTINGS OF PONGAMIA PINNATA TREES

Anis Khalida Binti Abd Halim

Bachelor of Science with Honours
(Plant Resource Science and Management)
2013

UNIVERSITI MALAYSIA SARAWAK

Grade: _____

Please tick (√)

Final Year Project Report ☑

Masters ☐

PhD ☐

DECLARATION OF ORIGINAL WORK

This declaration is made on the ....5th....day of....JULY....2013.

Student's Declaration:

I ANIS KHALIDA BINTI ABD HALIM, 25906, FACULTY RESOURCE SCIENCE AND TECHNOLOGY (FRST)

(PLEASE INDICATE STUDENT'S NAME, MATRIC NO. AND FACULTY) hereby declare that the work entitled, EFFECT OF ~~AGR~~ ON ROOTING OF CUTTINGS OF PONGAMIA PINNATA TREES is my original work. I have not copied from any other students' work or from any other sources except where due reference or acknowledgement is made explicitly in the text, nor has any part been written for me by another person.

05/07/2013
Date submitted

Anis Khalida 25906
Name of the student (Matric No.)

Supervisor's Declaration:

I Hamsani SANI (SUPERVISOR'S NAME) hereby certifies that the work entitled, ----------(TITLE) was prepared by the above named student, and was submitted to the "FACULTY" as a * partial/full fulfillment for the conferment of ....BSc.... (PLEASE INDICATE THE DEGREE), and the aforementioned work, to the best of my knowledge, is the said student's work

Received for examination by: _____ Date: 5/7/13

(Name of the supervisor)

Dr. Hamsani Sani
Professor
Fakulti Sains dan Teknologi Sumber
Universiti Malaysia Sarawak

I declare this Project/Thesis is classified as (Please tick (√)):

☐ **CONFIDENTIAL** (Contains confidential information under the Official Secret Act 1972)*
☐ **RESTRICTED** (Contains restricted information as specified by the organisation where research was done)*
☑ **OPEN ACCESS**

Validation of Project/Thesis

I therefore duly affirmed with free consent and willingness declared that this said Project/Thesis shall be placed officially in the Centre for Academic Information Services with the abide interest and rights as follows:

- This Project/Thesis is the sole legal property of Universiti Malaysia Sarawak (UNIMAS).
- The Centre for Academic Information Services has the lawful right to make copies for the purpose of academic and research only and not for other purpose.
- The Centre for Academic Information Services has the lawful right to digitise the content to for the Local Content Database.
- The Centre for Academic Information Services has the lawful right to make copies of the Project/Thesis for academic exchange between Higher Learning Institute.
- No dispute or any claim shall arise from the student itself neither third party on this Project/Thesis once it becomes sole property of UNIMAS.
- This Project/Thesis or any material, data and information related to it shall not be distributed, published or disclosed to any party by the student except with UNIMAS permission.

Dr. Hamsawi bin Sani
Profesor
Fakulti Sains dan Teknologi Sumber
Universiti Malaysia Sarawak

Student's signature _____ (Date) 05/07/2013

Supervisor's signature: _____ (Date) 5/7/13

Current Address:
LOT 54, RMR BE AIR HITAM CHEMBONG, 71300 REMBAU, NEGERI SEMBILAN

Notes: * If the Project/Thesis is **CONFIDENTIAL** or **RESTRICTED**, please attach together as annexure a letter from the organisation with the period and reasons of confidentiality and restriction.

[The instrument was duly prepared by The Centre for Academic Information Services]

APPROVAL SHEET

Name of candidate: Anis Khalida binti Abd Halim

Title: Effect of PGR on rooting of cuttings of *Pongamia pinnata* trees.

.....

Prof Dr Hamsawi Sani

Supervisor

Dr. Hamsawi bin Sani
Profesor
Fakulti Sains dan Teknologi Sumber
Universiti Malaysia Sarawak

.....

(Dr Rebicca Edward)

Coordinator

Plant Resource Science and Management Programme

Department of Plant Science and Environmental Ecology

Faculty of Resource Science and Technology

Universiti Malaysia Sarawak

Acknowledgement

Thankful to Allah S.W.T. finally, I finished my research for my final year project according to the schedule set. Firstly I would like to thanks my supervisor Professor Dr Hamsawi Sani for guidance, supervision, full support, critics and advice throughout the study. Also thanks to the staff of UNIMAS (especially Laboratory Assistant) who also help me during my experiment, my course mates Redzeque Ramza, Siti Jamaliah, Nur Syakira, and others who lends their hands during the early till the end of my experiment.

Most important person in my life, my family who has been supportive and encourage me when I am down during my final year project and always comfort me mentally and physically to finish my work.

Declaration

I, Anis Khalida binti Abd Halim third year student of student of Plant Resource Science and Management form the faculty of Resource Science and Technology hereby declare that all the work done in preparing this report and writing and producing of this report is done by me with the guidance of supervisor Prof. Dr. Hamsawi Sani until the report is finish.

Anis Khalida

.....

Dated: 05/07/2013

(Anis Khalida binti Abd Halim)

Table of content

List of Abbreviations

GLM- General Linear Model

IBA- Indole-Butyric- Acid

PGR- Plant Growth Regulator

SADEX- IBA 1000mg/L in powder form

UNIMAS- Universiti Malaysia Sarawak

List of Tables and Figures

Tables

Figures

Appendix

Effect of PGR on rooting of cuttings of *Pongamia pinnata* trees

Anis Khalida binti Abd Halim
Plant resource science and management
Faculty of Resource Science and Technology
University Malaysia Sarawak

ABSTRACT

The research study was aimed to determine the effect of PGR on rooting of cuttings of *Pongamia pinnata* trees. The study was carried out at the external lab of University Malaysia Sarawak (UNIMAS). The research used cuttings from the six month old potted seedling and the eight years old tree at external lab and west campus, UNIMAS. *Pongamia Pinnata* is leguminous tree belongs to the family *Fabaceae* and subfamily *Papilionoideae*. The species *P pinnata* is commercially important tree species used to produce biofuels, which known also for its multipurpose benefits and its role in agro-forestry in few countries especially in India. Due to very few study published on the rooting of cuttings of this species, this study examined the flexibility of vegetative propagation and effect of different age of tree of *P. pinnata* through rooting of stem cuttings treated with varying concentrations IBA. The concentration of IBA was manipulated to obtain the appropriate concentration of the hormone needed to grow different ages of cuttings trees. The significant different of the effectiveness of the concentration of the IBA on difference ages trees were analyzed using statistical method, General Linear Model (GLM). Among the PGR used, there were no significant difference between the concentrations. While between the two ages of stock cuttings it was observed that 6 month old seedling was significantly higher by 88.8% rooting success compared to 8 year old cuttings with only 18.2%. Therefore, it is recommended that young cutting stock were used in future research so that higher rooting success produced.

Keywords: Biodiesel - Pongamia - Stem cuttings - Vegetative propagation - PGR – Age

ABSTRAK

Tujuan penyelidikan ini adalah bertujuan menguji kesan kepekatan PGR terhadap pengakaran keratan pokok *Pongamia pinnata*. Kajian ini dijalankan di makmal luar Universiti Malaysia Sarawak (UNIMAS). Kajian ini akan menggunakan keratan dari anak benih berumur enam bulan dan pokok berumur lapan tahun di makmal luar dan kampus barat, UNIMAS. *P. pinnata* adalah pokok kekacang daripada famili *Fabaceae* dan subfamili *Papilionoideae*. Spesies *P. pinnata* adalah spesies pokok komersial penting, yang digunakan untuk menghasilkan biofuel, species ini juga dikenali sebagai pokok pelbagai guna dan peranannya dalam agro-perhutanan di beberapa negara khususnya di India. Disebabkan masih sedikit kajian yang diterbitkan tentang perakaran melalui keratan daripada spesies ini, kajian ini meneliti fleksibiliti pembiakan vegetatif dan kesan umur yang berbeza melalui pengakaran keratan batang dirawat dengan pelbagai kepekatan IBA. Kepekatan IBA dimanipulasikan untuk mendapatkan kepekatan yang sesuai hormon yang diperlukan untuk berkembang umur yang berbeza daripada keratan pokok. Perbezaan ketara keberkesanan kepekatan IBA pada perbezaan umur pokok akan dianalisis dengan menggunakan kaedah statistik, GLM. Di antara ke enam-enam kepekatan PGR yang digunakan didapati tiada pembezaaan nyata dalam eksperiment ini. Walaubagaimanapun pembezaan ketara dapat dilihat di antara dua umur, iaitu anak pokok berumur 6 bulan dengan 88.8% berjaya berakar lebih tinggi berbanding pokok berumur 8 tahun dengan hanya 18.2%. Oleh itu, adalah digalakan untuk kajian masa hadapan, mengunakan sumber keratan dari pokok yang muda agar pertumbuhan akar lebih banyak.

Keywords: Biodiesel - Pongamia - keratan Batang - pembiakan vegetative - PGR – Umur

2.0 INTRODUCTION

Pongamia pinnata, commonly known as Pongam or Karanja belongs to the family *Fabaceae* and subfamily *Papilionoideae* (Joshua, 1997). This species is a shrub; it can reach up to 18 m high and more than 50 cm trunk diameters (Joshua, 1997; Parthiban, Kumar, Subbulakshmi & Vennila, 2010). The alternate, compound *P. pinnata* leaves consist of 5 or 7 leaflets with single terminal leaflet. Flowers, borne on racemes, are pink, light purple, or white. Pods are elliptical, thick walled, and usually contain a single seed. Seeds are oblong and light brown in color (Joshua, 1997).

Pongam is basically grown for controlling soil erosion and binding sandbank because of its dense network of lateral roots (Sangwan, Rao & Sharma, 2010). Besides from that, *Pongamia* is also one of the nitrogen fixing tree that produced biofuel (Ramesh, 2007). In addition, *Pongamia* are commonly found along waterways, with whether its roots in fresh or saltwater (Joshua, 1997; Parthiban et al., 2010; Sangwan, Rao & Sharma, 2010).

Since Pongam is native to humid and subtropics environment, this species can grow under the shade of other trees yet they not a shade demander as it grow well even with full overhead light (Joshua, 1997). Other than that, matured Pongam trees are drought resistant and well adapted to extreme climatic conditions and soil moisture conditions (Ramesh, 2007; Parthiban et al., 2010; Divakara, Alur & Tripati (2010).

P. pinnata naturally regenerates profusely from seeds under frost condition. In addition of that, in natural conditions the tree also coppicing well and sends out root suckers (Ramesh, 2007). As for the artificial regeneration, Pongam easily established

by three methods which are, by direct seedling, by planting nursery raised seedling or by stem cuttings of 1 to 2 cm root collar diameter. Besides from that, propagation by branch cutting and root suckers is also possible (Parthiban et al., 2010).

Other than that, *P. pinnata* produce non edible oil that is very useful as the biofuel (Parthiban et al., 2010; Sangwan et al., 2010). Moreover, the other parts like the bark, roots, leaves, and the twigs have another function. From the bark, we can use as rope, while the roots can be used as fish poison (Divakara et al., 2010). Moreover, the Pongam dried leaves can be used as insect repellent and the twig can be consumed as toothpick (Joshua, 1997; Sangwan et al., 2010). In short, this tree has terrific economic value that very profitable for those who grow it (Divakara et al., 2010).

2.1 Problem statement and objectives

As we know nowadays the demands for biodiesel are extremely increasing, therefore some countries especially India has established large scale plantation for biofuel (Ramesh, 2007; Parthiban et al., 2010). Although *P. pinnata* produce large amount of seed and can be easily germinated, the need to produce true-type seedlings is important to ensure quality seeds for biofuel can be produced (Divakara et al., 2010). Thus, the need to raise large number of quality planting stocks is critical. Despite from that, the effective vegetative technique by cutting is not yet commercialized and analyzed (Ramesh, 2007; Divakara et al., 2010).

Vegetative propagation by rooting of cuttings can produce true-to-type seedlings for large scale plantation (Haapala, 2004). However, the ability to root is also determined by the cutting age, size of cuttings and retention of leaf (Dickens, Ikie

& Nnaji, 2009). There is still very few study published on the rooting of cuttings of this species. Thus, the objective of this study was to study the *P. pinnata* species through vegetative propagation method. In addition, this study was also carry out to determine the effect of different ages of tree and cutting position for the rooting of cuttings. Besides from that, optimum concentration of PGR for rooting of cuttings can be determined.

2.2 Hypothesis

- Cutting taken from physiologically juvenile tissue will root better compared to older tissue (Haapala, 2004). Therefore the younger part of trees and the younger age tree will show better result than the older one.
- IBA (PGR) enhances adventitious root formation. IBA induce cell division and the formation of callus; Later these calli acts as the precursors of adventitious roots (Palanisamy & Subramanian, 2000; Wiesman, Riov, & Epstein, 1989).

3.0 LITERATURE REVIEW

3.1 Botanical Description

Botanically Pongam tree has a medium size which about 8 m height and more than 50 cm trunk diameter (Kumar, Chaukiyal, Kasyap & Singh, 2009; Joshua, 1997; Divakara et al., 2010). Yet as mention by Joshua (1997) in his writing, these trees can reach up to 18 m high. The compound pinnate leaves are dark green spreading from short thick branches into hemispherical crown (Ramesh, 2007). Next characteristic, the flowers are racemes with white or light purplish pink in colour. Other than that, the thick walled pods only have single seed about 10 to 20 cm long in size (Joshua, 1997).

Pongamia trees can endure a wide range of soil types including saline, alkaline, sandy, heavy clay and rocky soils and waterlogged soils. However, it performs best in deep, well-drained, sandy loams with adequate moisture (Ramesh, 2007; Kumar et al., 2009). Native to the Asian subcontinent, this species has been introduced to humid tropical lowlands in the Philippines, Mascarene Islands, in tropical Asia, across Malaya to Australia and Polynesia (Joshua, 1997). The natural distribution of Karanja is along coasts and river banks in India and Burma.

3.2 Economic Importance

3.2.1 Nitrogen Fixing Tree

Generally nitrogen fixing tree is a leguminous tree. There are many tree species with nitrogen fixing ability, for instance, *Acacia* sp., *Albizia lebbeck*, *Leucaena leucocephala*, *Virgilia oroboides* and *Mimosa blanchetii* (Sprent & James,

2009). The role of the nitrogen fixing trees is enhancing soil fertility and increasing productivity of agricultural crops. *P. pinnata* is also one of the few the nitrogen fixing trees in the world (Joshua, 1997). The present of nitrogen fixing bacteria at the legume root nodules helps to fix nitrogen gas in the air into simpler nitrogen forms that can be used directly by plant for their nutrient.

3.2.2 Biofuel

Pongamia produce non edible oil from it seeds, which also knew as "karanja oil" that has the same properties to conventional diesel, biofuel (Ramesh, 2007). Moreover, the emissions of the combustion are cleaner, non-polyaromatic and with lesser toxic smoke and soot produce which in contrast with *Jathropha* sp. (Ramesh, 2007; Kumar et al, 2009). Therefore by producing massive yield potential of 800 to 1000 kg seed per tree annually, up to 15 years profitable term yield and the life span also can reach up to 100 years with 60 years productive oil seed lifespan, it is very suitable substitute tree to *Jathropha curcas* (Divakara et al., 2010).

3.2.3 Medicinal Purpose

Besides functioning as plant source for biofuel, *P. pinnata* also can be consume as medicinal purpose from all it parts for instance the oil, root, stem, leaf, fruit, and etc. (Arote, Dahikar & Yeole, 2009; Sangwan et al., 2010). The oil can be used as liniment for rheumatism (Arote et al., 2009). The root can medicinally use for gonorrhea treatment and for cleaning gums, teeth and ulcers (Sangwan et al., 2010). Other than that the leaves can be used as extract for medicinal aim to relieve cold, diarrhea, itches and herpes (Brijesh, Daswani, Tetali, Rojatkar, Antia, & Birdi, 2006; Arote et al., 2009; Sangwan et al., 2010).

3.3 Propagation techniques

Basically, plant can be propagated sexually or asexually. For *Pongamia* itself, both methods have the advantages and disadvantages.

3.3.1 Sexual propagation

Seed basically contain the combination of genetic material from both male and female gamete through fertilization process. The seed is germinated whether in polybags or directly in the ground, seedling forms may have better or worst characteristic than the parent tree (Kumar, Radhamani, Singh & Varaprasad, 2007). Because of that reason, the genetic quality is hard to be maintained using sexual method of propagation (Shivanna, Balachandra & Suresh, 2007). Besides from that, the storage of the seed also become problematic if not manages properly (Kumar et al., 2007). This is because variety seeds from difference parent tree have different morphological, physiological and biochemical composition (Shivanna et al., 2007).

3.3.2 Asexual propagation

Asexual method or the vegetative propagation is possible by the ability of plant to generate adventitious shoots and roots. By this properties of regeneration, unexpected locations from parts of plant due to respond towards stress or wounding are able to produce new individual of the same genetic qualities as the parent plant (Biswas & Kobayashi, 1995). This type of propagation is economically least expensive than sexual propagation in long term. Besides this techniques are less complicated to exercise. The best thing

is by vegetative propagation the juvenile phase is able to be shortened (Haapala, 2004).

3.3.2.1 Tissue Culture

For tissue culture the growing media must be prepared depending on the purpose of the cloning which is to produce shoot, root or callus (Haapala, 2004). This technique provides alternative to seedling heterozygosity, space, quantity and time consideration (Jaskani., Abbas, Sultana, Khan, Qasim & Khan, 2008). However, it is also expensive in term of preparation of cuttings and price of the cuttings produced (Haapala, 2004; Jaskani et al., 2008).

In addition from that professional skill and expertise labour also required to produce high quality of explants. Generally this type of propagation helps a lot in agriculture and forestry as it can produce genetic alteration to improve the quality of the crops and products like producing disease resistant, climate tolerable and high yield of fruits plants (Jaskani et al., 2008).

3.3.2.2 Air layering, grafting and cutting

Air-layering and grafting is a consistent and easy means of propagation, especially in species which are difficult-to-root on cuttings (Paul & Aditi, 2009). Application of root promoting substances during the layering process helps to get adventitious roots within a short period. While grafting combination of root stock and scion must be compatible and does not required hormone to promote its successfulness. Therefore these

types of propagations have potential means of production of high quality planting stock (Kumar et al., 2009).

There are three type of cuttings which are stem, root and leaf cuttings. Stem cuttings are the commonly used method of cuttings. The wound respond by forming callus at the cut, and then the neighbouring cells reorganizes and differentiate to form adventitious tissues (Biswas & Kobayashi, 1995). The succession of cutting is depending on various factors that play very important role to ability of rooting.

3.4 Factors affecting rooting of cutting

The successfulness of cuttings can be depending to many factors for instance amount of light, retention of leaves, age of the tree, PGR concentration, types of wood hardness and types of rooting media. Generally these factors respond differently depending on the plant species.

3.4.1 Light intensity

The main source of light for plant is most probably sunlight. According to Hossain and Kamaluddin (2011), the intensity if sunlight does affect the growth and morphology of shoots and rooting ability of cuttings in term of the height of the plant, leaf area, number of node and internode length of the seedling and also rooting ability to the cutting. Most of the plants required low light intensity and high shading to excel in rooting ability (Haapala, 2004; Hossain & Kamaluddin, 2011; Nelson & Mohn, undated). Hapala (2004) suggest that rooting should take place in low light intensity and that further

growth should then take place in a higher light intensity. This is most probably because higher light intensity in the experiments possibly caused more severe water stress and therefore poorer rooting results. Hossain & Kamaluddin (2011) also studied the effect of shade on rooting, found that cutting of *Artocarpus heterophyllus* rooted best under 100% shade compared to those rooted fewer than 67% shade without IBA.

.

3.4.2 Retention of Leaves

Leaves role in plant is as a source of carbohydrates by the process of photosynthesis, this process result in production of generally glucose and oxygen (Reuveni & Raviv, 1980; Haapala, 2004). The glucose is stored as starch for plant to get energy to carry out it biochemical processes. According to Reuveni and Raviv (1980), carbohydrates which accumulate at the base of the cuttings are an important factor in the rooting process. Other than that leaf also may contain different nutritional constituents which might influence rooting successfulness (Akinyele, 2010). Reuveni and Raviv (1980) stated that if all leaves were removed from cuttings of an easy-to-root, none of them rooted and they died after a short time.

3.4.3 Age of the tree

Generally, age of the cutting tree can be divided into two which are juvenile and mature (Bijalwan & Thakur, 2010). According to Awang, Sandrang, Mohamad and Selamat (2011), rooting ability of the Tabebuia heterophylla stem cutting decreases with increasing age of the plant. For instance, the percentage of root formation of *Cinnamomum kanehirae* cuttings from 14 years

old ortets was much lower than the rooting ability of terminal cuttings collected from newly formed sprouts as stated by Awang et al. (2011).

Therefore, the usage of juvenile tissues is common to achieve a better rooting of cuttings (Bijalwan & Thakur, 2010; Awang et al., 2011). Yet poor organogenesis also recorded when collecting young tissues from older trees due to high concentration compounds that can inhibit the process (Haapala, 2004).

3.4.4 PGR Concentration

PGR is synthetic plant hormone or plant growth regulators, basically PGR are not nutrient for plant but plant chemical at low concentration that promote and affect the growth, development and differentiation of cells and tissues (Wiesman, et al., 1989; Anon. 3, 2012). For stem, fully developed leaves and roots cutting is accomplished by using auxin (IBA) as the rooting compound applied to the cut surface.

In despite that auxin may also be toxic to young cuttings of certain species, IBA is the best hormone for general use as it has wide range of nontoxic concentration levels (Akwatulira, 2011). Bijalwan and Thakur (2010) determined that juvenile and mature cuttings treated with IBA at 1500 and 2000 ppm respectively showed highest rooting response for *Jatropha curcas L.* species. Basically the optimum concentrations of PGR on rooting vary depending on type of species and woods hardness (Hartmann, 1955).

3.4.5 Wood Hardness

Generally there are three type of wood hardness, which is softwood, semi hardwood and hardwood (Hartmann, 1955; Surendran., Parthiban, Vanangamudi, & Balaji, 2000). Each category has their specific characteristic, for softwood the shoot is easily snapped, while the tissues are physically fragile and easily wilted. Meanwhile for semi hardwood stem cutting are also known as green wood.

According to Surendran, et al. (2000), the physiology of semi hardwood stem is usually with complete growth flush and firm stem. In addition of that the leaves are mature and required wound treatment. Lastly the hardwood stem cutting is easy to handle with deciduous and evergreen broadleaf. However the rooting is usually a failure or required quite long time consideration (Hartmann, 1955). According to Hartmann (1955), the hardwood cutting rooting also required high concentration of rooting hormone treatment.

3.4.6 Type of Rooting Media

The type of rooting media is very crucial depending on the purpose of propagation whether for germination or rooting (Akinyele, 2010). According to Dickens et al. (2009) the successfulness of rooting and the quality of the roots produced are greatly influenced by the medium, which is a crucial part of the propagation system. Rooting media allows absorption of water, nutrient, and air (Dickens et al., 2009; Akinyele, 2010). Poor rooting might be a result from