

**EFFECT OF ORGANIC & BIOFERTILIZER ON THE GROWTH
OF PATCHOULI PLANT (*Pogostemon cablin*)**

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UNIVERSITI MALAYSIA PAHANG

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AHMAD FARHAN BIN MD. SHOKERI

**A thesis submitted in fulfilment of the
requirements for the award of the degree of
Bachelor of Chemical Engineering (Biotechnology)**

**Faculty of Chemical and Natural Resources Engineering
Universiti Malaysia Pahang**

APRIL 2008

I declare that this thesis entitled
**“EFFECT OF ORGANIC & BIOFERTILIZER ON THE GROWTH
OF PATCHOULI PLANT (*Pogostemon cablin*)”**

is the result of my own research except as cited in references.

The thesis has not been accepted for any degree and is not concurrently
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DEDICATION

Special dedication of this grateful moment to my...

Beloved parents;

Md. Shokeri bin Md. Zin

Siti Zalikhoo binti Hj. Abd. Ghani

Loving family member;

Ahmad Farizal bin Md. Shokeri

That always loves me,

My friends, my fellow colleague

and all faculty members

For all your Care, Support and Believe in me.

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Wassalam

ABSTRACT

Patchouli was first described by botanists in the Philippines in 1845. Today growing interest in its fragrance has led to patchouli's widespread cultivation throughout tropical Asia. The research is focused on the growth of patchouli plant (*Pogostemon cablin*) and its essential oil production. The objective of this research is to study the effect of organic and biofertilizer on the growth of patchouli plant (*Pogostemon cablin*). All the fertilizer that was used in this research was analyzed to determine its characteristic using DR 2800 Spectrophotometer (HATCH) and Polarized Zeeman Atomic Absorption Spectroscopy (HITACHI). There were 28 patchouli plants used in this research and hydro distillation was proposed to extract its essential oil. Patchouli oil accounted for about 2-3% of the patchouli dried leaves. The growth of plant had no significant effect by types of fertilizer. However the highest yield of oil was observed at 1.231 g/g using biofertilizer, which is 155% increase compared to control.

ABSTRAK

Patchouli telah di perkenalkan buat pertama kalinya oleh seorang ahli botani dari Filipina pada tahun 1845. Kini perkembangan minat terhadap keharumannya telah menyebabkan penanaman terhadap patchouli telah tersebar luas ke segenab Asia Tropika. Kajian yang di jalankan ini adalah tertumpu kepada mengkaji kadar pertumbuhan pokok patchouli (*Pogostemon cablin*) dan juga pati minyaknya. Objektif kajian ini adalah untuk mengkaji kesan penggunaan baja organik serta baja biologikal terhadap pertumbuhan pokok patchouli (*Pogostemon cablin*). Kesemua jenis baja yang di gunakan dalam kajian ini akan di analisis terlebih dahulu untuk mengetahui ciri-ciri isi kandungannya dengan menggunakan *DR 2800 Spectrophotometer (HATCH)* dan juga *Polarized Zeeman Atomic Absorption Spectroscopy (HITACHI)*. Terdapat 28 pokok patchouli yang di gunakan dalam kajian ini dan proses penyulingan berair di gunakan untuk mengekstrak keluar pati minyaknya. Minyak patchouli adalah kira-kira 2-3% daripada jumlah asal berat kering daun patchouli. Kadar pertumbuhan pokok hampir tidak di pengaruhi oleh jenis baja yang di gunakan. Walaubagaimanapun, hasil minyak terbanyak di perolehi menggunakan baja biologikal, iaitu 1.231 g/g, melebihi 155% daripada kawalan.

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LIST OF SYMBOLS/ABBREVIATIONS

ADP	-	adenosine diphosphate
ATP	-	adenosine triphosphate
cm	-	centimeter
DNA	-	deoxyribonucleic acid
g	-	gram
in	-	inch
kg	-	kilogram
L	-	liter
m	-	meter
mg/kg	-	milligram per kilogram
mg/L	-	milligram per Liter
min	-	minute
mL	-	milliliter
mL/min	-	milliliter per minute
mm	-	millimeter
pH	-	hydrogen ion concentration
RNA	-	ribonucleic acid
TKN	-	Total Kjeldahl Nitrogen
%	-	percentage
°C	-	degree Celcius
μm	-	micrometer
°/min	-	degree per minute

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CHAPTER 1

INTRODUCTION

1.1 Background of Study

Patchouli is native to tropical Asian regions and was widely used incense, perfumes and insect repellents. The west's introduction to patchouli began early in the 19th century when fascination grew about the layers of crushed herb sprinkled liberally in shipments of carpets, fabrics and clothing imported from India and the Middle East. The dried leaves were not placed for aesthetic purposes but as insect repellent, a use for which the east had always used patchouli. Patchouli became the signature scent of the hippie generation of the 1960's where it was used to mask the tarry odour of marihuana. The scent of patchouli is heavy and strong. It has been used for centuries in perfumes, and is grown in the East and West Indies. The word derives from the Tamil language, patchai (green) and ellai (leaf) [1].

The patchouli plant is a bushy herb reaching two or three feet in height and it grows well in southern climates. It enjoys hot weather but not direct sunlight [1]. If the plant withers due to lack of watering it will recover well and quickly once it has been watered. The seed-bearing flowers are very fragrant and bloom in late fall. The tiny seeds may be harvested for planting, but they are very delicate and easily crushed. Cuttings from the mother plant can also be rooted in water to produce further plants.

Patchouli is usually grown on small forest plots by individual farmers who harvest and dry the leaves, then sell them to distilleries throughout the growing

region. The still operators buy many lots of dried leaves and combine them into one steam distillation, the results of which may then be combined with successive distillations until they've obtained sufficient quantities of oil. Newly distilled patchouli oil has a fresh, green, slightly harsh aroma. As the oil ages it mellows considerably, becoming sweeter and more balsamic. Patchouli is one of very few oils that, like fine wine, improve with age. High quality patchouli oils emit a suave, fruity, wine-like top note when uncapped. Other oils that age well are sandalwood and vetiver, both of which blend quite nicely with patchouli.

Patchouli is most often used in aromatherapy. In aromatherapy patchouli is often used as a relaxant. The warmth and depth of its aroma make it comforting and relaxing. Patchouli's relaxing attributes, coupled with its rich and exotic nature, have led to its inclusion in sensual and amorous blends, particularly appropriate for products like massage oil.

In the process of plant the patchouli, it should have to provide optimum nutrients to the patchouli plant to growth well. To protect the environment from any side effect, it is safe to use the organic fertilizer and biofertilizer than a chemical fertilizer.

Organic fertilizers differ from chemicals fertilizers in that they feed the plants while adding organic material to the soil. Soils with lots of organic matter remain loose and airy, hold more moisture and nutrients, foster growth of soil organisms, and promote healthier plant root development. On the other hand, application of chemical fertilizer resulted in loses of organic matter and microbiotic activity. As organic matter is used up, the soil structure deteriorates, becoming compact, lifeless and less able to hold water and nutrients.

Biofertilizers are the most advanced bio technology necessary to support developing organic agriculture, sustainable agriculture, green agriculture and non-pollution agriculture. This biofertilizer can increase the output, improve the quality and it is responsible for agriculture environment [2]. It is well known that the

continue use and overuse of petrochemical based fertilizers and toxic pesticides have caused a detrimental effect to the soils, water supplies, foods, animals and even people. Biofertilizer contains a wide range of naturally chelated plant nutrients and trace elements, carbohydrates, amino acids and other growth promoting substances. Kelp acts as a soil conditioner by stimulating microbial activity in the soil which results in improved air-water relationships in soil, improved fertility and makes soil less prone to compaction and erosion.

1.2 Problem Statements

After the introduction of chemical fertilizers in the last century, farmers were happy for the increase of yield in agriculture. But slowly, chemical fertilizers is showing their ill-effects such as leaching out, and polluting water basins, destroying micro-organisms and friendly insects, making the crop more susceptible to the attack of diseases, reducing the soil fertility and thus causing irreparable damage to the overall system.

A number of intellectuals throughout the world started working on the alternatives and found that biofertilizers can help in increasing the yield without causing the damage associated with chemical fertilizers.

Biofertilizer, the name itself is self explanatory. The fertilizers are used to improve the fertility of the land using biological wastes, hence the term biofertilizers, and biological wastes do not contain any chemicals which are detrimental to the living soil. They are extremely beneficial in enriching the soil with those micro-organisms, which produce organic nutrients for the soil and help combat diseases. The farm produce does not contain traces of hazardous and poisonous materials. Thus those products are accepted across the world as organic ones.

1.3 Objective

The objective of this research is to study the effect of organic and biofertilizer on the growth of patchouli plant (*Pogostemon cablin*) and essential oil yield.

1.4 Scope of Study

The scopes for this study were:

- i. To characterize of organic and biofertilizer
- ii. To study the effect of various fertilizer on the growth of patchouli plant
- iii. To optimize the dosage fertilizer of the growth of patchouli plant
- iv. To extract essential oil using steam distillation method.

CHAPTER 2

LITERATURE REVIEW

2.1 Introduction

There is increasing concern regarding environmental problems related to fertilizers, as they can be a major source of non-point source pollution in soils and waters. Eutrophication, NO₃ contamination of groundwater, and the accumulation of heavy metals in soil and their release to waters, together with their potential bioaccumulation in the food chain, are among the main problems [3].

Although fertilizers provide nutrients to crops, they can contain elements, such as heavy metals, that are potentially harmful for the environment. In most cases, legislation regarding fertilizer composition affects only label statements concerning the nutrient contents, but does not regulate the concentration of other elements. In recent years, some countries have set tolerance limits for some elements [3].

Patchouli (*Pogostemon cablin*) oil is one of the important natural essential oils used as a base and provides lasting character to fragrance in perfumery industry. The dry leaves of patchouli on steam distillation yield an essential oil called the oil of patchouli. Indonesia is the major producer of patchouli oil in the world with an estimated 550 tons per year, which is more than 80% of the total [4].

2.2 Patchouli Plant (*Pogostemon cablin*)

2.2.1 Patchouli Historical

During the 18th and 19th century silk traders from China traveling to the Middle East packed their silk cloth with dried patchouli leaves to prevent moths from laying their eggs on the cloth. Many historians speculate that this association with opulent eastern goods is why patchouli was considered by Europeans of that era to be a luxurious scent. This trend has continued to the present day in modern perfumery [5].

The patchouli plant is a fragrant herb, native to tropical Asia and is cultivated in India, Indonesia, Malaysia, the Philippines and Singapore. It grows from 2 to 3 feet in height, and bears egg-shaped leaves, along with whitish flowers, tinged with purple. Harvested two or three times a year, the leaves are then dried in preparation of distillation [6].

Distillation produces thick oil, amber to dark orange in color, with a powerful, earthy sweet, somewhat musty, unmistakable fragrance - that improves as it ages. The plant and oil have a number of claimed health benefits in herbal folklore, and its scent is used with the aim of inducing relaxation.

The patchouli plant is a bushy herb reaching two or three feet in height. It grows well in southern climates and enjoys hot weather but not direct sunlight. If the plant withers due to lack of watering it will recover well and quickly once it has been watered. The seed-bearing flowers are very fragrant and bloom in late fall. The tiny seeds may be harvested for planting, but they are very delicate and easily crushed. Cuttings from the mother plant can also be rooted in water to produce further plants [5].


Patchouli	
	
Scientific classification	
Kingdom	Plantae
Division	Magnoliophyta
Class	Magnoliopsida
Order	Lamiales
Family	Lamiaceae
Genus	<i>Pogostemon</i>
Species	<i>P. cablin</i>
Binomial name	
<i>Pogostemon cablin</i>	

Figure 2.1: Scientific classification of patchouli

Patchouli is a tropical member of the mint family, grown in the East and West Indies. Leaves are harvested several times a year, dried, and exported for distillation of the oil, although the highest quality oil is usually produced from fresh leaves, distilled close to the plantation.

2.2.2 Morphology Characteristic

Patchouli is one of the herbal plants. Patchouli is categorizing as a small plant with a lot of branch and can growth about 1.0 m height. It has a leaves with oval shape, thick, jagged around it and dark green in color. Pressing and squeezing the patchouli leaves will produce a unique aroma. Patchouli stem and branch appear as a purple in color, square in shape and covered by soft fur.

Flower spikes of clusters of very tiny, pink lip flowers form on axillary and terminal stems. As the flowers fade, the fine, brown seeds form in small capsules, which look like tiny knots on the spike. Leaves, flowers and seeds freely give off their aroma, but even more so if crushed between the fingers. When the stems are cut, their aroma will permeate the room as they dry [7].

2.2.3 Patchouli Species

2.2.3.1 *Pogostemon cablin*

This species are easily can found in Brazil, Philippine, Malaysia, Madagascar and also Indonesia. *Pogostemon cablin* has the heart shape leaves and have furry under it leaves. Normally, this species of Patchouli are not contains a flower and can be produce about 2.5 - 5% of oil.

2.2.3.2 *Pogostemon heyneanus*

Pogostemon heyneanus is another species of Patchouli plant. This species are normally found in a jungle and also in small bushes. Because of that, this species are classified as a wild plant and peoples in Malaysia called it as 'Nilam hutan'. It has thin and also a sharp shape of leaves. Different from *Pogostemon cablin* species, *Pogostemon heyneanus* species have a flower. Extraction of *Pogostemon heyneanus*

can only produce about 0.5 - 1.5% of extraction oil. Moreover, the oil produced is low in quality compared to oil from *Pogostemon cablin*.

2.2.3.3 *Pogostemon hortensis*

The leaves of *Pogostemon hortensis* species have the same characteristic with the *Pogostemon heyneanus* and the difference is that this species does not flower. This species of Patchouli plant also have a low quality of its extraction oil and only can produce about 0.5 - 1.5% of extraction oil.

2.2.4 Special Properties and Effectiveness

2.2.4.1 Medical Use

Patchouli oil has had a long history of medicinal use in India, China and Japan. Patchouli has a sweet spicy aroma, with a hint of musk, used to stimulate the nervous system, lift depressed moods, relieve stress and give a feeling of elation and wellbeing. Patchouli is believed to help balance the endocrine system, which in turn balances the hormones of the body. The aroma assists the body to relax and promotes a feeling of peace. It also stimulates the pituitary gland, which secretes endorphins, which are known for their ability to relieve pain and induce euphoria as well as sexual feelings. Just sniffing the fresh leaves can give a feeling of rejuvenation when feeling worn out [7].

2.2.4.2 Aromatherapy Use

Patchouli essential oil is used as a topical remedy for skin problems such as acne, eczema, inflamed, cracked, chapped and irritated skin. It is known as a cell rejuvenator and helpful in healing wounds and scars. As an antifungal, patchouli oil

has been used to treat athlete's foot. For the hair, patchouli oil has been used for dandruff and to aid oily hair.

For the nervous system, patchouli essential oil helps to reduce tension, insomnia and anxiety. It is also known as uplifting fragrance that helps to soothe away everyday cares, and to bring about a sense of nourishment. In this way, and due to its wine-like intoxicating aroma, patchouli oil is also known as an aphrodisiac [8].

2.2.4.3 Spiritual Use

Patchouli is used in temples as incense. It is said to assist in grounding and centering the mind prior to meditation. It also produces a strong connection to the earth as such is an aid to connecting with the natural beauty of the planet [8].

2.2.4.4 Use in Perfumery

A base note and fixative par excellence. Patchouli oil is used in many famous perfumes such as Tabu and Shocking. A little patchouli oil, used as a fixative can be used in many natural perfume formulations. Patchouli oil mixes well with many essential oils including vetiver, sandalwood, frankincense, bergamot, cedarwood, myrrh, jasmine, rose and the citrus oils [8].

2.2.4.5 Other Uses

The essential oil is used as a room freshener, several drops added to the bath water, or applied on a handkerchief. The pure essential oil is very strong and can be tempered down by adding 10-20 drops of oil to 2 tablespoons of almond oil and 5 drops of wheat germ oil to preserve freshness. The oil is rubbed on pulse points,

temples or the mixture used as massage oil. It is also used to cool inflamed skin, and clear rough, cracked skin, sores, wounds and to treat minor skin disorders such as acne and dermatitis. Oil of patchouli is used in aromatherapy to clear lethargy and sharpen the wits. It is practical oil for releasing stress and tension in males. If a man wants to attract a woman, he could try patchouli perfume as a lure! Patchouli is cultivated extensively in India, Madagascar, Sumatra and the Seychelles for steam distillation of oil and used extensively in the manufacture of perfumes, incense, soaps, hair tonic, tobacco and cosmetics. Patchouli is known as one of the best fixatives in heavy perfumes. A fixative is a substance that helps the other ingredients hold their perfume longer. Its musty, earthy, scent is used as a base note in a third of all women's perfumes produced and half of men's perfumes [7].

2.2.5 Patchouli Extraction Oil

Patchouli (*Pogostemon cablin*) is an aromatic crop which yields an essential oil containing various sesquiterpenes and hydrocarbons such as; patchouli alcohol (patchoulol), patchoulene, bulnesene, guaiene, caryophyllene, elemene and copaene [9-11]. The essential oil is one of the most important naturally occurring perfumery raw materials because of its characteristic woody fragrance and fixative properties by which the scent is fixed and make it last longer on the skin [12]. Areas of commercial cultivation are mainly located in Indonesia, which accounts for over 80% of world patchouli oil production [13].

2.2.6 Essential Oil Content

The leaves contain 1-3.5% of essential oil. Hence it requires around 29-100 kg of leaves to produce 1 kg of oil [14].

2.3 Plant Nutrition

2.3.1 Major Element

Fresh plant material is usually made up of between 80 and 95% water. So far, 16 elements have been identified as essential for plant growth. The plant cannot complete its life cycle without the element. Action of the element must be specific - no other element can take its place. The element must be directly involved (structure, constituent, enzyme activator, etc.). Three elements absorbed in large amounts from the air, water and soil are carbon (C), oxygen (O) and hydrogen (H) [15].

Plant concentrations of essential elements may exceed the critical concentrations, the minimum concentrations required for growth, and may vary somewhat from species to species. Nonetheless, Table 2.1 gives the general requirements of plants:

Table 2.1: Typical concentrations sufficient for plant growth^a [16]

Element	Symbol	mg/kg	percent	Relative number of atoms
Nitrogen	N	15,000	1.5	1,000,000
Potassium	K	10,000	1.0	250,000
Calcium	Ca	5,000	0.5	125,000
Magnesium	Mg	2,000	0.2	80,000
Phosphorus	P	2,000	0.2	60,000
Sulfur	S	1,000	0.1	30,000
Chlorine	Cl	100	--	3,000
Iron	Fe	100	--	2,000
Boron	B	20	--	2,000

Table 2.1: Typical concentrations sufficient for plant growth^b [16]

Element	Symbol	mg/kg	percent	Relative number of atoms
Manganese	Mn	50	--	1,000
Zinc	Zn	20	--	300
Copper	Cu	6	--	100
Molybdenum	Mo	0.1	--	1
Nickel	Ni	0.1	--	1

Table 2.2: Major element of plant nutrient^a

Component	Sources	Deficiencies
Carbon	From carbon dioxide in the air. Converted into plant biomass by photosynthesis. All carbohydrates, proteins and fats are composed of a backbone of carbon atoms.	Not usually a problem. However, in a closed greenhouse in winter, or early in the morning before the vents open, plants can use up enough carbon dioxide to slow photosynthesis and reduce growth. Recommend: CO ₂ generation.
Oxygen	From the air, and as part of water molecules (H ₂ O) and fertilizers (e.g., MgSO ₄). Component of carbohydrates, proteins and fats; necessary for respiration	Can be a problem within the root zone root rot, and plant death.

Table 2.2: Major element of plant nutrient^b

Component	Sources	Deficiencies
Hydrogen	From water and fertilizers as above. Component of carbohydrates, proteins and fats.	Usually not a problem.

2.3.2 Macronutrient

Six macro nutrients absorbed in large amounts from the soil or hydroponic solution that are nitrogen (N), phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and sulfur (S) [15].

Table 2.3: Macronutrient of plant nutrient^a

Component	Sources	Deficiencies
Nitrogen	From fertilizers such as ammonium nitrate, potassium nitrate and/or calcium nitrate. Part of nucleic acids, chlorophyll and every "amino" acid and therefore every protein.	Restricted growth and yellowing (chlorosis) of older leaves.
Phosphorus	From fertilizers such as potassium phosphate. Part of the "energy currency" of cells (ATP, etc.), stimulates root/plant growth, maturity and blooming	Poor root/plant growth and flowering, "purplish" under leaves.

Table 2.3: Macronutrient of plant nutrient^b

Component	Sources	Deficiencies
Potassium	From fertilizers such as potassium nitrate and potassium phosphate. Acts as a catalyst or activator of enzymes promotes overall growth, critical for stomata turgor.	Poor growth, leaf chlorosis/necrosis (death), slowed gas exchange.
Calcium	From fertilizers such as calcium nitrate. Primarily a cross-linking agent in cell walls. Also involved in acid/base regulation during metabolism and as an enzyme activator.	Poor growth of meristems (growing tip), blossom end rot.
Magnesium	From fertilizers such as magnesium sulfate. The "heart" of chlorophyll, and an activator for ATP/ADP metabolism, photosynthesis, respiration & DNA/RNA formation.	Interveneal chlorosis/necrosis of lower mature leaves.
Sulfur	From fertilizers such as magnesium sulfate or potassium sulfate. Part of 2 amino acids and therefore all proteins, forms sulfur bridges to establish and maintain protein structure.	Reduced growth in mid/young leaves, thin brittle stems.

2.3.3 Micronutrient

Seven micro nutrients absorbed in small amounts from the soil or hydroponic solution that are iron (Fe), manganese (Mn), boron (B), zinc (Zn), copper (Cu), molybdenum (Mo) and chlorine (Cl). Besides that, a number of other elements have

been found in plant tissue and are most likely required by some plants including sodium, silicon, cobalt, vanadium, iodine, bromine, fluorine, aluminum and nickel [15].

Table 2.4: Micronutrient of plant nutrient^a

Component	Sources	Deficiencies
Iron	From ferrous sulfate, ferric chloride or iron chelate. Acts as a catalyst for enzymes involved in chlorophyll production, protein synthesis, respiration and other reactions.	Interveinal chlorosis of young leaves.
Manganese	From compounds such as manganese chloride. Involved in enzyme activation during carbohydrate reduction, chlorophyll and RNA/DNA synthesis and other reactions.	Interveinal chlorosis of young leaves, necrotic spots, leaf shed.
Boron	From compounds such as boric acid. Regulates carbohydrate metabolism; involved in RNA synthesis; probably related to the metabolism of calcium and potassium.	Poor growth, blackening then dies back of roots/shoots.
Zinc	From compounds such as zinc sulfate. Acts as an enzyme activator in protein, hormone and RNA/DNA synthesis and metabolism; aids in ribosome complex	General stunting esp. of young growth; interveinal chlorosis.

Table 2.4: Micronutrient of plant nutrient^b

Component	Sources	Deficiencies
Copper	From compounds such as copper chloride. Involved in chlorophyll synthesis; part of the photosynthetic electron transport pathway and of several oxidases, etc.	Stunting, tip death, new leaf twist, blue-green leaves, necrosis, loss of turgor.
Molybdenum	From compounds such as molybdenum trioxide. Involved in nitrogen and carbohydrate metabolism.	Interveinal chlorosis, mottling and marginal scorching or inward cupping of older leaves.
Chlorine	From compounds such as copper chloride or manganous chloride. Acts as an enzyme activator during photosynthesis; involved in respiration; regulation of cell turgor; etc.	Older leaves chlorotic then necrotic; wilt; overall stunting.

2.3.4 Nutrient Solution

Optimum plant growth and yield are the goals, thus watering and nutritional supply is critical. Optimum nutrient solutions begin with good quality and quantity water. Before starting any commercial operation, the water must be analyzed. The source of water should be fairly neutral pH (5-8) with low salt and heavy metal content. Low or high pH can affect nutrient uptake and salt and metals can affect plant growth.

Optimum plant growth is a function of nutrient concentration in the plant. There is a critical nutrient concentration below which growth is reduced/terminated. The adequate zone is above the critical concentration and provides maximum

growth. The toxic zone is above the adequate zone, again resulting in reduced growth or death [15].

Nutrients are available in several forms, including pre-mixed liquid concentrates and pre-mixed powder concentrates varieties. In liquid concentrated form, calcium compounds are mixed separately from phosphates & sulfates since they will form insoluble precipitates and become unavailable to plants. The nutrient recipes vary according to types of crop, life stage, environmental conditions, time of year, etc.

2.4 Fertilizer

After the introduction of chemical fertilizers in the last century, farmers were happy of the increasement of yield in agriculture. Unfortunately, the chemical fertilizers started displaying their ill-effects such as leaching out, and polluting water basins, destroying micro-organisms and friendly insects, making the crop more susceptible to the attack of diseases, reducing the soil fertility and thus causing irreparable damage to the overall system [17].

A number of intellectuals throughout the world started working on the alternatives and found that biofertilizers can help in increasing the yield without causing much damage associated with chemical fertilizers.

2.4.1 Biofertilizer

The name itself is self explanatory. The fertilizers are used to improve the fertility of the land by using biological wastes, hence the term biofertilizers, and biological wastes do not contain any chemicals which are detrimental to the living soil. They are extremely beneficial in enriching the soil with those micro-organisms,

which produce organic nutrients for the soil and help combat diseases. The farm produce does not contain traces of hazardous and poisonous materials [17].

Biofertilizers are ready to use live formulates of such beneficial microorganisms which on application to seed, root or soil mobilize the availability of nutrients by their biological activity in particular, and help build up the micro-flora and in turn the soil health in general [18].

2.4.1.1 Why Should Use Biofertilizer

With the introduction of green revolution technologies the modern agriculture is getting more and more dependent upon the steady supply of synthetic inputs (mainly fertilizers), which are products of fossil fuel (coal+petroleum). Adverse effects are being noticed due to the excessive and imbalance use of these synthetic inputs. This situation has lead to identifying harmless inputs like biofertilizers. Use of such natural products like biofertilizers in crop cultivation will help in safeguarding the soil health and also the quality of crop products [18].

2.4.1.2 Benefit and Advantages Biofertilizer

Usage a biofertilizer helps to increase crop yield by 20-30% and replace chemical nitrogen and phosphorus by 25%. Besides that, it can activate the soil biologically and restore natural soil fertility. It can also provide protection against drought and some soil borne diseases [18].

Advantages from using a biofertilizer are cost effective firstly. Next it can be a supplement to fertilizer besides as an eco-friendly (friendly with nature). Biofertilizer also can reduce the costs towards fertilizers use, especially regarding nitrogen and phosphorus [18].

2.4.1.3 A Group of Component with a Particular Skill of Biofertilizer

Table 2.5: Component of biofertilizer^a

Component	Function
Phospho	It releases insoluble phosphorus in soil and fix this phosphorus in clay minerals which is of great significance in agriculture [17].
Rhizo	Rhizo Bacterial plays a very important role in agriculture by inducing nitrogen fixings nodules on the root of legumes such as peas, beans clove and alfalfa.
Azobactor	Atmosphere contains 78% nitrogen which is a very important nutrient for plant growth. Azotobactor fixes the atmospheric nitrogen in the soil and make it available to the plants. It protects the roots from other pathogens present in the soil.
Trichoderma	It is a non- pathogenic and eco-friendly product. The product is antagonistic hyper parasitic against different pathogens in the field and economically well established biocontrol agent.
Tricho-Card	Trichogramma is an efficient destroyer of eggs of many leaf and flower eaters, stems, fruit, shoot borers etc. It can be used in a variety of crops as well as in horticultural and ornamental plants, such as sugarcane, cotton, brinjal, tomato, corn, jawar, vegetables, citrus, paddy apple etc.
Vermi Compost	It is 100% pure eco-friendly organic fertilizer. This organic fertilizer has nitrogen phosphorus, potassium, organic carbon, sulphur, hormones, vitamins, enzymes and antibiotics which help to improve the quality and quantity of yield. It is observed that due to continuous misuse of chemical fertilizer soil losses its fertility and gets salty day by day. To overcome such problems natural farming is the only remedy and Vermi compost is the best solution.

Table 2.5: Component of biofertilizer^b

Component	Function
Biocompost	It is eco-friendly organic fertilizer which is prepared from the sugar industry waste material which is decomposed and enriched of with various plants and human friendly bacteria and fungi. Biocompost consists of nitrogen, phosphate solubilizing bacteria and various useful fungi like decomposing fungi, <i>Trichoderma viridea</i> which protects the plants from various soils borne disease and also help to increase soil fertility which results to a good quality product.

2.4.2 Organic Fertilizer

Organic fertilizers differ from chemicals fertilizers. Organic fertilizer will feed the plants by adding organic material to the soil. Soils with lots of organic matter remain loose and airy, hold more moisture and nutrients, foster growth of soil organisms, and promote healthier plant root development. If only chemicals are added the soil gradually loses its organic matter and microbiotic activity. As organic matter is used up, the soil structure deteriorates, becoming compact, lifeless and less able to hold water and nutrients. An organic fertilizer is eco-friendly because it made from renewable resources [18].

This organic fertilizer is unequaled in its ability to nourish the beneficial micro-organisms in the soil greatly increasing the soil's humus content and improving its ability to sustain and nurture healthy, when planting flower beds or spread it around established plants and scratch it into the soil, more colorful plants. Use by the handful when planting individual plants, broadcast and mix it deeply into the soil.

There are millions of microscopic organisms near the plants that consist in a micro environment that provides nutrients to the plants and also it helps to keep the water and retain the nutrients in the soil for a plants uses [19].

When fill it with chemicals fertilizer, most of microscopic organisms will die forever and losing the capacity of the soil to be sustainable at long term.

2.4.2.1 Benefit Offers by Organic Fertilizer

Organic fertilizer is an easy product to apply to the plant. By using organic fertilizer also, it is excellent for pest and disease tolerance because at the same time it can protect plants. It is safe to use, non-toxic and extremely universal. It mean that organic fertilizer is safe for a children or any pets and can be apply to any plant in a yard [19]. Besides that, the result from using an organic fertilizer can be observed immediately. Organic fertilizer also is tremendous drought resistance by enhances plant root systems. Finally, it is unbeatably cost effective and user doesn't have to spend a fortune for a beautiful lawn and garden.

2.5 Patchouli Essential Oil Production

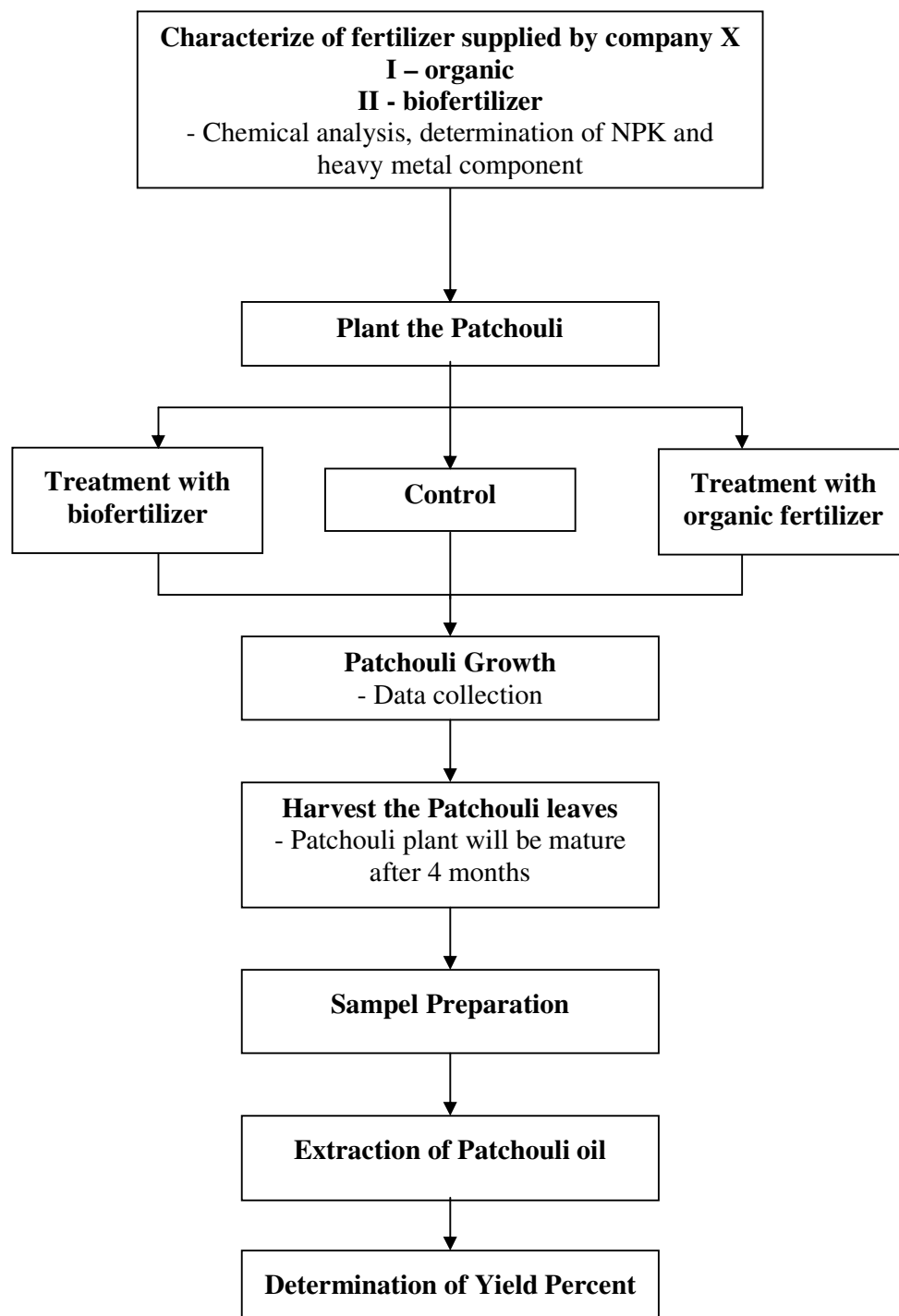


Figure 2.2: Overall process in production patchouli oil through hydrodistillation

CHAPTER 3

METHODOLOGY

3.1 Introduction

In order to plant the patchouli (*Pogostemon cablin*), the simplest step, like medium preparation, plant the patchouli's young, sprinkle with water and applying the fertilizer must be done before extraction process can be take place. In this experiment, 28 patchouli plants were used. Organic and biofertilizer is used as nutrients for the patchouli to growth well. The component of organic and biofertilizer also will be characterized first before apply it into the patchouli.

3.2 Pre-preparation

3.2.1 Preparation of Planting Area

All the patchouli will be planted in a suitable area to keep it growth within a good condition. 28 patchouli plants (planted in ceramic vases) were used in this study. All the ceramic vases will be arranged properly in a group and located in an open area.

3.2.2 Preparation of Medium

6.5 kg of soil was added with 1.3 kg of composite fertilizer and mixed well with a ratio 1:5 using a spade. Then the prepared medium was put into a ceramic vase. The same ratio between soil and composite fertilizer will be applied for each vase and it was leave for a week to make sure the soil and composite fertilizer are fully mixed with good aeration, loose and without any water sump on it.

3.3 Determination of Fertilizer Component

3.3.1 Pre-preparation

3.3.1.1 Sample Preparation for Determination of NPK Component

100 g of fertilizer was dissolved in a 900 mL of distilled water in a beaker. The solution was stirred with a glass rod. Then, the solution was blended to get a fully dissolved fertilizer solution. Next, the concentration of NPK in the solution was tested using DR 2800 Spectrophotometer (HATCH).

3.3.1.2 Sample Preparation for Determination of Heavy Metal Component

100 g of fertilizer was dried at temperature of 95°C for 24 hours until a constant weight was reached. The dried fertilizer was then crushed to powder and weighted in an analytical balance which was calibrated prior to use [20].

3.3.1.3 Open Acid Digestion

The fertilizer which was crushed dry were placed in a mixture of 1 mL concentrated HCl and 1 mL 65% HNO₃ and was left to preliminary digestion for 3

hours. After that, it was placed upon a hot plate in Teflon-beaker for digestion activity above 100°C until completely digested and the experiment was done in a fume chamber for 1 and half hours. After it is cooled from the previous reactions, 1.0 mL of H₂O₂ was added to the sample to break down any recalcitrant lipid material. A clean solution was obtained and the mixture was evaporated to dry. When it was cooled, the mixture contents were topped up to 50 mL with deionised double distilled (Milli-Q) water. To ensure that no error by contaminant occurred, blank runs (without sample) acid was done concurrently [20].

Fertilizer solutions were filtered in 0.45 µm syring filters before being tested using Polarized Zeeman Atomic Absorption Spectroscopy (HITACHI).

3.3.2 Determination of Nitrogen Content using Kjeldahl Method

20 mL of prepared sample were digested as described in the Digesdahl Digestion Apparatus Instruction Manual and 20 mL of deionized water were also digested as the blank. The 10 mL analysis volume was pipet from the sample and the blank into separate 25 mL mixing graduated cylinders. Next, one drop of TKN Indicator was added to each cylinder. If the aliquot is greater than 1 mL, drops of 8 N KOH were added to each cylinder until the first flash of blue color appears. When it is appears, stopper was used to invert the cylinder after each addition [21].

1.0 N KOH was added to each cylinder, one drop at a time and mixing after each addition until the first permanent blue color appeared. Then both cylinders were filled to the 20 mL with deionized water. Three drops of mineral stabilizer were added to each cylinder and stopper was used to invert several times to mix. Three drops of Polyvinyl Alcohol Dispersing Agent was added to each cylinder and stopper was used to invert several times to mix. After that, both cylinders were filled to the 25 mL mark with deionized water and stopper was used to invert several times to mix [21].

1.0 mL of Nessler's Reagent was pipetted to each cylinder. The solution should not be hazy to avoid incorrect results. After 2-3 minutes, the contents of each cylinder were poured into separate square sample cells and tested in the DR 2800 Spectrophotometer (HATCH) [21].

3.3.3 Determination of Total Phosphorus Content

5.0 mL of deionized water was pipetted into total phosphorus test 'N tube vial as a blank. 5.0 mL of sample was pipetted also using the same tube vial and a funnel was used to add the contents of one Potassium Persulfate Powder Pillow to each vial. The cap was tightly and shakes to dissolve. Then the vials were inserted in the DRB 200 Reactor and heated for about 30 minutes. Then, the hot vials were carefully removed from reactor and allowed to cool to room temperature (18-25°C).

2.0 mL of 1.54 N sodium hydroxide was added to a capped vial and invert to mix. 0.5 mL of Molybdovanadate Reagent was added using a polyethylene dropper to each vial and vortexed and leave for seven minutes. The prepared sample was wiped and inserts into the 16-mm cell holder and the result show by DR 2800 Spectrophotometer (HATCH) was recorded [21].

3.3.4 Determination of Potassium Content

25 mL of sample was filled with one Potassium 1 Reagent Pillow and one Potassium 2 Reagent Pillow and inverted several times to mix. The content of one Potassium 3 Reagent Pillow was added after the solution clears and shakes for 30 seconds. A white turbidity was formed if potassium is present. 10 mL of the solution from the cylinder was poured into a square sample cell as a prepared sample. After 3 minutes, the second square sample cell was filled with 10 mL of sample as a blank preparation. The prepared sample was wiped and inserts it into the 16-mm cell holder and the result show by DR 2800 Spectrophotometer (HATCH) was recorded [21].

3.3.5 Determination of Heavy Metal Content

Polarized Zeeman Atomic Absorption Spectroscopy (HITACHI) will be used to analysis and quantitatively measure the content of boron (B), copper (Cu), iron (Fe), manganese (Mn) and zinc (Zn) in the prepared sample.

For each heavy metal element analysis, the analytical condition for each heavy metal element was set up before measured the prepared sample. The standard solution for each heavy metal component was injected together with the prepared sample and analyzed using the specific column in order to get the actual concentration of each heavy metal element in the sample.

3.4 Planting the Patchouli (*Pogostemon cablin*)

The medium was prepared one week prior to cultivation. Planting area in the vase should be cleared from any unwanted weeds to reduce competitor. Spade is used to prepare a small hole in the middle of each vase. The young patchouli was transferred from poly bag into each vase. Next, 1 L tap water was sprinkled into patchouli and the vase was located at the open area with enough sunlight.

3.4.1 Watering Patchouli

1 L of tap water was sprinkled to plant every morning. The amount of water applied for each plant must be standardized to prevent from any error.

3.4.2 Fertilizing Patchouli

Patchouli plant was treated with organic fertilizer ((Hadzrin Landscap Nursery) and biofertilizer (Hadzrin Landscap Nursery). 5 g of organic fertilizer was weighed and apply it to 4 patchouli plant. Next, 10 g and 20 g of organic fertilizer were weighed and apply for the next 4 different patchouli plants. The total numbers of patchouli that is treated with organic fertilizer is 12 patchouli plants.

The same amount was applied to another 12 patchouli plants with the biofertilizer. The balance of 4 patchouli plants as a control will not be treated with any fertilizer. All the 24 patchouli plant was fertilized once a week.

3.5 Observation of Patchouli Growth

In this research, there are a few features of a plant that were measured during cultivation in order to determine the extent of plant growth [22]. Table 3.1 shows the types of observation and also how frequently it was observed during 4 months planted the patchouli.

Table 3.1: Types of observation^a

Measurement	Procedure	Frequency
Plant height	The height of the patchouli plant was measured from the border of the vase to the top of the patchouli plant stem.	Twice a week (Monday and Thursday)
Number of leaves (indicated a plant's physiological age)	The number of leaves on each patchouli plant were counted and recorded. Every visible leaf on the patchouli plant was counted including the tips of new leaves just beginning to emerge [23].	Once a week (Saturday)

Table 3.1: Types of observation^b

Measurement	Procedure	Frequency
Plant color	Any observations on changes or differences in patchouli plant color were recorded.	Once a week (Saturday)

3.6 Harvesting

After 6 months, the matured patchouli plants were harvested. The soil around the patchouli plant was clawed using a spade to loosen the soil and it was cleaned from traces of soil.

3.7 Weighing the Patchouli Plant

3.7.1 Measuring Fresh Weight

Patchouli plant was removed from traces of soil. The soft paper towel was used to blot the patchouli plant gently to remove any free surface moisture. The patchouli plant was weighed immediately using a scale and the data was recorded.

3.7.2 Measuring Dry Weight

Patchouli plants were dried in an oven at 34°C for overnight. On the day, the patchouli plant was removed and sealed in a Ziploc bag.

3.8 Extraction of Patchouli Leaves

3.8.1 Dried Leaves

200 g of patchouli dried leaves from each sample were cut using a scissor into small pieces to size 1 cm [24].

3.8.2 Extraction of Essential Oil using Hydrodistillation

3.8.2.1 Distillation Set-up

A 100 mL heating mantle was clamped to a ring stand above a magnetic stirrer and a 100 mL round bottom flask was placed to the ring stand. 200 g of patchouli leaves were weight and immersed in water with ratio 1:10 g and transfer to the flask using the weighing paper as a funnel.

A rubber band was used to hold the vacuum adapter on the end of the condenser. The mixture was stirred at 60 rpm. The heating voltage was lowered if foaming is occurred [25].



Figure 3.1: Hydrodistillation set-up

3.8.2.2 Hydrodistillation

The cooling water was turned on and made sure that water was flowing through the condenser. The mixture was not heated too intensely or it will bump over. The top of flask and the connector was wrapped with glass wool to avoid vapor from condensing. The heating was adjusted so that distillate dripped at a rate of one drop every 2-5 seconds. Water was added in small amounts via the separatory funnel to match the rate of distillate collecting. The ice was replenished in the beaker that is used to cool the collection flask as necessary. A Pasteur pipet was used to remove some of the water before added with more ice. The vapor shall be cloudy when the natural products are co-distilling with the water. When the vapor becomes cleared, then the process was stopped [25].

3.8.2.3 Extraction of Oil

The distillate was extracted with dichloromethane (3 x 10 mL portions) and the organic layer over Na_2SO_4 filter was dried and filtered.

3.8.3 Determination of Yield Product

The essential oils were weighed using a weighing scale and yield in wt% was calculated [26].

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Introduction

In order to study the effect of organic and biofertilizer on the growth of patchouli plant (*Pogostemon cablin*), this research was started with analysis of component in fertilizer and continue with applying it on the patchouli plant. As a result among this research, the results below are discuss to identify which fertilizer have a better benefit to produce more yield of patchouli essential oil.

4.2 Fertilizer Component

4.2.1 Component of NPK

From the analysis that was performed by using DR 2800 Spectrophotometer (HATCH), the result was tabulated in the Table 4.1.

Table 4.1: Component of NPK in Organic Fertilizer and Biofertilizer

Fertilizer Component	Organic Fertilizer	Biofertilizer
Nitrogen (N)	65.5 mg/L	53.9 mg/L
Phosphorus (P)	70.6 mg/L	66.5 mg/L
Potassium (K)	27.4 mg/L	41.5 mg/L

From the Table 4.1, concentration of nitrogen and phosphorus in organic fertilizer are higher than concentration in biofertilizer. Concentration of phosphorus is just about 70.6 mg/L in organic fertilizer and 66.5 mg/L in biofertilizer while concentration of nitrogen is 65.5 mg/L in organic fertilizer compare to 53.9 mg/L in biofertilizer. But its difference in a potassium component, there is about 41.5 mg/L concentration of potassium in biofertilizer compared to 27.4 mg/L in organic fertilizer.

Phosphorus plays important roles for enhancing the growth of plant by providing an energy currency such as ATP. Plants that grow without enough phosphorus have a poor root and growth. Lacking of phosphorus supply can be detected by finding a purplish color under leaves.

Potassium element is important to the biofertilizer because it acts as a catalyst and also an activator for the enzymes inside the biofertilizer. It has about 41.5 mg/L potassium in biofertilizer. This potassium will convert the nitrate (NO_3) inside the soil into the nitrogen to support the growth of the patchouli plant.

Nitrogen is the most important element in plant nutrient. Nitrogen involved in the growth of the plant, where it will grow normally if there is enough nitrogen concentration in the soil. The deficiencies of nitrogen component will restrict the patchouli growth and yellowing of older leaves [15].

4.2.2 Component of Heavy Metal

In heavy metal analysis, the results are proved that both organic and biofertilizer contain only a small quantities of heavy metal. That why all these components are classified as a micronutrient for a plant growth.

Table 4.2: Component of heavy metal in organic fertilizer and biofertilizer

Fertilizer Component	Organic Fertilizer	Biofertilizer
Boron (B)	3.66 mg/L	3.4 mg/L
Copper (Cu)	0.93 mg/L	3.8 mg/L
Iron (Fe)	3.20 mg/L	2.6 mg/L
Zinc (Zn)	2.70 mg/L	3.7 mg/L

From the analysis that had been done using the Polarized Zeeman Atomic Absorption Spectroscopy (HITACHI), the result in Table 4.2 shows that organic fertilizer have higher content of heavy metal, which are boron and iron compared to the biofertilizer which have more higher value for component copper and zinc. Even in small quantities, micronutrient such as boron and zinc are still important to the plant because these heavy metal components are involved in RNA synthesis and metabolism inside the plant [15].

4.3 Patchouli Growth Observation

4.3.1 Height of Plant

The heights of plant showed significant different between patchouli that was treated with a fertilizer and the patchouli in control. The height of the patchouli plant was measure twice in a month.

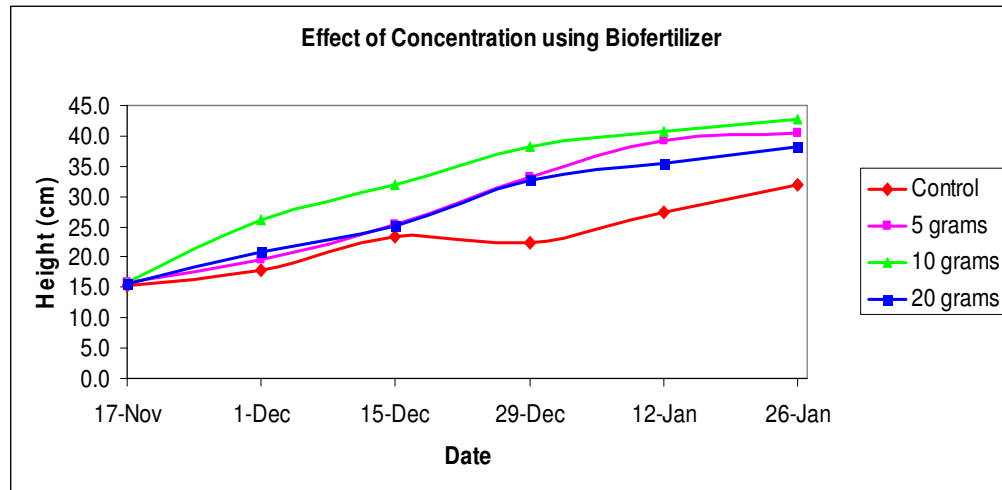


Figure 4.1: Graph of patchouli growth effect by concentration using biofertilizer

From the Figure 4.1, the result shows that the heights of patchouli plant that were treated with biofertilizer were above 35 cm. The control was growth below the 35 cm. This result proves that using a biofertilizer can help the plant to grow more rapidly than without using any fertilizer. The biofertilizer that apply into a plant can enrich the soil airing system with those micro-organisms, which produce organic nutrients for the soil and help combat diseases by converting the nitrate into a nitrogen.

Besides that, the graph also shows that all the patchouli plant that treats by biofertilizer have a constant rate of growth with a 10 grams amount of fertilizer apply is the most stable in growth compare to the others. It is because 10 grams is the optimum amount for a patchouli growth. 5 grams amount will growth the patchouli well if there are a lot of water apply to it and different with 20 grams amount, it will hinder the growth when a lot of water apply to it.

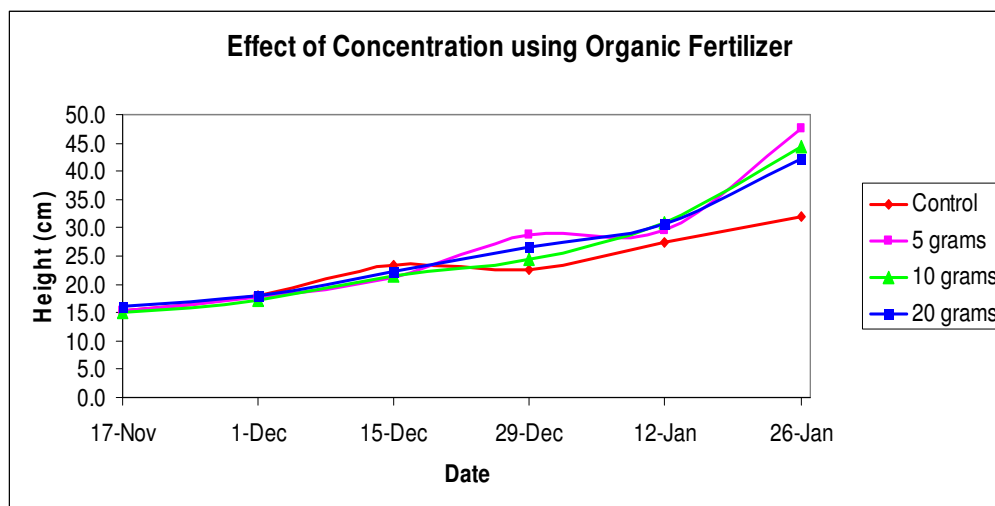


Figure 4.2: Graph of patchouli growth effect by concentration using organic fertilizer

Organic fertilizer provides ideal pH for the soil for the optimum growth because organic fertilizer was produced by a waste of animal and also from the plantation waste such as corn and sugar cane. Based on the Figure 4.2, it showed that there is no specific difference between the entire variant in amounts that apply into the plant. The shape of the graph just showed that there is a rapid increase in the growth of the patchouli plant from the reading on 29 December 2007 until the last reading. The effect of using an organic fertilizer can be seen after the patchouli plant becomes mature, so it will be a rapid increase at the end of the graph. The reason is because the organic fertilizer is more suitable for a mature plant to grow well in an optimum nutrient.

After the overall finding, it shows that an organic fertilizer has provided a group of patchouli plants with a higher height compared to the biofertilizer. But from the physical analysis, even the patchouli that treated by organic fertilizer has more height in growth but it has a less number of patchouli leaves compared to the patchouli that treated by biofertilizer.

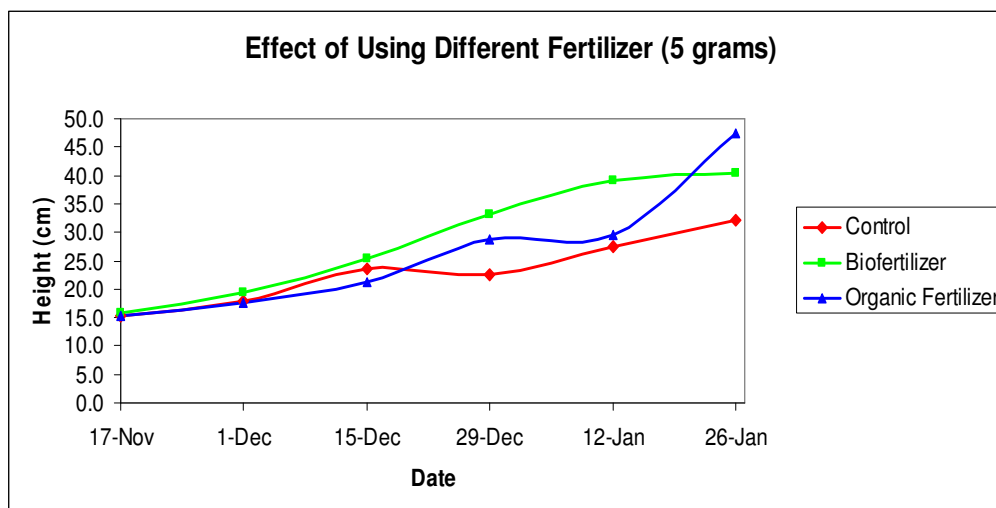


Figure 4.3: Effect of various fertilizers on the growth of patchouli plant at concentration 5 grams

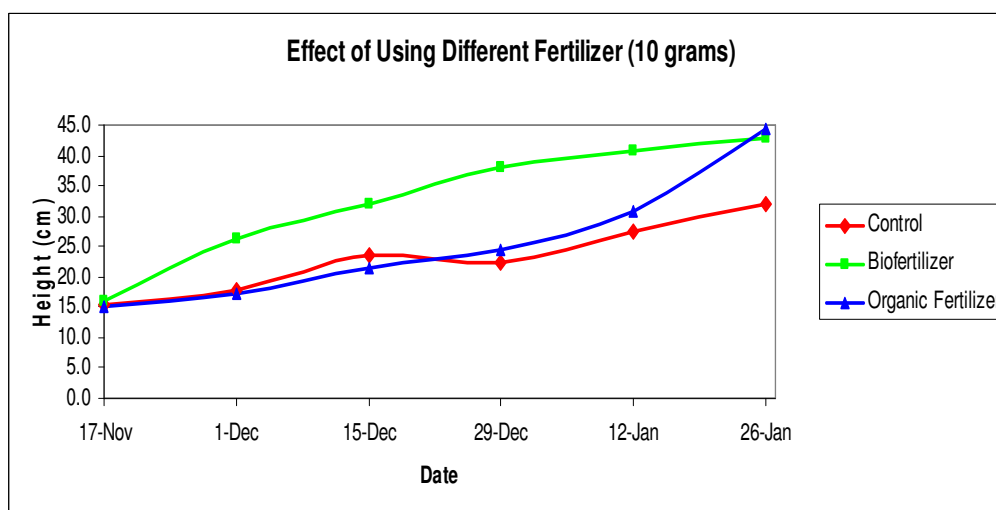


Figure 4.4: Effect of various fertilizers on the growth of patchouli plant at concentration 10 grams

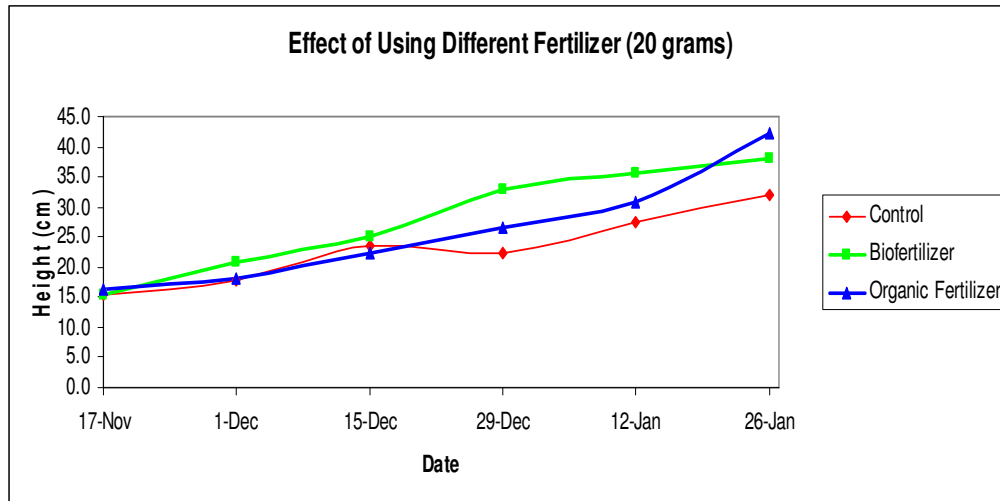


Figure 4.5: Effect of various fertilizers on the growth of patchouli plant at concentration 20 grams

Figure 4.3 until Figure 4.5 are the graph of effect using difference amount of fertilizer applied during the growth of patchouli plant. After doing the analysis, it can be say that organic fertilizer has more effective in a mature patchouli plant because it contains a lot of nitrogen and phosphorus contents inside it fertilizer. Different with biofertilizer, it is most suitable to be applying to the young patchouli plant with it content of potassium that can convert nitrate to nitrogen. This reaction can improve the airing system inside the soil and than it will be easy to the young root to growth. While doing this project there is a little bit deviation occur when there is a heavy rainy season at the end of December. It could be giving the bad result for the biofertilizer to provide a good condition to the patchouli plant. The excess of water can be washout the biofertilizer away from the vase.

As a result, from the graph it showed that the optimum amount should be applied to the patchouli plant is 5 grams twice a month for organic fertilizer and 10 grams twice a month for the biofertilizer.

4.3.2 Patchouli Plant Color

In this observation, all the patchouli leaves have an average color (light green) at the young stage. But the color of patchouli leaves start to differ two months after applying the fertilizer.



Figure 4.6: Leaves of patchouli that treat by biofertilizer

Figure 4.6 above has show that the patchouli that was treated with biofertilizer had a perfect green leaves with a fresh oval shape. This group of patchouli has growth with an enough nutrient.



Figure 4.7: Leaves of patchouli that treat by organic fertilizer

The picture of patchouli that was treated with organic fertilizer as showed in Figure 4.7. It has a totally different in the color of the leaves compare to the Figure

4.6. For this group of patchouli, it also has a normal oval shape leaves but it appears in a yellowing leaves.



Figure 4.8: Leaves of patchouli in control

Figure 4.8 shows of control patchouli plant. This group of patchouli plant have a black green leaves with an abnormal shape leaves. At the same time, the plant also infected with diseases making young leaves becomes twist. The color of the leaves also indicates that the plant are lacking of nutrient to growth.

4.4 Weighing of Patchouli Plant

4.4.1 Fresh Weight

Weighing the fresh patchouli was done immediately after harvesting. The result for the fresh weight is tabulated in Table 4.3.

Table 4.3: Fresh weight of patchouli plant

Fertilizer Apply At The Patchouli Plant		Fresh Weight
Control		1.490 kg
Organic Fertilizer	5 gram	1.860 kg
	10 gram	1.900 kg
	20 gram	2.090 kg
Biofertilizer	5 gram	1.780 kg
	10 gram	1.995 kg
	20 gram	2.100 kg

From the Table 4.3, patchouli in control had a least weight compare to the others. Without a nutrient from any fertilizer, control plant cannot grow very well. In overall analysis, all the patchouli that were treated with biofertilizer have more fresh weight value compare to the patchouli treated by organic fertilizer except for the 5 grams amount where organic fertilizer is heavier than biofertilizer.

Even patchouli in a group of organic fertilizer has growth more height than biofertilizer, but it contains less number of leaves compare to the patchouli plant in a group of biofertilizer. In fresh patchouli, the weight of patchouli was increase by the increase of the fertilizer amount applied for both fertilizers. It proves that, each patchouli plant will grow more if there is more fertilizer applies to each patchouli plant.

4.4.2 Dry Weight

The data from dry weight analysis was more reliable compare to fresh weight analysis. All the data collected are tabulate in the Table 4.4. From the Table 4.4, the trend for the dry weight is quite different from the fresh weight of patchouli. In the dry weight, patchouli in a group of biofertilizer has higher dry weight compared to the patchouli in a group of organic fertilizer as well as for control. As a conclusion,

10 grams is the best amount for both fertilizers because it will produce more dried patchouli leaves compare to the others. Patchouli that treated with 10 grams amount of fertilizer will produce more patchouli leaves compare to the others.

Table 4.4: Dry weight of patchouli plant

Fertilizer Apply At The Patchouli Plant		Dry Weight
Control		0.2275 kg
Organic Fertilizer	5 gram	0.1930 kg
	10 gram	0.2840 kg
	20 gram	0.2340 kg
Biofertilizer	5 gram	0.2760 kg
	10 gram	0.2900 kg
	20 gram	0.2760 kg

4.5 Extraction of Patchouli Leaves

4.5.1 Extraction of Essential Oil

The data for oil yield are recorded as in Table 4.5. Patchouli in control produced less oil compared to plant treated with biofertilizer and organic fertilizer. It just 1.1 g oil collected after 3 hours and half distillation process. At 5 grams, the yield produce is higher compared to other concentration because 5 grams is the optimum amount for the producing of oil contents inside the patchouli leaves. The highest increasement is 109% for organic fertilizer and 209% for biofertilizer as compared to control.

Table 4.5: Essential oil recovers by hydrodistillation

Fertilizer Apply At The Patchouli Plant		Oil Weight
Control		1.1 g
Organic Fertilizer	5 gram	2.3 g
	10 gram	1.7 g
	20 gram	1.4 g
Biofertilizer	5 gram	3.4 g
	10 gram	2.5 g
	20 gram	1.8 g

Compare between patchouli that treats by biofertilizer and organic fertilizer, patchouli in a group of biofertilizer has produce more patchouli essential oil. But for both fertilizer, the trend from the oil recovery showed that the weight of oil produce was decrease according to the increasingly of the fertilizer amount apply. In producing the oil inside the leaves, patchouli plant needs an optimum amount for it. Using a large amount of fertilizer will hinder the production of oil. The apparent of the oil also differ from each types of fertilizer. Oil from group of organic fertilizer is in cloudy form and oil from group of biofertilizer is in clear form.

4.5.2 Yield of Essential Oil

Table 4.6 shows the trend from the yield of product show that the both fertilizer were produce more yield of oil with a lowest amount apply to the patchouli plant. At the same time, patchouli in a group of biofertilizer was produce more higher yield of product compares to the organic fertilizer.

Table 4.6: Yield of essential oil

Fertilizer Apply At The Patchouli Plant		Yield of Product (g/g biomass)
Control		0.483
Organic Fertilizer	5 gram	1.190
	10 gram	0.598
	20 gram	0.598
Biofertilizer	5 gram	1.231
	10 gram	0.862
	20 gram	0.652

CHAPTER 5

CONCLUSION AND RECOMMENDATION

5.1 Conclusion

Base on the research result and observation, there is a different in the amount of macronutrient and micronutrient in both fertilizer. Every types of fertilizer have their own concentration of NPK and also heavy metal to support the growth of the patchouli plant in varies way. From the analysis, it prove that the biofertilizer is most suitable used for a young patchouli plant but an organic fertilizer is most suitable to used in a patchouli plant that have already in a matured phase. Biofertilizer is better in providing provide more nutrients in the earlier stage of the patchouli growth.

From the analysis, it is show that, biofertilizer is a better enhancer for growth of patchouli compared to organic fertilizer, with higher increasement observed at 10 grams, which was 33.5% higher compared to control.

In terms of oil production, maximum oil production (yield) was observed using biofertilizer at amount concentration of 5 grams, with 209% increasement compared to control.

5.2 Recommendation

For further research, it is suggest to improve a few steps in doing this research for a better result with a minimum deviation. It is suggested to:

- i) Plant the patchouli below the sun sheet area to reduce water loses in a hot day. At the same time, it can reduce the deviation from the water excess if there is a heavy raining in between the running of this research.
- ii) Use a steam distillation method for the extraction of patchouli essential oil because it is the most common method will be use for the patchouli. Using a hydrodistillation method will be expected can damage the oil structure and at the same time can reduce it oil content.
- iii) Run the extraction process for about 7 to 10 hours to make sure all the patchouli essential oil are fully extract from the dried patchouli leaves.

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APPENDIX A

EQUIPMENT



Appendix A.1: Weighing scale



Appendix A.2: Oven



Appendix A.3: Polarized Zeeman Atomic Absorption Spectroscopy (HITACHI)



Appendix A.4: DR 2800 Spectrophotometer (HATCH)



Appendix A.5: Hydrodistillation set-up

APPENDIX B

SAMPLE ANALYSIS



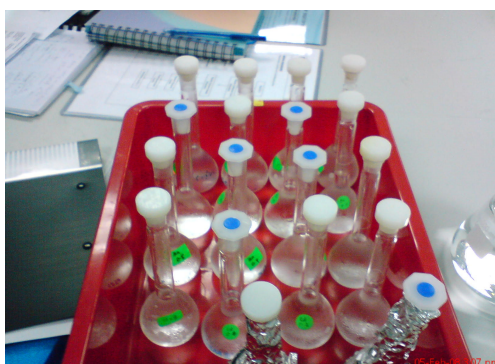
Appendix B.1: Organic fertilizer under drying process in oven



Appendix B.2: Biofertilizer under drying process in oven



Appendix B.3: Sample of both fertilizers after dilution



Appendix B.4: Standard solution of heavy metal component in AAS



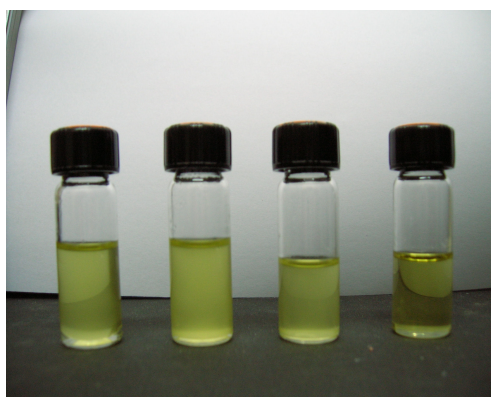
Appendix B.5: Sample preparation for HATCH analysis



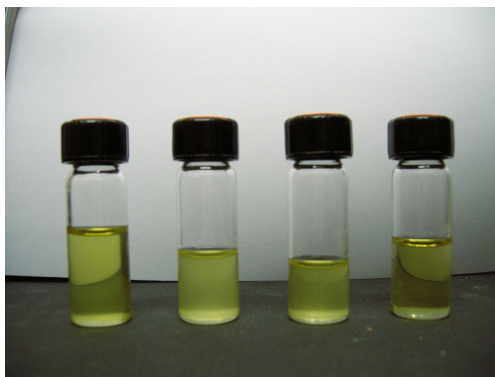
Appendix B.6: Young patchouli plant



Appendix B.7: Matured patchouli plant



Appendix B.8: Patchouli essential oil (Treat by biofertilizer with control)



Appendix B.9: Patchouli essential oil (Treat by organic fertilizer with control)

APPENDIX C

DATA COLLECTION

		Reading of Patchouli Growth (cm)						
		17 Nov	1 Dec	15 Dec	29 Dec	12 Jan	26 Jan	
Control	1	15.00	17.00	21.00	23.00	25.00	29.00	
	2	16.00	17.50	25.00	27.00	29.00	32.00	
	3	14.50	19.00	27.00	29.00	32.00	38.00	
	4	16.00	18.00	21.00	23.00	24.00	29.00	
Average		15.38	17.88	23.50	25.50	27.50	32.00	
Biofertilizer	5 grams	1	15.50	19.00	25.00	31.00	37.00	38.50
		2	16.50	18.50	24.00	29.50	36.50	37.00
		3	15.00	21.00	28.00	35.00	43.00	45.50
		4	16.50	19.50	25.00	37.00	40.00	41.00
			15.88	19.50	25.50	33.13	39.13	40.50
	10 grams	1	17.00	27.00	34.00	38.00	41.00	44.00
		2	15.00	26.00	32.00	38.50	42.00	44.00
		3	16.00	25.50	30.00	39.00	40.00	42.00
		4	15.50	26.50	31.50	37.00	40.00	41.00
			15.88	26.25	31.88	38.13	40.75	42.75
	20 grams	1	15.00	20.00	23.00	29.50	32.00	33.50
		2	14.90	21.00	24.50	35.00	38.00	41.00
		3	16.00	19.60	26.00	32.60	35.00	38.50
		4	16.00	23.00	27.00	34.00	37.00	39.50
	Average		15.475	20.9	25.125	32.775	35.5	38.125
Organic Fertilizer	5 grams	1	15.50	18.00	22.00	27.20	31.50	51.00
		2	16.00	18.10	21.90	26.90	30.00	50.00
		3	15.00	17.80	21.00	24.80	27.00	44.00
		4	14.80	16.90	20.00	24.50	29.40	45.00
			15.33	17.70	21.23	25.85	29.48	47.50
	10 grams	1	15.00	17.00	20.50	22.00	26.80	38.50
		2	15.50	17.00	21.70	25.20	30.00	45.50
		3	16.00	18.40	21.50	25.00	32.00	46.00
		4	14.00	16.80	22.00	26.00	35.00	47.00
			15.13	17.30	21.43	24.55	30.95	44.25
	20 grams	1	17.00	18.00	20.00	23.00	28.00	31.00
		2	16.80	17.70	23.00	27.00	35.00	47.00
		3	15.40	18.50	23.30	28.30	30.00	46.50
		4	15.50	17.90	22.90	27.80	30.00	44.50
	Average		16.175	18.025	22.3	26.525	30.75	42.25