WOUND HEALING PROPERTIES OF *Melastoma malabathricum* L. LEAVES EXTRACTION

LIM YEN YEN

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JUDUL: WOUND HEALING PROPERTIES OF Melastoma malabathricum L. LEAVES EXTRACTION.

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I hereby declare that this dissertation contains my original research work. Sources of finding reviewed herein have been duly acknowledged.

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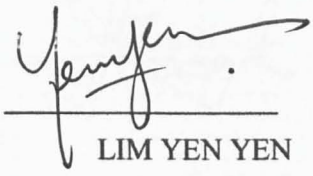

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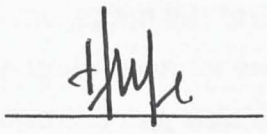
(PROF. DATIN DR. MARYATI MOHAMED)

2. CO-SUPERVISOR 1

(DR. YUSOF BIN DATUK HAJI AWANG DAMIT)



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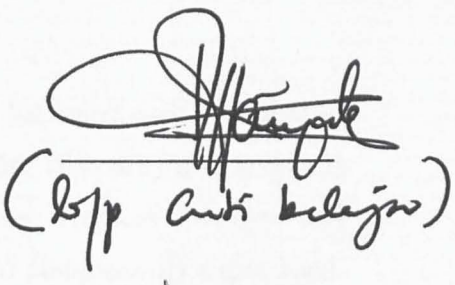

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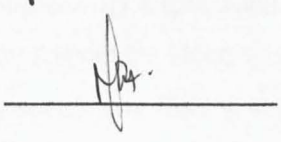

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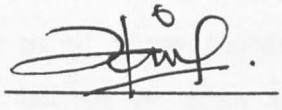

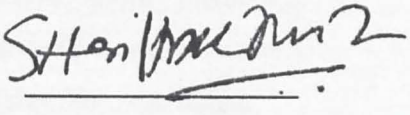

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(SUPT/KS, PROF. MADYA DR. SHARIFF A. K. OMANG, ADK)

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ABSTRACT

Melastoma malabathricum L. is commonly known as Straits Rhododendron or in Malaysia, it is locally known as 'senduduk', 'koduk' or 'kenduduk'. From the family of Melastomataceae, it is easily found in the tropical area, especially secondary forest or wasteland area. In this experiment, 15 mice were used to test on four different concentration of the leaves extract (test types): $1gml^{-1}$ of leaves paste, 5 gml^{-1} of leaves paste, 10 gml^{-1} of leaves paste and leaves paste with the mixture of saliva. Two different test were conducted which one is to test on the speed of blood coagulation, in which the blood was ejected from mice; and the second test is to test on the wound healing properties of this plant on mice (size of wound measurement); which the wounds were created on mice and different concentration of the leaves extract were applied on the wound of the mice. In the test of blood coagulation, it shows that there is a significant difference for the time taken for blood to coagulate on different test types. The most effective test type found in this test is the leaves solution with saliva; yet the most ineffective test type is the leaves extract of 1 ml. As for the test on size of wound measurement, though there is no significance found in this test, however it shows that the leaves paste of 10 gml^{-1} concentration is the most effective test types among all.

ABSTRAK

Melastoma malabathricum L. biasa dikenali sebagai 'Straits Rhododendron' atau di Malaysia, ia juga dikenali sebagai senduduk, koduk atau kenduduk. Berasal daripada famili Melastomataceae, ia mudah didapati di kawasan tropika, khasnya di hutan sekunder atau di lapangan terbiar. Dalam eksperimen ini, 15 ekor tikus telah digunakan untuk mengkaji empat jenis kepekatan daun ekstrak yang berlainan (jenis ujian): perekat daun 1 gml^{-1}, perekat daun 5 gml^{-1}, perekat daun 10 gml^{-1} dan perekat daun yang dicampur dengan air liur. Dua jenis kajian yang berlainan telah dikendalikan, satu daripadanya adalah untuk mengkaji kelajuan pembekuan darah, di mana darah perlu dikeluarkan daripada tikus; dan ujian yang ke dua adalah untuk mengkaji penyembuhan luka pada tikus (ukuran saiz luka), di mana luka dibuat pada badan tikus dan selepas itu daun ekstrak yang berlainan kepekatannya disapu pada luka-luka tikus. Dalam kajian pembekuan darah, didapati masa bagi pembekuan darah pada jenis ujian yang berlainan mempunyai perbezaan signifikan. Jenis ujian yang paling berkesan dalam kajian ini adalah daun ekstrak yang bercampuran dengan air liur; manakala yang paling tidak berkesan adalah daun ekstrak 1 ml. Bagi pengkajian ukuran saiz luka pula, walaupun tidak terdapat perbezaan signifikan, tetapi keputusannya didapati perekat daun yang berkepekatan 10gml^{-1} adalah yang paling berkesan antara semua jenis ujian.

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CHAPTER 1

INTRODUCTION

1.1 Background

The name of *Melastoma* is derived from Greek *melas*, meaning 'black', and *stoma*, meaning 'mouth', in reference to the staining of the mouth if the fruits are eaten (Jones, 1993). *Melastoma malabathricum* L. is commonly known as Straits Rhododendron or in Malaysia, it is locally known as 'senduduk', 'koduk' or even 'kenduduk'. This plant can be easily found in the tropical area, especially secondary forest or wasteland area (Asnuzilawati, 2002). It is also one of the most common weeds that grow abundantly in waste land area and open fields in Malaysia.

Belonging to the family Melastomataceae, *M. malabathricum* are shrubs which have small leaves, approximately one to five inches long, and flowers consisting of five petals. The flowers can be purple, white or pink in colour, base on the species (Asnuzilawati, 2002). The berries which contain seedbox are enclosed in the calyx cup and the fruit is soft, breaking open irregularly, with many seeds embedded in a purplish pulp. This plant has leaves that are rather narrow and pointed at both ends, with undersurface which are bristly and rough to the touch (Henderson, 1961).

Herbs are plants with annual above-ground shoots, including a flower or inflorescence, the plant itself being annual, biennial, or perennial (Simpson, 2006). In tropical developing countries, about 80 percent of the rural population (Hedberg, 1987 and Walters *et. al.*, 1993) relies on traditional practitioners using predominantly herbal medicines for their primary health care.

Though given the diversity and unexplored nature of the flora here and not to mention on Borneo Island (Leaman *et.al.*, 1990), such survey will no doubt reveal a great wealth of the undiscovered floras with their medicinal properties and potential economic value. It is believe that there is a wide usage of *M. malabathricum*, from the leaves, stem, until the root's part. The leaves, flowers and fruits are edible (Wong, 2004). From the family of Melastomataceae, only a few are cultivated for their ornamental value (Henderson, 1949). For instance, species like *M. beccarianum* have the potential value as an ornamental plant, as it is adapted to harsh soils in open conditions and so enable to ease cultivation, and fulfils important ecological role (Wong, 1997).

Both the leaves and roots of this plant have been used widely in traditional treatments which include dysentery, ulcers, haemorrhoids, and flatulence (Jones, 1993). Especially the leaves are used in skin treatments of scars and burns. The Beta-sitosterol, a pure biological component found in the leaves of this plant has the function of decreasing cholesterol in human's body (Asnuzilawati, 2002).

1.2 Justification Of Research

It is ought that there is an urgent need to document the valuable knowledge on medicinal plants due to the rapid disappearance of forest habitats in nowadays. Furthermore, the native healers who possess the traditional knowledge of medicinal plants often fail to pass their knowledge to the new generations who appears to have access to all the benefit and conveniences of the modern medicine and thus least interested to inherit the traditional knowledge of their forefathers (Fasihuddin & Ghazally, 2003). Although many local people in Sabah know the traditional usage of *M. malabathricum* as herbs, but not many research about this plant has been carried out in Sabah Borneo. Hence, this research aims to test the effect of wound healing properties of the *M. malabathricum*'s leaves on mice, in order to discover its property as medicine used in wound healing.

1.3 Objective Research

This research is carried out with the following objectives:-

1. To determine the best dosage used for wound healing on the mice.
2. To determine the most effective test type used on wound healing.
3. To know the speed of blood clotting based on different test type.

1.4 Hypothesis

The hypothesis of this research is that rate of wound healing of mice is dependent on the test types.

CHAPTER 2

LITERATURE REVIEW

2.1 Introduction

Medicinal plants constitute an important group among all of mankind's natural resources. Healing herbs have been traded and used since ancient times. The traditional medicines, no doubt, have developed over a long period of time through a close relationship between local people and the natural world (Kabilsingh, 1987). The utilization of plants in traditional medicines has been a common practice among the aborigines, particularly in the rural areas, as it is far less expensive compare to the modern medicine (Riswan *et al.*, 1990). Kabilsingh (1987), has pointed out that many of the medicines found in the pharmacy stores nowadays are the results and products of a natural screening process, resulting in thousands of plants being tested for medicinal contents all over the world.

However, not many surveys on medicinal plants had been conducted among the indigenous tribes in Sabah (Leaman *et al.*, 1990). Hence, this research is carried out to test the effect of wound healing properties of the *Melastoma malabathricum*'s leaves on mice, in order to discover its function as medicinal use in curing cuts and wound.

2.2 *Melastoma malabathricum* L.

Melastoma malabathricum is a very common herb or shrub found throughout the tropics, and is the best known bushes of open places, as well as waste ground in the lowland areas (Henderson, 1961). They may also be found in the forest at the edge of a stream, on landslips or in old clearings, and are evergreen with flowers blooming throughout the year (Burkill, 1966).

Below is the classification of *M. malabathricum* L.:-

Kingdom	: *Plantae*
Subkingdom	: *Tracheobionta*
Superdivision	: *Spermatophyta*
Division	: *Magnoliophyta*
Class	: *Magnoliopsida*
Subclass	: *Rosidae*
Order	: *Myrtales*
Family	: *Melastomataceae*
Genus	: *Melastoma* L.
Species	: *Melastoma malabathricum* L.

With its common name as *Senduduk,* or basically famous as Singapore Rhododendron, it is also known as *kodok-kodok* and *kemunting* in Brunei. *Senduduk* consists of three varieties, having large, medium and small size flowers with dark purple-magenta petals, light pink-magenta petals, and (the rare variety) white petals. The leaves are simple, opposite, narrow and pointed at both ends. Each leaf has three

distinct ribs, with its underside bristles and is rough (Henderson, 1961). The edible fruits are berry-like often surrounded by the persistent calyx. Seeds are numerous, with no endosperm (Keng, 1983). One of the most distinctive parts of the plant is the rounded basal portion of the flower, called the hypanthium, which is densely covered in red to crimson stiff bristles that each ends in a tuft of spreading hairs (Wong, 1997). Flowers are showy and large. It has a cup-shaped green calyx with five narrow reddish sepals and five purplish petals (Keng, 1983). *Melastoma malabathricum* with the white flower is reported to have miraculous healing properties (Corner, 1951).

The plants have been used in traditional Malay medicine for the treatment of diarrhea as an astringent, post-partum treatment and haemorrhoids (Burkill, 1966). In folk medicine, different parts of the plant have been used for the treatment of several diseases (Sulaiman, 2004). The decoctions of the leaves and shoots have been reported to have antiemetic and antispasmodic actions. The crude extract prepared from the leaves and roots is used to relieve toothache and also used externally against different types of inflamed wounds (Sulaiman, 2004). Scars left by small cuts are rubbed on with the plant's purple flowers and also used in a face wash (Fasihuddin, 2003). Also found in this plant that a tea of flowers is used to ease stomachache and root decoction is taken for measles (Fasihuddin, 2003).

2.3 Ethnobotanical Aspect

Health care in the village is mostly attended by the village medicinal practitioner. The knowledge of medicinal plants is normally passed from one generation to another, and

thus this knowledge not only survive but also grows and develops in the particular community as well (Riswan *et al.*,1990).

According to the estimation of the World Health Organisation, approximately 80 percent of the people in developing countries rely on traditional medicines for their primary health care, of which a major portion involves the use of plants extract or active principles originating from the parts of plants (Farnsworth *et al.*, 1985). The plant parts or whole plants are often used as remedies to cure different kind of ailments (Riswan *et al.*, 1990). The remedies for various ailments are normally involve in the use of roots, leaves and bark of the plants, while flowers, fruits and seeds are very seldom used as they are only available at certain times of the year (Fasihuddin & Hasmah, 1990).

The plant uses in medicine generally can be divided into two broad lines: for internal or external uses. According to Fasihuddin & Hasmah (1990), the external use plant materials are crushed with oil or water into poultice or paste and applied on affected areas. This method is suitable especially for skin complaints, stomachaches, headaches, etc. For internal treatment such as fever, gastric, high blood pressure, diabetes, etc. the plant parts or mixtures are boiled with water and the concoction taken as tea. The Orang Aslis use those plants in one form or another to cure or alleviate a variety of ills from bruises, bee or snake bites, tooth or stomach ache, rashes, kidney pains to healing haemorrhoids, tongue or mouth ulcers, bone fractures and malaria (Kumari *et al.,* 1998). There are also preparations and infusions for diabetes, diarrhea, rheumatism, coughs, anemia and smallpox (Kumari *et al.*, 1998).

For most villagers, the primary forest is the most important gathering site as certain medicinal plants species can only be found and to be collected (Jacob *et al.*, 2003). The collection is not done regularly but only when the need arises, and also because only very few of the plants can be stored for later use. Nowadays, many developing countries are very active in research and development of traditional medicines. For example in China, the state of traditional medicine recognizes traditional medicines and even to the extent of developing and marketing, together with the modern medicines (Lim, 1991).

CHAPTER 3

MATERIALS AND METHODS

3.1 Research Site

Peat swamp forest is the formation of a forest on peat soil in the coastal lowland and is periodically waterlogged. The peat matrix is a semi-liquid mass of dark brown material, where those partly decomposed remains of plants can be found (Berhaman & John, 1999). The peat swamps is able to support a kind of vegetation which is specially adapted to grow under the waterlogged condition. Prominent pneumatophores (breathing roots) are one of the adaptations to the specialized habitat condition of peat swamp forest (Berhaman & John, 1999).

In the 1980s, 86,000 ha of the peat swamp forest were estimated to be in Sabah of which approximately 60,000 ha of mixed peat swamp forest were in the Klias Peninsula, which includes some pristine peat swamp forests (Scott, 1989). The peat is acid with a pH of four or less, meanwhile the drainage water is tea-colored and is highly acidic as well. Here, detailed study on flora and ecology of peat swamp forests is less known compared to other dryland forests due to difficult working condition and poor accessibility (Berhaman & John, 1999). However, the Klias-

Binsulok Scientific Expedition 1999 (Maryati *et al.,* 2000) has successfully carried out botanical collection and inventory, mainly in the peat swamp forests. Klias is a peat swamp forests, situated on the western part of Sabah on the Klias Peninsula, with the size area of approximately 3620 hectares. Its Forest Reserve is located in the Beaufort district (Figure 3.2), with the main access from Kota Kinabalu-Beaufort highway. Klias Forest Reserve is classified under the Class I Protection Forest Reserve, as they are protected for climatic and ecological importance, as well as watershed areas. The climate in this area is typical wet tropical climate with a relatively constant temperature. The annual rainfall is high, between 2500 mm and 3000 mm. the rainfall is highest normally in the month of October till December (monthly rainfall >300 mm) and lowest in February till March with the monthly rainfall of lesser than 200 mm (Idris *et al.*, 2005).

On physical development, the Klias Peat Swamp Forest Field Centre was completed on the 25th August 2006. One of the most significant achievements in building this Klias Peat Swamp Forest Field Centre (Figure 3.3) is the establishment of the ecological monitoring process, to ensure that the ecosystems are adequately protected from further threats. Klias Peat Swamp Forest Reserve is rich in biological resources. The forest type here is characterized by the dominant association of several species, namely Kapur Paya (*Dryobalanops rappa*), Seraya Paya (*Shorea platycarpa*), Jongkong (*Dactylocladus stenostachys*), and Ramin (*Gonystylus bancanus*). Together with these species make up 60-70% of the standing basal area, particularly in Klias Peat Swamp Forest Reserve. It also contains thousands of plants species, many of which are still undefined such as algae, mushrooms, fungi, wild orchids and wild gingers. Based on the local people experiences, there are more than 60 plants species

with medicinal values. Some can be used to remedy cuts and wounds, skin and eye infections (Fasihuddin & Hasmah, 1990). Others can be used to treat muscular pains, fever and headaches, and remedies for health complaints affecting children. One the plants which is found and believe to have the medicinal values for cuts and wounds in this area is the *Melastoma malabathricum*.

Klias Discovery Trail
In Klias Peat Swamp Field Centre

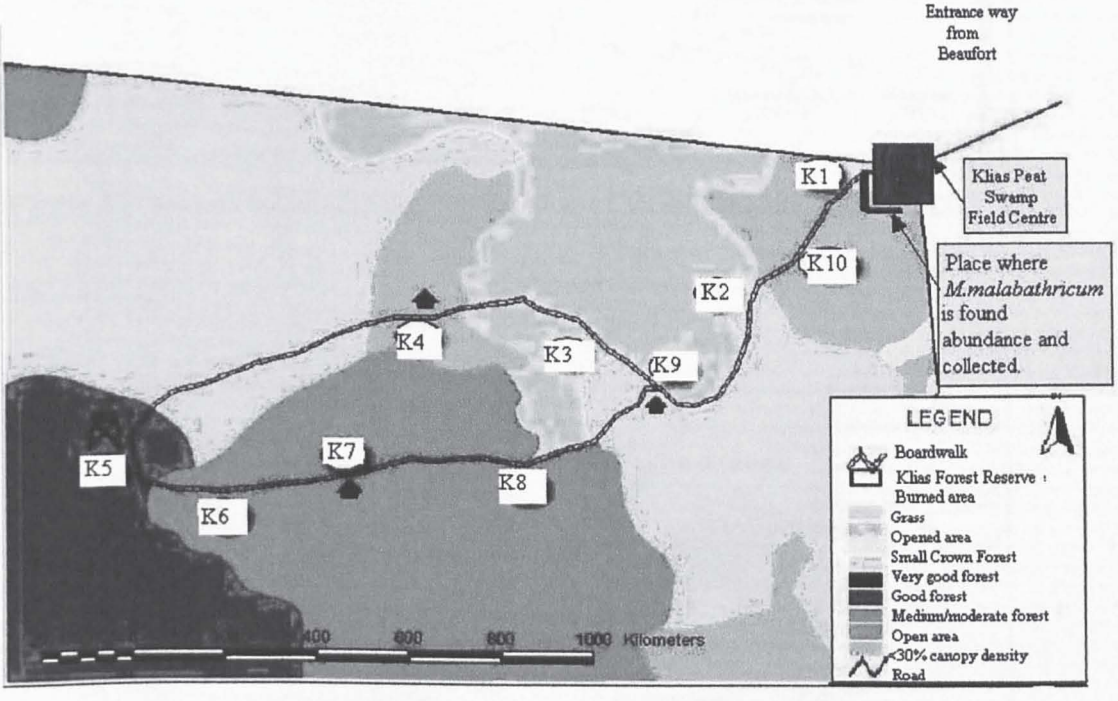

Figure 3.1 Map of Klias Peat Swamp Field Centre

The map as shown above (Figure 3.1) is the site where Klias Peat Swamp Field Centre is located (blue box). The thick red "L" shape line as shown on the map with arrow is the place where *M. malabathricum* is found abundance. It is also the area which the specimen is collected.

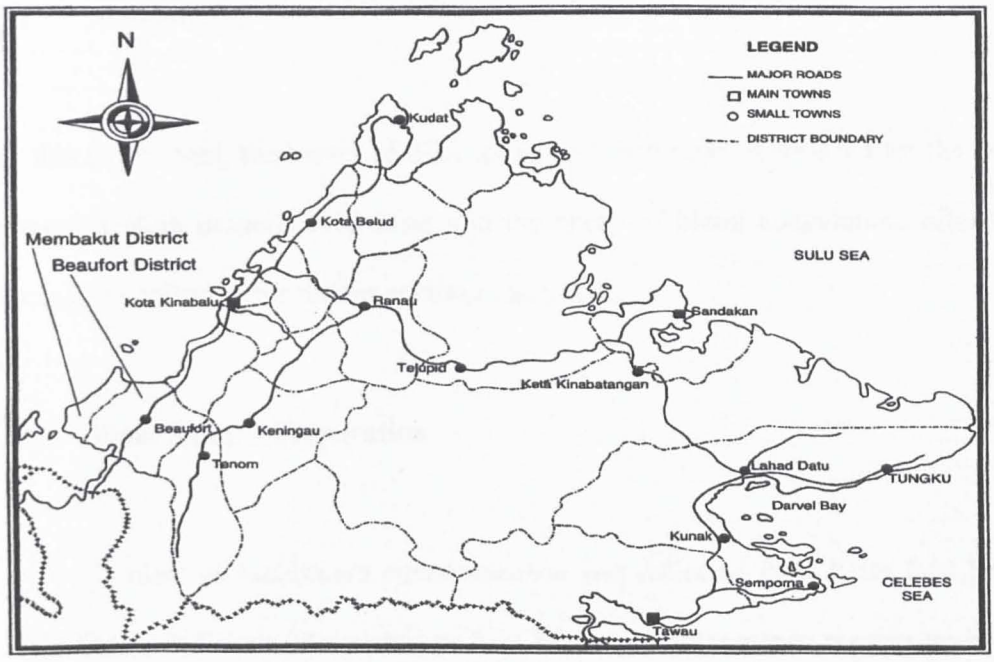

Figure 3.2 Map of Sabah showing the location of Membakut and Beaufort District.

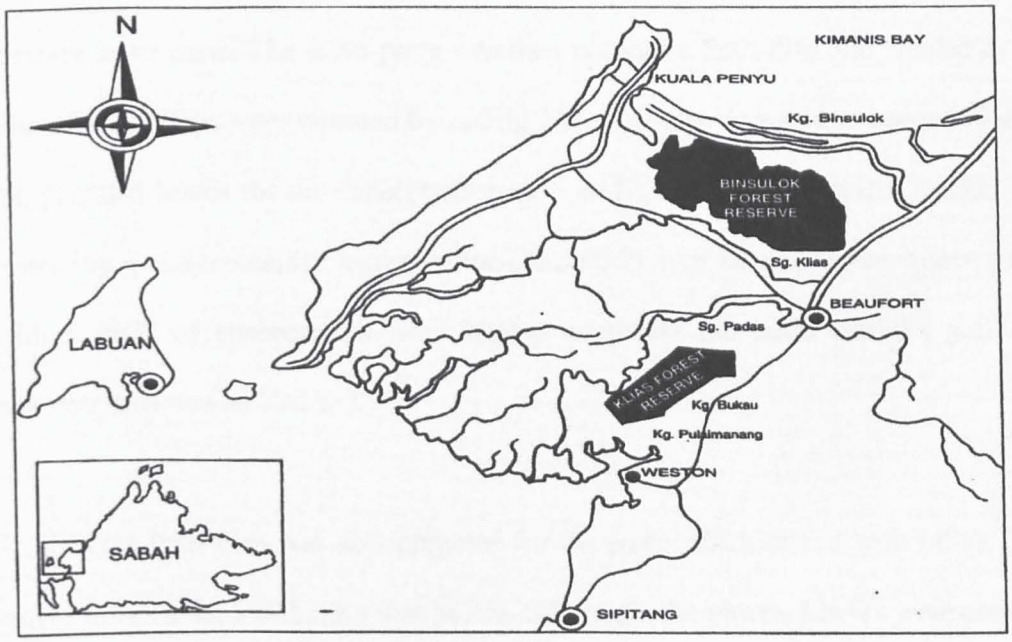

Figure.3.3 The Map showing the area of Klias Forest Reserve

3.1 Methodology

In this experiment, the leaves of *Melastoma malabathricum* were used for the test of wound healing properties on mice and the speed of blood coagulation, afterwhich datas were collected for further statistical analysis.

3.2.1 Plant Sample Preparation

A plant of *Melastoma malabathricum* was collected from Klias Peat Swamp Field Centre in Sabah. The plant was kept in a bucket of water to prevent drying out. Young leaves with light green in color were chosen and weighted until 100 g. The leaves were pounded using mortar and pestle. Meanwhile, 10ml of water will be added to every 10 g of the pounded leaves, in order to get a concentration of 1 gml^{-1} of mixture leave paste. The leave paste was then put into a Petri dish and labeled as A. The previous steps were repeated by adding 20 ml of water is added to every 100 g of the pounded leaves for the concentration of 5 gml^{-1}, and 10 ml of water is added to every 100 g of the pounded leaves for the concentration of 10 gml^{-1} paste. Leave paste with 5 gml^{-1} of concentration was labeled as B and the paste with 10 gml^{-1} of concentration was labeled as C.

One Petri dish was also prepared for the paste which mixed with saliva. The leaves were cleaned with tap water before chewing in the mouth. Leaves were chewed on mouth to combine the saliva with the leaves, until it becomes paste form. The paste was put into a Petri dish and labeled D.

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