



# OPTIMISATION AND CHARACTERISATION OF **BIODIESEL FROM BIODEGRADATION** OF ORGANIC WASTES BY BLACK SOLDIER FLY LARVAE



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# UNIVERSITI PENDIDIKAN SULTAN IDRIS

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### ABSTRACT

This research aimed to optimise and characterise environmental friendly biodiesel produced through biodegradation of organic wastes by black soldier fly (Hermetia illucens) larvae. This research is divided into four main studies, namely characterisation of organic wastes, cultivation of insect larvae, characterisation of insect larval lipid and determination of the properties of insect larval biodiesel. The organic wastes used for cultivation of insect larvae were food kitchen waste (FKW), soya residue (SR) and mixed waste (MW). The main scientific instruments used in this study were Fourier transform infrared (FTIR) spectrometer, nuclear magnetic resonance (NMR) spectrometer and gas chromatography-flame ionisation detector (GC-FID). For organic wastes and insect larvae characterisation, several analyses such as content of moisture, protein, carbohydrate, ash and fat, pH value and crude fiber were carried out. The extraction of larval lipid was conducted using Soxhlet method and underwent a two-step transesterification process to produce biodiesel. The highest yield of biodiesel (95.80%) was from black soldier fly larval lipid as a result of fed by FKW followed by SR (90.26%) and MW (90.25%). FTIR and NMR analyses confirmed the successful transformation of larval lipid to biodiesel by the appearance of the fatty acid methyl ester (FAME) functional groups in the spectra. While, GC-FID analysis showed the FAME composition of biodiesel comprised of both saturated (lauric acid, myristic acid, palmitic acid, capric acid and stearic acid) and unsaturated (oleic acid, palmitoleic acid and linoleic acid) FAME. The produced insect larval biodiesel met the value recommended by American Society for Test and the Materials (ASTM) D6751 and European (EN) 14214 standards. In conclusion, black soldier fly larvae were able to convert organic wastes studied to biodiesel. In implication, the insect larvae can be potentially applied as a low-cost biodiesel feedstock for reducing the operational cost of biodiesel production.

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### PENGOPTIMUMAN DAN PENCIRIAN BIODIESEL DARIPADA BIODEGRADASI SISA ORGANIK OLEH LARVA LALAT ASKAR HITAM

#### ABSTRAK

Kajian ini bertujuan untuk mengoptimumkan dan mencirikan biodiesel mesra alam terhasil melalui biodegradasi sisa organik oleh larva lalat askar hitam (Hermetia illucens). Penyelidikan ini dibahagikan kepada empat kajian utama, iaitu pencirian sisa organik, pemeliharaan larva serangga, pencirian lipid larva serangga dan penentuan sifat-sifat biodiesel larva serangga. Sisa organik yang digunakan untuk pemeliharaan larva serangga adalah sisa makanan dapur (FKW), hampas soya (SR) dan campuran sisa (MW). Peralatan saintifik utama yang digunakan dalam kajian ini ialah spektrometer inframerah transformasi Fourier (FTIR), spektrometer resonans magnet nukleus (NMR) dan kromatografi gas-pengesan nyala pengionan (GC-FID). Bagi pencirian sisa organik dan larva serangga, beberapa analisis seperti kandungan lembapan, protein, karbohidrat, abu dan lemak, nilai pH dan serat kasar telah dijalankan. Pengekstrakan lipid larva telah dilakukan menggunakan kaedah Soxhlet dan telah menjalani suatu proses transpengesteran dua langkah untuk menghasilkan biodiesel. Hasil biodiesel tertinggi (95.80%) adalah dari lipid larva lalat askar hitam hasil suapan FKW diikuti SR (90.26%) dan MW (90.25%). Analisis FTIR dan NMR mengesahkan kejayaan transformasi lipid larva kepada biodiesel dengan kehadiran kumpulan-kumpulan berfungsi asid lemak metil ester (FAME) di dalam spektra. Manakala, analisis GC-FID telah menunjukkan komposisi FAME biodiesel terdiri dari kedua-dua FAME, tepu (asid laurik, asid miristik, asid palmitik, asid kaprik dan asid stearik) dan tak tepu (asid oleik, asid palmitoleik dan asid linoleik). Biodiesel larva serangga yang dihasilkan menepati nilai yang disyorkan oleh piawai American Society for Test and the Materials (ASTM) D6751 dan piawai European (EN) 14214. Kesimpulannya, larva lalat askar hitam mampu menukar sisa organik yang dikaji kepada biodiesel. Implikasinya, larva serangga berpotensi digunakan sebagai suatu stok suapan biodiesel berkos rendah bagi mengurangkan kos operasi penghasilan biodiesel.









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# LIST OF ABBREVIATIONS

ANOVA	Analysis of Variance
AOAC	The Association of Official Analytical Chemists
АР	Adequate Precision
ASEAN	Association Southeast Asian Nations
ASTM	American Society for Test and the Materials Standard
ATR	Attenuated Total Reflectance
AV	Acid Value
Star of pustaka.upsi.edu.my	Black Soldier Fly Bainun PustakaTBainun Optbupsi
CCD	Central Composite Design
CDCI <sub>3</sub>	Deuterated Chloroform
CV	Coefficient of Variance
DSC	Differential Scanning Calorimeter
ECD <sub>F</sub>	Efficiency of Conversion of Digested Feed
EN	European Standard
FAEE	Fatty Acid Ethyl Ester
FAME	Fatty Acid Methyl Ester
FAO	Food and Agriculture Organization of the United
	Nations
FCR	Feed Conversion Ratio
FFA	Free Fatty Acid



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FKW	Food Kitchen Waste
FTIR	Fourier Transform Infrared
GC-FID	Gas Chromatography-Flame Ionisation Detector
GCMS	Gas Chromatography Mass Spectrometry
GDP	Gross Domestic Product
GHG	Greenhouse Gaseous
$H_2SO_4$	Sulfuric Acid
HCl	Hydrochloric Acid
HF	Housefly
HHV	High Heating Value
IEA	International Energy Agency
IPCC	Intergovernmental Danal on Climate Change
nee	Intergovernmental Panel on Climate Change
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6 IV pustaka.upsi.edu.my KeTTHA	Iodine Value Tuanku Bainun Abdul Jali Shah Pustaka TBainun Ministry of Energy Green Technology and Water
6 IV Opustaka.upsi.edu.my KeTTHA KOH	Iodine Value       Tuanko Bainon       Image: Construction of the property of
6 IV pustaka.upsi.edu.my KeTTHA KOH LOD	Iodine Value       Ministry of Energy Green Technology and Water         Potassium Hydroxide         Limit of Detection
KeTTHA KOH LOD LOQ	Iodine ValueDescent ControlMinistry of Energy Green Technology and WaterPotassium HydroxideLimit of DetectionLimit of QuantificationMinistry of Energy, Science, Technology, Environment
KeTTHA KoH LOD LOQ MESTECC	Iodine Value       Image: Descent Balow       Image: Descent Balow       Image: Descent Balow         Ministry of Energy Green Technology and Water         Potassium Hydroxide         Limit of Detection         Limit of Quantification         Ministry of Energy, Science, Technology, Environment and Climate Change
KeTTHA KoH LOD LOQ MESTECC	Iodine Value       Image: Descent Technology and Water         Anisistry of Energy Green Technology and Water         Potassium Hydroxide         Limit of Detection         Limit of Quantification         Ministry of Energy, Science, Technology, Environment and Climate Change         Methanol
KeTTHA KoH LOD LOQ MESTECC	Iodine ValueImage: Control of
KeTTHA KoH LOD LOQ MESTECC	Fodine ValueDescent ParticularMinistry of Energy Green Technology and WaterPotassium HydroxideLimit of DetectionLimit of QuantificationMinistry of Energy, Science, Technology, Environment and Climate ChangeMethanolMixed wasteMalaysian Palm Oil Board
KeTTHA KoH LOD LOQ MESTECC MeOH MW MPOB	Fodine ValueExcent BainMinistry of Energy Green Technology and WaterPotassium HydroxideLimit of DetectionLimit of QuantificationMinistry of Energy, Science, Technology, Environment and Climate ChangeMethanolMixed wasteSodium Hydroxide





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OECD	Organisation for Economic Cooperation and Development
OLF	Oriental Latrine Fly
OPM	Optical Polarisation Microscope
PV	Peroxide Value
RSM	Response Surface Methodology
SEDA	Sustainable Energy Development Authority
SIRIM	Standard and Industrial Research Institute Malaysia
SR	Soya Residue
SV	Saponification Value
TGA	Thermal Gravimetric Analyser
TLC	Thin Layer Chromatography
USEPA 6832 pustaka.upsi.edu.my	United States Environmental Protection Agency
WHO	World Health Organization
WRI	Waste Reduction Index
YMB	Yellow Mealworm Beetle









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# **CHAPTER 1**

## **INTRODUCTION**



#### 1.1 **Research Background**

Energy resources play important roles in the development of every country in the world, which significantly give impact to the political and economic stabilities (Gicquel & Gicquel, 2013; Baños et al., 2011). Several articles have discussed the factors that contribute to the world's energy growth including technologies, economic and population, availability of energy prices and fuel, and environmental regulations and standards (Gozgor, Lau, & Lu, 2018; Saad & Taleb, 2018; Acheampong, 2018; Shahbaz, Zakaria, Shahzad, & Mahalik, 2018; Arminen & Menegaki, 2019). A report entitled 'The World Energy Balance: Overview 2018' issued by International Energy Agency (IEA) has focused on total primary energy supply, worldwide. This report has described the energy supply was mainly consisted of oil (32%), coal (27%), natural





gas (22%), biofuels/waste (10%), nuclear (5%), hydro (2%) and others (2%) as presented in Table 1.1. For the past 50 years, there was a dramatic increment for the total global energy supply. For instance, the energy supply was increased from 5,523 million tonnes of oil equivalent (Mtoe) in 1971 to 13,761 Mtoe (2016), which oil resource remained the dominant energy supply (World Energy Balance, 2018; Zafar, Shahbaz, Hou, & Sinha, 2018; Abas, Kalair, & Khan, 2015). The world energy production and supply was contributed by several regions such as Organisation for Economic Cooperation and Development (OECD, 38%), non-OECD Asia (35%), non-OECD Europe and Eurasia (8%), non-OECD Americas (5%), middle east (5%) and bunkers (3%). Interestingly, non-OECD Asia becomes the second largest energy supply region after OECD, which involves China, India, Indonesia, Malaysia, Vietnam and several other countries.

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Moreover, IEA has also stated that the world energy consumption is comprised of industry (37%), transport (29%), residential (22%), commerce and public services (8%), agriculture/forestry (2%) and others (2%). The main consumptions of energy for each sector have been summarised by Gicquel & Gicquel (2013), as shown in Table 1.2. In addition, Chen & Wu (2017) has reported that the global energy consumption was increased up to 22% (2014) since 2004 that significantly promotes the energy growth.



# Table 1.1

## Total Primary Energy Supply

Energy sources	Production (%)
Fossil oil	32
Coal	27
Natural gas	22
Biofuel/waste	10
Nuclear	5
Hydro	2
Others	2

Note. Adapted from The World Energy Balance: Overview (2018).

As mentioned earlier, fossil fuel is the dominant world energy supply. Fossil

fuel has been widely used for production of liquefied gases, gasoline, naphtha, pustaka upsted uny production of liquefied gases, gasoline, naphtha, kerosene, chemical, lubricants and bitumen, and its demand forecasted to increase up to 53% by 2030 (IEA, 2017). Nevertheless, the big issue of fossil fuel consumption is its non-renewable properties. In recent years, debates continue among researchers on depletion of fossil fuel resources since these resources have almost reached the peak production. Moreover, the question has been also raised about the unfavourable outcome of prolong use of fossil fuel to the environment and health (Gielen, 2019; Chen & Wu, 2017; Wang et al., 2019a, Nguyen & Kakinaka, 2019; Sharvini, Noor, Chong, Stringer, & Yusuf, 2018; Gozgor et al., 2018; Abas et al., 2015; Tang, 2019).





# Table 1.2

Sector	Consumption	Energy
Agriculture	Mechanical energy	Oil and electricity
	Space heating	Natural gas and oil
	Fertilizer	Natural gas and oil
	Pesticides	Natural gas and oil
Industry	Electrolysis	Electricity
	Steel	Coal
	Chemistry	Oil
	Metallurgy	Natural gas and electricity
	Cement	Coal and oil
Residential/commercial	Perpustakaan Tuar Lighting	dul Jalii Shah
	Cooking	Natural gas, liquefied petroleum gas (LPG) and electricity
	Hot water	Natural gas, LPG and electricity
	Space heating	Natural gas, oil, coal and electricity
Transport	Office	Electricity
	Rail	Electricity
	Road	Oil
	Air/sea	Oil

Main Consumption of Energy

Note. Adapted from Gicquel and Gicquel (2013).

**(**) 05-450



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#### 1.2 Roles of Malaysia towards Green Energy Development

Malaysia is one of the oil producing countries and has been categorised as non-OECD Asia that contribute to the world energy supply and production. Over the years, Malaysia has made a great effort towards the revolution of the energy industry in order to reduce the dependency on fossil fuel resources. Moreover, several reviews also discussed on the Malaysia's energy and renewable energy policies that present its effort towards clean environment strategy as well as enhancing Malaysia's energy growth (Hannan et al., 2018; Oh, Hasanuzzaman, Selvaraj, Teo, & Chua, 2018). The Renewable Energy Act 2011 and National Energy Policy have been introduced to encourage the growth of the energy situation in Malaysia. Malaysia government has also set up the Ministry of Energy, Science, Technology, Environment and Climate <sup>05-4506</sup> Change (MESTECC) (previously known as Ministry of Energy Green Technology and Water (KeTTHA) and Sustainable Energy Development Authority (SEDA) in order to assist the implementation of the new energy policy. MESTECC is responsible for sustainable, renewable and conservation energy programme. Meanwhile, SEDA mainly focuses on promoting the sustainable energy as stated in the Renewable Energy Act 2011 (Abdul Manan, Baharuddin, & Chang, 2015).

Due to the disadvantages of the non-renewable energy resources, the interest on investigating the alternative energy resources known as renewable energy is keep on increasing. Moreover, Soliman, Lopez, & Biona (2018) stated that renewable energy provides energy security compared with non-renewable energy as well in order to promote the global energy growth. The renewable energy including solar, hydro, wind, tidal and biomass provides sustainable and environmental friendly







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characteristics as compared to non-renewable energy. Biodiesel is one of the potential renewable energies that has been widely reported by researchers and received high consideration due to several advantages such as engine combustion efficiency without engine modification, low sulphur content, high cetane number and biodegradable (Omidvarborna, Kumar, & Kim, 2016a; Ghazali, Mamat, Masjuki, & Najafi, 2015; Bhuiya, Rasul, Khan, Ashwath, & Azad, 2016; Knothe & Razon, 2017; Nongbe et al., 2017). Moreover, application of biodiesel was also able to reduce the greenhouse hazardous gas emission mainly from fuel combustion such as carbon dioxide, methane, nitrous oxide, ozone and chlorofluorocarbons (Omidvarborna, Kumar, & Kim, 2016b; USEPA, 2016).

# 05-4506 **1.3 Description of Biodiesel** Repustakaan Tuanku Bainun

Biodiesel is a mono alkyl ester of long chain fatty acid derived from renewable lipid that can be categorised into vegetable oils, animal fats, waste cooking oil and microorganisms (Knothe & Razon, 2017). The application of biodiesel for diesel engine consumption was firstly introduced by Rudolf Christian Karl Diesel in the late of 19<sup>th</sup> century (Abas et al., 2015). To date, vegetable oil is the main biodiesel feedstock by about 95% of the total biodiesel production (Bhuiya et al., 2016; Zheng et al., 2012; Wu et al., 2017). However, the application of vegetable oil as a biodiesel feedstock is commonly related to the high total biodiesel production cost (Knothe & Razon, 2017). This situation occurred due to the increment of raw material cost for about 60-75% of the total biodiesel production and also the requirement of large arable plant for extensive biodiesel production (Kirubakaran & Selvan, 2018a; Patel,



Arora, Mehtani, Pruthi, & Pruthi, 2017; Anuar & Abdullah, 2016; Yang, Li, Gao, Zheng, & Liu, 2014). Additionally, the usage of vegetable oils particularly edible oils may cause food crisis in the long term (Patel et al., 2015). Another issue is the seasonal harvest period for almost oilseeds which make vegetable oils are inefficient for biodiesel production process (Yang et al, 2014).

#### 1.4 Insect Larvae for Biodiesel Production

Previously, many studies have reported on application of insect larvae particularly for organic waste treatment (Mertenat, Diener, & Zurbrügg, 2019; Kumar, Negi, Mandpe, Singh, & Hussain, 2018; Lalander, Diener, Zurbrügg, & Vinnerås, 2019; Nguyen et al., 2015). Moreover, Čičková et al. (2015) and Diener et al. (2011) have also discussed on availability of insect larvae such as housefly, black soldier fly, face fly, green bottle fly and flesh fly to feed naturally in organic wastes as well as assisting in reducing the organic waste. While, studies on cultivation of insect larvae using organic wastes and harvested them mainly for livestock feed such as chicken, fish and pet receive high interest among researchers (Wang et al., 2019b; Makkar, Tran, Heuzé, & Ankers, 2014; Čičková, Newton, Lacy, & Kozánek, 2015, Hu et al., 2018; Henry, Gasco, Piccolo, & Fountoulaki, 2015; Barragan-Fonseca, Dicke, & van Loon, 2017; Cutrignelli et al., 2018; Bovera et al., 2015). Moreover, Henry et al. (2015) has stated on the capability of insect to be fish feed due to nutrient content is almost similar to fish meal. They also reported on application of insect as fish meal able to reduce the cost for aquaculture feed. In recent years, studies have also highlighted the feasibility of insect larvae as alternative feedstock for renewable energy production



(Nguyen et al., 2018a; Surendra, Olivier, Tomberlin, Jha, & Khanal, 2016; Niu et al., 2016).

Recently, Black soldier fly (BSF), Hermetia illucens (Diptera: Stratiomyidae) originally from Americas and is now widely spread to over the world has also get special attention for livestock feed and biodiesel production process. The advantage of BSF includes nonvector properties which does not transmit diseases as well as harmless to human and animal (Caliagi et al., 2018; Sheppard, Tomberlin, Joyce, Kiser, & Sumner, 2002). Moreover, BSF has also been reported to act as a biological control over housefly (HF) by avoiding HF oviposition thus leads to decrement of HF numbers (Harnden & Tomberlin, 2016; Myers, Tomberlin, Lambert, & Kattes, 2008; Čičková, Newton, Lacy, & Kozánek, 2015; Leong et al., 2015). Huge amount of 05-4506 organic waste is required for BSF larval growth development, which significantly due to a longer cultivation period (20-24 days) and larger size of BSF larvae (18-20 mm). This scenario promotes a better organic waste management (Mertenat, Diener, & Zurbrügg, 2019; Kumar, Negi, Mandpe, Singh, & Hussain, 2018; Čičková et al., 2015; Manzano-Agugliaro et al., 2012). Several studies have been carried out in order to determine the feasibility of BSF larvae as feedstock for biodiesel production (Nguyen et al, 2018a; Nguyen et al, 2018b; Surendra et al., 2016; Leong, Kutty, Malakahmad, & Tan, 2015; Zheng et al., 2012; Li, Zhen, Hou, Yang, & Yu, 2011b).

Other insect species have also been evaluated and reported for biodiesel production such as house fly (Zi-zhe et al., 2017; S. Wu et al., 2017; Niu et al. 2017; Yang et al., 2014), oriental latrine fly (*Chrysomya megacephala*) (Li et al., 2012; Yang & Liu, 2014) and mealworm beetle (*Tenebrio molitor*) (Zheng et al., 2013). In





recent years, insect larvae have received great attention among scientists for producing a low-cost biodiesel. The application of insect larvae offers low raw material cost which therefore able to reduce the total biodiesel production cost as compared to the cost to produce vegetable-based biodiesel. To date, the studies on the potential of insect larvae for biodiesel production are still remain scarce. Thus, it is imperative to investigate in further details the capability of insect larvae for biodiesel production. Therefore, this research was carried out in order to determine the potential of insect larvae cultivated on several organic wastes as promising biodiesel feedstocks for biodiesel production.

#### 1.5 Problem Statement

Currently, fossil fuel is the most growing energy demand over the world. However, the fossil energy is interfacing the depletion of its resources mainly due to non-renewable energy properties. The tremendous increase in energy demand promotes the application of renewable energy worldwide which provides a better energy security and sustainability. The potential of biodiesel as a promising renewable energy has been emphasised by researchers but its price stability is the main concern. This situation occurred due to the dependence of biodiesel production on oilseed, requirement of larger land area and the impacts of food crisis. Therefore, there is an urgent need to find a new non-food and cheap feedstock for biodiesel production. To date, there are several efforts to produce biodiesel using insect larvae have been reported. However, the quantity and quality of biodiesel production are still low. Thus, this research was devised as a direct response to the aforementioned issue. In

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this research, several factors affecting the optimisation of quality and quantity of biodiesel production through biodegradation of organic wastes by insect larvae were evaluated.

#### 1.6 Research Gap

Based on the literature reviews, although a number of studies had successfully developed and assessed the potential of several feedstocks for biodiesel production, several significant research gaps have been found and they are relevant to be investigated.

- 1. Vegetable oils are the most widely used feedstock for biodiesel production. Unfortunately, the application of vegetable oils involves high total operation cost which leads to high biodiesel price. Therefore, it is important to perform a study to evaluate alternative a low-cost biodiesel feedstocks. In this study, insect larvae have been used as potential feedstocks for biodiesel production.
- Researchers normally reared insect larvae using one type of organic waste. It is attractive important to study the growth of insect larvae on different organic wastes. Therefore, this study was carried out using different organic wastes including food kitchen waste, soya residue and mixed waste.
- 3. Researchers normally used conventional method in determination of the optimum parameters for transesterification. It is interesting to conduct the transesterification study using statistical analysis, Response Surface





Methodology (RSM). In this study, RSM was applied for optimisation of biodiesel production study.

- 4. Up to date, the physical and chemical characterisation studies of insect larval biodiesel are rarely reported in the literatures. It is imperative to conduct a characterisation study of insect larval biodiesel. Here, the produced insect larval biodiesel was characterised using several scientific instruments such as Optical Polarisation Microscope (OPM), Fourier Transform Infrared (FTIR) Spectrometer, and Nuclear Magnetic Resonance (NMR) Spectrometer, Thermal Gravimetric Analyser (TGA), Differential Scanning Calorimeter (DSC), CHNS-O elemental analyser and Gas Chromatography-Flame Ionisation Detector (GC-FID).
- 5. In recent years, the number of literatures reported on properties of insect larval pustaka.upsi.edu.my and propustakaan Tuanku Bainun biodiesel are still low. Therefore, the insect larval biodiesel produced from this study was determined and compared with American Society for Test and the Materials (ASTM) D6751 and European (EN) 14214 standards.

#### 1.7 Research Aim

The overall aim of this research is to produce environmental friendly and sustainable biodiesel by biodegradation of organic wastes by black soldier fly. It is hoped that this larva has great potential to be used as alternatives to expensive biodiesel feedstocks.





# 1.8 Research Objectives

The objectives of this research are:

- To analyse the nutrient content of organic waste from different source (food kitchen waste, soya residue and mixed waste) for BSF cultivation.
- 2. To observe the BSF larval growth development during cultivation process.
- 3. To extract and characterised BSF larval lipid.
- 4. To evaluate the quality of the insect larval biodiesel produced.

# 1.9 Research Scope

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This research consisted of four main studies. The first study was to characterise the organic wastes. Three types of organic wastes, namely food kitchen waste, soya residue and mixed waste were used for rearing insect larvae. These organic wastes were characterised based on several parameters including content of moisture, protein, carbohydrate, ash, pH and crude fiber.

The second study was focused on cultivation of insect larvae. Insect larvae of BSF was inoculated into the aforementioned organic wastes and left for natural biodegradation process. The growth development of BSF larvae was monitored by evaluating the color, individual weight and size. After the cultivation period was completed, the BSF larvae were characterised based on content of moisture, protein, carbohydrate, fat, ash and crude fiber. Meanwhile, degradation of selected organic





wastes during the growth of BSF larvae was evaluated based on growth rate of BSF larvae, overall degradation (OD), waste reduction index (WRI) and efficiency of conversion of digested feed ( $ECD_F$ ).

The third study was on characterisation of insect larval lipid. Initially, the insect larvae of BSF were oven-dried prior to extraction study. In the extraction study, the efficiency of three solvents namely petroleum ether, acetone and ethanol was evaluated based on insect larval lipid yield within certain extraction time. The produced insect larval lipid was characterised accordingly based on acid value (AV), free fatty acid (FFA), saponification value (SV), peroxide value (PV) and iodine value (IV).

The final study was focused on evaluation of the properties of produced larval based biodiesel. Due to high percentage of FFA for BSF larval lipid, the biodiesel production was conducted using a two-step transesterification process which involved acid-catalysed esterification and alkaline-catalysed transesterification. In order to determine the optimum conditions for each parameters, namely: (1) methanol:oil (molar ratio), (2) catalyst % (w/w), (3) reaction time (min) and (4) reaction temperature (°C) for transesterification process, a statistical analysis Response Surface Methodology (RSM) was applied. Furthermore, the properties of insect larval biodiesel were examined based on oxidation stability, water content, flash point, kinematic viscosity, cetane number, density, cloud point, acid number and high heating value. The properties of insect larval biodiesel were also compared with the American Society for Test and Materials (ASTM D6751) and European Standard (EN 14214) standards. Moreover, several instruments were used to clarify the successful

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transformation of insect larval lipid to biodiesel including FTIR spectrometer and NMR spectrometer. Meanwhile, the thermal properties of insect larval biodiesel were investigated using TGA, DSC and OPM. Additionally, GC-FID was applied to determine the fatty acid methyl ester (FAME) composition of insect larval biodiesel.

#### 1.10 **Research Significance**

This research emphasises the significance of the development of insect larval biodiesel to replace or decrease the dependence of fossil fuel-based diesel in engine consumption. Moreover, this research highlights the efficacy of biodegaradtion of organic wastes by insect larvae for biodiesel production.

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#### **Hypothesis** 1.11

The larval lipid of BSF larvae can be used as alternative feedstock to oilseeds in biodiesel production.

#### **Thesis Organisation** 1.12

This thesis contains five chapters. Chapter 1 is dedicated to the introduction which covers the research background, problem statement, research gap, research aim, research objectives, research significance and hypothesis. Chapter 2 explains the





development of energy covering topics related to energy situation, energy mix, challenges and crisis of energy, renewable energy and potential renewable energy source worldwide including Malaysia. This chapter also covers the description of biofuel and biodiesel which includes biodiesel production process, biodiesel feedstocks and the crisis and challenges of biodiesel production process. Chapter 3 describes a detailed methodology involved in the biodiesel production from insect larvae and the characterisation techniques used to determine the chemical and physical properties of organic wastes, insect larvae, insect larval lipid and insect larval biodiesel. This chapter also presents the biodiesel production using the transesterification process. Chapter 4 explains and discusses the findings of this research. The results include the chemical and physical characterisations of organic wastes, insect larvae, insect larval lipid and insect larval biodiesel. The findings obtained from optimisation of transesterification process of insect larval lipid for biodiesel production were also further explained in this chapter. The quality of insect larval biodiesel was also discussed and compared with the ASTM D6751 and EN 14214 standards. Conclusions drawn from this research and suggestions for future research are stated in Chapter 5.



