COMPARISON OF MICROWAVE-ASSISTED HYDRODISTILLATION WITH THE CONVENTIONAL HYDRODISTILLATION METHOD IN THE EXTRACTION OF ESSENTIAL OILS

HANIS FARZANA BINTI MOHD SADEK

A thesis submitted in fulfillment of the
requirements for the award of the degree of
Bachelor of Chemical Engineering

Faculty of Chemical & Natural Resources Engineering
University Malaysia Pahang

APRIL 2010

ABSTRACT

The production of essential oils from natural sources is highly profitable nowadays. People worldwide start to realize the important of healthy awareness. Highly demand of ginger *(Zingiber officinale Roscoe)* oil and citronella grass *(Cymbopogon nardus)* oil due to its uses. Here, a modern and an alternative method, microwave-assisted hydrodistillation (MAHD) is approach. The purpose of this study is to investigate the performance of MAHD on the extraction of essential oils. The essential oils from ginger and citronella grass were studied and results were compared with the conventional HD in terms of extraction yield, extraction time, cost of operation and chemical composition. MAHD was provided higher quantitative in short of time. MAHD resulted in similar citronella oil recovery for 60 min to that obtained by 90 min of HD (1.17%, v/w). Same as ginger essential oil, MAHD required extraction time period of 30 min for recovering 0.88% yield of essential oil of ginger, instead of HD required 90 min of extraction time for recovering same percentage yield. Gas chromatography-mass spectrometry analysis of the extracted essential oils indicated that the use of microwave irradiation recover high total peak area (%). MAHD was found to be a green technology.

ABSTRAK

Pengeluaran minyak pati daripada sumber semulajadi pada masa kini menjanjikan pulangan yang lumayan kepada para pengusaha. Ramai yang telah mula menyedari kepentingan penjagaan kesihatan. Permintaan yang tinggi bagi minyak pati halia *(Zingiber officinale Roscoe)* dan juga serai wangi *(Cymbopogon nardus)* berikutan penggunaanya yang semakin meluas. Bagi memenuhi permintaan tersebut, satu kaedah alternatif dan moden, hydrodistillation dengan bantuan gelombang mikro diperkenalkan. Dengan itu, kajian ini dijalankan bertujuan untuk mengenalpasti prestasi MAHD pada pengekstrakan minyak pati. Minyak-minyak pati daripada halia dan serai wangi telah dikaji dan dibandingkan dengan HD konvensional dari segi hasil pengekstrakan, masa penghasilan, kos operasi dan komposisi kimia. MAHD telah menghasilkan lebih tinggi kuantitatif dalam waktu yang singkat. MAHD mampu mengekstrak minyak serai wangi sebanyak 1.17 % pada minit ke 60 berbanding HD yang memerlukan 90 minit untuk menghasilkan jumlah minyak yang sama. Begitu juga bagi minyak pati halia, pada minit ke 30, MAHD telah mengekstrak seabanyak 0.88% hasil minyak pati halia berbanding HD pada minit ke 90. Hasil analisis *gas chromatography-mass spectrometry*, minyak pati yang dihasilkan dengan penggunaan penyinaran gelombang mikro mencapai jumlah puncak yang tinggi (%). MAHD merupakan satu teknologi yang mesra alam.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

bp	-	Boiling point
DEET	-	(N,N-diethyl-m-toluamide)
DNA	-	Deoxyribonucleic acid
etc	-	Et cetera (and so forth)
g	-	Gram
GC	-	Gas chromatography
GC-FID	-	Gas chromatography-flame ionization detector
GC-MS	-	Gas chromatography- mass spectrometry
GHz	-	Gigahertz
HD	-	Hydrodistillation
HPLC	-	High performance liquid chromatography
i.e.	-	Id est (that is)
MAE	-	Microwave-assisted extraction
MAHD	-	Microwave-assisted hydrodistillation
MHz	-	Megahertz
min	-	Minute
mL	-	Milliliter
Mw	-	Molecular weight
SFE	-	Supercritical fluid extraction
TLC	-	Thin layer chromatography
UV	-	Ultraviolet

LIST OF SYMBOLS

^{o}C	-	Degree Celsius
η	-	Refractive index
%	-	Percent
&	-	And
×	-	Multiply
v/w	-	Volume per weight
W	-	Watt

CHAPTER 1

INTRODUCTION

1.1 Research Background

Nowadays, people were eager to find something new, especially from herbs or medicinal plant. It is because people worldwide start to realize the important of healthy awareness. By using product base natural sources, they believe and realize there are no side effect to society and environment. Essential oil or also known as ethereal oil is a concentrated, hydrophobic liquid that contains hundreds of aromatic compounds, organic constituents, including hormones, vitamins and other natural elements. These compounds are extracted from leaves, stems, flowers, bark, roots or other elements of a plant; essential oil contains highly volatile components. These essential oils have many uses. Besides being medicinal herbs, most of them are produced for the perfume industry, handmade soap, bath salts, lotions and other natural bath and body products.

There are a lot of plants that can extract the essential oils from. For example, Ginger (*Zingiber officinale Roscoe),* Citronella Grass *(Cymbopogon nardus),* Lavender flowers (*Lavandula angustifolia Mill*, Lamiaceae), Gaharu (*Agarwood*), Thyme (*Thymus vulgaris L.),* and Misai Kucing (*Orthosiphon Stamineus)*.

Zingiber officinale Roscoe (common ginger), a member of *Zingiberaceae* family is an aromatic/medicinal plant. It is indigenous to the Asia South East. Ginger products such as essential oil and oleoresin, are internationally commercialized for

use in food and pharmaceutical processing (C. Z. Kelly et al., 2002). The name Ginger is derived from the Sanskrit word Sringavera (meaning shaped like a horn) and being commonly found in South East Asia. It is highly beneficial in combating depression. It has also proved its effectiveness in relieving pain. Ginger essential oil is sought after due to its aphrodisiac and stimulant properties. Ginger herb is extensively brought to use for its moisture retention characteristics. It helps a great deal in toning the body. It serves as the best remedy to fight against Malaria.

Cymbopogon nardus also known as citronella grass is a perennial of the *Poaceae* grass family, originating in tropical Asia. It is a genus of about 55 species of grasses, native to warm temperate and tropical regions of the Old World and Ocenia. It is a perennial tufted grass with long, sharp-edged blades. The fresh stalks and leaves have a clean lemonlike odour because they contain an essential oil, which is also present in lemon peel. Citronella grass cannot be eaten because of its unpalatable nature. This plant is an invasive species that renders pastureland useless as cattle will starve even in its abundance.

Orthosiphon stamineus, better known as Misai Kucing by the locals is rich in flavonoids. Most flavonoids are bioactive compounds due to the presence of phenolic group in their molecule (Khamsah et al., 2006). *Orthosiphon Stamineus* (Misai Kucing) is a medicinal herb found mainly throughout South East Asia. It is believed to have anti allergic, antihypertensive, anti-inflammatory and diuretic properties. It is used as a remedy for arteriosclerosis (capillary and circulatory disorders), kidney stones and nephritis.

Thymus vulgaris L. (thyme), a member of the Labiatae family, is an aromatic/medicinal plant of increasing economic importance for North America, Europe, North Africa and Asia (Letchamo & Gosselin, 1996). Thyme is one of many aromatic plants that have been utilized in variety of food products to provide a flavour specific to this herb.

The main methods to obtain essential oils from the plant materials are hydro distillation (HD), steam distillation, steam and water distillation maceration, empyreumatic (or destructive) distillation, and expression (Stahl-Biskup & Saez, 2002). Among these methods, HD has been the most common approach to extract the essential oils from the medicinal herbs/plants (Stahl-Biskup & Saez, 2002). However, in order to reduce the extraction time and possibly improve the extraction yield, to enhance the quality of the extracts and also to reduce the operation costs, new approaches such as microwave-assisted extraction (MAE), pressurized solvent extraction, supercritical fluid extraction, and ultrasound-assisted extraction have also been sought (Kaufmann & Christen, 2002; Wang & Weller, 2006).

1.2 Problem Statement

Currently, in Malaysia, essential oil and oleoresin is gaining popularity as herbal medication as it gave a lot of benefit to overcome the disease. Most of the essential oil has medicinal properties and it had been used since thousand years ago. Ginger is a well known as marketable spice. Today, the essential oil from ginger is widely used and the most important is that the ginger oil is used in medical field for a few sicknesses. Same as citronella oil, people widely used as insect repellent, aromatherapy, astringent and also perfume industry.

By using the conventional hydrodistillation, it was prolong the extraction time. For example, it was needed 7 days continuous heating in order to extract volatile material. So, in order to fulfil the demand and commercialize the product, Microwave-assisted hydrodistillation is an alternative and cost effective method to extract essential oil from ginger. This method will be useful in order to overcome the disadvantages of conventional hydrodistillation. Besides, this method is environmental friendly and cleaner process for isolation of essential oil using microwave energy. Regarding environmental impact, the calculated quantity of carbon dioxide rejected in the atmosphere is lower. The waste water also reduced (Asma Farhat et al., 2009).

1.3 Objectives

The objectives of this study are:-

1) To identify the chemical composition of the essential oil extracted from ginger *(Zingiber officinale Roscoe)* and citronella grass *(Cymbopogon nardus)* by using microwave-assisted hydrodistillation (MAHD) and conventional hydrodistillation (HD).
2) To investigate the performance of potential microwave-assisted hydrodistillation (MAHD) on the yield of essential oil.
3) To identify the effect of extraction time and operational cost.

1.4 Scopes of Study

The scopes of this research are listed as below:-

1) Study the effect of extraction time to the yield of essential oil. Every method consumed different time of extraction.
2) Comparing the extraction yield and efficiency between microwave-assisted hydrodistillation MAHD and conventional hydrodistillation HD method.
3) Study performance of microwave-assisted hydrodistillation MAHD in extraction of essential oil.
4) Analyze the chemical composition of essential oil by using Gas Chromatography-Mass Spectrometry (GC-MS)
5) Study the operational cost based on extraction time and yield.

CHAPTER 2

LITERATURE REVIEW

2.1 Essential Oils

An essential oil is a concentrated, hydrophobic liquid containing volatile aroma compounds from plant. Essential oils contain the DNA of the plant of herb they are extracted from. Essential oils or as they are sometimes called volatile or ethereal oils are believed to be that small portion of the plant material, which imparts the characteristic odour and flavour most closely associated with the vegetative matter which they are obtained. Essential oils do not as a group need to have any specific chemical properties in common, beyond conveying characteristic fragrances.

The advantages of essential oils are their flavour concentrations and their similarity to their corresponding sources. The majority of them are fairly stable (notable exception is the citrus oil) and contain a few natural antioxidants. Essential oils are usually colorless, particularly when fresh. Nevertheless with age essential oil may oxidize and resinify, which resulting the colour becomes darker. Therefore, essential oil need to be stored in a cool, dry place tightly stoppered and preferably full in amber glass containers.

The choice of a particular process for the extraction of essential oil is generally dictated by the following considerations:

1) Sensitivity of the essential oils to the action of heat and water.
2) Volatility of the essential oil.

3) Water solubility of the essential oil.

As most of the essential oils of commerce are steam volatile, reasonably stable to action of heat and practically insoluble in water hence are suitable for processing by distillation. They are used in perfumes, cosmetics and bath products, for flavouring food and drink, and for scenting incense and household cleaning products. Essential oils are derived from various sections of plants. For example, Lavender flowers (*Lavandula angustifolia Mill, Lamiaceae),* Gaharu *(Agarwood),* Misai Kucing *(Orthosiphon Stamineus),* Thyme *(Thymus vulgaris L.),* Lime *(Citrus latifolia Tanaka),* Citronella grass *(Cymbopogon nardus)* and Ginger (*Zingiber officinale Roscoe).*

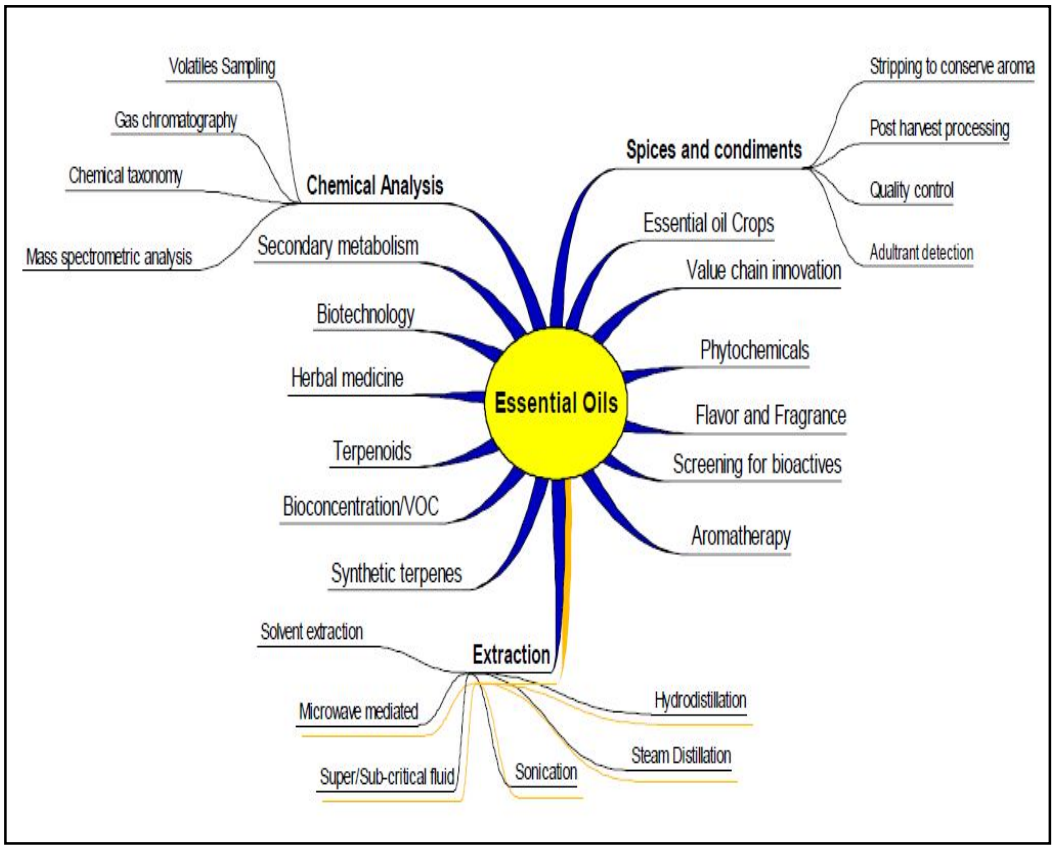

Figure 2.1: Tree diagram showing the wide branching of specializations in the field of essential oils

2.1.1 Lavender Flowers

The name lavender (*Lavandula angustifolia Mill*, Lamiaceae) comes from the Latin root *lavare*, which means "to wash." Lavender is one of the most useful medicinal plants. Commercially, the lavender is an important source of essential oil that is widely used in fragrance industry including soaps, colognes, perfumes, skin lotions and other cosmetics (Paul et al., 2004). In food manufacturing, lavender essential oil is employed in flavouring beverages, ice-cream, candy, baked goods and chewing gum. Recently, uses of lavender in aromatherapy as relaxant are becoming increasingly popular (Fakhari et al., 2005). The scent has a calming effect which may aid in relaxation and the reduction of anxiety.

Lavender is native to the mountainous zones of the Mediterranean where it grows in sunny, stony habitats. Today, it flourishes throughout southern Europe, Australia, and the United States. Lavender is a heavily branched short shrub that grows to a height of roughly 60 centimetres. Its broad rootstock bears woody branches with erect, rod-like, leafy, green shoots. A silvery down covers the gray-green narrow leaves, which are oblong and tapered, attached directly at the base, and curled spirally. Figure 2.2 below shows the lavender plant.

Human clinical studies have reported that lavender essential oil may be beneficial in a variety of conditions, including insomnia, alopecia (hair loss), anxiety, stress, postoperative pain, and as an antibacterial and antiviral agent. Lavender oil is also used together with other forms of integrative medicine, such as massage, acupuncture, and chiropractic manipulation. It may also help to relieve pain from tension headache when breathed in as vapor or diluted and rubbed on the skin. When added to a vaporizer, lavender oil may aid in the treatment of cough and respiratory infection.

Figure 2.2: Lavender plant

2.1.2 Gaharu

Gaharu or also known as ‘Agarwood’ ‘Aloeswood’, ‘Eaglewood’ in English and ‘Jinko’ in Japanese is a fragrant wood product that usually obtained from a pathological condition of the wood of standing trees of *Aquilaria (Thymelaeceae)* species and one of the most valuable non-timber products in Asian tropical forest. *Aquilaria* is a fast-growing, archaic tropical forest tree, which occurs in South and Southeast Asia, from the foothills of the Himalayas to the rainforests of Papua New Guinea. The tree grows in natural forests at an altitude of a few meters above sea level to about 1000 meters, and it grows best around 500 meters. It can grow on a wide range of soils, including poor sandy soil. Seedlings need a lot of shade and water. According to Dato Dr Abdul Rashid Abdul Malik there are 25 species of Agarwood worldwide and only about 12 of it were able to produce gaharu (Borneo Post, 2009). The major source of agarwood is Aquilaria Malaccensis.

The production of gaharu resins have a several potential such as used primarily in traditional Chinese and Korean medicines. The oil also used in perfumes and cosmetic product. Besides that the chips of agarwood are ground into powder can be used for special cigarettes. Its ethereal fragrance is demanding for incense which used in religious and spiritual ceremonies of Islam or Buddhism especially in Asia such as Thailand, Malaysia and Indonesia. Agarwood might have some effects

towards central nervous system (CNS) such as higher brain function, from traditional use as a sedative (Ueda J. et al, 2006). The odor of agarwood is complex and pleasing, with few or no similar natural analogues.

Formation of agarwood occurs in the trunk and roots of trees that have been infected by a parasitc ascomycetous mold, *Phaeoacremonium parasitica*, a dematiaceous (dark-walled) fungus. As a response, the tree produces a resin high in volatile organic compounds that aids in suppressing or retarding the fungal growth. While the unaffected wood of the tree is relatively light in colour, the resin dramatically increases the mass and density of the affected wood, changing its colour from pale beige to dark brown or black. In natural forest only about 7% of the trees are infected by the fungus. Figure 2.3 shows the picture of agarwood trees and Figure 2.4 shows pieces of agarwood.

Figure 2.3: Agarwood trees

Figure 2.4: Pieces of agarwood

2.1.3 Thyme

Thyme (*Thymus vulgaris L.)* is a low growing herbaceous plant, sometimes becoming somewhat woody. Figure 2.5 shows thyme plant. It is native to southern Europe. It is much cultivated as a culinary herb. Thyme is one of many aromatic plants that have been utilized in variety of food products to provide a flavor species to this herb (Golmakani & Rezaei, 2008). Thyme is a good source of iron and is used widely in cooking. Thyme is often used to flavour meats, soups and stews. It has a particular affinity to and is often used as a primary flavour with lamb, tomatoes and eggs.

From the medicinal point of view, the essential oil of thyme is made up of 20-54% thymol. Thymol, an antiseptic, is the main active ingredient in Listerine mouthwash. Before the advent of modern antibiotics, it was used to medicate bandages. It has also been shown to be effective against the fungus that commonly infects toenails. Medicinally thyme is used for respiratory infections. In traditional Jamaican childbirth practice, thyme tea is given to the mother after delivery of the baby.

Figure 2.5: Thymus vulgaris L.

2.1.4 Misai Kucing

One of the local herbs which is scientifically known as *Orthosiphon stamineus* Benth (*O. stamineus*) or locally called as Misai Kucing is rich in flavonoids (Surmayono *et al*., 1991). Most flavonoids are bioactive compounds due to the presence of phenolic group in their molecule (Apati, 2003: Havsteen, 2002). The leaves of *O. stamineus* are arranged in opposite pairs. The petiole is relatively short, about 0.3 cm in length and reddish purple in colour. The flowers are borne on verticals about 16 cm length, white to bluish in colour with long far-exerted filaments, making it look like cat's whispers (Wong FK, 2002). Figure 2.6 below shows how *Orthosiphon stamineus* look like.

Orthosiphon stamineus Benth (Lamiaceae) is a popular medicinal plant in Southeast Asia. It is widely used for the treatments of many diseases, especially those affecting the urinary tract, diabetes mellitus, hypertension, rheumatism, tonsillitis and menstrual disorder (Awale et al., 2003a,b). The methanolic extracts of this plant have shown the inhibitory activity on nitric oxide production in macrophage like cells (Awale et al., 2003a,b). *O. stamineus* is also found in other countries such as Thailand, Indonesia and Europe. In these countries, Misai Kucing is also known as Yaa Nuat Maeo, Rau Meo or Cay Bac (Thailand), Kumis Kucing or Remujung (Indonesia), moustaches de chat (French) and Java Tea (European) (Indubala & Ng, 2000).

Figure 2.6: *Orthosiphon stamineus* plants

2.1.5 Lime

The lime tree is a member of the Rutaceae family. The essential oil from the lime has a sweet, tangy scent. It is also used now medicinally to help enhance the nervous system. The oil also has antiseptic properties. They are very well known across the world and are extensively used in pickles, jams, marmalades, sauces, confectionaries, squashes, sorbets, desserts, beverages, cosmetics and a numerous other industrial products.

Citrus oils are mixtures of very volatile components as terpenes and oxygenated compounds. Limonene, a monoterpene, is the major component of lime and other related citrus essential oils. These oils are used in the pharmaceutical, perfumery and food industries, and the quality of the oils is related to the value of total aldehydes, basically citral content, which is between 4-5%. The common commercial methods to produce the oils from citrus fruits and peels are machine cold pressing and distillation (Atti-Santos, 2005).

2.1.6 Ginger

Zingiber officinale Roscoe also known as ginger consists of the fresh or dried roots of *Zingiber officinale*. The English botanist William Roscoe (1753-1831) gave the plant the name *Zingiber officinale* in an 1807 publication. The ginger family is a tropical group especially abundant in Indo-Malaysia, consisting of more 1200 plant species in 53 genera. The genus *Zingiber* includes about 85 species of aromatic herbs from East Asia and tropical Australia. The name of the genus, *Zingiber*, derives from a Sanskrit word denoting "horn-shaped," in reference to the protrusions on the rhizome (Foster, 2009). Today, it is cultivated all over tropic and subtropaic Asia (50% of the world's harvest is produced in India), in Brazil, Jamaica (whence the best quality is exported) and Nigeria, whose ginger is rather pungent, but lacks the fine aroma of other (Arnould, 1981). Figure 2.7 shows a picture of the fresh ginger rhizome and Figure 2.8 shows the ginger plant.

Ginger is a perennial herb and grows to about 3 - 4 feet high with thick spreading tuberous rhizome. Every year it shoots up a stalk with narrow spear-shaped leaves as well as white or yellow flowers growing directly from the root (Lawrence and Reynolds, 1984). The ginger of commerce consists of the thick scaly rhizomes (underground stems) of the plant. They branch with thick thumb-like protrusions, thus individual divisions of the rhizome are known as "hands."

Figure 2.7: Fresh ginger

Figure 2.8: Ginger plant

2.1.6.1 Historical Use

Ginger had a reputation for being the remedy that could cure some of the most difficult and chronic ailments. Some physicians regularly used it as a remedy to counter violent temper and hysteria and the treatment for such conditions involved making the patients smoke without their knowledge, dried, finely shredded ginger roots out of a hollowed reed instead of tobacco. Along with Cinnamon, Ginger was high on the list of culinary ingredients and was and still is a much appreciated addition to food in England. Although not quite as popular as it used to be, the consumption of ginger is still fairly high in the UK and this is reflected in traditional ginger beer, ginger brandy, ginger champagnes, ginger wines and gingerbread men (Perez, 2005).

Ginger is very widely used in Traditional Chinese Medicine. It is said to be a Yang/spleen remedy and is supportive to the spleen, stomach and kidneys (the latter is specially indicated for men and it is classified as an aphrodisiac and considered a good remedy for impotence). Both fresh and dried roots are official drugs of the modern Chinese pharmacopoeia, as is a liquid extract and tincture of ginger. Ginger is used in dozens of traditional Chinese prescriptions as a "guide drug" to "mediate" the effects of potentially toxic ingredients. In fact, in modern China, Ginger is believed to be used in half of all herbal prescriptions (Foster, 2009).

Meanwhile, in Arabic medicine, ginger is said to be hot in the second degree and moist in the first. It is warming and has a softening effect on the belly; it is beneficial to the body against digestive ailments such as flatulence, food poisoning and constipation (Perez, 2005).

Ginger holds just as an important place in Western medicine as it does in Eastern medicine (China, Japan and India to name but a few). It has been used on its own or included as an ingredient in specific herbal formula and also used as a ‘corrective remedy’ against the unwanted effects of other plants. It has been verified in recent research.

2.1.6.2 The Composition of Ginger Oil

Figure 2.9 shows the structures of zingiberene and β-sesquiphellandrene for ginger oil constituents. Ginger essential oil mostly consists of zingiberene, dexto-camphene, beta-sesquiphellandrene, bisabolene, dexto-phellandrene, betaphellandrene and 1,8-cineole. Ginger oil owes its aroma to bisabolene, zingiberene and zingiberol. The physical properties and characteristics of some of the constituents of the ginger oil are shown in Table 2.1 (Nurul Azlina, 2005).

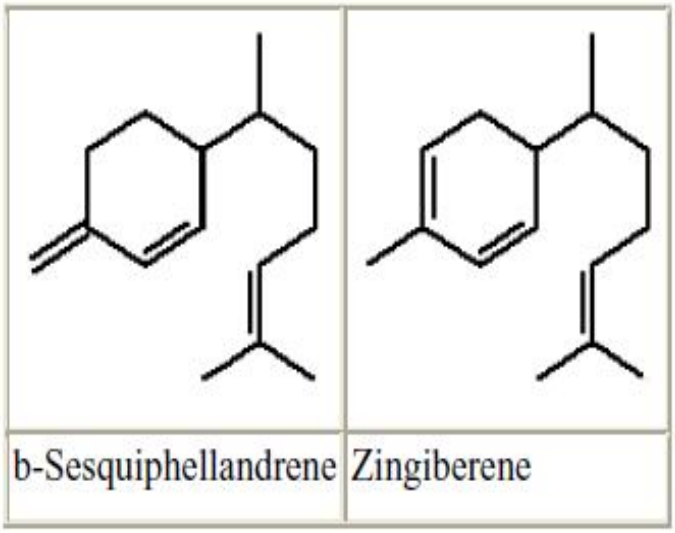

Figure 2.9: Structures of ginger oil constituents (Lawrence and Reynolds, 1984)