THE POTENTIAL OF MORINGA OLEIFERA (KELOR) AND CERBERA ODOLLAM (PONG-PONG) WASTE OIL EXTRACTION AS LOCUSTS ELIMINATION

MUHAMAD SHAHIRUL FAIZ BIN MOHD NASIR

Bachelor of Engineering Technology (Energy & Environment)

UNIVERSITI MALAYSIA PAHANG

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MUHAMAD SHAHIRUL FAIZ BIN MOHD NASIR

Thesis submitted in fulfillment of the requirements for the award of the degree of Bachelor of Engineering Technology

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ABSTRAK

Belalang adalah salah satu masalah utama di industri pertanian yang menyebabkan kerosakan tanaman yang besar. Serangan yang kerap mejurus kepada kerugian yang besar terhadap alam sekitar dan ekonomi. Racun perosak kimia yang digunakan dalam kebanyakan pertanian menyebabkan beberapa kesan negatif terhadap organisma bukan sasaran dan pencemaran alam sekitar. Racun serangga botani atau lebih dikenali sebagai biopestisid merupakan salah satu alternatif untuk mengatasi masalah ini. Oleh itu, potensi pengekstrakan kelor dan pong-pong untuk digunakan sebagai biopestisid dalam penghapusan belalang telah dikaji dalam penyelidikan ini. Biji dari kelor dan daun dari pong-pong diekstrak dengan menggunakan proses pengekstrakan etanol. Hasil minyak yang dihasilkan telah disiasat ke atas belalang dalam tempoh pemerhatian selama 150 minit. Jumlah belalang yang terselamat dikira untuk setiap 15 minit. Kelor dan pongpong, kedua-duanya merekodkan kadar kematian sebanyak 100%. Tumbuhan dan rumput mengekalkan keadaannya di dalam tempat simpanan belalang sepanjang tempoh pemerhatian ini. Unsur makro dan mikro dalam ekstrak kelor membantu melindungi tumbuhan dan alam sekitar tetapi juga bertindak sebagai biopestisid yang baik. Sementara itu, daun pong-pong kaya dengan flavonoid, saponin, cerberin dan sebatian alkaloid dan oleh itu, bermanfaat untuk menghapuskan belalang. Minyak kelor merekodkan pH 6.81 manakala minyak pong-pong mencatatkan pH 6.82. pH untuk kedua-dua minyak lebih hampir kepada pH semula jadi 7, mengesahkan sifat mesra alam. Minyak pong-pong merekodkan masa yang lebih pantas untuk menghapuskan kesemua 30 belalang disebabkan oleh kadar penyerapannya yang tinggi. Kehadiran pelbagai sebatian telah disahkan berdasarkan analisis FTIR pada minyak kelor dan pong-pong seperti alkohol dan hidroksil, alkana, amina tertier, amina aromatik, kumpulan C-O dan kumpulan C-H alkena. Kedua-dua minyak dianggap bermanfaat dan berkesan berdasarkan kepada beberapa kelebihan, iaitu, kaedah penyediaan yang mesra alam, mesra kos, mudah disediakan dengan kadar kematian belalang yang tinggi.

ABSTRACT

Locusts is one of the primary issues in agriculture industries causing major crop failure. Frequent outbreaks have led to tremendous environmental and economic losses. The chemical pesticides employed in most agriculture retain some negative impact on nontarget organisms and environmental contamination. Botanical pesticides or commonly known as biopesticides is one of the alternatives to overcome this problem. Thus, the potential of moringa oleifera and cerbera odollam extraction to use as biopesticides in elimination of locusts was studied in this current research. The seeds from moringa and leaves from cerbera was prepared to extract using ethanol extraction process. The oil extraction products were investigated into a number of locusts for 150 min observation period. The amount of locust survived were calculated for every 15 min. Both moringa and cerbera recorded 100% mortality rate. The plants and grass maintain its condition inside the locust's storage throughout this observation period. Macro and micro elements in the moringa extract help to protect the plant and environment but also acts as efficient biopesticides. Meanwhile, cerbera leaves is rich with flavonoids, saponins, cerberin and alkaloids compound and therefore, beneficial to eliminate locusts. Moringa oils recorded pH of 6.81 whereas, cerbera oils recorded pH of 6.82. The pH for both oils closer to natural 7 extend its confirmation of environmentally-friendly attributes. Cerbera oil recorded faster time to eliminate all 30 locusts credited its high absorption rate. The presence of multiple compounds was confirmed based on FTIR analysis on moringa and cerbera oil such as alcohol and hydroxyl, alkanes, tertiary amines, aromatic amines, C-O group and C-H group of alkenes. Both extraction oils are regarded beneficial and effective credited to several advantages, namely, eco-friendly, cost-friendly, simple preparation method with high mortality rate of locusts.

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LIST OF SYMBOLS

°C	degree Celsius
%	percentage
ml	Millimetre
hr	Hour
Km	Kilometre
min	minutes

LIST OF ABBREVIATIONS

FAO	Food and Agriculture Organizations
FTIR	Fourier-Transform Infrared (FTIR) Spectroscopy
GCMS	Gas Chromatography-Mass Spectrometry
ICP-OES	Inductively Coupled Plasma-Optical Emission Spectroscopy
UN	United Nations
XRF	X-Ray Fluorescence Spectroscopy

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CHAPTER 1

INTRODUCTION

1.1 Background of Study

Locusts are part of the Orthoptera order's Acrididae family, and they have a big hind leg. Locusts are a significant component of both natural and disturbed ecosystems because they stimulate plant growth, play an important role in the food chain byparticipating in the nutrient cycle, but they also alter ecological processes such as carbon cycles and water cycles, which can lead to increased runoff and soil erosion. Locusts are larger grasshoppers that form swarms and can travel a longer distance (Pandey et al., 2021). It is regarded as one of the most significant agricultural pests in the world due to its voracity, rapid reproduction, and wide range of flight, which can cause massive damage to all plant species (Githae & Kuria, 2021).

The key to reducing the extent to which locusts might harm agricultural areas, according to the United Nations (UN) Food and Agriculture Organization (FAO), is early warning and early intervention. Countries must conduct the necessary field surveys and maintain them on a regular basis following unusually heavy rains to track breeding conditions and locust infestations (Suteu et al., 2020).

Although conventional pesticides have become an indispensable tool for controlling some pests economically, instantly, and effectively, their widespread use may result in a number of unfavourable side effects, including the development of insect resistance and the resurgence of primary and secondary pest outbreaks. They can also have a negative impact on nontarget organisms and broad environmental contamination (Erdogan et al., 2012). Other issues with synthetic insecticides include pollution and insect resistance. According to Erler et al. there is a growing interest in the use of botanical pesticides for crop protection. Many scientists are exploring with and creating alternative plant extracts for use as insecticides against pest insects (Erler et al., 2010) Plants are the most abundant source of renewable natural insecticides. Plant extracts, in particular, offer a safe and viable alternative to synthetic pesticides and are consistent with the use of beneficial organisms, pest-resistant plants, and the preservation of a healthy ecosystem in an effort to reduce reliance on synthetic pesticides. There are numerous advantages of employing botanical pesticides, including less environmental degradation, increased safety for farm workers, increased food safety, reduced pesticide resistance, and increased production profitability (Erdogan et al., 2012).

The neem plants extraction products were recommended by several research as alternatives to the currently used harmful pesticides for the control of desert locust. Neem biopesticides containing azadirachtin have been reported to control approximately 400 species of insects, including important pests, such as armyworms, aphids, and whiteflies. In addition to controlling pests, most neem biopesticides have negligible effects on beneficial insects, and low environmental impacts (Mansour et al., 2015). However, it was reported that neem extracted pesticides despites its eco-friendly characteristic, it has slow action and usually resulted in incomplete kill of the locusts (Pandey et al., 2021).

Thus, it is important to explore different types of plants to extract biopesticides especially in the locust management control. In recent years, plant oils have received more and more attention as alternative, potentially valuable compounds that can be used locust pests. Several studies have confirmed that plant oils could be used as natural pesticides to control agricultural pests (Aly et al., 2012; Tak et al., 2017; Tripathi et al., 2009) and plant diseases (Arshad et al., 2014; Elshafie & Camele, 2017; Parikh et al., 2021). Most natural plant oils are suitable for organic agriculture because they are non-residual, non-toxic to humans and vertebrate animals, easy to prepare and apply, cheap, effective and natural (Elshafie & Camele, 2017). One of the plants that are suitable for this purpose is moringa oleifera. It is originates from the Moringa cease family, is a single genus of bush tree families cultivated throughout the tropics and is used for various purposes (Manzoor et al., 2015). Moringa can be considered as alternatives due to its various properties such as its oil can be suitable replacement for olive oil in the diet and moringa seed benefits human health. The efficiency of commercial moringa plant oil

produced by produced by Egyptian Natural oil Co was studied and resulted in 96% of mortality rate of grasshoppers (Soltan, 2020).

In addition, poisonous plants also known to be an effective as biopesticides since it naturally containing bioactive toxin that used successfully as insecticides and fungicides (Erdogan et al., 2012). Cerbera odollam (pong-pong) is a dicotyledonous angiosperm, a plant species in the family Apocynaceae and commonly known as the suicide tree. The bioactive toxin in the plant known as cerberin has a mechanism of action similar to digoxin. Hence, its seeds contain a toxic and lethal potent compound called glycosides cerberin that suitable for biopesticides alternatives. The past researches on another type of Apocynaceae such as *nerium oleander* (oleander) and *calotropis procera* (Sodom apple) showed high percentage of mortality rate against locust with its high toxicity and inhibit development of locust (Githae & Kuria, 2021).

1.2 Problem Statement

Locusts are considered very harmful pests that feed on every edible substance available. One single mature locust can consume crops equal to its weight. In present condition where various world nations are struggling with the problem of food scarcity, this desert locust invasion has emerged as a considerable setback in achieving the universal goal of food security. Frequent outbreaks have led to tremendous environmental and economic losses. So, different control strategies are introduced to overcome these issues.

Chemical insecticides used for locust control are insecticides, which means they kill not only just locusts but also other insects and arthropods present in the environment, some of which could be beneficial ones such as honeybees, other pollinators or natural enemies of locusts. The chemical pesticides that are used in locust control pose some risks to human health, although certain products are less hazardous than others (Sabarwal et al., 2018). Hence, it is very crucial to develop more environmentally sustainable methods as alternatives to toxic pesticides in order to reduce toxic ecosystem and agricultural impacts from the use of current chemical pesticides.

Natural plant protection products (known as biopesticides), commonly derived from plants can be used as eco-friendly alternatives due to less toxic to humans and the environment, no release/leaching of harmful residues, and usually much specific to the target pests. Therefore, the extraction of plants such as moringa oleifera and cerbera odollam to produce valuable biopesticide is evaluated in this study to investigates the potential use of extracts of cerbera odollam and moringa oleifera to eliminate locusts.

1.3 Objectives of the Study

This study aims to:

According to the outcomes from this study, the following recommendations may be suggested for future research:

- 1. To synthesize biopesticides using extraction methods from moringa oleifera seeds and cerbera odollam leaves.
- 2. To evaluate the mortality and repellents effects of moringa oleifera seeds and cerbera odollam leaves oil on biological control of locusts.
- 3. To compared the performance of moringa oleifera seeds and cerbera odollam leaves extraction oil in order to eliminate locust.

1.4 Scope of Study

The first scope is to synthesize biopesticides from moringa oleifera seeds and cerbera odollam leaves by using ethanol extraction technique. Moringa seeds and cerbera leaves was left in ethanol for 2 days and oil extraction was collected by separating the oil with filter paper. The second scope, in order to evaluate the mortality and repellence effects of both biopesticides are sprayed on two target species of locust and the grasshoppers that test according the pH and percentage of mortality rates based on Schneider-Orelli formula. The observation period was done for every 15 min for total of 150 min. The comparative study between the effect of both moringa oleifera seeds and cerbera odollam leaves biopesticides are done using the same method to achieve percentage of mortality rates of locusts.

1.5 Significant of Study

During dry spells, solitary locusts are forced together in the patchy areas of land with remaining vegetation. A small swarm of locusts contains thousands of individuals that spread over several hundred square meters, but large swarms contain up to 80 million individuals per km² and can infest more than 1000km². Since a swarm can covera distance of 100km per day, farmers regard gregarious locusts as one of the most destructive plagues on earth. When the locusts start attacking crops and thereby destroy the entire agricultural economy, it is referred to as locust plague/locust invasion. Plagues of locusts have devastated societies since they still wreak havoc today. This study identifies several methods to eliminate locusts.

The most significance method is biopesticide generated using the moringa oleifera seeds and cerbera odollam leaves. Several studies have confirmed that plant oils could be used as natural pesticides to control agricultural. Most natural plant oils are suitable for organic agriculture because they are non-residual, non-toxic to humans and vertebrate animals, easy to prepare and apply, cheap, effective and natural (Elshafie & Camele, 2017). This method considers as non-chemical methods for controlling pests knownas the locusts. Utilizing the biopesticide in the agriculture field, it can enhance the growth of the plants where it can contribute to the economic increase.

CHAPTER 2

LITERATURE REVIEW

2.1 Overview of Locusts

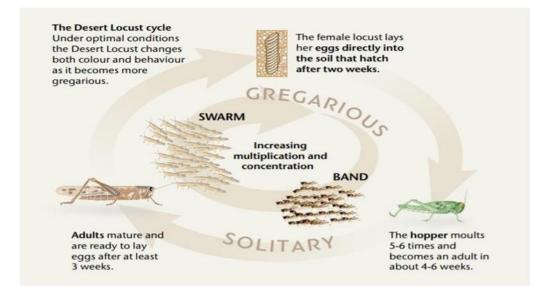
Locusts are large insectivorous insects that, because of their ability to create thick and rapidly mobile swarms, can be serious agricultural pests. They are short-horned grasshoppers that form large colonies in dense migration groups on a regular basis, with individuals differing in many ways from those living separately (Zhang et al., 2019). It is critical that countries conduct the requisite field surveys and sustain them on a regular basis after extremely heavy rainfall to track breeding conditions and locust infestations. The discovery of large infestations necessitates control operations to prevent a further increase in locust numbers. In order to establish effective strategies for controlling harm, an integrated approach to understanding the conditions that lead to locust build-up and migration is needed (Pandey et al., 2021). The growth and mitigation of locust occur due to 3 factors that arewind, rainfall and temperature.

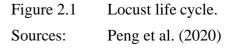
Controlling locusts is complicated by a number of factors. First, the swarm is highly mobile, migrating from 50 to more than 100 km in a day. Second, the total invasion period frequently occurs in a relatively brief time, sometimes as short as a month but rarely longer than three months. Then, the swarms are unevenly distributed in time, so that very large swarms may beavailable for only a few days, followed by relatively long periods when none is present. Next, swarms are variable in size and can extend up to thousands of hectares. Lastly, campaign experience, funds and supplies are often lacking in affected countries because of the irregular occurrence of locust upsurges and plagues (Branson, 2017).

The large number of scientific articles published on locust outbreaks and pests reflects the historical importance of this subject, which has received significant interest from researchers in the domains of physiology, biochemistry, ecology, and evolution. The majority of this information pertains to the locust plague and insect physiology, andit is inferred that the exponential increase in the species population growth will result in catastrophic environmental consequences such as the complete destruction of cropfields, resulting in food shortages and famine, as well as socioeconomic losses. It is critical to find a solution that can mitigate and prevent the outbreaks that are causing significant concern in order to avoid serious crop damage and reduce the massive losses in agriculture, forestry, and animal husbandry. Finding a solution that can mitigate and prevent the outbreaks that are causing significant concern is critical. Green preventative methods that are less hazardous to the environment when compared to traditional pesticides are getting more attention as we moving forward.

2.1.1 Locusts Life Cycle

Locusts can live for three to five months, but this is highly unpredictable and mostly dependent on weather and environmental factors. There are three stages of the lifecycle: embryo, hopper (nymph), and adult (Figure 2.1). The lifetime of a locust is about three months but this can be extended to up to six months under cold conditions. Depending on the temperature, eggs hatch in around two weeks (the range is10–65 days). Hoppers shed their skins five to six times, increasing in size each time. This is known as moulting, and the stage between moults is known as an instar. Hoppers mature in about three weeks to nine months, but more often in two to four months, depending on environmental conditions, especially temperature. Adults will stay immature for up to six months if the weather is dry and cold. Adults do not moult, so they do not rise in bulk, but they do gain weight steadily. Any day, an adult locust will consume 2.5 g of its own body weight. Adults that can fly are sexually immature at first, but they mature over time and can copulate and lay eggs. Solitary individuals are still present in the desert, waiting for the right circumstances to mate (Peng et al., 2020).





There are more than 10,000 grasshopper species in the world, which are in a variety of habitats including tropical, temperate grassland, and desert environments. Locusts are a collection of 18–21 species that have the ability to swarm over large distances, making them one of the greatest migratory pests that cause harm to agricultural crops. It is believed that the desert locusts are the most prevalent species of locust in the world, with approximately ten subspecies found in Europe, Asia, Africa, and Australia. When it comes to crop damage and food supply population, these species comprise the largest and most damaging insect groups on the planet. It is particularly concerning since the desert locust may migrate over great distances and rise in numbers quickly, but the migratory locust is the most widely distributed species (Zheng et al., 2020).

The desert locust has a life cycle of 3–6 months, and a mature female locust can produce approximately 100 eggs each day at its highest. As the larvae develops quickly, it only takes around 20 days for them to reach maturity and become the source of the next generation of insect pests (see Figure 2.2). The most favourable conditions for locust growth are 25–32 °C at relative humidity levels ranging from 85 % to 92 %, with soil humidity levels ranging from 15 percent to 18 percent, respectively. Drought limits the amount of vegetation cover, which helps to protect the eggs until rainy weather conditions are favourable for spawning. The massive movement of locusts is triggered by changes in the neurotransmitter

serotonin levels in the brain. It is resulted in this hyperpolarization, post-synaptic potentials in the locust's legs muscle fibres are inhibited, resulting in a weakening of the depolarized excitatory post-synaptic response, allowing the locust to jump one hundred times their body length. Locusts havea tremendous flight capacity that allows them to fly continuously for more than 10hours a day and travel up to 150 km. When compared to other locust species, the desert locust can breed one generation in less than a month, which is six times faster than the other locust species' reproduction rate (Meynard et al., 2017; Yang et al., 2020).

The massive movement of locusts is triggered by changes in the neurotransmitter serotonin levels in the brain. As a result of this hyperpolarization, post-synaptic potentials in the locust's legs' muscle fibres are inhibited, resulting in a weakening of the depolarized excitatory post-synaptic response, allowing the locust to jump one hundred times their body length. Locusts have a tremendous flight capacity that allows them to fly continuously for more than 10 hours a day and travel up to 150 km. When compared to other locust species, the desert locust can breed one generation in less than a month, which is six times faster than the other locust species' reproduction rate This, combined with their exponential development, allows the specimens to survive for up to three months and so expand the population by a factor of 20 for each generation (Githae & Kuria, 2021; Peng et al., 2020).

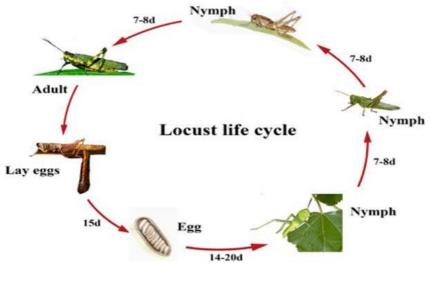
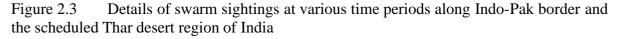


Figure 2.2The life cycle of locustsSources:Peng et al. (2020)

2.1.2 Locusts Swarm Data

The Locust Hub portal provides users with access to location-based data on Hoppers, Bands, Adults, Swarms, ecological settings, and specifics of control activities conducted in various nations. The swarm's dataset includes a date-time stamp, geolocation, metadata, place name, and affected area. The transit paths of swarms were studied utilising locust attack incidents from the national media. The Locust Hub portal was utilised to retrieve a subset of data regarding swarms thatoscillated in the Indian subcontinent before, during, and after the Amphan Storm cyclonic occurrences (Pandey et al., 2021; Zhang et al., 2019).

Region of locust oscillations	Time Period	No. of locust swarms reported
Indo-Pak border and the scheduled Thar desert region	1 Jan to 09 May, 2020	2192
Scheduled Thar desert region in India	10 May to 15 May, 2020	54
Swarms transited through the first conduit	16 May to 21 May, 2020	36
Entry of fresh swarms into the Thar desert region from Pakistan during the living period of Amphan	16 May to 21 May, 2020	22
Swarms transited through the second conduit	22 May, to 28 May, 2020	49



Sources: Locust Hub (2020)

Figure 2.4 shows locations of Locust swarms in and around the Thar Desert during various phases of Amphan Storm. In this map, archival sightings of the Desert locust till the period of Amphan Storm are shown in black dots. Note the movement of swarms towards the Southeast direction during the living period of the Amphan Storm and towards the Eastern side after the storm episode (Pandey et al., 2021).

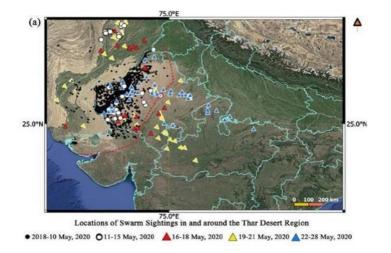


Figure 2.4Map of Locust swarms in and around the Thar DesertSources:Locust Hub (2020)

Figure 2.5 illustrates the swarm conduits formed during the living period of Amphan Storm (green colour conduit) and post cyclonic episode (blue colour conduit). The first conduit favoured the migration of swarms to the south-central parts of India and second conduit favoured migration of swarms to the Indo-Gangetic plane (Pandey et al., 2021).

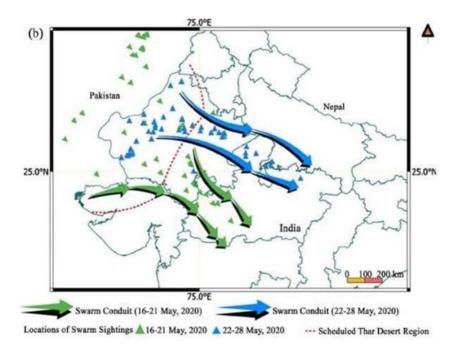


Figure 2.5Map of swarm conduitsSources:Locust Hub (2020)

2.2 Pesticides

Pesticides are a class of chemical elements used to control and prevent pest populations. Pesticides are chemical and natural chemicals used to control or destroy pests such as insects, organisms that cause plant diseases, and weeds. It is also used to manage other living species, including as nematodes, arthropods, insects, andvertebrates, which ruin our food sources and cause a variety of health concerns. Pesticides are chemical chemicals that are used to protect crops from insects, weeds, and pests while also nourishing and boosting crop productivity and efficiency (Rani et al., 2021).

Pesticides are classified widely based on their usage, target organism, and chemical composition. Pesticides are classified into three types based on their application: agriculture (used to protect crop pests, insects, and weeds), public health (used to kill disease vectors), and domestic pesticides (used to kill insects like cockroach, bacteria, protozoa, mice etc). Pesticides are classified into four types based on their target organism: insecticides (chemicals used to kill insects), fungicides (chemicals used to inhibit or kill fungi), herbicides (chemicals used to control or kill weeds), rodenticides (pesticides used to kill rodents), fumigants (gaseous pesticides used to kill or control pests such as bedbugs), and insect repellents (which are applied to kill or cloth to keep insects away from the skin and cloth). Pesticides are classified chemically as organochlorines, organophosphates, carbamates, pyrethroids, phenyl amides (carbanilates, acylanalides, toluidines, and acetamides), phenoxyalkonates, trazines, benzoic acid derivatives, benzonitriles, phtalimide derivatives, dipyrids, and others (El-wakeil, 2013; Jayaraj et al., 2016).

2.2.1 Advantages of Pesticides

Pesticides are widely used in crop production zones to reduce insect infestations and hence safeguard crops from potential harvest losses and product quality declines. There are a wide range of positive outcomes following the application of various pesticides. Reduced crop loss as a result of fungicide spraying is an obvious benefit but, some are less obvious, either because they occur in the medium or longterm, or because they are subtle or modest incremental assistances dispersed across a large area. The impacts are the immediate results of pesticide use. Pesticides have three key effects: they manage farm pests and plant disease vectors, they control vectors of human and livestock sicknesses and irritation creatures, and they prevent or control creatures that damage other human acts and constructions (Damalas & Koutroubas, 2018). The key benefits are the direct gain predicted after employing pesticides as a result of their effect.

Primary advantages of pesticides (see Figure 2.6) can be sorted into firstly, controlling pests and plant disease vectors, secondly, controlling disease vectors of animals and thirdly, harmful organisms and controlling organisms which harm other human activities and structures. Controlling pests and plant disease vectors can lead to better crops or livestock yields and quality, reduced fungal toxins, better self-life of products, reduced fuel use and labour of weeding and controlled invasive species. In addition, by controlling disease vectors of animal, it will reduce human disturbance, reduced animal suffering, improved livestock yields and quality and contained diseases geographically. Controlling organisms which harm other human activities and structureshelp protection of plant area, prevention of moots or moisture damage and protection of civic ornaments.

Secondary advantages can be put into communities, national and worldwide perspectives. Based on communities, it can improve nutrition, health, farm/agribusiness revenues, food safety, food security, range if viable crops and reduce stress and maintenance costs. In terms of national advantages, it can help to enhance export revenue, workforce productivity, or reduce soil erosion and reduce moisture loss along with pleasant urban areas. From worldwide perspective, safe and diverse food supply can be improved, reduced civil unrest, less pressure in uncropped land, less greenhouse gases, reduce pest introduction, improve tourism and habitable areas and reduce environmental effects like global warming to conserve more biodiversity (Bonner & Alavanja, 2017; Damalas & Koutroubas, 2018; Zhang, 2018)

PRIMARY ADVANTAGES

Controlling pests and plant disease vectors

- o Better crop/livestock yields
- o Better crop/livestock quality
- o Reduced fungal toxins
- Better self-life of product
 Retailer networks
- establishedReduced labour of weeding
- D L LC L C
- Reduced fuel use for weeding
- Reduced soil disturbance
- Pests contained locally
- o Invasive species controlled

Controlling disease vectors of animals and harmful organisms

- Human lives saved
- Reduced human sufferingReduced human
- disturbance
- o Animal lives saved
- o Reduced animal suffering
- o Increased livestock yields
- Increased livestock quality

 Diseases contained geographically

Controlling organisms which harm other human activities and structures

- Drivers view unobstructed
- Tree/bush/leaf hazards prevented
- Prevention of roots/moisture damage
- o Protection of turf
- o Garden plants protected
- o Civic ornaments protected
- Wooden builds protected

 Masonry/paint/plastic/fuel etc. protected

SECONDARY ADVANTAGES							
Communities Farm and agribusiness revenues Improved nutrition and health Food safety Food security Better quality of life Wider range of viable crops Labour freed for other tasks Increased life expectancy Reduced medical costs	National• National agricultural economy• Increased export revenues• Increased workforce productivity• Agronomic advice improves cropping• Reduced soil erosion/moisture loss• Reduced migration to	World-wide • Safe and diverse food supply • Reduced civil unrest • Less pressure on uncropped land • Less greenhouse gases • Fewer pest introductions • International tourism • Increased habitable areas • Biodiversity conserved • Less global disease spreade					
 Fitter population Reduced stress Reduced maintenance costs 	 cities Pleasant urban areas Fewer transport accidents 	 Reduced global warming Timber viable for construction 					

Figure 2.6 Primary and secondary advantages of pesticides

Sources: Rani et al. (2021)

2.2.2 Risks of Using Pesticides

Pesticides have a variety of purposes, including increasing crop output, controlling vector illnesses, and killing or inhibiting dangerous pests. Pesticides, on the other hand, have unavoidable negative consequences. Because they cause considerable harm to the environment as well as humans, they degrade the quality of water and soil, posing a risk to animals, birds, plants, and humans. Pesticides also disrupt biodiversity, and long-term direct

or indirect pesticide exposure can pose serious health risks. They can cause serious health problems such as cancer, diabetes, reproductive problems, respiratory problems, and neurological problems. Today, there is a need to restrict pesticide use and identify substitutes, as well as to educate people about safety precautions to take when applying pesticides, and to urge farmers to practise organic farming, so that pesticide use can be minimised. In the future, research will be required to generate creative concepts in current farming that will reduce the use of pesticides. Adsorption, bioremediation, advanced oxidation, and other remediation strategies have been documented for the removal of pesticides from contaminated environments (Rani et al., 2021).

However, adsorption and bioremediation are reported to be the best treatments sincethey are ecologically benign, economically effective, and produce fewer harmful by-products. Farmers, environmental protection organisations, health officials, pesticide manufacturers, and governments should work together to lessen the risk of pesticide poisoning. Pesticides should be managed efficiently by enforcing tight rules and toxicity restrictions (Rani et al., 2021). Pesticide application and pesticide control can both benefit from integrated pest management. Pesticides should be manufactured with precision and accuracy, as well as a higher safety profile, in order to have a lower detrimental influence on the environment and humans. In addition to this, future research should focus on developing relatively safe chemicals for the same purpose.

Thus, it is obvious that the inevitable use of pesticides may approach some danger to human health and the environment, and that these risks must be analysed in order to determine control the negative impacts. The first chemical agents used as pesticides were of natural origin, such as plant extracts like pyrethrum or mineral derivatives like arsenic and copper compounds.

2.3 Biopesticides

Biopesticides can be used instead of chemical pesticides since they have been proved to be effective for pest management and the production of sustainable agricultural goods (Prabha et al., 2016). However, technical problems and barriers have restricted their efficacy to date. The impact of Integrated Pest Management (IPM) policies in the European Union gives an incentive for novel pest management tactics, particularly the use of biopesticides, which include living microbesor natural materials (Chandler et al., 2011).

Biopesticides enable a more sustainable approach to crop improvement, which should expand their use and appeal in the coming years. Furthermore, attitudes toward the usage of biopesticides have begun to shift as a result of the new realisation of the environmental repercussions of chemical pesticides. Other factors influencing their greater use are advancements in activity spectra locations and enhanced delivery and application options (Mishra et al., 2017). Several criteria suggest that biopesticides are superior to synthetic pesticides. Figure 2.7 outlined the benefits of biopesticides, such as economic effectiveness and decreased toxicity to beneficial pests (Gupta & Dikshit, 2010).

Factors	Benefits of biopesticides Costlier but reduced number of applications			
Cost effectiveness				
Persistence and	Low, mostly			
residual effect	biodegradable and self perpetuating			
Knockdown effect	Delayed			
Handling and Bulkiness	Bulky: Carrier based			
	Easy:Liquid formulation			
Pest resurgence	Less			
Resistence	Less prone			
Effect on beneficial flora	Less harmful on beneficial pests			
Target specificity	Mostly host specific			
Waiting time	Almost nil			
Nature of control	Preventive			
Shelflife	Less			

Figure 2.7Benefits of biopesticidesSources:Gupta & Dikshit (2010)

Biopesticides can be divided into three categories. The first category is known as microbiological pesticides, and it includes items derived from microorganisms such as bacteria, fungi, viruses, protozoa, and algae. Fungi, for example, can manage weeds and

remove particular insects. Bacillus thuringiensis (Bt) is also frequently used to control insects on potato, cabbage, and other crops.

Microbial insecticides often control pests by producing certain toxic metabolites that cause pests to become ill, and they can also hinder the growth of other microbes. Pesticidal compounds in plants that result from plant genetic alteration are included in the second group. The transgenic expression of the Bt gene, which allows plants to produce a bacterial protein that kills pests on their own, is a great example. Finally, nontoxic biochemical pesticides that occur naturally to suppress pests, such as plant growth regulators that disrupt pest development, mating, or appealing pheromones, are available (Gupta & Dikshit, 2010).

Botanical insecticides, also known as plant secondary metabolites, are predicted to be viable locust-control alternatives to standard chemical insecticides. They exhibit broad-spectrum activity, relative specificity in mechanism of action, and are simple to process and apply. They are also generally safe for animals and the environment (George et al., 2015).

Several studies have reported the use of botanical extracts as potential bio- pesticides against locust. Around 27 plant species belonging to 20 families were identified as tested against locust (Table 2.1) but had varied results. *Azadirachta indica* and *Melia volkensii* were the most studied plant species, both belonging to Meliaceae, which is known to have biologically active limonoids. These were followed by *Calotropis procera*, *Fagonia bruguieri*, and *Peganum harmala*. The most represented plant family was *Apiaceae* (Githae & Kuria, 2021).

Plant extraction have been reported to eliminate locust in different ways. For instance, azadirachtin from *A.indica* has an anti-feeding product that controls a wide range of pant pests without harming beneficial insects. Anti-feeding activity of A. indica against desert locust has also been well studied (Bashir, 2014; Hamadah et al., 2013; Kearney et al., 1994). The activity varied *in A. indica* (79.62%), *Jatropha curcas* (78.92%), and *Solenostemma argel* (56%) against desert locust with a significant mortality of 43.39%, 40.54%, and 20.70%, respectively, after 7 days of treatment. In addition, cold-pressed *A. indica* seed oil resulted in molting disturbances and morphogenetic defects of the wings, legs, and antennae. When *Nigella sativa* extracts were compared with those of *A. indica*, *Nigella sativa* caused a decrease in the body weight but with no mortality (Schmutterer et al., 1993).

The effects of *Calotropis procera*, *Zygophyllum gaetulum*, and *Peganum harmala* on survival, feeding, and reproduction in desert locust showed that all the extracted plants reduced food intake, increased weight loss, and caused a significant mortality (Abbasi et al., 2015). Plants extracted from *C. procera* and *Z. gaetulum* prevented sexual maturity both in males and female while those of *P. harmala* reduced female fecundity and hatching rate. Both *C. procera* and *P. harmala* generated 100% mortality within a few days after treatment and portrayed symptoms similar to those of insects treated with insecticides (Abdellah et al., 2013; Kaidi et al., 2017). There is another study on the effect of *C. procera*, *Schouwiapurpurea*, and *Zizyphus lotus* on the locust observed morphological changes, molting inhibition and antifeeding effects with a significant mortality ranging from 45% to 53% on day five after treatments (Messgo-Moumene et al., 2015).

In a different experiment, fruit extracts of *Ammi visnaga* promoted the acidic phosphatase (AcP) activity and reduced alkaline phosphatase (AlP) activity in fat bodiesof adult desert locust. This was attributed to presence of couramins and furocoumarins (Ghoneim et al., 2014). Extracts of *Fagonia bruguieri* also inhibited growth and development of locust by intervening with the process of metamorphosis (Aly et al., 2010). Meanwhile, desert locust nymphs treated with *Nerium oleander* leave extracts could not molt, had reduced food intake, and had a cumulative mortality rate greater than 50% from the fourth day of application and 100% mortality at the 12th day of application (Bagari et al., 2013). Extracts derived from different parts of the *Rhizophoramucronata* have insecticidal and antifeedant activity against the locust (Kabaru & Gichia, 2001). *Melia volkensii* extracts also have a growth inhibiting anti-feeding properties against desert locust and other insect pests (Jaoko et al., 2020). Field tests have shown that the crude powder of *M. volkensii* has effective control resulting from acute toxicity, retarded growth, and 80% malformations, which led to 100% mortality after 14 days (Diop & Wilps, 1997).

Several studies have confirmed that extracted oils are effective against locusts and could be used as natural controls. A novel mixture of plant oils that had high toxic effects on desert locust after a single spray treatment was developed between combination of three plant oils of *Carum carvi*, *Citrus aurantium dulcis*, and *Gaultheriaprocumbens*. Interestingly, a mean mortality rate of 80% was observed within 24 h after treatment (Abdelatti & Hartbauer, 2019). Furthermore, oils extracted from different plant species were tested against desert

locust. Oil from *Allium cepa* proved to be the most toxic to the locusts with 67.76% mortality rate followed by *Petroselinum sativum* with 60.03% Other plants studied were *Pelargonium radula* (56.21%) *Cuminum cyminum* (42.42%) *Ocimum basilicum* (51.31%), *Origanum vulgare* (38.55%) and *Matricaria chamomilla* (31.66%) which showed different mortality rate against the locust (Mansour et al., 2015).

These botanicals have shown their potential as bio-pesticides but are still at the experimental stage as far as locust control is concerned. Hence, for this current study, the extraction of moringa oleifera and ceberra odullam from family of *Moringaceae* and *Apocynaceae*, respectively are studied towards mortality rate of locusts. Table 2.1 summarized the lists of plants species that have been tested as biopesticide for locusts.

Scientific name	Common name	Family	Mortality rate	Duration of treatment	Reference
			(%)	(h)	
Azadirachta indica (A. Juss.) Brandis	Neem	Meliaceae	43.39	168	(Hamadah et al., 2013)
Jatropha curcas L.	Physic nut	Euphorbiaceae	40.54	168	Kearney et al. (1994)
Solenostemma argel (Delile) Hayne	Argel	Asclepiadaceae	20.70	168	Bashir et al. (2014)
Nigella sativa L.	Black seed/cumin	Ranunculaceae	-	-	Schmutterer et al. (1993)
Calotropis procera (Aiton) W.T. Aiton	Sodom apple	Apocynaceae	_	72 - 360	Abbassi et al. (2015)
Calotropis procera (Aiton) W.T. Aiton	Sodom apple	Apocynaceae	100	5 - 7	Abdellah et al. (2013)
Peganum harmala L.	Wild rue	Nitrariaceae	100		
Peganum harmala L.	Peganum harmala L.	Nitrariaceae	100	168	
Calotropis procera (Aiton) W.T. Aiton	Sodom apple	Apocynaceae	100	192	Kaidi et al. (2017)

Table 2.1List of plant species that have been tested as biopesticide for locusts

Table 2.1	Continued				
Scientific name	Common name	Family	Mortality rate	Duration of treatment	Reference
			(%)	(h)	
Schouwia purpurea	Saharan amaifan	Dressionen	42.2		
(Forssk.) Schweinf	Saharan crucifer	Brassicaceae	43.3		
Zizyphus lotus (L.) Desf.	Jujube	Rhamnaceae	40	-	Messgo-Moumene et
Calotropis procera (Aiton)	Sodom onnio	A n o o y n o o o o o	93.3	360	al. (2015)
W.T. Aiton	Sodom apple	Apocynaceae	93.5		
Ammi visnaga (L.) Lam	Toothpick weed	Apiaceae	34.9 - 95.0	-	Ghoneim et al.
					(2014)
Fagonia bruguieri D.C	Fagonia	Zygophyllaceae	40 - 50	24	Aly et al. (2010)
Nerium oleander L.	Oleander	Apocynaceae	50 - 100	288	Bagari et al. (2013)
Rhizophora mucronata Lam.	Mangrova	Managana	96	48	Kabaru & Gichia
	Mangrove	Rhizophoraceae	90	40	(2001)
Melia volkensii Giirke	Melia	Meliaceae	100	336	Diop & Wilps (1997)

Table 2.1	Continued				
Scientific name	Common name	Family	Mortality rate	Duration of treatment	Reference
			(%)	(h)	
Carum carvi L.	Caraway	Apiaceae			
Citrus aurantium L.	Orange	Rutaceae		24	Abdelatti &
Gaultheria procumbens L.	Wintergreen	Ericaceae	80-100	24	Hartbauer (2019)
Allium cepa L.	Onion	Amaryllidaceae	67.76		
Petroselinum sativum	Parsley	Apiaceae	60.03	-	
Hoffm.	T arsiey	Aplaceae	00.05		
Pelargonium radens H.E.	Radula	Geraniaceae	56.21	-	
Moore	Kauula	Gerainaceae	50.21		
Cuminum cyminum L.	Cumin	Apicaceae	42.42	- 25 - 50	Mansour et al. (2015)
Ocimum basilicum L.	Basil	Lamiaceae	51.31	25 - 50	wansour et al. (2013)
Origanum vulgare L.	Oregano	Lamiaceae	38.55	-	
Matricaria chamomilla L.	Chamomile	Asteraceae	31.69	-	

2.4 Moringa Oleifera

Moringa oleifera (M. oleifera) is a tropical and subtropical plant that belongs to the Moringaceae family. Commercial cultivation occurs in India, Africa, South and Central America, Mexico, Hawaii, and throughout Asia and Southeast Asia. It is known as the drumstick tree because of the shape of its immature seed pods, the horseradish tree because of the flavour of ground root preparations, and the ben oil tree because of the oils generated from its seeds. Immature seed pods are consumed in some locations, while the leaves are commonly used as a basic food because to their high nutritional content (Stohs & Hartman, 2015). Moringa oil may also be a good replacement for oliveoil in the diet, as well as for non-food applications such as biodiesel, cosmetics and equipment lubricant. Furthermore, following oil extraction, the seed cake can be utilised as a natural coagulant in waste water treatment or as an organic fertiliser to increase agricultural production (Stohs & Hartman, 2015). Moringa oleifera is a versatile plant that not only offers sustenance to animals but also serves as a fantastic alternative forthe treatment of a variety of ailments. Moringa treatment in the lab has proven to be quite effective and has numerous favourable effects on crops such as increased growth, resulting in vigorous plant development, and resistance to pests and diseases (Manzoor et al., 2015).



Figure 2.8 Moringa oleifera

Moringa extract can improve plant metabolism due to the presence of zeatin, ascorbic acid, phenolic compounds, and vitamin E. Moringa oleifera extract has been

shown to be an effective water purifier and has been utilised as a natural coagulant, medicinal plant, green manure, fresh vegetable, biogas, livestock feed, and bio-pesticide (Shah et al., 2017). It is also involved in cell elongation and division and is utilised as a seed priming agent for maize and sunflower as well as a plant growth promoter (Manzoor et al., 2015; Samada & Tambunan, 2020). Furthermore, moringa extracts demonstrated bioinsecticidal potential by altering the morphology of the digestive system, inhibiting trypsin activity, misbalancing digestive enzymes and interfering with egg hatchability (Gbaye et al., 2018).

The study on extraction of moringa has been evaluated in various field especially as biopesticide against insects or its effect on the plants itself. Manzoor et al. investigated the potential of moringa oleifera as a biopesticide against wheat aphids and possible impacts as growth regulator in wheat crop. Results showed statistically significant increase in crop growth traits and reduction in aphid infestation (booting, milk, and heading stage). The authors concluded that Moringa oleifera leaf and root extracts are effective plant growth regulators and biopesticides against different chewing and sucking insect pests, especially wheat aphids. Moringa leaves contain several macro and micro elements which are important plant growth enhancers leave no hazardous residual effects.

Shah et al. 2017 compared the performance of neem seed extract and moringa leaf extract in the control of aphid in the wheat fields. The combination of moringa extract with chemical pesticides imidacloprid reduced significantly aphids infestation compared to the other treatments, hence resulting in higher yield. The percentages of yield increase in imidacloprid and moringa treated plots over the control were 19.15–81.89% for grains per spike and 27.59–61.12% for yield kg. The authors suggested further studies should consider the side effects of biopesticides on non-target organisms in order to provide better management practices in the field. It also necessary to study moringa extract as sole biopesticide without combining with other type of commercial chemical pesticides (Shah et al., 2017).

The combination of commercial moringa and jojopa extraction oils was tested against grasshoppers within 12 days of treatment by Soltan (2020). Mortality percentages were calculated between time interval of 2 days for total of 12 days. The results indicated that the mortality percentages of grasshoppers were 96% for moringa and 65% for jojopa oils. The efficacy of moringa oil can be further study for different part of moringa such as its leaves and seeds for locust's elimination (Soltan, 2020).

2.5 Cerbera Odollam

Cerbera odollam is a poisonous dicotyledonous angiosperm in the Apocynaceae family. Cerbera odollam thrives in coastal salt swamps and creeks in southern India, as well as along riverbanks in southern and central Vietnam, Cambodia, Sri Lanka, Myanmar, Madagascar, and Malaysia (Kansedo & Lee, 2013). The poisonous portion of the plant is the kernel, which is found at the centre of the fruit. Cerberin, a cardiac glycoside of the cardenolide class, is the plant's bioactive toxin. Cerberin has a similar mechanism of action to digoxin, hence Cerbera odollam intoxication generates symptoms comparable to acute digoxin poisoning. When its kernel is consumed, it produces nausea, vomiting, hyperkalemia, thrombocytopenia, and ECG abnormalities. Other active glycosides found in the kernel, besides cerberin, include odollin and cereberoside. However, these compounds have received little attention, and their mode of action is based on their structure. Cerberin, a cardiac glycoside of the cardenolide class, appears to be the major toxin (Menezes et al., 2018).



Figure 2.9 Cerbera Odollam

Cerbera odollam has few domestic uses. In India, the plant's milky white latex is known for its laxative, emetic, and irritating effects. Some of the plant's components are used to make fibre. Furthermore, there have been instances of humans consuming the leaves and bark for their cathartic qualities in several regions. Burmese burn oil is extracted from its seed and is known for its bright flame and nice nut-like aroma. They also use the oil as a cosmetic and combine it with other oils to make an insect repellent or insecticide (Menezes et al., 2018).

Although considerable research has been conducted on this plant, it is still regarded limited because some of the research has concentrated on the toxicity of this plant. Somsroi and Chaiyong investigated the effect of cerbera odollam extract on common cutworm, an insect pest that has spread and harmed many key crops. Within 24 hours of observation, the extraction of cerbera odollam leaf dipping process revealed that the proportion of death at concentrations of 1, 5, 10, and 30% was 13.33, 80.00, 93.33, and 100%, respectively. Because of its high insecticidal efficiency, the authors suggested that this species of plant be used more extensively in the management of insect pests (Somsroi & Chaiyong, 2016).

Furthermore, Purwani, et al. studied cerbera odollam leaf extract as bioinsecticide in several concentration to control *Spodopteralitura F* (*S. litura F*) knownas one of the chili pests with its polyfag character. *S.litura. F* mortality and its developments were observed for 20 days. *S. litura F* were reached 75 % in mortality level and got weight loss, with 2% leaf extract dose of cerbera odollam for 8 days observation time (Purwani et al., 2014).

Both moringa oleifera and cerbera odollam are considered an attractive alternative for extraction of biopesticides due to its availability and characteristics. Hence, this work is focused on the extraction of both plants to control and manage locusts' population.

CHAPTER 3

METHODOLOGY

3.1 Introduction

This chapter presents the detailed description of materials, equipment, extraction methods and data analysing of locusts' elimination. The overall experimental work in this study is summarized in Figure 3.1. Firstly, both cerbera odollam and moringa oleifera is collected and continued with the extraction process to produce pesticides. The detailed procedure of extraction process is explained in Section 3.3. Then, about 90 number of locusts are captured at local community for research purpose. Data collection and analysis conducted on the effectiveness of pesticides extracted from these in terms of mortality rate.

The overall experimental work in this study is summarized in Figure 3.1. The collection of both cerbera odollam and moringa oleifera was prepared around Kuantan, Pahang before employing oil extraction process using ethanol as solvent. Then, a number amounts of locusts was collected and evaluation on the effect of aforementioned oil extraction on locusts was conducted. Based on the performance of cerbera odollam and moringa oleifera oil extraction, the analysis was performed and lastly, conclusion and recommendation were evaluated.

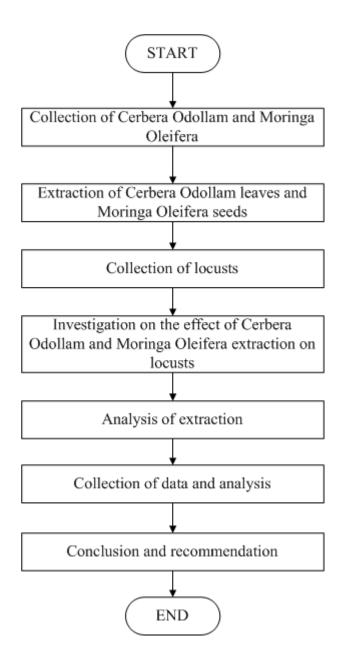


Figure 3.1 Overall experimentl flow chart for this study

3.2 Materials

3.2.1 Chemicals

The chemical used in this study is listed below as shown in Table 3.1. The part of cerbera odollam and moringa oleifera was used in this study is leaves and seeds,

correspondingly. Meanwhile, ethanol was employed as the main solvent for the oil extraction process in this research.

Table 3.1List of materials and chemicals

Chemicals/Materials	Application
Cerbera Odollam leaves	Extraction process
Moringa Oleifera seeds	Extraction process
Ethanol	Extraction process

3.2.2 Equipment

There is several equipment involved in to evaluate the outcome of this research along with its application as shown in Table 3.2. The equipment listed are available in FTKA and FTKKP laboratory UMP.

Equipment	Application	
Oven	Drying leaves and seeds	
Blender	Blend leaves and seeds	
Beaker	Storage of sample	
Aluminium foil	Sample cover	
Filter paper	Extraction process	
Round flask	Extraction process	
Rotary evaporator	Extraction process	
pH meter	Checking pH	

Table 3.2List of equipment

3.3 Experimental

3.3.1 Collection of Locusts

Locusts were collected at Lorong Mawar Kampung Mahkota Kuantan, Pahang area. About 100 locusts kept in a clear plastic container with fresh grass, soil, water and supplied with air. The container is divided into three section namely, distilled water, moringa and cerbera filled with approximately 30 locusts each.

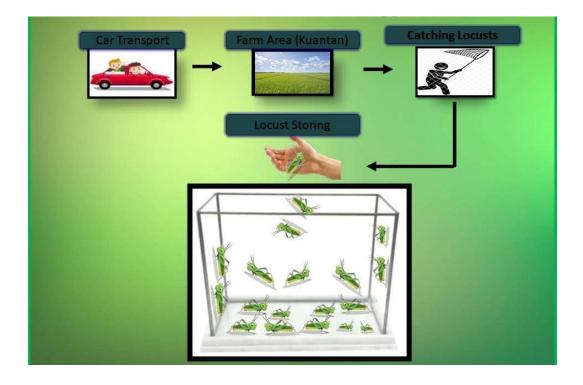


Figure 3.2 Schematic diagram of locusts collection and storage

3.3.2 Extraction of Cerbera Odollam and Moringa Oleifera

The cerberra odollum were collected at location near Taman Gelora, Kuantan. The Cerberra odollam crude extraction method was done by following the procedure described by Somsroi and Chaiyong. 2016. The leaves were washed with water and cut into small pieces. The leaves were left under sunlight for 3 h and later dried in the hot air oven at 70°C until it completely dried. The sample was blend with blender until it turns into powder. The extraction was carried out by soaking with 200 ml of ethanol (95% purity) for 2 days. After 2 days, the oil extraction was collected by separating the oil with filter paper. Then, the extracts were evaporated to remove the solvent under reduced pressure using rotary evaporator for 2 hr. The leaves crude extracts collected were kept under room temperature before further evaluation.

The moringa oleifera seeds were obtained around UMP Gambang. The extraction of moringa were following with same method as extraction of cerbera odollam. After the seeds had been cleaned thoroughly, the seed are dried in an oven at 70°C. After that, the seeds were put in a blender to get powder form. The powder was then put in a beaker and filled with 200ml ethanol and left for 2 days. The oil was collected by using filter paper after 2 days. The sample was then placed in rotary evaporator for 2 hr. The oil extracts were retained before testing under room temperature.

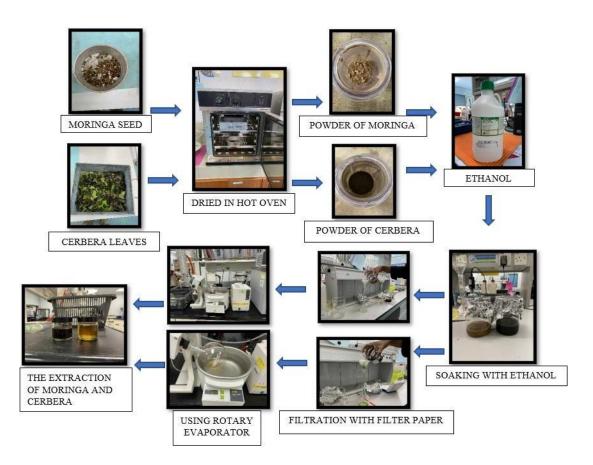


Figure 3.3 Extraction process of Moringa and Cerbera oils

3.4 Analysis of Extraction Oil

3.4.1 Fourier-Transform Infrared (FTIR) Spectroscopy

The presence of functional groups product extraction from cerbera odollum and moringa oleifera was identified by Fourier-Transform Infrared (FTIR) spectroscopy measurement. The FTIR spectra of support and catalysts was recorded in the range of 400-4000 cm⁻¹ and spectral resolution of 4 cm⁻¹. The number of scans of 100 was employed on a Thermo Fisher Scientific FTIR spectrometer. Prior to measurement, a background check was conducted for each run on the ATR sample holder and the crystal plate was cleaned with acetone and dried for 15 min to completely remove contamination. The spectra for the samples were interpreted using OMNIC software.

3.5 Testing Method

To evaluate the potential of cerbera and moringa extracts as biopesticides, a container filled with 30 locusts in three main section and labelled as blank, moringa and cerberra. Each section was spray with around 10 ml of distilled water, moringa and cerberra extracts. The mortality of the locusts was observed for every 15 minutes. The number of locusts survived, and dead were recorded accordingly until 150 min. The data was tabulated based on its mortality rate and means rate.

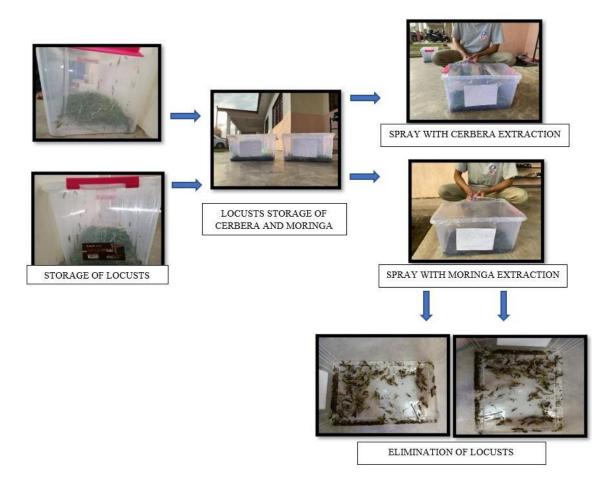


Figure 3.4 Testing Method

3.6 Equation and Data Analysis

3.6.1 Testing and Unit of Method

The study undergoes few methodologies which involved parameters. Each parameter has its own unit listed as below.

Table 3.3Unit for each method

Method	Unit	
Mortality rate calculations	%	
• Percentage observed mortality		
	• Being 7 are neutral	
pH	• Less than 7 are acidic	
	Greater than 7 are alkaline (basic)	

3.6.2 Mortality Rate Calculation

The mortality rates were calculated using Eq. (3.1) where percentage of observed mortality individuals was estimated from total number of dead divide by total number of treated individuals (Abdelatti & Hartbauer, 2019).

% of observed mortality individuals =
$$\frac{Total \ number \ of \ dead}{Total \ number \ of \ treated \ individuals} \times 100$$
 3.1

3.6.3 T-test Analysis

The t-test is a statistical test for comparing the means of two groups. It's frequently used in hypothesis testing to see if a method or treatment influences the population of interest, or if two groups vary from one another. The ratio of the difference in group means over the pooled standard error of both groups is used in the t-test to evaluate the genuine difference between two group means. This t-test analysis is also called a correlated pairs t-test, a paired samples t-test or dependent samples t-test is where you run a t test on dependent samples. Dependent samples are essentially connected, and they are tests on the same person or thing. The null hypothesis for the independent samples t-test is $\mu 1 = \mu 2$. In other words, it assumes the means are equal. With the paired t test, the null hypothesis is that the pairwise difference between the two tests is equal (H0: $\mu d = 0$). The difference between the two tests is very subtle; which one you choose is based on your data collection method. The significant acceptance value is alpha = 0.05. This t-test analysis is carried out by using Microsoft Excel to determine the results.

In this project the null hypothesis is there are a difference in the value of distilled water, Moringa and Cerbera. The alternate hypothesis is there are no difference in the value of distilled water, Moringa and Cerbera.

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Introduction

The performance of moringa and cerbera oil extraction was evaluated in this Chapter 4. The analysis and assessments of oil extraction was divided into 3 categories namely, distilled water, moringa and cerbera. The observation period was within 15 min time interval for 150 min. The data was presented according to its mortality and means rate. The FTIR analysis for both oil extraction also explained in this chapter.

4.2 Evaluation of Moringa Oleifera and Cerbera Odollam Oil Extraction

Table 4.1 and Figure 4.1 illustrates the observation of locusts for 150 min with introduction of distilled water. This sample recorded no mortality rate since all 30 locusts were survived for 150 min observation period. Distilled water acts as blank tests for this study to ensure that no outside contamination in the container (Shanmugavel et al., 2018). Similar trend was reported by Moodley et al. as water was used as blank sample in against antimicrobial potential where not changes were observed within the evaluation interval (Moodley et al., 2018).

Distilled Water				
Time (m	in) Locu	ist Survived		
15		30		
30		30		
45		30		
60		30		
75		30		
90		30		
105		30		
120		30		
135		30		
150		30		

Table 4.1Locusts survived with distilled water within 150 min

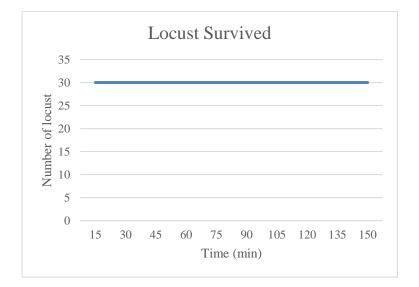


Figure 4.1 Graph of locust survived with distilled water within 150 min

The performance of moringa oleifera oil extraction was summarized in Table 4.2 and Figure 4.2. The changes in the amounts of locusts survived with every 15 min observation period are as follows; 27 > 24 > 20 > 19 > 17 > 15 > 10 > 4 > 0. No locusts were recorded survived after 135 min when exposed to moringa oleifera oil extraction.

This sample also achieved 100% mortality rate within 150 min observation period. In addition, the performance of oils extraction by using ethanol is comparable with the commercialized moringa essential oils against grasshoppers reported by Soltan. (2020). Moringa oils also reported effective to reduce or inhibit other types of pests managements such as wheat aphid (Manzoor et al., 2015; Shah et al., 2017). It was worth noted that even though moringa achieved complete mortality rate on locusts for this study, moringa oil preserved the plants inside the locust's storage. Moringa extract possessed macro and micro elements that important to conserved the plants but acts as efficient biopesticides. Moringa oils also has pH of 6.81 close to 7. This phenomenon further confirms its characteristic as eco-friendly biopesticides (Farooq et al., 2010; Manzoor et al., 2015).

Locust Survived
27
24
20
19
17
15
10
4
0
0

Table 4.2Locusts survived with moringa oleifera extraction within 150 min

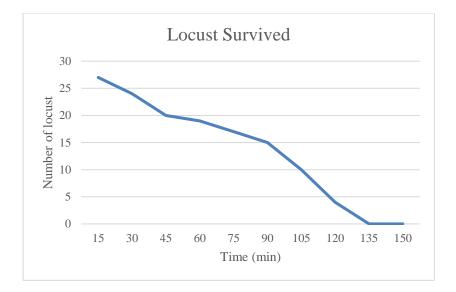


Figure 4.2 Graph of locust survived with moringa oil extraction within 150 min

Table 4.3 and Figure 4.3 illustrates the effect of cerbera oil extraction against locusts. The number of locusts were reduced every 15 min for 150 min observation period with this particular order; 27 > 22 > 18 > 13 > 7 > 2 > 0. Cerbera oils recorded 100% mortality rate after 105 min. Cerbera oil extraction also reported high performance to control other insect pests such as common cutworm with a complete mortality rate (Somsroi & Chaiyong, 2016). The conditions of plants inside locust's storage recorded similar observation with moringa oil. Toxic effects of cerbera oils are absorbed by the insects by stomach and contact toxin. Stomach toxin is a type of insecticide that is eaten by insects and kills the insect in particular by destroying or absorbing the digestive system. Contact toxin is a type of insecticide that is absorbed through the body wall so that the insect has to feel in direct contact with the insecticide (Sholahuddin et al., 2018). Moreover, it was reported that cerbera leaves contain flavonoids, saponins, cerberin and alkaloids compound which is multi-benefits to eliminate pests or insects but not effecting the green environments (Menezes et al., 2018; Sholahuddin et al., 2018). The pH recorded for cerbera oil is 6.82 which is close to natural state of 7 to further validate its environmentally friendly characteristics.

Cerbera Extraction				
Time (min)	Locust Survived			
15	27			
30	22			
45	18			
60	13			
75	7			
90	2			
105	0			
120	0			
135	0			
150	0			

Table 4.3Locusts survived with cerbera odollam extraction within 150 min

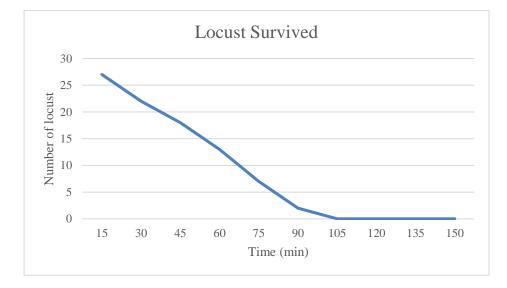


Figure 4.3 Graph of locust survived with cerbera oil extraction within 150 min

The comparisons of distilled water, moringa and cerbera was recorded in Table 4.4. Both moringa and cerbera recorded a complete mortality rate with 100%. Moringa and cerbera oils regarded as eco-friendly biopesticides since it preserved the condition of plants and grass in which only effecting locust's mortality. This situation most likely

because of macro and micro elements in moringa and cerbera to improve biopesticides effectiveness. The close to natural pH (6.81 and 6.82 for moringa and cerbera, correspondingly) also contribute to its green-friendly pesticides. Furthermore, cerbera extraction showed faster mortality rate in contrast to moringa. This is owing to higher adsorption rate of cerbera extraction oils than moringa. The active toxins and elements in cerbera and moringa facilitated the mortality rate of locusts in this study. Nevertheless, both extraction sample demonstrated excellent performance in locust's elimination within short observation period.

Time (min)		Locust Survived	
1 mie (mm)	Distilled water	Moringa	Cerbera
15	30	27	27
30	30	24	22
45	30	20	18
60	30	19	13
75	30	17	7
90	30	15	2
105	30	10	0
120	30	4	0
135	30	0	0
150	30	0	0

Table 4.4Locusts survived between distilled water, moringa and cerbera oilextraction within 150 min

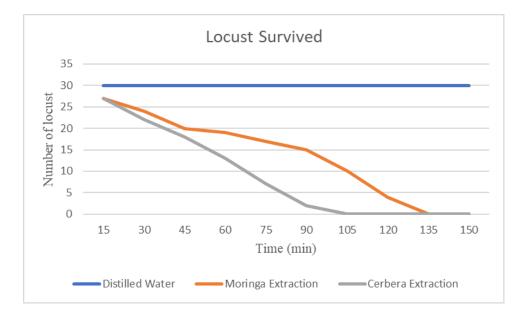


Figure 4.4 Graph of locust survived with distilled water, moringa oil and cerbera oil extraction within 150 min

4.3 T-test Analysis

The results analysis for the testing experiments using the t-test paired two samples for means. This type of analysis was used because it is the simplest analysis and suitable for all the testing samples. The t-test analysis paired two samples for means is also, capable to determine the efficacy and accuracy of the system and the sensors used for this system.

For the distilled water, Moringa extraction and Cerbera extraction the testing was carried out with 10ml of sample. The samples were tested with the 30 locusts for the period of 150 minutes with every 15 minutes interval of collected data. The collected data was then analysed by using t-test analysis to determine the effectiveness of the different sample.

The paired-samples t-test was conducted to compare the locusts survived from the different sample which are distilled water, Moringa extraction and Cerbera extraction. There was a significant difference in the locusts survived from the testing results. These results suggest that Cerbera extraction is more effective than Moringa extraction for

locusts elimination because the Cerbera extraction absorb faster than Moringa to the locusts.

Table 4.5T-test pair with distilled water and moringa extraction for means

Ho: There is no significant different in locusts survived between distilled water and Moringa Ha: There is a significant different in locusts survived between distilled water and Moringa

Elements	T-test: Paired two samples for means		
Elements	Distilled water	Moringa	
Mean	30	13.6	
Variance	0	94.0444444	
Observations	10	10	
Person Correlation	-		
Hypothesized Mean			
Difference	1		
df	9		
t Stat	5.02173913		
P (T=t) one-tail	0.000358694		

Conclusion: Since P value < 0.05, fail to accept the null hypothesis. There are strong statistical evidence shows that there is a difference in locusts survived between distilled water and moringa.

Table 4.6 T-test pair with distilled water and cerbera extraction for means

Ho: There is no significant different in locusts survived between distilled water and Cerbera

Ha: There is a significant different in locusts survived between distilled water and Cerbera

Elements	T-test: Paired two samples for means			
Elements	Distilled water	Cerbera		
Mean	30	8.9		
Variance	0	107.4333333		
Observations	10	10		
Person Correlation	-			
Hypothesized Mean				
Difference	1			
df	9			
t Stat	6.132344324			
P (T=t) one-tail	8.6179E-05			

Conclusion: Since P value < 0.05, fail to accept the null hypothesis. There are strong statistical evidence shows that there is a difference in locusts survived between distilled water and Cerbera.

Table 4.7 T-test pair with distilled water and cerbera extraction for means

Ho: There is no significant different in locusts survived between Moringa and Cerbera

Ha: There is a significant different in locusts survived between Moringa and Cerbera

Elements	T-test: Paired two samples for means		
	Moringa	Cerbera	
Mean	13.6	8.9	
Variance	94.0444444	107.4333333	
Observations	10	10	
Person Correlation	0.887197987		
Hypothesized Mean			
Difference	1		
df	9		
t Stat	2.433251158		
P (T=t) one-tail	0.018890277		

Conclusion: Since P value < 0.05, fail to accept the null hypothesis. There are strong statistical evidence shows that there is a difference in locusts survived between distilled Moringa and Cerbera.

4.4 Fourier-Transform Infrared (FTIR) Spectroscopy

The FTIR spectra for both cerbera odollam and moringa oleifera are shown in Table 4.8 And Figure 4.5, respectively. FTIR spectra illustrates similar trend for both sample with broad absorption band ranging from 3379 to 3255 cm⁻¹ was assigned to alcohol and hydroxyl group (N-H, O-H) stretching. The peaks observed at 2976 cm⁻¹ indicates the presence of alkanes group (C-H). The peaks around 1643 to 1644 cm⁻¹ attributed to tertiary amines (N-H). In addition, the peak at 1382 cm⁻¹ for both samples shows the presence of aromatic amines (C-N). There is also visible peak at 1085 and a long and sharp peak at 1043 cm⁻¹ assigned to C-O group in alcohol hydroxyl. Another sharp peak was detected around 877 cm⁻¹ represent stretch C-H group of alkenes (Ashokkumar & Ramaswamy, 2014; Deepashree et al., 2012). The presence of these bond on FTIR analysis also in agreement with other studies on extraction of moringa (Moodley et al., 2018; Shanmugavel et al., 2018).

	•	•	
Wavenumber	Cerbera	Moringa Oleifera	Bond
	Odollam		
3450-3550 3340.0	2240.0	3345.59	Alcohol and Hydroxyl
	5545.59	group (N-H, O-H)	
2918-2990	2976.77	2976.64	Alkanes group (C-H)
1535-1660	1644.54	1643.08	Tertiary Amines (N-H)
1036-1385	1382.87	1382.8	Aromatic Amines (C-N)
1020-1220	1085.4	1085.72	C-O Group
1020-1220	1043.62	1043.69	C-O Group
805-897	877.74	877.74	Stretch of Alkenes (C-H)

Table 4.8Summary of bond existed in moringa and cerbera oil extraction

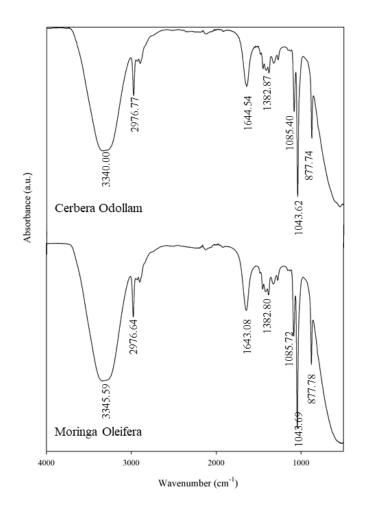


Figure 4.5 FTIR spectra for moringa oleifera and cerbera odollam oils extraction

CHAPTER 5

CONCLUSIONS AND RECOMMENDATIONS

5.1 Conclusions

Botanical pesticides are considered as an attractive alternative to synthetic chemical pesticides for locusts' management credited to its reduction of the significant effect towards environments and human health. Thus, the oil extraction from moringa oleifera and cerbera odollam in elimination of locusts was investigated in this study. Moringa seeds and cerbera leaves were extracted using ethanol extraction technique. The oil extraction was then tested to a number of locusts based on its mortality rate. Both moringa and cerbera recorded 100% mortality rate. Moringa and cerbera also preserved the plants and grass inside the locust's storage. Both extracts have toxins and elements that significant to protect the plants but acts as efficient biopesticides. The naturality of both moringa and cerbera based on pH measurements further confirming its eco-friendly qualities. Cerbera oils recorded faster time to eliminate all 30 locusts in around 105 min, whereas, moringa oils eliminate all locusts within the first 135 min with total of 150 min of observation period. The slight difference in the performance of both plants due to high absorption rate of cerbera oils compared to moringa. The results analysis for the testing experiments is using the t-test paired two samples for means. For the distilled water, Moringa and Cerbera the testing is carried out on 30 locusts. The paired-samples t-test was conducted to compare the number of locusts survived. Results from the statistical analysis shows that there is strong statistical evidence that there is a difference in number of locusts survived between sample distilled water, Moringa and Cerbera. It indicates that Cerbera more effective compared between to Moringa because of high absorption rate.

Summary from FTIR analysis conducted on both oils' extraction confirm the presence of several components namely, alcohol and hydroxyl, alkanes, tertiary amines, aromatic amines, C-O group and C-H group of alkenes. The extraction of oils from

moringa and cerbera is considered very valuable due to its eco-friendly, cost-friendly, simple preparation method with high mortality rate of locusts.

5.2 Recommendations

According to the outcomes from this study, the following recommendations may be suggested for future research:

For this work, moringa and cerbera were extracted using ethanol as solvent. In fact, selection of solvent is reported as one of factors that influence the oil extraction process. Thus, several solvents such as methanol and water are recommended in the future study.

Concentration of extracted oil has been reported before in biopesticides of pest managements. Hence, the extraction of different moringa and cerbera concentration are recommended in future research to discover its optimal concentration and improve this current work.

Further analysis of oils extraction products should be conducted for future review to study the elements existed in moringa and cerbera oils. Analysis such as X-ray fluorescence spectroscopy (XRF), inductively coupled plasma-optical emission spectroscopy (ICP-OES) and gas chromatography-mass spectrometry (GCMS) are suggested to extend the validity of this research.

Both Moringa oleifera and cerbera odollam plants reported high performance for elimination of locusts with high mortality rate. Therefore, other part of these plants most likely contributes to similar impact. Other parts of plansts such as moringa leaves, cerbera seeds or kernels could be taken into consideration for future research.

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APPENDICES

Appendix A:

Locusts Collection



Appendix B:

Locusts Storage



Appendix C:

Mortality rate calculation

% of observed mortality individuals = $\frac{Total \ number \ of \ dead}{Total \ number \ of \ treated \ individuals} \times 100$ B.1

B.1 Moringa oil extraction

 $\frac{Total \ number \ of \ dead}{Total \ number \ of \ treated \ individuals} \times 100$ $= \frac{30}{30} \times 100$ $= 100 \ \% \ mortality \ for \ moringa \ oil$

B.2 Cerbera oil extraction

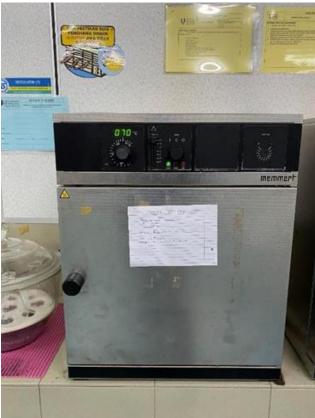
$$\frac{Total number of dead}{Total number of treated individuals} \times 100$$
$$= \frac{30}{30} \times 100$$
$$= 100 \% mortality for cerbera oil$$

Appendix D:

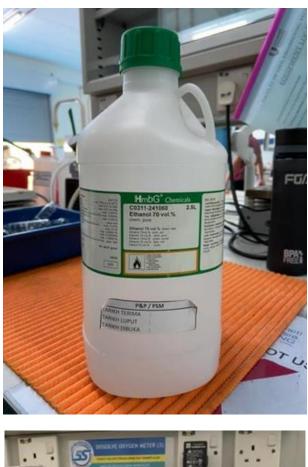
Extraction Process

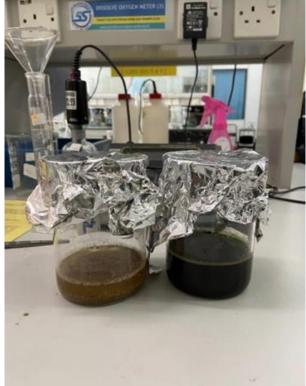










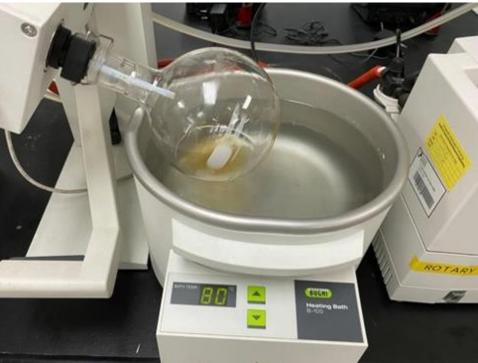




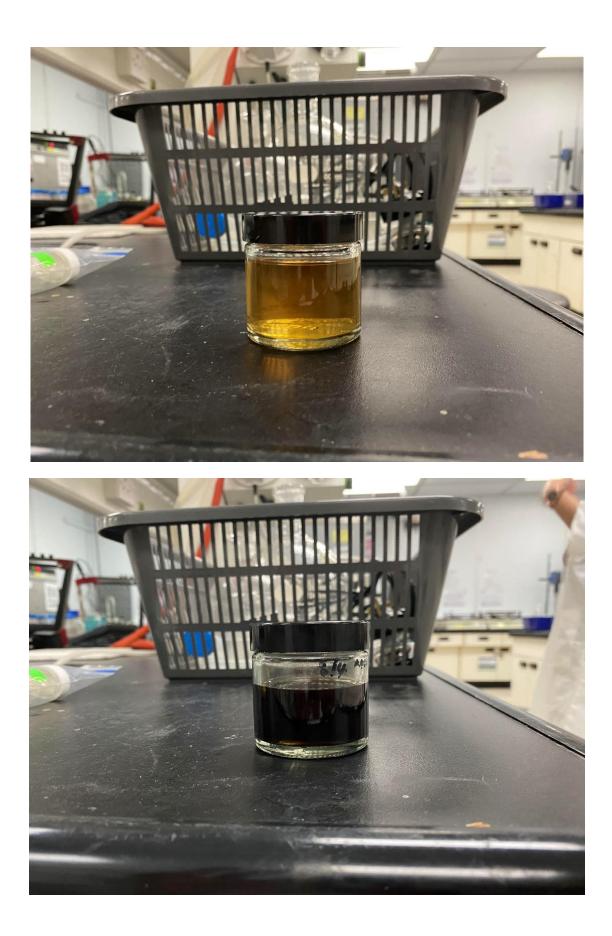












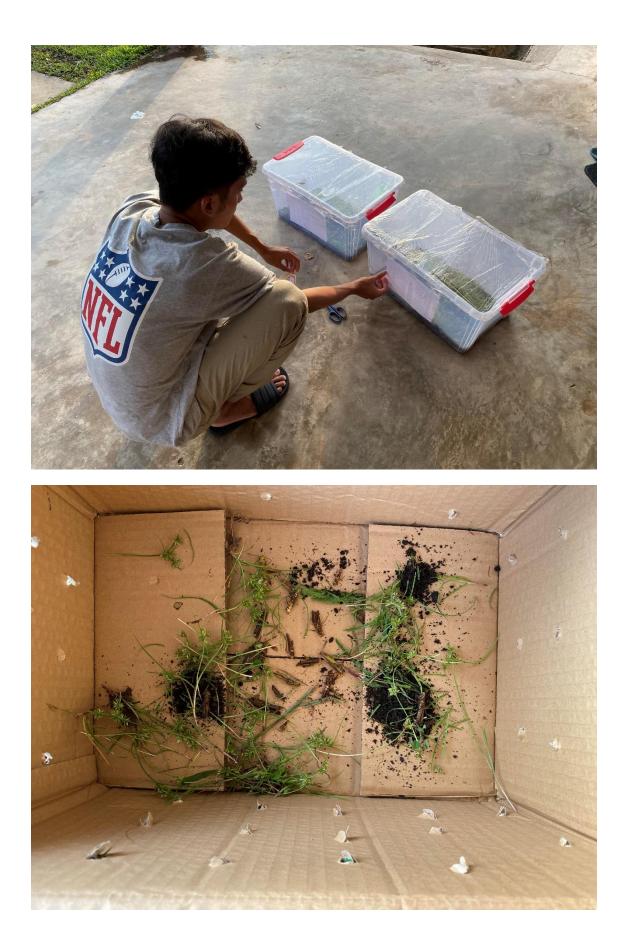


Appendix E:

Testing Method











Appendix F:

pH Meter





