Ethnobotanical Profiles and Phytochemical Constituents of *Barringtonia racemosa* L. for Potential Scrutiny of Bioactive Compounds through Plant Biotechnology

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Abstract

This paper reviews the traditional uses and customs of people in the use of *Barringtonia racemosa* L., a type of plant mangrove species in their daily life and medicinal approaches. In addition, the phytochemical constituents and the studies in plant biotechnology carried out on this species were also reviewed and summarized by referring to the available documented literatures. It is aimed to provide a systematic review of such topics to facilitate understanding and providing information regarding that particular species. From the current review of literature, it has been clearly depicted that *B. racemosa* is having promising potentials for natural product discovery through plant biotechnology to be further scrutinized in this species. The information gathered from the ethnobotanical uses of this species as well as its phytochemical constituents are useful to provide a significant background for future works regarding plant secondary metabolites from this species to be further explored by the application of plant biotechnology.

Keywords: *Barringtonia racemosa* L., Ethnobotany, Ethnopharmacology, Plant Bioactive Compounds, Phytochemistry

1. Introduction

Our mother nature has a great diversity of plant species and the existence of plant kingdom provides various benefits towards mankind. Plants may beneficially used for psychological, spiritual, cultural and medicinal elements¹. Traditional uses of plants and its relationships with human are termed ethnobotany and are very well associated with cultural practice and belief. Indeed, the ethnobotanical approach which is based on the indigenous uses of plants has been the basis for modern scientific research in pharmacological studies and phytochemical screening. Furthermore, the usefulness of plants not only lies in its medicinal properties used in herbal treatment but in fact, plants have been among the most significant element in daily household needs and being incorporated in industrial setting for instance in the production of agrochemicals, flavours, colourants and fragrances due to the presence of bioactive compounds in plant species².

Highly valued as a natural pharmaceutically-important reservoir, plants are also known as an important source of industrially-valuable compounds due to plant bioactive constituents which are inherently exist in plant species. There are numerous studies have

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been documented so far reporting the importance of plant bioactive compounds in natural product discovery. However, productions of these compounds are critically affected by environmental factors such as climatic changes, nutrient availability, pathogenic threats and cultivation difficulties. Therefore, in addition to the studies in the area of natural product chemistry, the application of plant biotechnology to produce bioactive agents by manipulating in vitro growth conditions and requirements has emerged as an exciting area of discovery. For instance, the research on cell suspension cultures establishment could be potentially used as a viable alternative to natural plant extraction and functions as a platform provider for high-value secondary metabolite production.

One of the most widely distributed plant species with myriad functional uses is *Barringtonia racemosa* or commonly known as powder puff tree or fish poison tree. It is a type of mangrove species and has been used in various tribes around the world due to its wide geographical distribution from eastern Africa to northern Australia. One of its well known functions is it could be used as fish poison and thus reflects its common name as the ‘fish poison tree’. Since *B. racemosa* is widely distributed geographically, therefore it has so many vernacular names and being called differently in different regions of the world.

Ethnobotanically used as effective remedy for certain ailments, *B. racemosa* had been scientifically proven to have medicinal properties through various pharmacological activity studies as well. Previously, a record on *B. racemosa* pharmacological properties had been documented and the pharmacological effectiveness of this species therefore could serve as an important clue for further scrutiny of natural drug discovery of lead compounds to be carried out in this species. Even though *B. racemosa* had been used ethnobotanically in various tribes and had been proven to have significant pharmacological properties, however the existing literatures documenting plant biotechnology applications on *B. racemosa* are quite limited. Therefore, more studies could be possibly undertaken in this regard involving the applications of plant tissue culture.

The investigation pertaining to propagation potential through plant tissue culture had been documented in earlier reports through which the leaves were used as the explant to regenerate the production of in vitro plantlets. In addition to that, the plant biotechnology approach had been incorporated in a very few studies which include the establishment of callus culture and cell suspension culture of the species. Due to the scarcity of investigations in plant biotechnology-related areas conducted on *B. racemosa*, therefore, there is a great potential for the species to be further studied in such a field.

This review was presented by fragmenting the information gathered on *B. racemosa* into ethnobotanical uses which encompassing its non-medicinal and medicinally-related uses respectively as well as its phytochemical constituents. The reviews compiled in this paper are meant to facilitate better understanding and to provide systematic documentation on ethnobotanical uses of this plant species and its phytochemical constituents.

2. Botanical Description

2.1 Taxonomy and Nomenclature

*Barringtonia racemosa* is one of the species in Plantae kingdom from genus Barringtonia which are classified under the family of Lecythidaceae. The Barringtonia genus got its name after Hon. Daines Barrington, 1727-1800, an English nobleman, lawyer, antiquary and naturalist who wrote a book on English trees. Botanically, the ‘racemosa’ term at the later part of the scientific name are reflecting the racemes structure of the species that it possesses which depicts a string-like arrangement of stalked flowers. The taxonomic hierarchy of *B. racemosa* could be arranged in the following order:

- Kingdom: Plantae.
- Subkingdom: Viridiplantae.
- Infrakingdom: Streptophyta.
- Superdivision: Spermatophytina.
- Class: Magnoliopsida.
- Superorder: Asteranae.
- Order: Ericales.
- Family: Lecythidaceae.
- Genus: Barringtonia.
- Species: *Barringtonia racemosa* (L.).
2.2 Vernacular Names

Owing to its widespread geographic distribution, the vernacular names of *B. racemosa* exist in various tribes and regions of the world. Different vernacular names are tabulated in Table 1.

2.3 Botanical Features

The trees of *Barringtonia racemosa* is a small to medium-sized evergreen shrub of around 4 to 8 m in height or occasionally large up to 20 to 27 m tall or even up to 30 to 47 m in height. The bole may reach up to 50 cm to 100 cm in diameter and branch-less for up to 18 m. Sometimes the bole having buttresses, smooth bark or fissured with grey or yellow hue. According to Orwa et al., no aboveground roots are present but they may have spreading surface roots. However, other record claimed that knee roots (pneumatophores) are sometimes present.

Leaves of *B. racemosa* are alternate, crowded towards the ends of twigs, simple; with the stipules which are very small and caducous. The petioles are approximately up to 1.5 cm long, slightly winged with elliptical to obovate-oblong or oblanceolate blade of 5-42 cm × 2-16 cm in size. The characteristic of leaf base is cuneate and narrowly tapering running into very short petiole, while the apex is deep green, acuminate, broadly tapering. The leaf appears to be entire to shallowly toothed or scalloped margin.

Flowers of *B. racemosa* are attractive and fragrant with the colours of white to pale pink arranged in many-flowered pendulous sprays of up to 60 cm in length or even more, hanging on axillary inflorescence as presented in Figure 1. The pendulous raceme could reach up to 70 to 100 cm long. Flowers are bisexual and sepals joined at the base, separating in 3-4 lobes. The flower petals are elliptic, up to 3 x 1 cm, attached to the staminal tube with numerous, long, white or pinkish stamens forming a large central mass of 3.5 cm in diameter. The ovary is 2 to 4 chambered and the style is red.

**Table 1:** The vernacular names of *B. racemosa* according to geographical regions

| Language/Country/Region | Vernacular Name                                                                 | Reference |
|-------------------------|---------------------------------------------------------------------------------|-----------|
| English                 | Barringtonia, brack-water mangrove, common putat, hippo apple, powderpuff tree, wild guava. | 6, 14     |
| Malaysia                | Putat kampung, putat padi, putat darat, putat kedul.                            | 15, 16    |
| Brunei                  | Angas gimpalang, angas gimplang, putat aying.                                   | 6, 17     |
| Indonesia               | Butun, alakan, butun darat, kungkungan, putat.                                  | 6, 17     |
| Myanmar                 | Kye-bin, kyi                                                                     | 6         |
| Cambodia                | Dawn troiekbres, pchek tekbray                                                  | 6         |
| Philippines             | Botong, ulam, kasouai, putat, tuba-tuba, kutkut-timbalon, nuling, paling         | 6, 17     |
| Thailand                | Chik                                                                             | 17        |
| Laos                    | Som pawng                                                                       | 6         |
| Papua New Guinea        | Paniak, paopao                                                                  | 6         |
| China                   | Yu rui                                                                          | 6         |
| India                   | Samudrapandu                                                                    | 15        |
| East Africa             | Mtmomondo                                                                       | 6         |
| Southern Africa         | Poeierkwasboom, iBhoqo, iBoqo (Zulu)                                            | 6         |
| Tanzania                | Mkuvumkuvu, mtomondo                                                            | 6         |
| Madagascar              | Fotatre, mag nondro, manondro, manontro                                         | 6         |
The distinctive features of the fruit are presented as having conical to ovate chicken egg-sized of about 3 to 9 cm x 2 to 5.5 cm (Figure 2) and crowned by the remains of the persistent calyx. The style is fleshy at first and turns hard, fibrous and yellowish-brown as maturity is attained. The fruits are green with purple and dark red tinge and usually one-seeded. The seed lies in the inner part of the fruit with the size of approximately 2 to 4 cm x 1 to 2.5 cm.

**Fig. 2.** Fruits of *B. racemosa* which are green with purple and dark red tinge.

### 2.4 Geographical Distribution and Ecology

The Barringtonia genus comprises about 40 to 50 species worldwide. South-East Asia especially the Malesian region is the centre of diversity in which there are approximately 30 to 40 species could be found in the region. The species of *Barringtonia racemosa* is distributed from eastern Africa and Madagascar to Sri Lanka, India, Myanmar, southern China, Taiwan, the Ryukyu Islands (Japan), Thailand, the Andaman and Nicobar Islands (India), throughout the Malesian region towards Micronesia, Polynesia (east to Fiji and Samoa) and northern Australia. Well known as a mangrove species, *B. racemosa* grows well in tropical and subtropical regions in primary and secondary forest, mostly restricted to inundated flood plains on tidal river banks, or in less saline swampy areas. Although being well recognised as mangrove associate, this plant species can also be found in tropical rainforest, open lowlands and thickets. Nevertheless, it cannot tolerate even light frost and thus can only grow in wet and moist tropical and subtropical climates at the height of 500-900 m altitude.

### 3. Ethnobotanical Profiles

#### 3.1 Construction

The bole and wood from *B. racemosa* had been widely used for construction purposes. It has been documented that the wood derived from this species are used in local house building, general planking, flooring and moldings. In addition, they are also useful in light and temporary construction and used to produce veneer and plywood to form a layer of decorative lining of fine wood to cover the coarse surfaces in construction. Due to these properties, its uses in constructive fabrication and furnishing have been extended to produce household utensils and included as an element in interior finishing and in the manufacturing of wooden pallets.

Apart from that, it is also suitable to be used for carving and turnery owing to the properties of the wood which is light and soft yet moderately resistant to pressure impregnation. In Nicobar Island, New Guinea and Pacific islands, the bole of *B. racemosa* had been utilized in the manufacturing of canoe. Meanwhile, in South India, the durable characteristic of the wood makes it suitable to be used in cabinet making, boat building, carts and rice pounders. The bark of *B. racemosa* had been recorded to be used as a source of fibre and hence suitably used as tying materials. Upon being treated with preservatives, the timber of *B. racemosa* serves as a suitable candidate to make good ties and paving blocks in Philippines.

#### 3.2 Animal Poisons

Being commonly named as fish-poison tree, fish-killer tree and fish-poison wood, it is called as such due to its potential in poisoning fish. It is used to stun fishes and octopus in the area of Pacific islands and many regions of the world where the species are native to. When the pounded materials of *B. racemosa* passes through the gills of fish or being directly ingested, the fishes are therefore stunned. The grated and pounded parts of *B. racemosa* are introduced into stagnant pool or slow-flowing stream and with that, the poisons are concentrated and not easily being washed away by the water current. The uses of this species as animal poisons are attributed to its tannin and saponin content. In stunning fishes, the saponins from *B. racemosa* are taken...
in directly into bloodstream through gills. Even though the saponins effects are powerful, the results on fishes are not fatal. In addition, the toxins which act on fishes’ respiratory organs are not affecting their edibility. *B. racemosa* and other species of Barringtonia genus are widely used for this purpose and most frequently, the seeds\(^1\) and leaves parts are being used\(^2\) in addition to bark and fruits\(^3\).

Apart from effectively used as fish poison, the species is also used to poison wild pigs in Philippines\(^1\). The effectiveness of this species to poison animals had also been verified scientifically. The toxicity of *B. racemosa* extract had been proven to be useful in combating agricultural pest threats. It had been proven that this species exerted toxic effects against golden apple snail by using the seed kernel\(^2\). In addition to that, the ethnobotanical records of this plant species in Palawan Island, Philippines had shown that it is used as insecticide\(^2\). *B. racemosa* was reported to have insecticide potential against citrus aphids\(^1\). For instance in certain district of Bangladesh, it is used as insect repellant\(^3\) and snake repellant\(^4\). Besides, the potency of this species to produce toxic effects towards mosquitoes, snails and brine-shrimp had been documented as well\(^5\). Interestingly, scientific studies had proven that fruit and seed extracts of this mangrove species are having molluscidal, cercariacidal, larvicidal, antiplasmodial, and piscicidal properties\(^6\). The possible role of *B. racemosa* to control snail intermediate hosts of schistosomiasis and dengue fever had been proven by Adewunmi et al.,\(^7\) in a research conducted on *Biomphalaria glabrata* snails and *Aedes aegypti*. In such assay, the pericarp extracts were more potent in molluscidal activities than those of seed extracts while the seed extracts had shown promising larvicidal activities.

### 3.3 Culinary

The shoots and young fruits are usually eaten raw and mostly favoured by the elderly. The young fruits are consumed and favoured due to its crunchy texture\(^8\). Its leaves are usually served as salad\(^9\) especially in Malay custom and eaten together with shrimp paste\(^8\) in main meals of lunch and dinner. Additionally, it is also being added in Malay’s cooking as sauté paste\(^8\). In certain region of the world, its use as an element in culinary is also manifested in which the seeds are milled to produce edible flour. They are pounded to extract the starchy content whereby the pounded seeds are used to make foods such as cakes\(^1,11\).

### 3.4 Household and Other Daily Uses

According to Kitalong et al.,\(^12\) *B. racemosa* is used for cleaning purpose of personal hygiene such as soap by the people of Republic of Palau. The suitability of *B. racemosa* to be used as soap might be owing to its saponin content whereby the presence of saponin in this plant species had been identified and validated by a number of previous studies\(^13,14\). The presence of saponin is useful in soap making since saponin has been recognized to be able to exhibit distinct foaming properties and feasible to be added to shampoos, liquid detergents, toothpastes as emulsifier and long-lasting foaming agent\(^15,16\).

Other than being used as cleaning agent, the pulp of *B. racemosa* is used in paper industry\(^17\). This species is also used as a good source of dye which produces reddish brown dye due to its tannin content which has been associated to its bark of stems and roots. The tannins found in the species belong to the group of condensed proanthocyanidins which produce reddish leather and used to dye vegetable fibres.

In addition, the wood of this plant species is used as a source of fuel and firewood\(^17\). It had been reported that oil from the seeds is used for lighting\(^18\). Apart from being plucked and extracted for multiple ethnobotanical purposes, this species also being appreciated as ornamental plant species and planted for decorative purposes\(^1,14,17\) which might be due to its attractive characteristics of delicate white flowers with masses of pinkish white stamens and its unique hanging fruits found on the hanging racemes.

### 3.5 Ethnopharmacological Uses

Ethnopharmacological term is used to refer to the traditional uses of plant species specifically for medicinal purposes. *Barringtonia racemosa* L. has been traditionally used in various tribes around the world as a type of medicinal approaches. The ethnomedicinal properties of the species according to the related areas and ethnicity were presented in Table 2.
### Table 2: Ethnopharmacological uses of *B. racemosa* in various regions of the world

| Ethnomedicinal properties                                                                 | Area of use /Ethnic involved                                      | Parts used / Method of Application                                                                 | References |
|----------------------------------------------------------------------------------------|-------------------------------------------------------------------|-----------------------------------------------------------------------------------------------------|------------|
| Used as deobstruent, in the treatment of cough, asthma and diarrhoea. Also applied for skin diseases, colic and parturition | Sri Lanka                                                        | The fruits are used for the treatment of cough, asthma and diarrhoea. It is powdered to be used in skin diseases. The aromatic seeds are used in colic and parturition. | 38         |
| Treatment of otitis media                                                              | Palau, Micronesia                                                 | Young leaves are crushed to produce herbal mixtures and injected into the ears of patients.          | 39         |
| Treatment of hallucination                                                             | Eastern part of former Zaire by Nyindu people                    | The pounded bark is soaked in water and the solution is used to wash patient’s head.                  | 40         |
| Treatment of hypertension, itchiness and chicken pox                                    | Malaysia                                                         | Its leaves are used to reduce high blood pressure and used as a depurative, in which the pounded leaves, roots and barks are used to reduce itchiness and chicken pox. | 5, 41      |
| Treatment of cancer-like diseases, jaundice, dog-bite wounds                           | Kerala, India                                                    | Not specified                                                                                       | 42         |
| Treatment of malaria, cough, asthma, diarrhoea, eye inflammation, sores, rheumatism, fever and used for women in parturition | Africa (South Africa, Eastern Africa)                            | The Zulus use the fruit to treat malaria while the pulverized fruit is used as snuff. Seeds are used for the treatment of eye inflammation. Meanwhile, midwives use them for parturition. Bark decoction is applied externally for rheumatism treatment. Root decoction is used as febrifuge. | 5, 14      |
| Treatment of snake bite                                                                 | Bagerhat district and Balidha village, Jessore district, Bangladesh by kavirajes | Leaves are used as a snake repellent and in Jessore and Bagerhat districts of Bangladesh, the leaves are smashed and the decoction is orally consumed for snake bite treatment. | 26-27, 43  |
| Treatment of heartburn, bleeding and abortion                                          | Palauan archipelago, Western Micronesia                           | Not specified                                                                                       | 33         |
| Treatment of cough                                                                     | Buton, Indonesia                                                 | Not specified                                                                                       | 44         |
| Used as anti-diabetic, anti-malaria, antidote                                           | Tamil Nadu, India                                                | Seed is used for the therapeutic purposes                                                           | 45         |
| Treatment of cough, asthma, diarrhoea, stomachache, ophtalmia and jaundice             | Uttara Kannada, India                                            | Aromatic seeds used in stomach ache and opthalmia; seed kernel with milk for treating jaundice. Also used in the preparation of skin ointment. | 46         |
| Treatment of scabies, tetanus and for placental apposition in women                    | Southeastern Madagascar                                          | Leaves are used for scabies, tetanus and placental apposition.                                      | 47         |
| Treatment of pain, joint pain and bone fracture                                        | Kagrachari, Bangladesh                                           | Murong tribes use the leaves to be applied externally.                                             | 48         |
| Treatment of body ache, fever, joint pain, snake bite                                   | Nancowry group of islands (Andaman and Nicobar Island) by Nicobarese | Applied topically to the affected area                                                              | 49         |
| Treatment of snake poisoning                                                            | Songkhla, Thailand                                               | The root is prepared to make decoction                                                             | 50         |
4. Phytochemical Constituents

4.1 Terpenoids

Terpenoid metabolites are one of the largest and most diverse classes of chemical compounds derived from plants. Plants use terpenoids for both basic functions in growth and development as well as for more specialized plants chemical interactions and protection. For instance, monoterpenes and diterpenes generally act as allelopathic agents, attractants in plant-plant or plant-pathogen/herbivore interactions or repellants. Through scientific discoveries, a great number of terpenoid metabolites had been isolated as phytochemical constituents in B. racemosa.

Saponins for instance are classified as triterpene glycosides which are very well known to be inherently present in B. racemosa. The presence of such compounds had been acknowledged to be the reason of its suitability to be used as cleaning agents due to its surface-active properties and may produce long-lasting foam. In addition, saponins in B. racemosa serve as the key factor of its insecticidal and piscicidal activities to stun fish and to stupefy animals and pests. Biologically, the mechanism by which saponins exert its toxicity as animal poison could be understood by the occurrence of hemolysis that they cause. When saponins are in the proximity of animal cell membrane, their interaction with cholesterol may create pores and resulting in hemolysis. The effects of hypersecretion due to such interaction will eventually cause toxicity to the fishes being stunned.

To date, several types of saponins had been identified from B. racemosa extract which are baringtonin and sapogenins. Three different metabolites had been further isolated from triterpenoid sapogenins group which are R1-barringenol, barringtonenol and barringtonic acid. Other two new triterpenoids had been successfully isolated in which the presence of olean-18-en-3beta-O-E-coumaroyl ester and olean-18-en-3beta-O-Z-coumaroyl ester from stem bark of B. racemosa had been discovered along with five known compounds which were germanicol, germanicone, betulinic acid, lupeol and taraxerol.

In year 2000, a research had been documented which reported the presence of two novel neo-clerodane type diterpenoids which were methyl-15, 16-epoxy-12-oxo-3,13(16), 14-neo-clerodatrien-18, 19-olide-17-carboxylate (nasimalun A) and dimethyl-15,16-epoxy-3,13(16), 14-neo-clerodatrien-17, 18-dicarboxylate (17-carboxymethylhardwickiic acid methyl ester (nasimalun B). First documentation on the presence of bartogenic acid had been recorded by Sun et al. from the extract of stem bark. In 2008, Nurul-Maryam et al. reported that great amount of β-carotene and lycopene were found in the ethanolic extract of B. racemosa leaf. Later on, further studies had been published which verified the presence of bartogenic acid done by Patil et al. In such discovery, they confirmed the isolation of bartogenic acid from the fruits of this plant species. A terpene compound classified under tetraterpenoid had been studied by Behbahani et al. and her findings verified the presence lycopene in callus and cell suspension cultures of B. racemosa.

4.2 Plant Sterols

Plant sterols are the components occur in plant cells which are generally functions to control membrane fluidity and permeability. The presence of sterol in plants is associated with a number of benefits for instance it has the potential to be used as natural preventive dietary product in lowering plasma cholesterol level. Phytosterols had been proven to play a key role in plant innate immunity against bacterial pathogens as well. In addition, they also have functions in temperature adaptation of plants. Considered as membrane reinforces which sustain the domain structures of cell membrane, plant sterols also involve in embryonic growth of plants and in fundamental biological process. In B. racemosa, the presence of stigmasterol had been isolated and identified from the ethyl acetate extract of stem bark.

4.3 Phenolics

Phenolics are compounds possessing one or more aromatic rings with one or more hydroxyl groups. They are broadly diversified ranging from simple molecules to highly polymerized substances with more than 8000 compounds being categorized into the group. Plant phenolics can be further classified into several subgroups which are phenolic acids, flavonoids, tannins.
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Tannin is another group of plant polyphenolic compounds under the phenolic groups which are characterized by having polymerized structure. The term ‘tannin’ comes from French word which reflects the tanning properties of this species. To date, very little is known about the validated composition of tannin in B. racemosa and no data on structural elucidation of the compounds have ever been reported. However, the total tannin content of the bark has been estimated to be 18% which belongs to the group of condensed proanthocyanidins and contributes to its reddish brown dye production.

5. Application of Plant Biotechnology in B. racemosa

Plant biotechnology offers advantageous alternative to the conventional breeding methods in the propagation of plant. In addition, plant biotechnology can also be utilized in the in vitro production of important plant secondary metabolites. The application of plant tissue culture as one of the pertinent elements in plant biotechnology could confer benefits owing to the totipotency property of plants. Totipotency is a special characteristic possessed by cells in young tissues and meristems. By adopting the practises of plant tissue culture, total genetic potential of the parent plant could be maintained through totipotency. The adoption of micropropagation technology through plant tissue culture method has broadened the potential of greater plant production of selected traits rapidly which can overcome the slow and labour-intensive process with the advantages of season-independent plant breeding.

Nevertheless, the application of plant biotechnology especially in the adoption of plant tissue culture in B. racemosa are still very few. The regeneration of this species through plant tissue culture was only recorded in a study published in 2007 whereby they managed to establish a protocol for in vitro propagation of B. racemosa from leaf explant. They found that the leaf explants which were cultured on Woody Plant Medium (WPM) supplemented with 30 g/l sucrose and treated with 2 mg/l kinetin and 0.2 mg/l IBA had successfully induced optimum shooting response. Meanwhile, the supplementation of 3 g/l activated charcoal and 0.8 mg/l IBA had been used as a good rooting induction medium in this species. Excellently acclimatized, the in vitro plantlets of B. racemosa were survived after being transferred to mixed soil and were grown well in the glass house.

Meanwhile, the effort in establishing callus culture in B. racemosa was previously recorded in a number of studies. The earliest documented protocol for the establishment of callus culture was made in 2011. In such experiment, the researchers found that the fast growing and friable calli formation were initiated in the culture of leaf explant treated with 2.0 mg/l 2,4-D in WPM medium. Later on, in 2013 another research had been documented in describing the protocol of callus culture establishment from leaf and endosperm explants.
of *B. racemosa*. Upon callus induction percentage assessment, the highest record was identified in the culture of endosperm explant treated with 1.5 mg/l 2,4-D and 0.5 mg/l kinetin in Murashige and Skoog’s (MS) medium whereby the calli formed from such treatment were found to be friable and grew intensively. In the most recent study of callus culture establishment which was published in 2016, the endosperm explant of *B. racemosa* was proven to be productive in yielding callus with desirable friable morphology. However, the optimal combination of Plant Growth Regulators (PGR) was different as compared to the previously documented research whereby the latter found that 1.0 mg/l 2,4-D in the presence of 1.5 mg/l kinetin was the optimal treatment for callus induction.

Research in the area of plant biotechnology which involves the establishment of cell suspension culture and secondary metabolite analysis was extremely limited and only found in a study documented in 2011. In such a study, they established optimum cell suspension culture by using friable calli cultured in liquid B5 medium. Further, they determined the content of lycopene in the cultures by chromatographic analysis. The effect of light towards lycopene level was investigated and it had been determined that the accumulation of lycopene in cell suspension culture was higher in the presence of light than in the darkness.

Due to the rarity of the research in the area of plant biotechnology conducted in this mangrove species, especially in the efforts of producing as well as enhancing its phytochemical constituents, therefore there’s tremendous potential for further investigation to be carried out in *B. racemosa*. The involvement of intense studies in such particular area for instance through the application of plant tissue culture, mass production through bioreactor as well as enhancement of phytochemical compounds through the action of elicitor could be made possible by harnessing the resources and competency in plant biotechnology.

### 6. Conclusion

Considering the usefulness of this plant species and its versatile functions in human lives, *B. racemosa* is indeed having great potentials to be further scrutinized owing to its bioactive compounds. With respect to its phytoconstituents, the discovery pertaining to the compounds present in the species could be further explored to produce fruitful productions of not only medicinally important compounds, but also those with tanning and colouring properties. Due to the scantiness of research in the area of plant biotechnology in this species, hence the research in the respective area on *B. racemosa* should be intensified and diversified to ensure the benefits of *B. racemosa* could be fully harnessed through further exploratory investigations.

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