EFFECT OF LEAF POSITION ON PHYTOCHEMICAL CONTENT, ANTIOXIDANT AND ANTIMICROBIAL ACTIVITIES OF SENDUDUK (*Melastoma malabathricum*)

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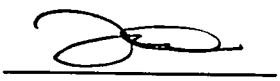

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ABSTRACT

Melastoma malabathricum (*M. malabathricum*) is a shrub that have been used in herbal remedies and folk medicine to treat human ailments. The local name of this medicinal herb is *Senduduk*. An experiment on this herb was conducted at the Faculty of Sustainable Agriculture Laboratory in University Malaysia Sabah, Sandakan, Sabah. The objective of the study was to study the effect of leaf position on phytochemical content (flavonoid, phenolic, alkaloid, tannin, saponin, glycoside and terpene), antioxidant and antimicrobial activities of *M. malabathricum*. Results were analysed using SPSS and treatment means were compared by using Tukey's test at 5% significance level. In this study, the phytochemical contents (flavonoid and phenolic), antioxidant activity and antimicrobial activity were found to be the highest in upper leaves (young leaves) compared to middle leaves and basal leaves (old leaves). The phenolic and flavonoid contents in upper leaves were 1619.19 ± 304.45 mg GAE/g and 1162.66 ± 34.70 mg QE/g respectively. The highest scavenging activity was shown in the extract from the upper leaves, with the IC_{50} value of 409.37 ± 74.52 µg/mL. The extracts were tested against *Escherichia coli*, *Bacillus cereus*, *Salmonella typhi* and *Staphylococcus aureus* and showed inhibition against these bacteria. The minimum concentration of upper leaves extract (0.039 mg/disc) was found to be against *Escherichia coli*. Therefore, the upper leaves are suggested to be best part to be consumed due to the high pharmaceutical quality.

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KESAN POSISI DAUN KE ATAS KANDUNGAN FITOKIMIA, AKTIVITI ANTIOKSIDAN DAN ANTIMIKROB POKOK SENDUDUK (Melastoma malabathricum)

ABSTRAK

Melastoma malabathricum (M. malabathricum) adalah herba perubatan yang telah digunakan dalam perubatan tradisional untuk merawat beberapa penyakit manusia. Nama tempatan herba perubatan ini adalah Senduduk. Satu uji kaji pada herba ini telah dijalankan di Makmal Fakulti Pertanian Lestari di Universiti Malaysia Sabah, Sandakan, Sabah. Objektif kajian ini adalah untuk mengkaji kesan posisi daun ke atas kandungan fitokimia (flavonoid, fenolik, alkaloid, tanin, saponin, glikosida dan terpene), aktiviti antioksidan dan antimikrob M. malabathricum. Keputusan dianalisis dengan menggunakan SPSS dan purata rawatan dibandingkan dengan menggunakan ujian Tukey's pada aras 5%. Dalam kajian ini, kandungan fitokimia (flavonoid dan fenolik), aktiviti antioksidan dan aktiviti antimikrob didapati tertinggi dalam daun atas (daun muda) berbanding dengan daun pertengahan dan daun basal (daun tua). Kandungan fenolik dan flavonoid dalam daun atas adalah 1619.19 ± 304.45 mg GAE/g dan 1162.66 ± 34.70 mg QE/g. Aktiviti antioksidan adalah tertinggi dalam ekstrak dari daun atas dan IC_{50} nilai adalah 493.01 ± 405.14 ug/mL. Ekstrak telah diuji terhadap Escherichia coli, Bacillus cereus, Salmonella typhi dan Staphylococcus aureus dan menunjukkan perencatan terhadap semua bakteria tersebut. Kepekatan minimum ekstrak dari daun atas (0.039 mg/disk) adalah terhadap Escherichia coli. Oleh itu, daun atas telah dicadangkan sebagai bahagian pokok yang terbaik untuk dimakan disebabkan oleh kualiti farmaseutikal yang tinggi.

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LIST OF SYMBOLS, UNITS AND ABBREVIATIONS

%	Percentage
°C	Celsius
µg/disc	Microgram per disc
µg/mL	Microgram per millilitre
µg/mL	Microgram per millilitre
µL	Microliter
µL/disc	Microliter
ANOVA	Analysis of variance
cm	Centimetre
cm^2	Centimetre square
CVD	Cardiovascular disease
g	Gram
GA/g	Gallic acid per gram
IC_{50}	Inhibit Cellular Proliferation by 50%
L	Litre
M	Mole
mg	Milligram
mg/disc	Milligram per disc
mg/mL	Milligram per millilitre
MIC	Minimum Inhibitory Concentration
mL	Millilitre
mm	Millimetre
mol/L	Mole per Litre
nm	Nautical mile
QE/g	Quercetin per gram
SD	Standard Deviation
SPSS	Statistical Package for the Social Sciences
TFC	Total flavonoid content
TPC	Total phenolic content
UMS	Universiti Malaysia Sabah
UV	Ultraviolet
w/v	Weight per volume
WHO	World Health Organization

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CHAPTER 1

INTRODUCTION

1.1 Introduction

According to World Health Organization (WHO), almost 80% of people relies on and utilize traditional medicines for their primary health care in the developing countries and also found out that 85% of such medicines in these regions are derived from plant extracts (Omar *et al.*, 2012). *Melastoma malabathricum* (*M. malabathricum*) or locally known as "Senduduk", is a shrub that belongs to Melastomataceae family which can be easily found in cleared land, waste places, and roadside throughout the Tropical Asia (Karupiah and Ismail, 2013). The genus Melastoma comprises twenty-two species, two subspecies and three varieties in Southeast Asia region including Malaysia and they are classified by the colour of the flower petals– light pink magenta, dark purple magenta and rare variety of white petals (Rajenderan, 2010).

Different parts of *M. malabathricum* such as leaves, flowers, shoots and roots have been used in herbal remedies and folk medicines to treat human ailments and diseases. Generally, leaves are more commonly used by Malay population to treat cuts and wounds, post-natal care, prevent scarring from small pox infections, diarrhea, and dysentery (Sulaiman *et al.*, 2004). Besides of the leaf, root and flower are used to treat toothache, anti-infection, and post-partum recovery (Jofrry *et al.*, 2012). This plant has been reported to possess antioxidant and anti-cancer, anti-viral, anti-inflammatory, anti-nociceptive, anti-pyretic, anti-ulcerogenic and hypotensive effects (Rajenderan, 2010). Besides, Omar *et al.* (2012) has revealed that the crude flower and fruit extracts of *M. malabathricum* exhibit anti-microbial properties that can treat infectious diseases caused by microorganisms.

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Phytochemicals are a large group of plant-derived compounds that responsible for disease protection. Common examples of phytochemicals are flavonoid, phenolic acid, and alkaloid (Lindsay and Astley, 2002). The levels of secondary metabolites such as phenolics and flavonoids in leaves may be affected by the differences in developmental ages of the leaves present at different positions, genetic, and agronomic factors (Bhakta and Ganjewala, 2009). Researchers believe that phenolic and flavonoid compounds are able to contribute towards antioxidant activity (Bixby *et al.*, 2005). Antioxidants can be defined as substances that help fight the harmful effects of unstable molecules in the body called free radicals.

Plants that rich in natural antimicrobial agents are more efficient to treat infectious disease than synthetic antibiotics (Mathur *et al.*, 2011). Scientists also believe that tropical plants have possibility of developing new and natural drugs as the metabolites discovered in these plants may useful against the new disease that continuously emerging as well as avoiding the side effect of synthetic drugs (Choudhury *et al.*, 2011). For instance, Veerapur *et al.* (2004) found out that *M. malabathricum* produces bioactive molecules which inhibit and control microbial colonization and proliferation. The antimicrobial properties are due to compounds synthesized in the secondary metabolism as well as antioxidant activity of the plants (Moyo *et al.*, 2012).

Suzuki and Waller (2006) have proved that the upper leaves (young leaves) contained high level of phytochemical contents compared to basal leaves (older leaves) and these findings are also in line with Achakzai *et al.* (2009) that reported level of flavonoids increased in premature leaves. Li *et al.* (2013) also reported the total flavonoid content in younger leaves of *Cinnamomum cassia* is doubled those of the older leaves. Besides, the leaf position affected the antioxidant activity in plants as well. The premature leaves of *Lantana camara* have exhibited greater antioxidant activity compared to that in older leaves as accumulation of secondary metabolites is higher in young leaves (Bhakta and Ganjewala, 2009). Akyon (2002) believes that high secondary metabolites and antioxidant activity will directly affect the antimicrobial activity in the plants as they inhibit bacteria to infections.

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1.2 Justification

This research was conducted to study the effect of leaf positions on phytochemical contents, antioxidant and antimicrobial activities of *M. malabathricum* leaves. *Melastoma malabathricum* was chosen for this study due to its high market value and high demand in current market as various parts of the plants are used to treat diseases and ailments and this herb is popular especially among Malay and Indian populations.

Phytochemicals are plant chemicals that have disease properties and thus deserves to draw more attentions from the public. Examples of plant chemicals with disease properties are flavonoid which functions against microbial attack and phenolic responsible for anti-inflammatory and antioxidant properties. Moyo *et al.* (2012) reported that these phytochemicals and antioxidant activity are correlated. Therefore, quantitative analysis and antioxidant activities analysis are carried out in order to determine the relationships of these properties.

Several studies have been done on effect of leaf position on other species of plants. However, there is limited and not much scientific study was carried out on *M. malabathricum*. For example, Do Thi and Hwang (2014) has claimed that young leaves of *Aronia melanocarpa* possess the good antioxidant and antimicrobial activity. Moreover, Raya *et al.* (2015) claimed that the leaf maturity and extraction solvents would probably affect the phytochemical contents and antioxidant properties of *Clinacanthus nutans* leaves. Therefore, it is necessary to find out the most suitable part to be harvested in order to obtain the highest pharmaceutical quality.

1.3 Objective

To study the effect of leaf position on the phytochemical contents, antioxidant and antimicrobial activities of Senduduk *(M. malabathricum)*.

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1.4 Hypothesis

H_o : There is no significant difference between the effect of leaf position on the phytochemical contents, antioxidant and antimicrobial activities of Senduduk *(M. malabathricum)*.

H_A : There is significant different between the effect of leaf position on the phytochemical contents, antioxidant and antimicrobial activities of Senduduk *(M. malabathricum)*.

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CHAPTER 2

LITERATURE REVIEW

2.1 *Melastoma malabathricum*

Melastoma malabathricum is a very common herb or shrub that mostly found in India, Thailand and Malaysia. It comes from family of Melastomataceae and the member of this family are largely herbs, shrubs and climbers. It is a well-known plant in Malaysian traditional medicine (Sunilson *et al.*, 2008). This plant has many common names including Senduduk (Malaysia), Straits Rhododendron (Singapore), Mang Kre (Thailand), Malabar melastome (Australia), Indian-rhododendron, Lutki (India), Keduduk, Senggani, Lingangadi (Murut) and Mang Kre (Thailand) (Alnajar *et al.*, 2012). *Melastoma malabathricum* is normally distributed in valleys near the streams up to an elevation of 1800m.

2.1.1 Botanical description of *M. malabathricum*

Melastoma malabathricum is a beautiful bushy and tall shrub that available throughout Malaysia and can grow up to a height of 1.5 - 5 meter or 4 – 16 feet, occasionally even up to 20 feet (Susanti *et al.*, 2007). The branches are four-angled or quadrangular in shape and twigs are densely covered with paleaceous hair. The bark is grey or brown in colour.

The leaves of the plant are broadly elliptic to lanceolate in shape. The size of leaves is generally 6 – 15 cm by 2 - 6.5 cm. The leaves are opposite and entire, generally having prominent longitudinal veins as shown in Figure 2.1 (b). The leaf blades of *M. malabathricum* are narrow and covered with rough hair on both surfaces. Besides of leaf blade, the petiole is also covered with flat hairs.

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Flowers are in colour bright mauve-purple and arranged in cluster at the ends of twigs Figure 2.1 (c). The flowers are commonly grow in 5 to 10 clusters and 5 petals for each clusters (Zakaria, 2007). The flower's size is mostly 15 – 35 mm by 10 – 22 mm. The petal is narrow at the base and ovoid at the top. Style is cylindrical and pinkish in colour. Pedicel is distinct, cylindrical and covered with flat hairs which similar to calyx that also covered with flat hairs.

Fruits are en-capsulated and contain numerous of non-endospermous and minute seeds with small embryos inside purplish pulps. The fruits are broadly ovoid and is 6.5 – 11.5 mm by 5 – 10.5 mm in size. The plant consists of fleshy capsule and the hypanthium becomes brittle, breaks and rupture transversely at maturity (Rajenderan, 2010).

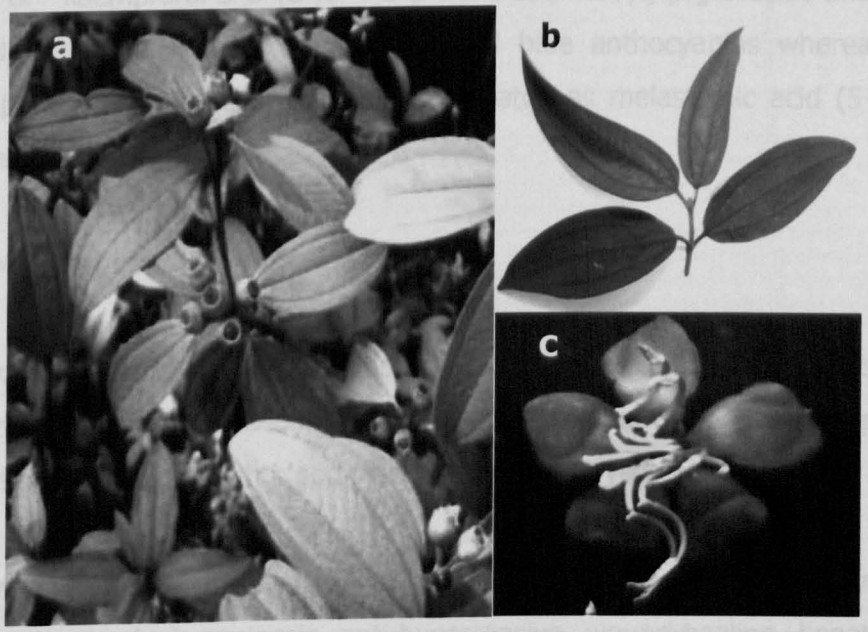

Figure 2.1 Morphology of plant. Growth habit (a); Leaves (b); Flowers (c)
Source: Janna *et al.*, 2006

2.1.2 Chemical constituents of *M. malabatricum*

Generally, *M. malabathricum* consists of various phytochemicals and most commonly found in the leaves are flavonoids, triterpenes (ursolic acid, 2-hydroxyursolic acid and Asiatic acid, glycerol-1,2-dilinolenyl-3-O-β-D-galactopyranoside and glycerol 1,2-dilinolenyl-3-O-(4,6-di-O-isopropylidene)-β-D-galactopyranoside), saponins, tannin (nobotanin B, dimers named malabathrins B, malabathrins C, and malabathrins D and monomers named 1,4,6-tri-O-galloyl-β-D-glucoside, 1,2,4,6-tetra-Ogalloyl-β-D-

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glucoside, strictinin, casuarictin, pedunculagin, nobotanin D, pterocarininand, nobotanin G, nobotanin H and nobotanin J) and steroids (Yoshida *et. al.,* 1992; Zakaria, 2007; Rajenderan, 2010).

Others including ellagic acid, cyanidin-3- glucoside, cyanidin-3,5-diglucoside, malvidin-3,5-diglucoside, *β*-sitosterol, ursolic acid, 2-hydroxyursolic acid, gallic acid, kaempferol, kaempferol-3-*O*-*α*-L-rhamnopyranoside, kaempferol-3-*O*- (2",6"-di-*O*-*p*-*trans*-coumaroyl), malabathrin B, malabathrin C, malabathrin D, strictinin, *α*- amyrin, quercetin, quercitrin and rutin (Susanti *et al.*, 2007).

Furthermore, the other parts of this plant also contains biochemical constituents such as the flowers and roots. According to Rajenderan (2010), flowers consists of kaempferol-3-O-(2",6"-i-O-*p*-*trans*-coumaroyl)-β-glucoside and kaempferol -3-OD-glucoside as well as orange, red and blue anthocyanins whereas the roots yielded β-sitosterol and a triterpenoid designated as melastomic acid (5-hydroxylup-20(29)-en-28-oic acid).

2.1.3 Medical uses of *M. malabathricum*

Melastoma malabathricum are traditionally used to heal cuts and wounds, treatment for toothache, sore, diarrhoea, stomach-ache, diabetes, piles, post-partum and haemorrhoids, lowering high blood pressure as well as face wash (Susanti *et al.*, 2007).

In scientifically researches and investigations, *M. malabathricum* claimed to possess anti-microbial, analgesic, anti-hypertensive, wound-healing, hepatoprotective, anti-nociceptive, anti-diarrhoeal, anti-bacterial, anti-fungal, anti-nociceptive, anti-inflammatory, anti-pyretic, anti-lipid peroxidation, anti-oxidant and anti-cancer activity as well as having anti-helminthic and anti-spasmodic actions (Sulaiman *et al.*, 2004; Zakaria *et al.*, 2006; Susanti *et al.*, 2007).

The most recent research on *M. malabathricum* revealed that its bioactive constituents exhibited free radical scavenging activity and anti-inflammatory effects on mouse ear edema, gastro-protective effects against ethanol-induced gastric ulcers in rats (Hussain *et al.*, 2008). Furthermore, various findings have shown positive

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therapeutic activity such as anti-coagulant activity, antiviral and cytotoxic effect against murine cell lines (Lohezic-Le Devehat *et al.*, 2002).

2.1.4 Pharmacological study in *M. malabathricum*

a. Anti-inflammatory activity

In Mazura *et al.* (2007) findings, they claimed that some of the natural compounds are useful against anti-inflammatory activity including *α*.-amyrin, betulinic acid, and isolated flavonoids. These compounds were assessed in vitro by determining their inhibitory effects on platelet activating factor (PAF) binding to rabbit platelets using ^{3}H-PAF as a ligand and the results showed that natural compounds from *M. malabathricum* possess selective antagonistic activity toward PAF and could be a potential natural anti-inflammatory compound which in line with Zakaria *et al.* (2006) study.

b. Anti-pyretic activity

Antipyretic properties of the aqueous extract of *M. malabathricum* leaves in experimental animals was studied by Zakaria *et al.* (2006) and the antipyretic activities were measured using brewer's yeast-induced pyrexia tests. The extract was found to exhibit antipyretic activities at all concentration tested (10%, 50% and 100%).

c. Anti-proliferative activity

In vitro anti-proliferative activities of various extracts from the leaves of *M. malabathricum* was determined using various established in vitro assays. Two different extracts were used to test for the anti-proliferative activity against various cell lines. The chloroform extract exhibited anti-proliferative activity against the Caov-3, HL-60, and CEM-SS cell lines while the methanol extract demonstrated anti-proliferative activity against more cell lines, including the MCF-7, HeLa, Caov-3, HL-60, CEM-SS, and MDA-MB-231 cancer cell lines (Zakaria *et al.*, 2011).

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d. Antimicrobial activity

Leaves of *M. malabathricum* have been tested against *Stapylococcus aureus* by disc diffusion method. The results showed that *M. malabathricum* extract inhibited the different clinical wound isolates of this bacteria (Sunilson *et al.*, 2008). Meanwhile, the result in the study of Choudhury *et al.* (2011) also showed significant zone of inhibition against three test bacteria including *Staphylococcus aureus*, *Streptococcus sp.* and *Escherichia coli*, which reflects the potentiality of *M. malabathricum* leaves as antimicrobial agent. Moreover, various species that belonging to Melastomataceae family showed significant antimicrobial activity against different Gram positive and Gram negative bacteria (Hullatti and Ravishankar, 2004).

e. Antioxidant activity

Previous study on antioxidant activity of *M. malabathricum* was carried out to evaluate the activity in different parts of plants using DPPH scavenging activity method. The result showed flower extract have the highest antioxidant activity with lowest concentration to achieve IC_{50}. In contrast, the stem showed lowest antioxidant activity among the plant parts (Suleiman *et al.*, 2015). Besides, in study of Susanti *et al.* (2007), antioxidant activity of *M. malabathricum* also being tested by DPPH radical scavenging method as well.

2.2 Phytochemicals

Phytochemicals can be defined as any compound found in plants. Phytochemicals are certain non-nutritive plant chemicals which have some disease preventive properties. Phytochemicals provide plants with colour, flavour and natural protection against pests (Chedes, 2013).

2.2.1 Flavonoid

Flavonoids are hydroxylated phenolic compounds and are produced as a result of microbial attack. This may be due to ability of flavonoids to form complexes with soluble proteins and extracellular material. These metabolites are also able to disrupt cell wall and cell membrane of microorganism (Harborne and Williams, 2000).

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Flavonoids are often synthesized at cytosols and the main precursors for the production of flavonoids are phenylalanine and malonyl-CoA (Cushine and Lamb, 2005).

Flavonoid also shields the plant from UV radiations, biotic stress and abiotic stress. It is able to withstand harsh environment such as high salinity and draught in order to protect the plant. According to Rathee *et al.* (2009), flavonoids are phenolic compounds that are synthesised universally in medicinal plants and can induce antibacterial response due to the presence of carbonyl group. Flavonoids that lacks hydroxyl groups on their beta rings are known to be more active in antimicrobial activity than those with hydroxyl groups in their structures (Maria *et al.*, 2012).

2.2.2 Phenolic

Phenolic acids are a type of aromatic acid compound. They are also a type of phytochemical called polyphenol. They comprise of large group of plant biologically active ingredients such as tannins, phenolic acids, lignans, lignins and stilbenes (Ho *et al.*, 2012). They are plant metabolites that are widely spread throughout the plant kingdom. There are many different phenolic acids found in nature and they can be divided into two categories which are benzoic acid derivatives, such as gallic acid and cinnamic acid derivatives, including caffeic acid and ferulic acid.

Presence or occurrences of phenolic compounds in medicinal plants indicates that these plants might be responsible for various biological activities including anti-microbial, anti-inflammatory, anti-clotting, anti-carcinogenic, anti-hypertensive and antioxidant agents. This can be proven by Okwu (2001) that phenolic compounds have been used in disinfections and disease preventives. Besides, phenols are also responsible in blocking specific enzymes that causes inflammation (Okwu, 2005).

2.2.3 Alkaloid

Alkaloids are one of the most diverse group of secondary metabolites of a class of naturally occurring organic nitrogen-containing bases. Oxygen and sulphur may present apart from the carbon, hydrogen and nitrogen compounds in certain alkaloid structure. At a rare case, it might carry elements such as bromine, chorine and

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phosphorus (Akira *et al.*, 2011). Alkaloids such as physostigmine, galantamine have been scientifically reported to exert an in vitro anti-acetylcholinesterase activity.

Alkaloids are low molecular weight and are often colourless if they contain oxygen in their structure. Most alkaloids has a bitter taste and poisonous upon consumption. Higher plants are known to consist around 10-25% of alkaloids in them (Tabasum *et al.*, 2016). They are very important in medicine because of their strong biological activities such as antimalarial and anticancer. Furthermore, many alkaloids have physiological effects against multiple diseases such as cardiac dysfunction and diabetics (Singh *et al.*, 2006).

2.2.4 Saponin

In early research, saponin have been shown to possess both beneficial and deleterious properties (Price *et al.,* 1987). The beneficial properties are lowering the cholesterol level whereas deleterious effects are permeabilization of intestine and possess cytotoxic activity. Saponin in plants functioned as a performed defense against attack by their enemies. Therefore, the aforementioned plant species might use their saponins as defense mechanisms against predation by microorganisms, insects and herbivores.

Recently, saponins have been found to be used in veterinary vaccines as adjuvant helping to enhance immune response. Saponins have relationship with oxytocin which is the sex hormones in controlling the onset of labour pain in women and the subsequent release of milk (Rajput *et al.*, 2007).

2.2.5 Glycoside

A glycoside is a molecule within which a sugar is absolute to another functional group through a glycoside bond. Glycosides are important and vital to living organisms including human being. Roles of glycosides include repairing incised wounds and stimulating collagen in human skin fibroblast cells (Rajenderan, 2010). There are several plants that store inactive glycosides and only may be activated as the sugar half broken off and some researchers believed the wound healing property may probably be due to the presence of the glycosides (Sunilson *et al.*, 2008).

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REFERENCES

Achakzai, A. K. K., Achakzai, P., Masood, A., Kayani, S. A. and Tareen, R. B. 2009. Response of Plant Parts and Age on the Distribution of Secondary Metabolites on Plants Found in Quetta. *Journal of Botanical* Science **41(5)**: 2129-2135 http://www.pakbs.org/pjbot/PDFs/41(5)/PJB41(5)2129

Agati, G., Brunetti, C., Di-Ferdinando, M., Ferrini, F., Pollastri, S. and Tattini, M. 2013. Functional Roles of Flavonoids in Photoprotection: New Evidence, Lessons from the Past. *Plant Physiology and Biochemistry* **72**: 35–45 http://www.sciencedirect.com/science/article/pii/S0981942813001022

Akira, N., Hiromichi, M. and Hidehiko, K. 2011. A Bacterial Platform for Fermentative Production of Plant Alkaloids. *Nature Communications* **2(1)**: 326-329 http://www.nature.com/articles/ncomms1327

Akiyama, H., Fujii, K., Yamasaki, O., Oono, T. and Iwatsuki, K. 2001. Antibacterial Action of Several Tannins against *Staphylococcus aureus*. *Journal of Antimirobial Chemotherapy* **48**: 487-491 http://jac.oxfordjournals.org/content/48/4/487.long

Akyon, Y. 2002. Effect of Antioxidants on the Immune Response of *Helicobacter pylori*. *Clinical Microbiology and Infection* **8**: 438–441 http://www.sciencedirect.com/science/article/pii/S1198743X14632572

Alnajar, Z. A. A., Abdulla, M. A., Ali, H. M., Alshawash, M. A. and Hadi, A. H. A. 2012. Acute Toxicity Evaluation, Antibacterial, Antioxidant and Immunomodulatory Effects of *Melastoma malabathricum*. *Molecules* **17**: 3547-3559 https://umexpert.um.edu.my/file/publication/00013091_115784

Alwash, M. S. A., Ibrahim, N., Yaacob, W. A. and Din, L. B. 2014. Antibacterial, Antioxidant and Cytotoxicity Properties of Traditionally Used *Melastoma malabathricum* linn Leaves. *Journal of Food Science Technology* **6(1)**: 6–12 http://www.maxwellsci.com/print/ajfst/v6-6-12

Anesini, C., Turner, S., Cogoi, L. and Filip, R. 2012. Study of Participation of Caffeine and Polyphenol on the Overall Antioxidant Activity of Mate (*Ilex paraguariensis*). *Food Science* Technology **45**: 299-304 http://ac.els-cdn.com/S0023643811001988/1-s2.0-S0023643811001988

Anna, S., Cezary, P. and Max, H. 2011. Influence of Environmental Abiotic Factors on the Content of Saponins in Plants. *Phytochemistry Reviews* **10(4)**: 471-491 http://link.springer.com/article/10.1007/s11101-010-9177

Ashfaq, M., Shah, K. W., Showket, A. and Dharmendra, S. 2012. Preliminary Phytochemical Screening and Aqueous Extracts of *Mentha Longifolia* Linn. Leaves. *International Journal of Biological and Pharmaceutical Research* **3(3)**: 384-386 http://www.ijbpr.com/cadmin/article/147_384-386

Bamishaive, E. L., Olayemi, F. F., Awagu, E. F. and Bamshaiye, O. M. 2011. Proximate and Phytochemcial Composition of *Moringa oleifera* Leaves at Three Stages of Maturation. *Journal of Food Science and Technology* **3(4)**: 233-237 http://maxwellsci.com/print/ajfst/v3-233-237

Bassani, D. C., Nunes, D. S. and Granato, D., 2014. Optimization of Phenolics and Flavonoids Extraction Conditions and Antioxidant Activity of Roasted Yerba-mate Leaves (*Ilex paraguariensis*) Using Response Surface Methodology. *Anais Da Academia Brasileira De Ciencias* **86**: 923–934 http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0001-37652014000200923

Bhakta, D. and Ganjewala, D. 2009. Effect of Leaf Position on Total Phenolics, Flavonoids and Proanthocyanidins Content and Antioxidant Activities in *Lantana Camara* L. *Journal of Scientific Research* **1(2)**: 363-369

UMS
UNIVERSITI MALAYSIA SABAH

http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.657.4454

Bixby, M., Spieler, L., Menini, T. and Gugliucci, A. 2005. *Ilex paraguariensis* Extracts are Potent Inhibitors of Nitrosative Stress: A Comparative Study with Green Tea and Wines Using a Protein Nitration Model and Mammalian Cell Cytotoxicity. *Life Science* **77**: 345–358
http://ac.els-cdn.com/S0024320505001505/1-s2.0-S0024320505001505

Blum-Silva, C. H., Chaves, V. C., Schenkel, E. P., Coelho, G. C. and Reginatto, F. H. 2015. The Influence of Leaf Age on Methylxanthines, Total Phenolic Content, and Free Radical Scavenging Capacity of *Ilex paraguariensis* Aqueous Extracts. *Brazilian Journal of Pharmacognosy* **25**: 1-6
http://www.sciencedirect.com/science/article/pii/S0102695X15000332

Boo, H. O., Heo, B. G., Gorinstein, S. and Chon, S. U. 2011. Positive Effects of Temperature and Growth Conditions on Enzymatic and Antioxidant Status in Lettuce Plants. *Plant Science* **181**: 479–484
http://www.sciencedirect.com/science/article/pii/S0168945211002159

Bottone, E. J. 2010. *Bacillus cereus*, A Volatile Human Pathogen. *Clinical Microbiology Reviews* **23(2)**: 382-398
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2863360

Busani, M., Masika, Julius, P. and Muchenje, V. 2012. Antimicrobial Activities of *Moringa oleifera* Lam Leaf Extracts. *African Journal of Biotechnology* **11(11)**: 2797-2802
http://www.academicjournals.org/article/article1380818242_Moyo%20et%20al

Cecilia, R. J. R., Lyle, E. C., Ma, N. R. M. and Juan, A. A. C. 2011. Humic Substances and Moisture Content in the Production of Biomass and Bioactive Constituents of *Thymus vulgaris* L. *Reviews Phytotechnology* **34(3)**: 183-188
http://www.revistafitotecniamexicana.org/documentos/34-3/5a

Celotto, A. C., Nazario, D. Z., Spessoto, M. D. A., Martins, C. H. G. and Cunha, W. R. 2003. Evolution of the In Vitro Antimicrobial Activity of Crude Extracts of Three Miconia species. *Brazilian Journal of Microbiology* **34(4)**: 339-340
http://www.scielo.br/pdf/bjm/v34n4/v34n4a10

Chedes, P. S. 2013. Phytochemical Analysis of *Citrus sinensis* Peel. *International Journal of Pharmacology and Biology Science* **4(1)**: 339-343
http://impactfactor.org/IJPPR/4/IJPPR,Vol4,Issue4,Article11

Choudhury, M. D., Nath, D. and Talukdar, A. D. 2011. Antimicrobial Activity of *Melastoma malabathricum* L. *Journal of Science and Technology* **7**: 76–78
http://www.inflibnet.ac.in/ojs/index.php/AUJSAT/article/viewFile/506/477

Chua, L. Y. P., King, P. J. H., Ong, K. H., Sarbini, S. R. and Yiu, P. H. 2015. Influence of Light Intensity and Temperature on Antioxidant Activity in *Premna serratifolia* L. *Journal of Soil Science and Plant Nutrition* **15(3)**: 605-614
http://www.scielo.cl/scielo.php?script=sci_arttext&pid=S0718-95162015000300005

Cremer, D. R. and Eichner, K. 2000. Formation of Volatile Compounds during Heating of Spice Paprika (*Capsicum annuum*) Powder. *Journal of Agriculture and Food Chemistry* **48**: 2454-2460
http://pubs.acs.org/doi/pdf/10.1021/jf991375a

Cushine, T. P. T. and Lamb, A. J. 2005. Antimicrobial Activity of Flavonoids. *International Journal of Antimicrobial Agents* **26(5)**: 343-356
http://www.idpublications.com/journals/PDFs/IJAA/ANTAGE_MostCited_1

Dalila, Z. D., Hafsah, J. and Manaf, A. A. 2015. Antioxidant Properties and Total Phenolic Content in Different Development Stages of *Barringtonis racemosa* and *Barringtonia spicata* Leaves. *Walailak Journal of Science and Technology* **12(5)**: 449-458

UMS
UNIVERSITI MALAYSIA SABAH

http://wjst.wu.ac.th/index.php/wjst/article/view/867/481

Dartora, N., De Souza, L. M., Santana, A. P., Iacomini, M., Valduga, A. T., Gorin, P. A. J. and Sassaki, G. L. 2011. Evaluation of Bioactive Compounds from Leaves of *Ilex paraguariensis* with Different Growth Conditions, Treatments and Ageing. *Food Chemistry* **129**: 1453–1461
http://www.sciencedirect.com/science/article/pii/S0308814611008156

Do Thi, N. and Hwang, E. S. 2014. Bioactive Compounds Content and Antioxidant Activities in Aronia (*Aronia melanocarpa*) Leaves Collected at Different Growth Stages. *Preview Nutrition Food Science* **19(3)**: 204-212
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4195626/

Elena, V., Juha-Pekka, S. I., Julia, K. and Lalevi, P. I. J. 2004. Changes in Leaf Trichimes and Epicuticular Flavonoids during Leaf Development in Three Birch Taxa. *Annals of Botany* **94**: 233-242
http://aob.oxfordjournals.org/content/94/2/233

Esmelindro, A. A., Girardi, J. D., Mossi, A., Jacques, R. A. and Dariva, C., 2004. Influence of Agronomic Variables on the Composition of Mate Tea Leaves (*Ilex paraguariensis*) Extracts Obtained from CO_2 Extraction at 30°C. *Journal of Agriculture and Food Chemistry* **52**: 1990–1995
http://pubs.acs.org/doi/pdf/10.1021/jf035143u

Federica, M., Rosa, T., Marco, B., Monica, R. L., Filomena, C., Gianearlo, S., Bruno, D. C., Peter, J. H. and Francesco, M. 2009. The Influence of Fruit Ripening in the Phytochemical Content and Biological Activity of *Capsicum chinense*. *Food Chemistry* **114**: 553-560
http://www.sciencedirect.com/science/article/pii/S0308814608011771

Firdouses, S. and Alam, P. 2011. Phytochemical Investigation of Extract of *Amorphophallus campanulatus* Tubers. *International of Phytomedicine* **3**: 32-35
http://www.arjournals.org/index.php/ijpm/article/view/273/270

Ghasemzadeh, A., Nasiri, A., Jaafar, H. Z. E., Baghdadi, A. and Ahmed, I. 2014. Changes in Phytochemical Synthesis, Chalcone Synthase Activity and Pharmaceutical Qualities of Sabah Snake Grass in Relation to Plant Age. *Journal of Molecular* **19**: 17632-17648
http://www.mdpi.com/1420-3049/19/11/17632/htm

Ghosal, S., Krishna, P. B. N. and Lakshmi, V. 1996. Anti-amoebic Activity of *Piper longum* Fruits against *Entamoeba histolytica* In Vitro and In Vivo. *Journal of Ethnopharmacology* **50**: 167–170
http://ac.els-cdn.com/0378874196013827/1-s2.0-0378874196013827

Gonçalves, J. L. S., Lopes, R. C., Oliveira, D. B., Costa, S. S., Miranda, M. M. F. S., Romanos, M. T. V., Santos, N. S. O. and Wigg, M. D. 2005. In Vitro Anti-rotavirus Activity of Some Medicinal Plants Used in Brazil against Diarrhea. *Journal of Ethnopharmacology* **99**:403–407
http://www.sciencedirect.com/science/article/pii/S0378874105000814

Grujic, N., Lepojevic, Z., Srdjenovic, B., Vladic, J. and Sudji, J., 2012. Effects of Different Extraction Methods and Conditions on the Phenolic Composition of Mate Tea Extracts. *Molecules* **17**: 2518–2528
file:///C:/Users/cheong/Downloads/molecules-17-02518

Harborne, J. B. and Williams, C. A. 2000. Advances in Flavonoids Research since 1992. *Phytochemistry* **55**: 481–504
http://www.sciencedirect.com/science/article/pii/S0031942200002351

Harriet, U. and Nandita, D. 2014. Mechanisms of Antibiotic Resistance in *Salmonella typhi*. *International Journal of Current Microbiology and Applied Science* **3(12)**: 462-476
http://www.ijcmas.com/vol-3 12/Harriet%20Ugboko%20and%20Nandita%20De

UMS
UNIVERSITI MALAYSIA SABAH

Harris, L. G., Foster, S. J. and Richards, R. G. 2002. An Introduction to *Staphylococcus aureus* and Techniques for Identifying and Quantifying *S.aureus* Adhesins in Relation to Adhesion to Biomaterials: Review. *European Cells and Materials* **4**: 39-60
http://www.ecmjournal.org/journal/papers/vol004/pdf/v004a04

Hartmann, T. 1996. Diversity and Variability of Plant Secondary Metabolism: A Mechanistic View. *Entomology Experiment Applied* **80**: 177-188
http://onlinelibrary.wiley.com/doi/10.1111/j.1570-7458.1996.tb00914

Hemm, M. R., Rider, S. D., Ogas, J., Murry, D. J. and Chapple, C. 2004. Light Induces Phenylpropanoid Metabolism in *Arabidopsis* roots. *Journal of Plant* **38**: 765–778
http://onlinelibrary.wiley.com/doi/10.1111/j.1365-313X.2004.02089

Ho, Y. L., Huang, S. S., Deng, J. S., Lin, Y. H., Chang, Y. S. and Haung, G. J. 2012. In-vitro Antioxidant Properties and Total Phenolic Contents of Wetland Medicinal Plants in Taiwan. *Botanical Studies* **53**: 55-66
http://ejournal.sinica.edu.tw/bbas/content/2012/1/Bot531-06

Hong, Y., Lin, S., Jiang, Y. and Ashraf, M. 2008. Variation in Contents of Total Phenolics and Flavonoids and Antioxidant Activities in the Leaves of 11 Eriobotrya Species. *Plant food Human Nutrition* **63**: 200–204
https://www.ncbi.nlm.nih.gov/pubmed/18726693

Hullatti, K. K. and Ravishankar, R. 2004. Antimicrobial Activity of *Memecylon malabaricum* Leaves. *Fitoterapia* **75(4)**: 409-411
http://www.klepharm.edu/media/uploads/publications/2004_mem_malb_1

Hussain, F., Abdulla, M., Noor, S., Ismail, S. and Ali, H. 2008. Gastroprotective Effects of *Melastoma malabathricum* Aqueous Leaf Extract against Ethanol-induced Gastric Ulcer in Rats. *Journal of Biochemistry and Biotechnology* **4**: 438–441
http://thescipub.com/PDF/ajbbsp.2008.438.441

Islam, M. S., Yoshimoto, M., Ishiguro, K., Okuno, S. and Yamakawa, O. 2003. Effect of Artificial Shading and Temperature on Radical Scavenging Activity and Polyphenolic Composition in Sweetpotato (*Ipomoea batatas* L.) Leaves. *Journal of American Society for Horticultural Science* **128**: 182–187
http://journal.ashspublications.org/content/128/2/182

Janna, O. A., Khairul, A., Maziah, M. and Mohd, Y. 2006. Flower Pigment Analysis of *Melastoma malabathricum*. *African Journal of Biotechnology* **5(2)**: 170-174
https://tspace.library.utoronto.ca/bitstream/1807/6658/1/jb06027

Jofrry, S. M., Yob, N. J., Rofiee, M. S., Meor Mohd. Affandi, M. M. R., Suhaili, Z., Othman, F., Abdah, M. A., Desa, M. N. M. and Zakaria, Z. A. 2012. *Melastoma malabathricum* (L.) Smith Ethnomedicinal Uses, Chemical Constituents, and Pharmacological Properties: A Review. *Evidence- Based Complementary and Alternative Medicine* **48:** 258-306
https://www.hindawi.com/journals/ecam/2012/258434

Karupiah, S. and Ismail, Z. 2013. Antioxidative Effect of *Melastoma Malabathticum* L Extract and Determination of its Bioactive Flavonoids from Various Location in Malaysia by RP-HPLC with Diode Array Detection. *Journal of Applied Pharmaceutical Science* **3(2)**: 19-24
http://www.japsonline.com/admin/php/uploads/781

King, D., Fan, M. Z., Ejeta, G., Asem, E. K. and Adeola, O. 2000. The Effects of Tannins and Nutrient in White Pekin Duck. *Brazilian Poultry Science* **41(5)**: 630-639
http://www.tandfonline.com/doi/abs/10.1080/713654982

Koupai-Abyazani, M. R., McCallum, J., Muir, A. D., Bohm, B. A., Towers, G. and Gruber, M. Y. 1993. Developmental Changes in the Composition of Proanthocyanidins from Leaves of Sainfoin (*Onobrychis viciifolia* Scop.) as Determined by HPLC Analysis. *Journal of Agriculture and Food Chemistry* **41**: 1066-1070

UMS
UNIVERSITI MALAYSIA SABAH

http://pubs.acs.org/doi/abs/10.1021/jf00031a010?journalCode=jafcau

Lako, J., Trenerry, V. C., Wahlqvist, M., Wattanapenpaiboon, N., Sotheeswaraan, S. and Premier, R. 2007. Phytochemical Flavonols, Carotenoids and the Antioxidant Properties of a Wide Selection of Fijian fruit, Vegetables and Other Readily Available Foods. *Food Chemistry* **101**: 1727-41
http://www.sciencedirect.com/science/article/pii/S0308814606000872

Lee, W. C., Mahmud, R., Pillai, S., Perumal, S. and Ismail, S. 2012. Antioxidant Activities of Essential Oil of *Psidium guajava* L. Leaves. *Procedia* **2**: 86-91
http://www.sciencedirect.com/science/article/pii/S2212670812000875

Lepedus, H., Gaca, V., Viljevac, M., Kovac, S., Fulgosi, H., Simic, D., Jurkovic, V. and Cesar, V. 2011. Changes in Photosynthetic Performance and Antioxidative Strategies during Maturation of Norway Maple (*Acer platanoides* L.) Leaves. *Plant Physiology and Biochememistry* **49**: 368-376
http://ac.els-cdn.com/S0981942811000362/1-s2.0-S0981942811000362

Li, Q. and Kubota, C. 2009. Effects of Supplemental Light Quality on Growth and Phytochemical of Baby Leaf Lettuce. *Journal of Environmental and Experimental Botany* **67(1)**: 59-64
http://ac.els-cdn.com/S0098847209001348/1-s2.0-S0098847209001348

Li, Y., Kong, D., Huang, R., Liang, H., Xu, C. and Wu, H. 2013. Variations in Essential Oil Yields and Compositions of *Cinnamomum cassia* Leaves at Different Developmental Stages. *Industrial Crop Production* **47**: 92-101
http://www.sciencedirect.com/science/article/pii/S0926669013001118

Lin, Y. L., Juan, I. M., Chen, Y. L., Liang, Y. C. and Lin, J. K. 1996. Composition of Polyphenols in Fresh Tea Leaves and Associations of Their Oxygen-Radical-Absorbing Capacity with Anti-proliferative Actions in Fibroblast Cells. *Journal of Agriculture and Food Chemistry* **44**:1387–1394
http://pubs.acs.org/doi/abs/10.1021/jf950652k

Lindsay, D. G. and Astley, S. B. 2002. European Research on the Functional Effects of Dietary Antioxidants. *Molecular Aspects of Medicine* **23**: 1–38
http://www.sciencedirect.com/science/article/pii/S0098299702000055

Liu, W., Liu, J., Yin, D. and Zhao, X. 2015. Influence of Ecological Factors on the Production of Active Substances in the Anti-cancer Plant *Sinopodophyllum hexandrum* (Royle). *Journal of PLos One* **10(4)**: 1-22
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4398539

Liu, Y., Qian, C., Ding, S., Shang, X., Tang, W. and Fang, S. 2016. Effect of Light Regime and Provenance on Leaf Characteristics, Growth and Flavonoid Accumulation in *Cyclocarya paliurus*. *International Journal of Botanical Study* **57**: 28
https://as-botanicalstudies.springeropen.com/articles/10.1186/s40529-016-0145-7

Lohezic-Le Devehat, F., Bakhtiar, A., Bezivin, C., Amoros, M. and Boustie, J. 2002. Antiviral and Cytotoxic Activities of Some Indonesian Plants. *Fitoterapia* **73**: 400–405
http://www.sciencedirect.com/science/article/pii/S0367326X02001259

Madsen, H. L., Nielsen, B. R., Bertelsen, G. and Skibsted, L. H. 1996. Screening of Antioxidant Actvity of Spices: A comparison between Assays Based on ESR Spin Trapping and Elestrochemical Measurement of Oxygen Concumption. *Food Chemistry* **57**: 331-338
http://www.sciencedirect.com/science/article/pii/0308814695002480

Mamun, O. R., Amin, M. R., Chowdhury, M. M. H., Rahman, M. A. and Shamim, M. 2014. Comparative Study on the Antibacterial Activity of Four Medicinal Plants

UMS
UNIVERSITI MALAYSIA SABAH

Leaves of Different Ages. *International Journal of Pharmaceutical Science Invention* **3(3)**: 26-32
http://www.ijpsi.org/Papers/Vol3(3)/E033026032

Maria, L. F. F., Sebastian, P. R. and Paula, C. 2012. Flavonoids: Biosynthesis, Biological Functions and Biotechnological Applications. *Front Plant Science* **3**: 222-232
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3460232/

Marjorie, M. C. 1999. Plant Products as Antimicrobial Agents. *Clinical Microbiology Reviews* **12(4)**: 564-582
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC88925

Mathur, R., Singh, S., Yousuf, A., Bhardwaj, S. K., Verma, P., Babu, V., Gupta, G. B. K. S. and Prasad, V. K. 2011. Antifungal Activity of Some Plant Extracts against Clinical Pathogens. *Advanced Applied Science Research* **2**: 260-264
file:///C:/Users/cheong/Downloads/Advances%20in%20applied%20Science%20Research%20pHd

Mazura, M., Susanti, D. and Rasadah, M. 2007. Anti-inflammatory Action of Components from *Melastoma malabathricum*. *Pharmaceutical Biology* **45**: 372–375
http://www.tandfonline.com/doi/full/10.1080/13880200701214797?scroll=top&needAccess=true

Mboto, C. I., Eja, M. E., Adegoke, A. A., Iwatt, G. D., Asikong, B. E., Takon, I., Udo, S. M. and Akeh, M. 2009. Phytochemical Properties and Antimicrobial Activities of Combined Effect of Extracts of the Leaves of *Garcinia Kola*, *Vernonia amygdalina* and Honey on Some Medically Important Microorganisms. *African Journal Microbiology Resource* **3(9)**: 557-559
http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.842.349&rep=rep1&type

Mediani, A., Abas, F., Ping, T. C., Khatib, A. and Lajis, N. H. 2012. Influence of Growth Stage and Season on the Antioxidant Constituents of *Cosmos caudatus*. *Plant Foods Human Nutrition* **67(4)**: 344– 50
http://link.springer.com/article/10.1007/s11130-012-0317

Meyer, S., Cerovic, Z. G., Goulas, Y., Montpied, P., Demotes-Mainard, S., Bidel, L. P. R., Moya, I. and Dreyer, E. 2006. Relationships between Optically Assessed Polyphenols and Chlorophyll Contents, and Leaf Mass per Area Ratio in Woody Plants: A Signature of the Carbon–Nitrogen Balance within Leaves. *Plant Cell Environment* **29**: 1338–1348
http://onlinelibrary.wiley.com/doi/10.1111/j.1365-3040.2006.01514.x/full

Mondolot, L., Fisca, P. L., Buatois, B., Talansierm, E., Kochko, A. D. and Campa, C. 2006. Evolution in Caffeoylquinic Acid Content and Histolocalization during *Coffea canephora* Leaf Development. *Annals of Botany* **98(1)**: 33-40
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2803532/

Moyo, Busani, Masika, Patrick, J. and Muchenje, V. 2012. Antimicrobial Activities of *Moringa oleifera* Lam Leaf Extract. *African Journal of Biotechnology* **11(11)**: 2797-2802
http://www.academicjournals.org/article/article1380818242_Moyo%20et%20al

Mutalib, L. F. 2015. Effect of Growth Age Period on Biochemical Composition of *Plantago major* Plant. *International Journal of Current Resource Review* **7(19)**: 6-10
http://search.proquest.com/openview/e7f3f2ca772bf6b6fe857d6fd4d34775/1?pq-origsite=gscholar

Noriham, A., Dian-Nashiela, F., Kerni, H. B., Nooraain, H. and Azizah, A. H. 2015. Influences of Maturity Stages and Extraction Solvents on Antioxidant Activity of

PERPUSTAKAAN UNIVERSITI MALAYSIA SABAH

UMS
UNIVERSITI MALAYSIA SABAH

Cosmos Caudatus Leaves. *International Journal of Research Studies in Biosciences* **3(12)**: 1-10 https://www.researchgate.net/publication/284714633_Influences_of_Maturity_Stages_and_Extraction_Solvents_on_Antioxidant_Activity_of_Cosmos_Caudatus_Leaves

Okwu, D. E. 2001. Evaluation of the Chemical Composition of Indigenous Spices and Flavouring Agents. *Global Journal of Pure and Applied Science* **8**: 455-459 http://www.ajol.info/index.php/ajb/article/viewFile/137808/127371

Okwu, D. E. 2005. Phytochemicals, Vitamins and Mineral Contents of Two Nigerian Medicinal Plants. *International Journal of Molecular Medicine and Advance Sciences* **1(4)**: 375-381 http://docsdrive.com/pdfs/medwelljournals/ijmmas/2005/375-381

Omar, S. N. C., Abdullah, J. O., Khairoji, K. A., Chin, S. C. and Hamid, M. 2012. Potentials of *Melastoma malabathricum* Linn. Flower and Fruit Extracts as Antimicrobial Infusions. *American Journal of Plant Sciences* **3**: 1127-1134 http://file.scirp.org/pdf/AJPS20120800013_42553380

Ozgen, S. and Sekerci, S. 2011. Effect of Leaf Position on the Distribution of Phytochemicals and Antioxidant Capacity among Green and Red Lettuce Cultivars. *Spanish Journal of Agricultural Research* **9(3)**: 801-809 http://revistas.inia.es/index.php/sjar/article/view/2033/1540

Padda, M. S. and Picha, D. H. 2007. Antioxidant Activity and Phenolic Composition in "Beauregard" Sweetpotato are affected by Root Size and Leaf Age. *Journal of the American Society for Horticultural Science* **132(4)**: 447-451 http://journal.ashspublications.org/content/132/4/447.full

Paananen, A., Mikkola, R., Sareneva, T., Matikainen, S., Hess, M., Anderrson, M., Julkunen, I. and Salkinoja-Salonen, M. S. 2002. Inhibition of Human Natural Killer Cell Activity by Cereulide, An Emetic Toxin from *Bacillus cereus*. *Clinical and Experimental Immunology* **129**: 420-428 https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1906479

Pasko, P., Barto, H., Zagrodzki, P., Gorinstein, S., Fota, M. and Zachwieja, Z. 2009. Anthocyanins, Total Polyphenols and Antioxidant Activity in Amaranth and Quinoa Seeds and Sprouts during Their Growth. *Food Chemistry* **115**: 994–998 http://www.sciencedirect.com/science/article/pii/S0308814609000624

Prashant, T., Kumar, B., Kaur, M., Kaur, G. and Kaur, H. 2011. Phytochemical Screening and Extraction: A Review. *International Pharmaceutical Sciencia* **1(1)**: 98-106 https://www.scribd.com/doc/94036813/Phytochemical-Screening-and-Extraction-a-Review

Price, K. R., Johnson, I. T. and Fenwick, G. R. 1987. The Chemical and Biological Significance of Saponins in Foods and Feedstuffs. *Critical Review in Food Science and Nutrition* **26**: 27-135 https://www.ncbi.nlm.nih.gov/pubmed/3308321

Pui, C. F., Wong, W. C., Chai, L. C., Tunung, R., Jeyaletchumi, P., Noor Hidayah, M. S., Ubong, A., Farinazleen, M. G., Cheah, Y. K. and Son, R. 2011. Salmonella: A Foodborne Pathogen. *International Journal* of *Food Research* **18**: 465-473 http://psasir.upm.edu.my/24060/1/24060

Rajenderan, M. T. 2010. Ethno Medicinal Uses and Antimicrobial Properties of *Melastoma Malabathricum*. *Segi Review* **3(2)**: 34-44 https://www.segi.edu.my/onlinereview/chapters/vol3_sr2_art4

Rajput, Z. I., Hu, S. H., Xiao, C. W. and Arijio, A. G. 2007. Adjuvant Effects of Saponins on Animal Immune Responses. *Journal of Zhejiang University of Science Botany* **8(3)**: 153-161

UMS UNIVERSITI MALAYSIA SABAH

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC1810383

Rathee, P., Chaudhary, H., Rathee, S., Rathee, D., Kumar, V. and Kohli, K. 2009. Mechanism of Action of Flavonoids as Antiinflammatory Agents. *Inflammatory Allergy Drug Targets* **8(3)**: 229-35
http://www.eurekaselect.com/84604/article

Raya, K. B., Ahmad, S. H., Farhana, S. F., Mohammad, M., Tajidin, N. E. and Parvez, A. 2015. Changes in Phytochemical Contents in Different Parts of *Clinacanthus nutans* Lindau due to Storage Duration. *Bragantia* **74(4)**: 445-452
http://www.scielo.br/scielo.php?script=sci_arttext&pid=S0006-87052015000400445

Riaz, T., Abbasi, A., Aziz-Ur-Rehman, Shahzadi, T., Ajaib M. and Khan, K. M. 2012. Phytochemical Screening, Free Radical Scavenging, Antioxidant Activity and Phenolic Content of *Dodonaea Viscosa. Journal of the Serbian Chemical Society* **77(4)**: 423-435
http://www.doiserbia.nb.rs/img/doi/0352-5139/2012/0352-51391100183R

Rodrigues, J., Michelin, D. C., Rinaldo, D., Zocolo, G. J., Dos-Santos, L. C., Vilegas, W. and Salgado, H. R. 2008. Antimicrobial Activity of *Miconia* species (Melastomataceae). *Journal of Medicinal Food* **1(1)**: 120-126
http://repositorio.unesp.br/bitstream/handle/11449/8785/WOS000257000900017

Samiullah, Aslam, S., Saba, N., Taher, K., Raof, F., Alam, F., Khalid, S., Khan, N., Achakzai, A. K. K. and Athiq-ur-rehman. 2015. Qualitative and Quantitative Phytochemical Investigation of Leaves of *Narium indicum* Collected from Panjgur, Balochistan*Lasbela. Journal of Science Technology* **4**: 101-108
http://www.luawms.edu.pk/lujstvolume4/lujst412

Sang, G. L., Chang, S. C., Jun, G. L., Yoon, A. J., Hye, J. L., Won, B. C. and Yeong, C. U. 2013. Influence of Air Temperature on Yield and Phytochemical Content of Red Chicory and Garland Chrysanthemum Grown in Plant Factory. *Horticulture, Environment and Biotechnology* **54(5)**: 399-404
http://link.springer.com/article/10.1007/s13580-013-0095

Scalbert, A. 1991. Antimicrobial Properties of Tannins. *Phytochemistry* **30**: 3875–3883
http://www.sciencedirect.com/science/article/pii/003194229183426L

Sener, A. and Dulger, B. 2009. Antimicrobial Activity of the Leaves of Verbascum sinuatum L. on Microorganisms Isolated from Urinary Tract Infection. *African Journal of Microbiology Research* **3(11)**: 778-781
http://www.academicjournals.org/article/article1380371881_Sener%20and%20Dulger

Senguttuvan, J., Paulsamy, S. and Karthika, K. 2014. Phytochemical Analysis and Evaluation of Leaf and Root Parts of the Medicinal Herb, *Hypochaeris radicata* L. for In Vitro Antioxidant Activities. *Asian Pacific Journal of Tropical Biomedicine* **4(1)**: 359-367
https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4025295

Sharma, H. K. and Kumar, A. 2011. Evaluation of Total Phenol, Flavonoid and In Vitro Antioxidant Activity of Methanolic Extract of Leaves of *Melastoma malabathricum* L. *Asian Journal of Chemistry* **23(1)**: 434-438
http://www.asianjournalofchemistry.co.in/User/ViewFreeArticle.aspx?ArticleID=23_1_103

Shetty, K. and Wahlqvist, M. 2004. A Model for the Role of the Proline-linked Pentose-phosphate Pathway in Phenolic Phytochemical Biosynthesis and Mechanism of Action for Human Health and Environmental Applications. *Asian Pacific Journal of Clinical Nutrition* **13(1)**: 1-24
http://apjcn.nhri.org.tw/server/markwpapers/Papers/Papers%202004/P327

UMS
UNIVERSITI MALAYSIA SABAH

Shuib, N. H., Shaari, K.,Khatib, A., Maulidiani, Kneew, R., Zareen, S., Raof, S. M., Lajis, N. H. and Neto, V. 2011. Discrimination of Young and Mature Leaves of *Melicope ptelefolia* Using HNMR and Multivariate Data Analysis. *Food Chemistry* **126**: 640-645 http://www.sciencedirect.com/science/article/pii/S0308814610012902

Siddiqui, Z. S. and Zaman, A. U. 2004. Effect of Systematic Fungicide (Benlate) on Germination, Seedling Growth, Biomass and Phenolic Contents of Two Different Varieties of *Zea mays. Pakistan Journal of Botany* **36(3)**: 577-582 http://www.pakbs.org/pjbot/PDFs/36(3)/PJB36(3)577

Singh, V. K., Singh, K. L. and Singh, D., 2006. Multidimensional Uses of Medicinal Plant Kachnar (*Bauhinia variegata* Linn.). *American Journal of Phytomedicine and Clinical Therapeutics,* **4(2):** 58-72 http://www.imedpub.com/articles/multidimensional-uses-of-medicinal-plantkachnar-bauhinia-variegata-linn

Sousa, C. P. 2006. The Versatile Strategies of *Escherichia coli* Pathotypes: A Mini Review. *Journal of Venomous Animal Toxins including Tropical Diseases* **12(3)**: 363-373 http://www.scielo.br/pdf/jvatitd/v12n3/31226

Sreelatha, S. and Padma, P. R. 2009. Antioxidant Activity and Total Phenolic Content of *Moringa oleifera* Leaves in Two Stages of Maturity. *Plant Foods Human Nutrition* **64(4)**: 303–11 http://link.springer.com/article/10.1007%2Fs11130-009-0141-0

Stamp, N. E. and Bowers, M. D. 1994. Effects of Cages, Plant Age and Mechanical Clipping on Plantain Chemistry. *Oecologia* **99**: 66-71 http://link.springer.com/article/10.1007/BF00317084

Sulaiman, M. R., Somchit, M. N., Israf, D. A., Ahmad, Z. and Moin, S. 2004. Antinociceptie Effect of *Melastoma Malabathricum* Ethanolic Extract in Mice. *Fitoerapia* **75:** 667-672 http://www.sciencedirect.com/science/article/pii/S0367326X04002011

Suleiman, D., Amirah, W. A., Yahaya, N. S., Khamsah, S. M., Mahadeva, R. U., Sharif, M. M. and Saravanan, D. 2015. Phytochemcical Screening, Antioxidant Potential and Cytotoxic Activity of *Melastoma Malabathricum* Linn. From Different Locations. *International Journal of Pharmacy and Pharmaceutical Sciences* **7(7)**: 408-413 http://innovareacademics.in/journals/index.php/ijpps/article/view/5673/2651

Sunilson, A. J., James, J., Thomas, J., Paulraj, J., Rajavel, V. and Muthu Paliniappan, M. 2008. Antibacterial and Wound Healing Activities of *Melastoma malabathricum* Linn. *African Journal of Infectious Disease* **2**: 68–73 http://journals.sfu.ca/africanem/index.php/AJID/article/view/391/370

Susanti, D., Sirat, H. M., Ahmad, F. and Ali, R. M. 2007. Bioactive Constituents from the Leaves of *Melastoma malabathricum* L. *Journal of Pharmacy* **5(1)**: 1-8 http://irep.iium.edu.my/1227/1/Bioactive_Constituents_from_the_leaves_of_Melastoma_Malabathricum._L

Suzuki, T. and Waller, G. R. 2006. Total Nitrogen and Purine Alkaloids in the Tea Plant throughout the Year. *Journal of Science of Food and Agriculture* **37**: 862-866 http://onlinelibrary.wiley.com/doi/10.1002/jsfa.2740370907/e

Tabasum, S., Khare, S. and Jan, K. 2016. Spectrophotometric Quantification of Total Phenolic, Flavonoid, and Alkaloid Contents of *Abrus Precatorius* L. Seeds. *Asian Journal of Pharmaceutical and Clinical Research* **9(2)**: 371-374 http://innovareacademics.in/journals/index.php/ajpcr/article/viewFile/11018/408

UMS
UNIVERSITI MALAYSIA SABAH

Veerapur, V. P., Palkar, M. B., Srinivasa, H., Kumar, M. S. Patra, S., Rao, P. G. M. and Srinivasan, K. K. 2004. Effect of ethanol extract of *Wrightia tinctoria* Bark on Wound Healing in Rats. *Journal of Natural Remedies* **4(2)**: 155-159 file:///C:/Users/cheong/Downloads

Wang, S. Y. and Lin, H. S. 2000. Antioxidant Activity in Fruits and Leaves of Blackberry, Raspberry, and Strawberry varies with Cultivar and Developmental Stage. *Journal of Agriculture and Food Chemistry* **48**:140–146 http://pubs.acs.org/doi/full/10.1021/jf9908345

Wang, S. Y. and Zheng, W. 2001. Effect of Plant Growth Temperature on Antioxidant Capacity in Strawberry. *Journal of Agriculture Food Chemistry* **49**: 4977–4982 http://pubs.acs.org/doi/abs/10.1021/jf0106244

Wongsen, W. Bodhipadma, K., Noichinda, S. and Leung, D. W. M. 2013. Relationship between Leaf Position and Antioxidant Properties in Three Basil Species. *International Journal of Food Research* **20(3)**: 1113-1117 http://www.ifrj.upm.edu.my

Wu, N., Fu, K., Fu, Y. J., Zu, Y. G., Chang, F. R., Chen, Y., Liu, X. L., Kong, Y., Liu, W. and Gu, C. B. 2009. Antioxidant Activities of Extracts and Main Components of Pigeonpea (*Cajanus cajan*) Leaves. *Molecules* **14**: 1032-1043 http://www.mdpi.com/1420-3049/14/3/1032/htm

Wu, S. C., Su, Y. S. and Cheng, H. Y. 2011. Antioxidant Properties of Lactobacillus-fermented and Non-fermented *Graptopetalum paraguayense* E. Walther at Different Stages of Maturity. *Food Chemistry* **129(3)**: 804–809 http://www.ncyu.edu.tw/files/list/fst/food_chemistry

Yoshida, T., Nakata, F., Hosotani, K., Nitta, A. and Okuda, T. 1992. Dimeric Hydrolysable Tannin from *Melastoma malabathricum. Phytochemistry* **31**: 2829-2833 http://ac.els-cdn.com/003194229283641B/1-s2.0-003194229283641B

Zakaria, Z. A. 2007. Free Radical Scavenging Activity of Some Plants Available in Malaysia. *Iranian Journal of Pharmacology and Therapeutics* **6**: 87-91 http://www.sid.ir/en/VEWSSID/J_pdf/101020070116

Zakaria, Z. A., Mohd, R. N., Mohd, N. R., Kumar, G. H., Ghani, Z. D. F. A., Sulaiman, M. R., Devi, G. R., Jais, A. M. M., Somchit, M. N. and Fatimah, C. A. F. 2006. Antinociceptive, Anti-Inflammatory and Antipyretic Properties of *Melastoma malabathricum* Leaves Aqueous Extract in Experimental Animals. *Canadian Journal of Physiology and Pharmacology* **84**: 1291–1299 http://www.nrcresearchpress.com/doi/pdf/10.1139/y06-083

Zakaria, Z. A., Rofiee, M. S., Mohamed, A. M., Teh, L. K. and Salleh, M. Z. 2011. In-vitro Anti-proliferative and Antioxidant Activities and Total Phenolic Contents of the Extracts of *Melastoma malabathricum* Leaves. *Journal of Acupuncture Meridian Studies* **4(4)**:248–56 http://www.jams-kpi.com/article/S2005-2901(11)00031-8

Zhang, L., Gao, Y., Zhang, Y., Liu, J. and Yu, J. 2010. Changes in Bioactive Compounds and Antioxidant Activities in Pomegranate Leaves. *Scientia Horticulturae* **123**: 543-546 http://www.sciencedirect.com/science/article/pii/S0304423809004920

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