CHEMICAL COMPOSITION, ANTIOXIDANT AND ANTIBACTERIAL ACTIVITY OF ESSENTIAL OIL FROM LEAF OF *SYZYGIUM POLYANTHUM* (Wight) Walp.

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ABSTRACT

Syzygium polyanthum (Wight) Walp. is a plant widely consumed as raw vegetables or used in culinary in Malaysia. This study was performed to determine the chemical composition, antioxidant and antibacterial activity of *Syzygium polyanthum* essential oil from Malaysia. The essential oil was extracted from the leaf by hydrodistillation method. The percentage of the essential oils obtained, for the fresh and the dried leaf were 0.67 % and 1.50 %, respectively. Analysis by GCMS revealed a total of 26 and 29 compounds, representing 71.71 % and 90.92 % of the overall compositions, were detected in the fresh and the dried leaf essential oils. Both oils were characterized by high amount of monoterpene hydrocarbons with α-pinene was the major compounds for the fresh leaf (28.78 %) and the dried leaf (34.15 %). The antioxidant activities of the fresh and dried leaf oils were evaluated using three different assay; DPPH free radical scavenging, β-carotene bleaching and ferrous-ion chelating. Results showed that the oils exhibited a potential antioxidant activity. The antibacterial activity was evaluated using the disc diffusion and microdilution methods against three gram-positive bacteria (*Bacillus subtilis, Enterococcus faecalis*) and three gram-negative bacteria (*Escherichia coli, Pseudomonas aeruginosa, Salmonella typhii*). The oils exhibited antibacterial activity against all of the tested bacteria. The minimum inhibitory concentration (MIC) values for the fresh leaf oil were in the range of 0.03 mg/ml to 0.25 mg/ml while the MIC values for the dried leaf oil were in the range of 0.03 mg/ml to 0.12 mg/ml. The chemical composition, antioxidant and antibacterial activity of essential oil of *S. polyanthum* from Malaysia is presented here for the first time.

ABSTRAK

Syzygium polyanthum (Wight) Walp. adalah sejenis tumbuhan yang digunakan secara meluas sebagai ulaman atau digunakan dalam masakan di Malaysia. Kajian ini telah dijalankan untuk menentukan komposisi kimia dan aktiviti antioksida dan antibakteria minyak pati *Syzygium polyanthum* dari Malaysia. Minyak pati ini diekstrak dari daun melalui kaedah penyulingan hidro. Peratusan minyak pati yang diperolehi adalah 0.67 % (daun segar) dan 1.50 % (daun kering). Analisis oleh GCMS mendedahkan sejumlah 26 dan 29 sebatian, yang mewakili 71.71 % dan 90.92 % daripada komposisi keseluruhan, dikesan dalam minyak pati daun segar dan daun kering. Kedua-dua minyak telah disifatkan oleh hidrokarbon monoterpene dengan α-pinene adalah sebatian utama untuk daun segar (28.78 %) dan daun kering (34.15 %). Aktiviti antioksida minyak dari daun segar dan kering diuji menggunakan tiga uji kaji berbeza; penghapusan radikal bebas DPPH, pelunturan β-karotena dan pengkelat ion ferus. Keputusan menunjukkan bahawa minyak pati mempamerkan potensi terhadap aktiviti antioksidan. Aktiviti antibakteria dinilai menggunakan menggunakan penyebaran cakera dan kaedah microdilution terhadap tiga bakteria gram-positif (*Staphylococcus aureus*, *Bacillus subtilis, Enterococcus faecalis*) dan tiga bacteria gram-negatif (*Escherichia coli, Pseudomonas aeruginosa, Salmonella typhi*). Minyak pati ini mempamerkan aktiviti antibakteria terhadap semua bakteria yang diuji. Nilai MIC daripada minyak daun segar adalah di antara 0.03 mg/ml hingga 0.25 mg/ml manakala nilai MIC untuk minyak daun kering di antara 0.03 mg/ml hingga 0.12 mg/ml. Komposisi kimia, aktiviti antioksida dan aktiviti antibakteria untuk minyak pati dari *Syzygium* polyanthum dari Malaysia dilaporkan disini buat pertama kalinya.

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LIST OF SYMBOLS

α	Alpha
β	Beta
cm	Centimetre
^{o}C	Degree Celsius
δ	Delta
=	Equals
Fe^{2+}	Ferrous-ion
γ	Gamma
g	Gram
>	Greater-than Sign
≥	Greater-than or Equal to Sign
<	Less-than Sign
L	Liter
m	Meter
µl	Microliter
mg	Milligram
ml	Mililiter
mm	Millimeter
mM	Millimolar
x	Multiplication Sign
nm	Nanometer
-	Negative Sign

%	Percentage
±	Plus-minus Sign
+	Positive Sign

LIST OF ABBREVIATIONS

A	Absorbance
ANOVA	Analysis of Variance
ATTC	American Type Culture Collection
BC	Before Century
CFU	Colony-forming Unit
GCMS	Gas Chromatography Mass Spectrometry
KI	Kovats Index
MAE	Microwave-assisted Extraction
NCCLS/CLSI	National Committee for Clinical Laboratory Standards
NIST	National Institute of Standards and Technology
RI	Retention Indices
SD	Standard Deviation
SPSS	Statistical Package for Social Science
SPME	Solid-phase Microextraction
USNRC	United States Nuclear Regulatory Commission

CHAPTER 1

INTRODUCTION

1.1 BACKGROUND OF STUDY

In this modern era, people are more aware of their health status that leads them to find natural-based products to substitute the role of various synthetic products which are available in market. According to the World Health Organization (WHO), about 60-80 % of people from developing countries consume traditional medicine for treatment of various diseases (Prabuseenivasan *et al.*, 2006). Therefore, herbs and medicinal plants are gaining interest because of their potential to produce natural products, which are known to be good sources to provide better health. Continuous research and studies of potential herbs and medicinal plants are important as natural products from plant origin will continue to be in demand.

Malaysia, a tropical Asian country, is gifted with a rich diversity of flora and fauna, particularly, herbs and medicinal plants. Malay people, especially, like to consume traditional vegetables, also known as *ulam* in their daily diet. This is due to the taste (Abas *et al*., 2006) and aroma which enhance their appetite and promote better health. There are more than 120 species of traditional *ulam*, varying from shrubs to large trees, which can be found in Malaysia. They are usually grown at home and can also be found abundantly in local market. One of the favorite *ulam* that have been consumed for ages is *Syzygium polyanthum* (Wight) Walp. *Syzygium polyanthum* (Wight) Walp. (syn. *Eugenia lucidula* miq and *Eugenia polyantha* Wight) is a member of Myrtaceae, mainly distributed in Malaysia, Indonesia, Myanmar, Thailand, Indochina and also in Suriname (Wartini, 2009). This deciduous tropical tree (Raden *et al*., 2009) is called differently in each country or province: serai kayu, salam, kelat (Malaysia), meselangan (Sumatra), salam (Java, Sunda, Madura), gowok (Sunda), manting (Java) or kastolam (Kangean) (Dalimartha, 2005 and Suganda and Ruslan, 2007). In Indonesia, it

is commonly called as Indonesian bay leaf or Indonesian laurels (Heyne, 1987) because the function in cooking resembles the original European bay leaf (*Laurus nobilis* L.).

Fresh and dried aromatic leaf of *S. polyanthum* are useful in culinary (Katzer, 2004) because of its scent, colour and flavour. It is often used as flavouring spice for meat, fish, and vegetable dishes or in rice (De Guzman and Siemonsma, 1999). In Malaysia, the leaf are an important ingredient in *nasi kerabu* and *kerabu perut* or eaten raw as *ulam*. It is believed that the leaf can treat haemorrhoids, stomach-ache, diarrhea (Kloppenburg-Versteegh, 1983), diabetes, itchiness, gastritis, astringent and scabies (Wijayakusuma, 1995), hypertension, high cholesterol (Suharti *et al.*, 2008) and others.

Figure 1.1: *Syzygium polyanthum* (Wight) Walp.

The presence of the essential oils and their compositions determine the characteristic aroma of the plant and flavour of the condiments (Chalchat and Ozcan, 2008). Essential oil (or volatile oil) is a concentrated hydrophobic mixture of chemical compounds with distinctive odor and flavor, which can be obtained from different parts, namely leaf, stems, barks, flowers, fruits, seeds and rhizomes (Munir and Oliver, 2007) varying according to plant species. Extraction is by physical means only (Ferhat, 2007), either by suppression, fermentation or extraction (Prabuseenivasan *et al.*, 2006). The common methods used are conventional hydro-distillation and steam-distillation and further analyzes using gas chromatography mass spectrometry (GCMS) for the chemical constituents. *Syzygium polyanthum* also produces essential oil.

Previous studies on the essential oils of *S. polyanthum* leaf shows that the main components are citric acid, eugenol, methyl chavicol (Sumono and Agustin, 2008). The essential oil of the leaf obtained from steam-distillation was found to contain cis-4-decenal (27.12 %), octanal (11.98 %), α-pinene (9.09 %), farnesol (8.84 %), β-ocimene (7.62 %) and nonanal (7.60 %) (Wartini, 2009). The essential oil is also reported to have potential use in dentistry (Sumono and Agustin, 2008). There are other previous studies which report that *S. polyanthum* also possesses biological activities such as antioxidant (Wong *et al.*, 2006 and Raden *et al.*, 2009), antibacterial (Hendradjatin, 2004 and Sumono and Agustin, 2008), antimicrobial (Srimuwarni *et al.*, 2005), anti-inflammation (Wientarsih *et al.*, 2007) and antifungal (Guynot *et al.*, 2005 and Noveriza and Miftakhurohmah, 2010).

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1.2 STATEMENT OF PROBLEM

The compositions of essential oil from *S. polyanthum* have been reported in earlier studies, but no studies have been done on the essential oil of the species grown in Malaysia. The lack of information on the phytochemical aspects of the tree is in great contrast with the widespread use in Malaysian cuisine. Thus, it would be an interesting subject for study. Based on these facts, it is our interest to perform the screening and analysis of the chemical compounds of the essential oil from *S. polyanthum* leaf grown in Malaysia and its antioxidant and antibacterial properties in order to provide information for potential utilization in future.

1.3 RESEARCH OBJECTIVES

1. To extract the essential oil from fresh and dried leaf of *Syzygium polyantum* (Wight) Walp. using hydrodistillation method.

2. To analyze the chemical compositions of the essential oil from fresh and dried leaf of *Syzygium polyantum* (Wight) Walp. using Gas Chromatography Mass Spectrometry (GCMS).

3. To determine the antioxidant and antibacterial activities of the essential oil extracted from fresh and dried leaf of *Syzygium polyantum* (Wight) Walp.

CHAPTER 2

LITERATURE REVIEW

2.1 HERBS AND MEDICINAL PLANTS

2.1.1 General

The oldest form of maintaining good health was practiced by the Egyptians, Chinese, Greeks and Romans back in 1500 B.C, where they utilized the traditional herbs and medicinal plants for medicine and also remedies for various usages (Juri *et al*., 1999). The natural products possessed by these plants are believed to improve and maintain health because they have minimal side effects, effective and safe (Katarina, 2005) compared to chemically synthesized and pure drugs. The return of herbalism and ancient medicines in this modern world are prior to the recognition that chemicals or synthetic products may cause harmful side effect in the long term. Currently, studies on medical plants are extensively done in order to improve the productivity of medicinal plants (Runham, 1996). Various scientific tests have been performed to prove that many traditional herbs possess interesting healing properties. With the rise of scientific tests on herbs and medicinal plants for their potential natural products, the limitation of sources is overcome and we can assess plants from around the world (Katarina, 2005).

2.1.2 Herbs and Medicinal Plants in Malaysia

Malaysia, as a tropical rainforest country, is gifted with great diversity of flora and fauna. About 12,000 species of flowering plants can be found in Malaysian rainforest and about 1,300 species are acknowledged as medicinal plants. Various chemical structures possessed by herbs and medicinal plants that can be found in Malaysian flora make natural products the excellent candidate for any screening interest (Ismail, 2000). The rapid increase of interest in herbs and herbal products, not only amongst scientists but also consumers has lead to the increased in demand, thus

increasing the price of raw materials and production cost. The utilization of local plants, either wild or introduced, by herbal industries has risen. This will give benefit to the local farmers and also provides profit to the country (Khatijah and Ruzi, 2006).

2.2 FAMILY *MYRTACEAE*

The family Myrtaceae comprises of around 121 genera and over 3800-5800 species (Stefanello *et al.*, 2011). It is widely distributed in Australia, South East Asia and America (tropical to southern temperate) and small occurrence in Africa. The genera *Eugenia, Syzygium,* and *Myrcia* (c. 1000, 1050, and 770 species, respectively) are among the predominant in the tropical rainforest trees, with the first two genera sharing much morphological similarity that has lead to their repeated synonymy (Lucas *et al.*, 2011) . The distinguished features of the family are leaf containing oil glands, half inferior to inferior ovary, numerous stamens, internal phloem, and xylem vessels with vestured pits (Wilson *et al*, 2001). Plants of Myrtaceae are evergreen trees or shrubs, usually with presence of essential oil in foliage, branches and flowers. All over the world, Myrtaceae is used as medicinal hygenic, edible and ornamental purposes (Ghannadi and Dezfuly, 2011). The uses of Myrtaceae as traditional medicine have led to the screening of some species for the essential oil and their biological activities (Stefanello *et al.*, 2011).

2.2.1 The Genus *Syzygium*

Syzygium, the genus of flowering plants, is one of the important genus in Myrtaceae family, mostly distributed in the tropical and sub-tropical region of the world, with the greatest diversity of species taking place in South East Asia such as Indonesia, Malaysia and also in East India. About 175 species can be found in Sabah and Sarawak (Ashton, 2006). It is represented by around 140 genera and 1100 species (Ayyanar and Babu, 2012), occurring as evergreen trees, shrubs and suffrutices.

Several species of the genus *Syzygium* were formerly classified as *Eugenia* and many references may still list these species as *Eugenia.* Most *Eugenia* species originated from America and the West Indies whereas *Syzygium* species originated from the Indo-Malaysian region. S*yzygium cuminutesi, Syzygium aromaticum, Syzygium polyanthum, Syzygium jambos, Syzygium malaccense,* and *Syzygium aqueum* are some key species of the genus. Several species of the *Syzygium* are grown and consumed for their edible fruits (*Syzygium jambos, Syzygium aqueum, Syzygium samarangense)* and some are used in traditional medicine to treat inflammation (Chauduri *et al.*, 1990), asthma (Alma *et al.*, 2007), various allergic disorders by oral administration (Kim *et al,* 1998), sore throat, bronchitis, thirst, dysentery and ulcers (Ayyanar and Babu, 2012). Some species also exhibited antiseptic properties and the commercial antiseptic from this genus is Clove (Mohanty and Cock, 2010).

Phytochemical studies on *Syzygium* species have led to the identification and isolation of main compounds such as eugenol, eugenol acetate, β-caryophyllene (Alma *et al.*, 2007), flavonoids, caryophyllene (Gao *et al.*, 2012) and others. Several species were reported to have antioxidant, antibacterial (Tsakala *et al.*, 1996; Corine *et al.*, 2000 and Shafi *et al*., 2002), antifungal (Chandrasekaran and Venkatesalu, 2004), anti-inflammatory activities (Muruganandan *et al.*, 2001). The chemical compositions of the essential oils and their biological activities from some *Syzygium* species have been previously evaluated, however little is known for the Malaysian *S. polyanthum* species.

2.2.2 Essential Oils of *Syzygium* Species

Table 2.1: Summary on major chemical constituents of essential oils from the leaf of some *Syzygium* species

Species	Region	Extraction method	Major Compounds	References
S. aromaticum (L.) Merr and Perry	Madagascar	Hydro distillation	Eugenol (82%), β-caryophyllene (13%), α-humulene (1.5%), eugenyl acetate (0.4%) and caryophyllene oxide (0.5%)	Srivastava *et al.* (2005)
S. samarangense (Blume) Merr. And L.M. Perry	India	Steam distillation	β-salinene (11.61%), α-salinene (11.40%), γ-terpinene (10.68%), β-caryophyllene (10.20%) and β-gurjunene (9.48%)	Reddy and Jose (2011)
S.polyanthum (Wight) Walp.	Indonesia	Steam distillation	*Cis-4-decenal* (27.12%), octanal (11.98%), α-pinene (9.09%), farnesol (8.84%), β-ocimene (7.62%) and nonanal (7.60%)	Wartini (2009)
S. cuminui L.	Egypt	Hydro distillation	α-pinene (17.53%), α-terpineol (16.67%),alloocimene (13.55%), α-bornyl acetate (6.27%) and caryophyllene (5.41%)	Elansary *et al.* (2012)
S. malacenses Merr. et Perry	Nigeria	Hydro distillation	p-cymene(13.5%),β-caryophyllene (9%), β-pinene (8%), α-terpineol (7.5%) and α-pinene (7.3%)	Karioti and Skaltsa (2007)
S.guineense (Willd.) DC. var *guineense*	Benin	Hydro distillation	α-cadinol(12.7%), cis-calamenen-10-ol (7.1%), epi-α-muurolol (5.7%), caryophyllene oxide (5.5 %) and cubenol (5.3 %)	Noudogbessi *et al.* (2008)

2.3 *SYZYGIUM POLYANTHUM* (WIGHT) WALP.

2.3.1 Introduction

Syzygium polyantum (Wight) Walp., (synonym *Eugenia polyantha*) is known as *Serai Kayu* or *Salam*, *Daun Kelat, Kelat Samak, Kelat Putih, Kelat Merah, Serah, Daun Salam Manting, Mantang, Ubah Laut, Pokok Palong, Jambu Hutan* in Malaysia and bay-leaf in Indonesia. It is native to tropical Asian countries which are Malaysia, Indonesia (Java, Sumatra, Kalimantan), Myanmar (Burma), Indo-China and Thailand. This medium-sized tree can reach up to 30 to 60 m high and grows wild in the rainforests but it can also be grown in gardens (Sumono and Agustin, 2008). Wee

(2003) reported that this plant can attract birds when fruiting and this helps to spread the seeds to other places. The chemical properties of *S. polyanthum* are the factors that contribute to the uses of this plant in dentistry for therapy and treatment (Utami and Lentera, 2005).

2.3.2 Description of Plant

Table 2.2: Description of *Syzygium polyanthum*

Plant Parts	Morphology and Descriptions
Leaf 	Leaf is opposite, simple, with 2.5-8 cm long and flat margins. The tip is blunt and the base of the leaf stretches along length and tights (Utami and Lentera, 2005) with presence of small oil glands (De Guzman, 1999). The dried brown leaf are aromatic, slightly sour and astringent.
Flowers 	The small flowers are in loose bunches that arise from twigs behind the leaf (Wee, 2003). The flowers are cream and later turning pink or reddish and have fragrance with the fruits around.
Fruits 	Fruit is a one-seed berry (12 mm). It is green and turns red to brown when mature (POM, 2004). The seeds are small (9-10 mm). The ripe fruits are quite sweet mixed sour.
Bark 	The grey round bark is fissured and scaly.

2.3.3 Uses

Culinary

Despite having a slightly sour taste and astringent, the fresh and dried leaf are often used as flavor condiment and food additive (Dalimartha, 2005) especially in Malaysian and Indonesian culinary. The crushed or ground leaf releases more desired fragrance than the whole leaf. In order to release the fragrance, it is suggested to leave the leaf to cook for some time. Ripe fruits are edible, although slightly astringent. In Northern Thailand, they consume and sell fresh ripe fruits during fruiting season (Suksri *et al.*, 2005).

Medicine

The leaf and bark are used traditionally to treat diarrhea (Very *et al.*, 2000). The chemical compounds present in the leaf have the effect of inhibiting the growth of bacteria that can cause diarrhea (Very *et al*., 2000 and Setiawaty, 2003). Poultices (combination of crushed leaf, bark and roots) are applied for curing itches and scabies. The leaf is used to stop gastritis, diabetes mellitus and can treat patient with high uric acid. The roots and fruits can be used to neutralize drunken people (Sumono and Agustin, 2008). The plant also has other benefits such as diuretic and analgesic effect (Utami and Lentera, 2005). *Syzygium polyanthum* is stated to have less side effect than synthetic drugs. It can be consumed as drugs by boiling or crushed as ointment and applied to the affected skin (Dalimartha, 2005).

Tannin or dyeing stuff

The bark consists of tannin, often used for dyeing fishing-nets to enhance the strength of the nets (Suksri *et al.*, 2005), bamboo matting and others.

Timber

Timber (trade name) of *S. polyanthum* belongs to the trade group *kelat*, which is a medium-weight to heavy hardwood. It is used for house building and furniture (Noorma Wati, 1995).

2.3.4 Phytochemicals

The many uses of *S. polyanthum* in traditional treatment are contributed mainly by the presence of the secondary metabolites possessed by the plant, referred to as essential oils and plant extracts. The essential oil of *S. polyanthum* can be obtained from the leaf, stems, fruits and barks. Previous studies have reported the composition of *S. polyanthum* essential oil in samples collected from Indonesia. The leaf contained essential oils, triterpenoids, saponins, flavonoids and tannins (Davidson and Branen, 1993).

The percentage yields and the main chemical compounds of the essential oil from previous studies of *S. polyanthum* have been reported previously. The highest yield was reported by Agusta (2000) with 1.0 % oil while Sumono and Agustin (2008) reported the essential oil yield of the dried leaf is about 0.17 %. The evaluation by Sembiring *et al*. in 2003 showed that the essential oil content of Indonesian bay leaf in Sukabumi (0.02 %) was higher than in Bogor (0.01 %).

Wartini (2009) reported that the essential oils obtained from steam-distillation without using n-hexane as solvents contained 27 compounds while the essential oil extracted from steam distillation using n-hexane yielded 25 compounds. The main compounds were *cis*-4-decenal, octanal, α-pinene, farnesol, β-ocimene, α-caryophyllene, trans-caryophyllene, citronellol and nerolidol. Another study reported that citric acid, eugenol and methyl chavicol (Sumono and Agustin 2008) were the main compounds of *S. polyanthum* oils. Sembiring (2003) have reported the presence of octanal, 3, 7-dimethyl-1-octene and cyclohexane while Agusta (2000) also reported 3, 7-dimethyl-1-octene but with the presence of n-decanal, patchoulin, D-nerolidol and caryophyllene oxide. Unfortunately, no eugenol, citric acid and methyl chavicol have

been identified in the present study and most of the previous studies. Eugenol is the major compound in the essential oils of clove, a member of the genus *Syzygium*. The main class of compounds listed above was aliphatic compounds and oxygenated sesquiterpenes.

Davidson and Branen (1993) reported that besides essential oils, the leaf also contained triterpenoids, saponins, flavonoids and tannins. The pytochemical analysis of leaf and stems of *S. polyanthum* done by Liliwirianis *et al.* in 2011 have reported the presence of alkaloid, saponin, steroid, phenolic and flavonoid with absence of terpenoid in both leaf and stems. It has also been reported that this plant consists of tannins (Dalimartha, 2005). Earlier research on the chemical constituents of the essential oil from Indonesian *S. polyanthum* has already been reported but no previous investigations have been done on the essential oil constituents of *S. polyanthum* cultivated in Malaysia.

2.3.5 Bioactivities

Some past studies supported the worth of the traditional use of *S. polyanthum*, providing several biologically active constituents especially main constituents in the essential oils. Studies involving bioactivities of the essential oil and plant extracts such as the antioxidant, antibacterial and antifungal properties from *S. polyanthum* have been previously demonstrated. Antioxidant activities of aqueous *S. polyanthum* leaf showed relatively high radical scavenging activity and high total polyphenol content (Wong *et al.*, 2006). Raden *et al.* (2009) reported that three bark extracts using methanol, methanol-water and water exhibit potential antioxidant activities, ranking in order: methanol-water extract > water extract > methanol extract. They proved that the total phenolic content has positive correlation with total antioxidative capacity in bark extracts.

The chemical compositions of *S. polyanthum* leaf also have the ability in the inhibition of pathogenic bacteria such as *Salmonella* sp., *Bacillus cereus*, *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas fluorescens and Bacillus subtilis* (Setiawan, 2002). Infusion and decoction of *S. polyanthum* leaf showed inhibition towards *E.coli* and *V. cholera* (Hendradjatin, 2004 and Srimurwarni *et al.*, 2005) and the extract of *S.*

polyanthum showed inhibition against *Candida albicans* growth in acrylic resin denture base (Sumono and Agustin, 2008). *Streptococcus* sp. colony was reduced in samples rinsed with (100 %, 75 % and 50 %) *S. polyanthum* solution. Noveriza and Miftakhurohmah (2010) demonstrated that the methanol extract of the leaf inhibited the growth of *Fusarium oxysporum* in solid media while for liquid media, the extract decreased conidia production and germination and hifa weight significantly. The essential oils of the leaf also possess antifungal properties. This was confirmed by Guynot *et al*. in 2005, where the essential oils inhibited the growth of the common fungus associated with the production of bakery which are *Eurotium* spp., *Aspergillus* spp. and *Penicillium* spp.

Tannin is believed to be responsible for the antibacterial activity of *S. polyanthum.* The mechanisms of inhibition of the bacteria growth involved precipitation forming and denaturing of the bacteria protein. The leaf of *S. polyanthum* also have potential in reducing cholesterol. Suharti *et al.* in 2008 concluded that *S. polyanthum* leaf meal could replace the use of antibiotic in broiler ration and reduces carcass cholesterol. The leaf of *S. polyanthum* showed strong *in vitro* anti-tumor promoting activity when assayed using Raji cells (Ali *et al.*, 2000) and significantly reduced the triglyceride serum level in hyperlipidemic rats (Hardhani and Suhardjono, 2008). Studies on fruit extract revealed that the fruit diet decreases blood glucose level, total cholesterol, triglycerides, cholesterol LDL and increases cholesterol HDL level of wistar mice (Ariviani, 2012).

2.3.6 Past Studies

The overview of past studies on the chemical compositions and bioactivities of *Syzygium polyanthum* is shown in Table 2.3.