PRODUCTION AND COMBUSTION OF WASTE ANIMAL FAT BIODIESEL AS ALTERNATE FUEL TO FOSSIL DIESEL

Cheruiyot, Edwin Kosgei.

218076625

Supervisor: Prof. Freddie, L. Inambao.

Thesis submitted in fulfillment of the requirement for the degree of Master of Science in Engineering (Mechanical)

College of Agriculture, Engineering and Science, University of KwaZulu-Natal, Durban, South Africa

As the candidate's supervisor, I have approved this dissertation for submission.

Signed…… Date 20th March, 2020.

Name: Prof. Freddie L. Inambao

Declaration 1 -Plagiarism

I, **Kosgei, Edwin Cheruiyot** declare that:

1. The research reported in this thesis, except where otherwise indicated is my original research.

2. This thesis has not been submitted for any degree or examination at any other university.

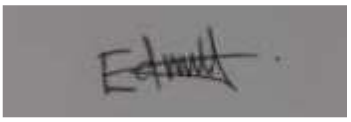
3. This thesis does not contain other persons' data, pictures, graphs or other information, unless specifically acknowledged as being sourced from other persons.

4. This thesis does not contain other persons' writing, unless specifically acknowledged as being sourced from other researchers. Where other written sources have been quoted, then:

a) Their words have been re-written but the general information attributed to them has been referenced

b) Where their exact words have been used, then their writing has been placed in italics and inside quotation marks, and referenced.

5. This thesis does not contain text, graphics or tables copied and pasted from the internet, unless specifically acknowledged, and the source being detailed in the thesis and in the References sections.



Signed…… Date: 20th March, 2020.

Declaration 2 -Publications

This section presents the articles that form part and/or include the research presented in this thesis. The following papers have been published or are have been accepted for publication:

DoHET Accredited Journals

1. **Cheruiyot, K. E** and Inambao F. L (2019). A comprehensive review of low cost biodiesel production from waste beef tallow. *International Journal of Mechanical Engineering and Technology. Volume 10, Issue 8, August 2019, pp. 285-305; ISSN Print: 0976-6340 and ISSN Online: 0976-6359. (***published***)*

2. **Cheruiyot, K. E** and Inambao F. L (2019). Current trends in enzymatic transesterification of waste animal fats**.** *International Journal of Mechanical Engineering and Technology. Volume 10, Issue 12, December, 2019, pp. 541-558; ISSN Print: 0976-6340 and ISSN Online: 0976-6359 (***published***)*

3. **Cheruiyot, K. E** and Inambao F. L (2019). Development and Characterization of Waste beef tallow methyl esters. *International Journal of Mechanical Engineering and Technology. Volume 10, Issue 11, November 2019, pp. 245-258; ISSN Print: 0976-6340 and ISSN Online: 0976-6359 (***published***)*

4. **Cheruiyot, K. E** and Inambao F. L (2019). Experimental study of performance and emission characteristics of beef tallow biodiesel. *International Journal of Mechanical Engineering and Technology. Volume 10, Issue 11, November 2019, pp. 371-381; ISSN Print: 0976-6340 and ISSN Online: 0976-6359 (***published***)*

For all the publications the candidate is the main author while Prof. Freddie L. Inambao is the supervisor.

Dedication

This work is dedicated to the Almighty God who helped me overcome all challenges and obstacles during my research period.

Acknowledgement

Success is seldom achieved alone. It is a tiresome journey that requires hard work and determination throughout the learning process. Learning something new and challenging helps one reach highest goals on this ever changing world. Since learning can never undertake alone, I would like to recognize efforts of people who made it possible to climb the academic ladder. The effort of my Supervisor Prof. Freddie Inambao is the first which is worth mentioning first. His words of encouragement and advise kept me working hard in every step. Thanks also goes to Prof. Glen who is the leader of discipline for providing serene environment to study with other members of staff. I also extend my hand of gratitude to Mr. Suren, senior lab technician for helping repair fume hood chamber when it broke down during period when it was most needed for performing experiments. I will be biased if I do not mention efforts of Buxsons Butchery management-Clairwood, Durban who assisted in donating beef tallow, thank you so much. I will not forget to mention members of the research Group-Green Energy Solutions. May God bless you abundantly for the guidance you gave me during research period. I would also like to thank Dr. Richard Steele for helping in editing of all the published work. Lastly, I thank family members for their encouragement and financial support during this important period more so My brother Erick who is also working towards his completion of doctoral degree at DUT.

Table of Contents

List of figures

Abbreviations/acronyms

KOH	Potassium hydroxide
H_2SO_4	Sulphuric acid
ASTM	American Society of Testing and Materials
HC	Hydrocarbon
NOx	Oxides of nitrogen
CO	Carbon Monoxide
EGT	Exhaust gas temperature
GCMS	Gas Chromatography Mass Spectroscopy
TGA	Thermogravimetric Analyses
FTIR	Fourier Transform Infrared
BTE	Brake thermal efficiency
BSFC	Brake Specific Fuel Consumption
IC	Internal Combustion
CI	Compression Ignition
CFPP	Cold Filter Plugging Point
UN	United Nations
WAFs	Waste Animal Fats
WBT	Waste Beef Tallow
B20	20 % biodiesel and 80 % diesel.
GHG	Greenhouse gas
GDP	Gross domestic product
EJ	Exajoules

Abstract

Fossil diesel is finite and is projected to be depleted by 2050. The growing population in the world consumes an increasing amount of animal products which generates waste products that can be converted to biodiesel. Biodiesel is a renewable fuel that is an alternative to petroleum diesel in compression ignition engines that can alleviate noxious gas emissions which contribute to global warming and climate change. An opportunity exists in the transport sector for biodiesel synthesized from waste animal fats (WAFs) (beef tallow), especially in developing countries. The main aim of this study was to produce beef tallow biodiesel by the transesterification method and to determine its suitability in an engine application. During this study fat was extracted by dry rendering in an open vessel before two step transesterification was performed using a potassium hydroxide (KOH) catalyst and methanol. Three parameters were varied to obtain optimum biodiesel yield. These parameters were reaction time, methanol to fat molar ratio, and catalyst loading concentration. Biodiesel physical properties measured were density, kinematic viscosity, flash point, calorific value, cloud point, pour point, cold filter plugging point, and water content. Chemical properties were characterized using Fourier transform infrared spectroscopy, thermogravimetric analysis and gas chromatography mass spectroscopy analysis. The pure biodiesel and diesel blends were evaluated using a stationary 3-cylinder tractor engine for performance and emission parameters. The brake thermal efficiency was lower by 22.54 % while exhaust gas temperature and brake specific fuel consumption for B100 biodiesel were 48.4 ℃ and 0.055 kg/Kwh higher than diesel fuel at maximum load. Carbon monoxide (CO), hydrocarbons (HC) and oxides of nitrogen (NO_X) showed an increasing trend with increase in load.

CHAPTER 1: INTRODUCTION

1.1 Introduction

There is rising public interest in the adoption of renewable energy and clean energy technologies so as to mitigate a temperature rise in the earth's atmosphere. The combustion of fossil diesel in compression ignition (CI) engines generates high levels of unburned hydrocarbons (UHC), NO_X and particulate matter (PM) that settles in the lower part of the atmosphere thus trapping harmful gases and reflecting the sun's rays in ways that contribute to climate change. The search for feasible alternate fuels is ongoing regarding cost of production and its life cycle benefit to the environment. Biodiesel application in diesel engines is safe for the existing engine geometry and is readily available. A well-to-wheel (WTW) analysis investigating emissions from soybean and renewable fuels showed soybean derived fuels can reduce greenhouse gas (GHG) emissions from 64 % to 174 % compared to petroleum diesel [1]. The use of biodiesel is beneficial compared to electric vehicles in developing countries since biodiesel can be stored in the existing infrastructure of fuel pumps without addition of new sophisticated technologies [2]. Biodiesel fuel use in sub-Saharan Africa can improve GDP by reducing importation of fossil diesel which in some cases is substandard.

Vehicle manufacturers and government inspectors have developed and implemented strict laws on the production of vehicles to meet the set emission standards. Manufacturers that release vehicles flouting set emission values face heavy fines as was seen in the case of the Volkswagen company scandal [3]. This case portrays authorities' commitment to a cleaner environment and to support for a clean biodiesel fuel for diesel engines, particularly in agricultural applications. Unlike petrol engines, diesel engines fuel spraying, mixing and combustion takes place spontaneously which leads to soot formation which contributes to black smoke in exhaust for most vehicles when it mixes with leaked oil. The quality of biodiesel depends on the quality of the raw material source. With the rapid growth and population increase in Africa, a lot of waste is generated from inedible livestock fats. This provides a rich source of raw material for biodiesel production which can modify the current energy portfolio in developing countries.

Traditionally, biodiesel has been sourced from edible vegetable oil, with about 93 % of global biodiesel produced being from edible oil sourced from agricultural industries. However, it is becoming increasingly clear to most stakeholders that this trend is unsustainable in the longer term because of the food versus fuel dispute it has triggered. Biodiesel production uses around 4.4 million hectares of arable land in the European Union (EU). Replacing 10 %

of diesel used in the EU with edible oil based biodiesel would account for around 19 % of world edible oil production in 2020 which would mean more land being planted with food crops, thereby releasing more GHGs. The alternative is to use WAF for energy generation.

A study conducted by Kesteloot et al. [4] links saturated fats to the increase in cancer cases in the world. The study showed a strong correlation between lard fat and dairy fat intake and cancer cases especially breast, prostate, colon and rectal cancer in men and women and lung cancer in men. The authors concluded that the findings are confirmatory evidence that saturated fats play a role in carcinogenesis. Therefore, saturated fats which are normally discarded as waste can be converted to biodiesel.

1.2 Statement of Problem

Global primary energy consumption growth has risen from 200.45 exajoules (EJ) in 1990 to 576.03 EJ in 2015 which amounts to 200.45 EJ in 25 years. This growth is projected to decline by 168.8 EJ in the next 25 years due to evolving policies. Historically, no major fuel source has seen its overall level of demand decline globally. Rather than energy transitions, the world has undergone a series of energy additions. Fossil fuels contribute 82 % and liquid fuels 32 % of global primary energy in 2015. The contribution of renewable energy and nuclear energy is 23 % and 11 % for electricity generation respectively [5]. Petroleum and other liquid fuels are the dominant sources of transport fuel with diesel and biodiesel accounting for 13.715 EJ. Motor vehicle industry fuel consumption and emissions vary by country. For instance, the EU and India have standards that aim to reduce carbon dioxide (CO_2), Canada and Brazil have standards that aim to increase fuel economy by using light duty vehicles while the US and Mexico aim to reduce both fuel consumption and emissions [6]. A report from the US Department of Energy states that 2.2 million workers in America are employed in energy efficient sectors compared to 1.1 million workers in fossil fuel stations [7] which emphasizes the application of clean energy.

Globally, fossil fuel supplies 87 % of energy used [8], which is a finite source of energy subject to political instabilities [9, 10] which raise the lending rates for countries that rely on importing crude oil. Recently, US sanctioned Iran over oil and threated sanctions against any country that trades with Iran for oil, which is affecting oil pump prices and weakening international relations [11]. Besides, the use of fossil fuel has a negative impact on the environment. The use of petroleum fuels increases emissions of PM, lead, aldehyde, sulfur

dioxide and poly-aromatic HCs into the atmosphere [12]. Fine particles with a diameter around 2.5 µm suspended in breathed air can pass into the human respiratory system and be deposited in the lungs [13]. The emission of NO_X has caused 10 000 premature deaths in Europe and contributed to the breaching of critical eutrophication levels [14]. Such outcomes can be avoided by phasing out fossil fuels. Thus, to safeguard our environment and satisfy fuel demand for developing countries, it is imperative to find sources of fuel that are available, economical, renewable and sustainable for economic development.

1.3 Research Motivation

Fossil fuels such as crude oil, coal and natural gas used in transport sector vehicles are regarded as dominant contributors of air pollution and cause an increase in global gases emissions [15]. China, a country with one of the fastest growing economies in the world, is the largest consumer of energy and producer of emissions in the world [16, 17], but has embarked on a campaign for clean energy. In 2015, China had a total primary energy consumption of 3152.60 million tons of oil equivalent which was 1.38 times higher than the US according to a report produced by BP [18]. At the domestic level, a developing country like Kenya has an energy demand of 1600 MW with a projection increase of up to 3600 MW by the year 2020 [19]. Biomass is the leading energy source at 68 % as shown in Figure 1. With the increase in population, there are fears that slow energy development could stagnate economic growth.

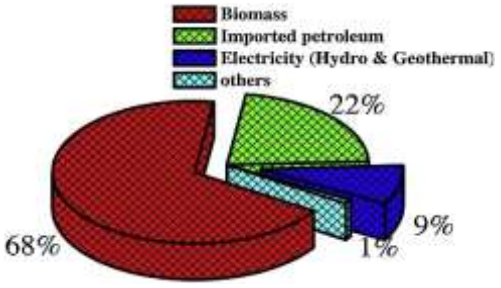


Figure 1 Percentage of energy production in Kenya [19]

Biodiesel can be utilized to mitigate the high level of emissions and reduce reliance on importation of petroleum products used in internal combustion engines. Biodiesel has gained wide acceptance as a substitute for diesel fuel in compression ignition engines. Biodiesel is produced from plant/animal sources which are renewable and clean burning. A range of

research conducted on engines shows that biodiesel has low CO_2, HC and smoke density compared to petroleum diesel [20]. Variations in results for NO_X [20] have been reported to depend on feedstock sources. Authors have recommended further testing to search for ways to improve knowledge and understanding of biodiesel benefits.

Vegetable oil and animal fats are the primary sources of biodiesel. Biodiesel can easily be produced from edible vegetable oils because of their low acid value. Countries like Malaysia, Brazil, Canada, United States and Germany among others have a surplus of edible oils for export and hence they blend biodiesel from edible oil with petroleum fuel [21]. Recently, vehement debate has emerged between energy experts and food organization groups on damage that biodiesel causes to exploitation of food reserves and food insecurity [22]. Growing feedstock requirements for biodiesel have led to soaring food prices in developed nations while developing countries which are food insecure may be faced with malnutrition and hunger as reported in research outcomes [23]. As a result, non-edible feedstock sources have been identified and are currently being used for biodiesel production. This includes waste cooking oil, algae and WAF. Although research shows algae can be used as a total replacement for petroleum diesel, its production capacity is hampered by the high technology required for cultivation and harvesting.

From the foregoing it is clear that biodiesel usage for transportation faces many challenges regarding feedstock source, combustion and performance in diesel engines. Thus, knowledge of a new source of feedstock, namely WAFs, can help in development of its biodiesel properties and its benchmarking with internationally recognized standards.

1.4 Research Questions

The research questions that can help address problems facing energy availability in diesel engines using WAF biodiesel are as follows:

1. To what extent is the production method for the WAF biodiesel aligned with previous work and the set standards for biodiesel application?
2. Are the performance and emission characteristics in compression ignition engine comparable to petroleum diesel?

1.5 Aims and Objectives

The aim of this research was to evaluate viability and characterization of WAF biodiesel in a compression ignition engine. At the end of the study, WAF biodiesel properties and experimental for performance and emission were compared with international set standards and remediation proposed where possible. The objectives of the study were:

1. To produce by acid-base transesterification method WAF biodiesel that meets ASTM standards for application in a compression ignition engine.
2. To characterize produced WAF biodiesel for physical and chemical properties.
3. To evaluate performance and emission of the test WAF biodiesel fuel compared with diesel fuel to check if it meets international regulation standards.

1.6 Research Significance

Africa as a continent is struggling to meet its energy demand for transportation which impacts negatively on its economic development. Most countries rely on the importation of fuels which makes oil exporters richer. Africa has the highest population growth and lowest technological development for conversion of waste products into profit making enterprises. Slaughterhouses, for instance, generate massive waste that is traditionally discarded in open landfills. These wastes include hoofs, skin, bones and fats. Waste fats are rich sources of feedstock for biodiesel preparation which is currently underutilized in Africa with no processing plants at all. Thus, studies are necessary on the performance of WAF biodiesel in diesel engines to benchmark this fuel with existing international fuel standards.

1.7 Thesis Outline

The chapters contained in this thesis are structured to reflect the aims and objectives of the research undertaken. The majority of the thesis comprises papers and publications as stipulated in the University of KwaZulu-Natal's requirements for the award of postgraduate degrees. The thesis consists of six chapters.

Chapter 1 is the introductory part for this thesis and provides the introduction, aims, objectives, research questions, significance, thesis outline and research scope.

Chapter 2 reviews the literature relating to biodiesel production and testing as well as WAF feedstock as a low cost biodiesel raw material.

Chapter 3 reviews the application of enzymes/lipases as catalysts during transesterification because the high free fatty acids in WAF leads to saponification and requires lengthy purification steps during processing.

Chapter 4 presents the first experimental work on WAF conversion to biodiesel. A two-step, acid-base method transesterification method is explained in this chapter. It also contains physical and chemical properties characterization of WAF biodiesel.

Chapter 5 presents the second experimental work on engine performance testing and compares the outcomes with other published work.

Chapter 6 presents the conclusions and findings of the research and makes recommendations for future work.

1.8 Research Scope

This thesis covers work that is related to sourcing, extraction, production and experimental engine testing of WAF biodiesel.

REFERENCES

[1] H. Huo, M. Wang, C. Bloyd, and V. Putsche, "Life-cycle assessment of energy and greenhouse gas effects of soybean-derived biodiesel and renewable fuels," ANL/ESD/08-2, Argonne National Laboratory, Illinois2008.

[2] J.-H. Ng, H. K. Ng, S. J. C. T. Gan, and E. Policy, "Recent trends in policies, socioeconomy and future directions of the biodiesel industry," *Clean Technologies and Environmental Policy*, vol. 12, pp. 213-238, 2010.

[3] B. Blackwelder, K. Coleman, S. Colunga-Santoyo, J. S. Harrison, and D. Wozniak, "The Volkswagen scandal," Robins School of Business, University of Richmond, 2016.

[4] H. Kesteloot, E. Lesaffre, and J. V. Joossens, "Dairy fat, saturated animal fat, and cancer risk," *Preventive Medicine*, vol. 20, pp. 226-236, 1991/03/01/ 1991.

[5] R. G. Newell, D. Raimi, and G. Aldana, "Global Energy Outlook 2019: The Next Generation of Energy," 2019.

[6] J. Conti, P. Holtberg, J. Diefenderfer, A. LaRose, J. T. Turnure, and L. Westfall, "International energy outlook 2016 with projections to 2040," USDOE Energy Information Administration (EIA), Washington, DC, 2016.

[7] B. Obama, "The irreversible momentum of clean energy," *Science*, vol. 355, pp. 126-129, 2017.

[8] R. Lyman, "Why renewable energy cannot replace fossil fuels by 2050,"Friends of Science, Calgary, Alberta, Canada, 2016.

[9] M. Rouhi, "US–Iran tensions and the oil factor," *Survival*, vol. 60, pp. 33-40, 2018.

[10] C. A. Bollino, B. Efird, F. Hasanov, and E. Hatipoğlu, "Iran sanctions: implications for the oil market," Research Report. KAPSARC (King Abdullah Petroleum Studies and Research Center), Riyadh, 2019.

[11] M. Rafieisakhaei, B. Barazandeh, A. Moosavi, M. Fekri, and K. Bastani, "Supply and demand dynamics of the oil market: A system dynamics approach," in *The 34rd International Conference of the System Dynamics Society*, 2016.

[12] A. T. Hoang, "Experimental study on spray and emission characteristics of a diesel engine fueled with preheated bio-oils and diesel fuel," *Energy*, vol. 171, pp. 795-808, 2019/03/15/ 2019.

[13] P. Penttinen, K. Timonen, P. Tiittanen, A. Mirme, J. Ruuskanen, and J. Pekkanen, "Ultrafine particles in urban air and respiratory health among adult asthmatics," *European Respiratory Journal*, vol. 17, pp. 428-435, 2001.

[14] J. E. Jonson, J. Borken-Kleefeld, D. Simpson, A. Nyíri, M. Posch, and C. Heyes, "Impact of excess NOx emissions from diesel cars on air quality, public health and eutrophication in Europe," *Environmental Research Letters*, vol. 12, p. 094017, 2017.

[15] R. K. Pachauri, M. R. Allen, V. R. Barros, J. Broome, W. Cramer, R. Christ, *et al.*, "Climate change 2014: synthesis report," *Contribution of Working Groups I, II and III to the fifth assessment report of the Intergovernmental Panel on Climate Change*: Ipcc, 2014.

[16] F. Fu, L. Ma, Z. Li, and K. R. Polenske, "The implications of China's investment-driven economy on its energy consumption and carbon emissions," *Energy Conversion and Management*, vol. 85, pp. 573-580, 2014/09/01/ 2014.

[17] Y. Fan and Y. Xia, "Exploring energy consumption and demand in China," *Energy*, vol. 40, pp. 23-30, 2012.

[18] B. PLC, "BP Statistical Review of WorldEnergy 2016," ed: London, UK: BP plc https://www. bp. com/content/dam/bp/pdf/energyeconomics …, 2017.

[19] A. George, S. Boxiong, M. Arowo, P. Ndolo, C. Chepsaigutt, and J. Shimmon, "Review of solar energy development in Kenya: Opportunities and challenges," *Renewable Energy Focus*, vol. 29, pp. 123-140, 2019/06/01/ 2019.

[20] Y. Devarajan, R. kumar Jayabal, D. Ragupathy, and H. Venu, "Emissions analysis on second generation biodiesel," *Frontiers of Environmental Science & Engineering*, vol. 11, p. 3, 2017.

[21] I. Ambat, V. Srivastava, and M. Sillanpää, "Recent advancement in biodiesel production methodologies using various feedstock: A review," *Renewable and Sustainable Energy Reviews*, vol. 90, pp. 356-369, 2018/07/01/ 2018.

[22] R. Herrmann, C. Jumbe, M. Bruentrup, and E. Osabuohien, "Competition between biofuel feedstock and food production: Empirical evidence from sugarcane outgrower settings in Malawi," *Biomass and Bioenergy*, vol. 114, pp. 100-111, 2018/07/01/ 2018.

[23] A. Aksoy and B. Hoekman, "Food prices and rural poverty," CEPR and the World Bank, 2010.

CHAPTER 2: A COMPREHENSIVE REVIEW OF LOW-COST BIODIESEL PRODUCTION FROM WASTE BEEF TALLOW

This chapter reviews production methods, parameters affecting transesterification process and properties of beef tallow biodiesel for its suitability in diesel engine. This work was published in the International Journal of mechanical Engineering and Technology.

Cite this Article: Cheruiyot Kosgei and Prof. Freddie L. Inambao, A Comprehensive Review of Low-Cost Biodiesel Production from Waste Beef Tallow. *International Journal of Mechanical Engineering and Technology* 10(8), 2019, pp. 285-305.

Link to the article:
http://www.iaeme.com/IJMET/issues.asp?JType=IJMET&VType=10&IType=8

International Journal of Mechanical Engineering and Technology (IJMET)
Volume 10, Issue 08, August 2019, pp. 285-305, Article ID: IJMET_10_08_023
Available online at http://www.iaeme.com/ijmet/issues.asp?JType=IJMET&VType=10&IType=8
ISSN Print: 0976-6340 and ISSN Online: 0976-6359

A COMPREHENSIVE REVIEW OF LOW-COST BIODIESEL PRODUCTION FROM WASTE BEEF TALLOW

Cheruiyot Kosgei and Prof. Freddie L. Inambao*
Department of Mechanical Engineering,
University of Kwazulu-Natal, Durban, South Africa
inambaof@ukzn.ac.za

ABSTRACT

Biodiesel as a renewable and biodegradable fuel has great potential to mitigate the problem of soaring fuel price demand. Biodiesel has excellent properties comparable to diesel hence can substitute diesel in compression ignition engines without any modification of engine geometry. Edible vegetable oil when used as feedstock increases retail price of biodiesel. Waste animal fats (WAFs)have gained popularity recently as cost effective feedstock for biodiesel production. This paper discusses the merits of WAFs as a diesel fuel substitute particularly from waste beef tallow. This article is a detailed review of the various production methods with emphasis on transesterification processes. The factors that affect transesterification such as temperature, time and molar ratio have been included. The study reveals that waste beef fat would be a suitable replacement of vegetable oils due to low cost, its role in protecting the environment, and beef tallow esters are close to ASTM D 6751 biodiesel standards. Research work needs to be carried out on heterogeneous catalysts and low-cost lipase catalysts to improve productivity.

Key words: Waste beef tallow fat and beef tallow methyl esters, Transesterification, Biodiesel, Physico-chemical properties

Cite this Article: Cheruiyot Kosgei and Prof. Freddie L. Inambao, A Comprehensive Review of Low-Cost Biodiesel Production from Waste Beef Tallow. *International Journal of Mechanical Engineering and Technology* 10(8), 2019, pp. 285-305.
http://www.iaeme.com/IJMET/issues.asp?JType=IJMET&VType=10&IType=8

1. INTRODUCTION

Globally, the rapidly rising human population growth has increased demand for food, energy, water and materials which considerably increases the amount of pollutants and green-house gases emitted in the environment [1]. Government agencies and policy makers are major players in ensuring transport, energy and environment are sustainable. The main aim is to reduce green-house gases especially in the transport sector. Due to high power required in transportation vehicles, compression ignition engines are used utilizing diesel as fuel. Diesel fuel is obtained from fossil sources mined underground has been associated with pollutant emissions which can cause cancer and increase in temperatures on earth. Research for an

alternative fuel is ongoing in many academic institutions and industries. Biodiesel has been accepted as petroleum diesel substitute.

Biodiesel, which originates from animal fat or vegetable oil, has already absorbed carbon as carbon dioxide from air hence it will contribute less to global warming, thus, mitigating climate change. Biodiesel chemical formulae have more oxygen present which is considered an important property in biodegradation [2]. Therefore, feed-stocks from these renewable sources will produce less emissions. Vegetable oils (edible and non-edible) and waste animal fats are known renewable sources. Non-renewable economical sources include pyrolyzate from tyres though it requires desulphurization and quality improvement [3]. Biodiesel has several advantages than other fuels in compression ignition engine. Biodiesel has acceptable limit of sulfur content and no aromatics present. Also cetane number is higher than diesel fuel [4], good lubricity property [5] and it is non-toxic. Disadvantage of biodiesel is increased nitrogen oxides at high temperatures [6], lower viscosity, lower calorific value [7] and poor oxidation stability [8]. Biodiesel blending with diesel is done in any ratio. These two fuels have similar characteristics. Continuous use of biodiesel can reduce pollutant output and carcinogen causing agents. Usage of blended biodiesel in engines reduce SO_2 emissions which are a primary cause of acid rain. Biodiesel can be produced anywhere in the world (hot or cold climatic regions) based on availability of feed-stocks [9].

Since the periods of energy revolutions and advancements in biodiesel as alternate fuel for diesel engines, plant source has often been used as feedstock. Over 350 plants species have been identified as oil rich for biodiesel with edible plants oil accounting for 95 % [9]. USA is the leading producer of soybean and corn with surplus for export and biodiesel [10]. Malaysia produces palm oil, Canada has large rapeseed fields, safflower in south Africa and coconut in Philippines. Other edible oil feed-stocks are sunflower, milkweed, linseed and rice bran. Non-edible oil sources for biodiesel are *Jatropha curcas* [11], *calophyllum inophyllum* [11], *Acrocomia aculeata* (*macaúba*) [12], desert date [13], jojoba [14] and among many others. Edible oils when used for biodiesel creates unfair competition with food [10]. Non-edible oils for biodiesel contributes to conversion of arable land to plant trees as well as increase use of fertilizers for faster tree growth. Microalgae on the other hand can lead to production of biodiesel 30 times more than other feed-stocks utilizing a smaller area. The main limitation on microalgae is the sensitivity of strains to contamination, high production cost and limited technology for biomass harvesting [15]. The above mentioned challenges raises the cost of feedstock which eventually increases retail price of biodiesel. Thus a new feed-stock source, waste animal fats (WAFs) composed of triglyceride is cheap, economical and easily available can totally replace vegetable oils and microalgae. Commonly used WAFs are chicken fat [16], lard [17], leather tanning [18], waste beef tallow [19] and fish oil [20].

Beef is the most extensively used meat all over the world [15]. During rendering of beef, residual materials like beef tallow are produced in slaughterhouses, and primarily utilized in the soap production industry. However, when this industry is overloaded, the extra fats are often disposed of in landfills or incinerated, which can pollute the environment. Therefore, the integrated use of bio-residues produced in slaughterhouses can prevent pollution induced by accumulation of these residues in natural resources. Beef tallow fat can be used as a very low-cost renewable source to produce biodiesel of comparable price to conventional petroleum diesel.

This paper reviews the current literature on the use of waste beef tallow for biodiesel production from the published research articles. Conventional and modern techniques available for biodiesel synthesis by transesterification, factors affecting transesterification and properties of beef tallow methyl esters have been discussed.

2. WASTE ANIMAL FATS FEEDSTOCK FOR BIODIESEL

Massive amount of wastes is generated in slaughter houses in rendering of various animal products. Poultry industry for chicken generates massive waste as by-products. Chicken rendering for instance, produces abdominal fats, skins and feathers. Chicken skin occupies larger portion of waste comprising 30 % fats. Fats in chicken abdomen are around 73%. Saturation of chicken fats are lower than other animal fats due to high percentage of oleic and linoleic acids. Enzymes can catalyze waste fats with high biodiesel yield output. Antonio et al. [21] obtained 96 % yield employing CALB enzyme and ethanol as acyl acceptor. Seffatti et al. [22] studied process parameters using heterogeneous $CaO/CuFe_2O_4$ nanocatalyst and obtained 94.52 % biodiesel yield. A two-step catalytic process employed by Keskin et al. [23] produced 87.4 % yield with fat to methanol molar ratio of 1:6 at 63 deg. Purandaradas et al. [24] compared biodiesel from rooster and chicken feather meal using NaOH and methanol. Authors concluded that rooster fat has better properties and quality glycerol than chicken fat. Rendering of pigs produces white semisolid/solid substance known as lard. Lard contains palmitic, stearic, oleic and linoleic as dominant fatty acids for biodiesel synthesis. Mikulski et al. [25] tested swine lard in a four cylinder engine at varying engine speeds of 1500 to 3000 rpm. Authors observed decrease in harmful gas emissions of carbon oxides and hydrocarbons except an increase in nitrogen oxides. Similar results were obtained by Duda et al. [26]. Pyrolysis of swine fat by Lee et al. [27] using environmentally friendly dimethyl carbonate as acyl acceptor produced 97.2 % yield fatty acid methyl ester at 380^0C without thermally cracking fats.

Fish processing plant effluents contains numerous wastes of heads, fins, bones and skin mixed with disposal water. Substantial percentage (6-11%) contains reasonable oil for biodiesel synthesis. de Almeida et al. [28] obtained biodiesel from waste fish oil and other mixtures where overall biodiesel properties is dependent on fatty acid composition in the mixture. Kara et al. [29] obtained biodiesel from high fatty acid waste fish oil by acid pretreatment followed by KOH transesterification. The obtained biodiesel contained no glycerol hence satisfied international standards. Another low cost important feedstock is waste beef tallow generated as waste in cattle slaughterhouses. Beef tallow and waste cooking oil biodiesel mixture were tested on 4-cylinder diesel engine which gave reduction on CO, NO and hydrocarbon emissions by Hazrat et al. [30]. Ghazavi et al. [31] assessed feasibility of beef tallow for biodiesel. The coworkers concluded that beef tallow is readily available feedstock in slaughterhouses at low price for biodiesel. Adewale et al. [32] produced biodiesels on beef tallow assisted by ultrasound. This method proved reduction in time and alcohol spent in reaction which directly lowers cost of production. Vafakish et al. [33] investigated amberlyst catalyst on beef tallow and authors concluded that final product had properties that meets ASTM standards. Table 1 is an overview of waste animal fats conversion methods to biodiesel. Waste beef tallow is less expensive, processed at low cost and easy to obtain from slaughter houses, hence it was preferred compared to other WAFs.

2.1 Advantages of waste beef tallow for biodiesel production

- There is no competition with food for fuel. Waste beef tallow is obtained as inedible waste during rendering process. Instead of being dumped in landfills to create nuisance in environment it is cheap source for biodiesel than edible oils.
- Acts as a lubricant and a solvent. Biodiesel from animal fats has excellent lubrication properties hence can be blended with diesel and alcohol, and acts as a solvent to improve properties.

- Easily available. Beef is widely consumed all over the world as a favourite food and waste generated can be recycled in slaughterhouses for an additional income. Biodiesel producers can buy the tallow at low cost.
- Tallow when converted to biodiesel under optimal reaction conditions results in high biodiesel yield. A yield of 98% was obtained by microwave assisted method for 30 minutes reaction time [34].
- Cetane index of beef tallow was 60.35 is higher than petro-diesel 46.52 which makes it attractive as cetane enhancers in diesel-tallow blend [19].
- The lowest heating value of 36500 kJ/kg of beef tallow makes it acceptable as diesel fuel substitute, though lower than diesel fuel energy content [35].

3. BIODIESEL PRODUCTION METHODS

Waste animal fats have high viscosity and density which limits their applications on diesel engines. Viscosity can be reduced by four different methods namely pyrolysis, emulsification, blending and transesterification.

3.1 Pyrolysis

Pyrolysis of animal fats decomposes triglycerides in oxygen deficient environment to form bio-oil and bio-char. Fast pyrolysis ensures high mass transfer within few seconds in a reactor. Khammasan et al. [78][36] investigated beef fat thermal cracking with ZSM-5 catalyst. The light liquid fuel formed at temperature of 443 ℃ had close properties to diesel though cost implication of the process has to be addressed. Pyrolysis of waste fish produced bio-oil with calorific value 9391 kca/kg at 500 ℃ as reported by Kraiem et al. [37]. Ben Hassen-Trabelsi et al. [38] obtained pyrolyzate of poultry, lamb and swine which had yield of 45.8 to 61.6 %. Co-workers concluded that resultant bio oil requires further enhancement on fuel properties before it is used on diesel engine.

3.2 Microemulsification

Microemulsion creates thermodynamically stable microstructure having diameter of 100 Å 1000 Å. Microemulsion is dynamically stable at constant pressure and temperature hence less agitation is necessary for it to remain in a single phase. Amphiphiles (n-butanol and surfactants) are added to lower viscosity of animal fats. Bora et al. [39] prepared hybrid emulsion of nonedible oil and butanol-ethanol. Diglyceride and FFA acts natural surfactants in the hybrid biofuels giving the diesel like fuel properties similar to biodiesel prepared from base feedstock. Aghbashlo et al. [40] prepared an emulsion fuel of water, diesel and waste cooking oil biodiesel containing nano-cerium oxide additive. The fuel emulsion outperformed pure petro-diesel in pollutant emission with $B5W3_m$ emulsion fuel giving better exergetic and environmental performance.

3.3 Direct use and Blending of fuels

High viscosity of waste animal fats make its direct use impractical in engine. Mixing of diesel and methyl esters gives a blend with a low viscosity than esters. Blends of tallow methyl esters and diesel was used on four stroke engine by Nautiyal et al. [41]. Reduction in exhaust emissions of carbon oxides and hydrocarbon were recorded except a slight increase in nitrogen oxides. Similar result was obtained by Shahir et al. [42] on beef tallow-diesel blend for CRDI engine. Authors concluded that the use of waste fats methyl esters on large scale will solve environmental pollution. A blend of chicken methyl ester, bioethanol and diesel tested for performance and emission by Alptekin et al. [43] showed fuel consumption decreased for low to high test load and emissions increased linearly with engine speeds. In

general, the blend performed well in diesel engine. Chicken fat biodiesel and fish methyl ester blended with pure diesel produced blend with low viscosity and good atomization, as well as reduction of engine emissions of CO, NO and smoke as reported by Behcet et al. [44].

3.4 Transesterification

Transesterification is a simple process for conversion of waste beef tallow to biodiesel which is preferred than other methods. Transesterification also called alcoholysis breaks down triglyceride in presence of alcohol and suitable catalyst to yield glycerol and ester as by products as shown in Figure 1. Most animal fats have high fatty acid content (>1 %) which makes reactions difficult due to formation of soaps [45]. Transesterification of animal fats have been synthesized from various catalysts grouped as alkaline, acidic and enzymatic. Non-catalytic transesterification was performed on duck tallow by kwon et al. [46] using thermochemical process of carbon dioxide and activated alumina as support material. Authors reported high conversion rate of 93.5 % biodiesel using less methoxide at ambient temperatures. Microwave and ultrasound transesterification are assisted methods to increase conversion rates within a short time [47]. Table 2 shows a review of transesterification reactions of beef tallow and other animal fats.

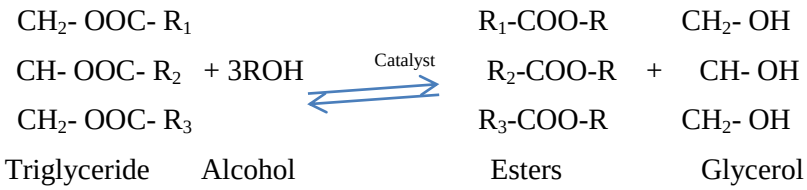


Figure 1 transesterification chemical equation

3.4.1 Homogeneous transesterification

Homogeneous transesterification common in industrial scale uses alkali or acid catalysts. Feedstock of high quality favours use of alkali catalysts but in low quality feedstock having high free fatty acid, acid catalysts are used. Acid catalysts can catalyse both esterification and transesterification simultaneously. da Cunha et al. [19] utilized alkaline catalyst KOH and methanol on beef tallow of low acid content 1.2 to 1.8 mgKOH/g which was within the acceptable limit for yield of 96.4 %. Similar results was reported by Ghazavi et al. [31]. Fadhil [48] produced biodiesel from beef tallow with NaOH and KOH catalysts. A two-step alkaline process produced more biodiesel yield than one step.

3.4.2 Heterogeneous transesterification

Heterogeneous catalysts both acidic and basic when used in reaction facilitates ease of products separation, minimizes waste water for purification and non-corrosive to engine fuel system. Hydrotalcites, alkali oxides, zeolites and activated carbon supported on alkali metals are common heterogeneous catalysts. Liu et al. [49] studied performance of Mg-Al hydrotalcite poultry fat biodiesel synthesis. At optimal conditions of 120 ℃ reaction temperature and 1:6 methanol to fat molar ratio, authors achieved 94 % biodiesel yield. Fadhil et al. [50] investigated PA/CaO catalyst on waste fish oil and bitter almond oil at 60 ℃ and 9:1 molar ratio for 2 h reaction time which produced over 91.22 % biodiesel yield. Gogul et al. synthesized biodiesel from HCA Immobilized AuNPS AMINE grafted SBA-15 catalyst on waste animal fats at 1:3 molar ratio, 60 ℃ reaction temperature for 90 minutes reaction time. Authors achieved 94 % yield biodiesel. Stojkovic´ et al. [17] produced lard methyl ester using quicklime (CaO) as cheap catalyst under 6:1 methanol-to-lard molar ratio, 60 min reaction time and 60 ^{0}C reaction temperature. Heterogeneous catalysts possess challenges for re-use in some reactions, more alcohol consumption from high alcohol to fat ratio, reactions take longer time and reactions occur at elevated temperatures.

3.4.4 Enzymatic transesterification

Enzymes have ability to catalyse both esterification and transesterification simultaneously of animal fats with high acid value. Immobilized enzymes confined to a support material may be affected by mixing technique of transesterification. Adewale et al. [32] employed ultrasound mixing at varying amplitudes on waste tallow and CALB enzyme catalyst. Authors observed that an increase in enzyme concentration increased biodiesel yield for all ultrasound amplitudes tested. Aryee et al. [51] examined shaking water bath mixture for animal fat catalysed by *Mucor miehei* (Lipozyme-IM). Authors reported low yield of 50 % at reaction time of 96 h. Lu et al. [52] while using reciprocal shaking mixer on lard fat catalyzed by immobilized *Candida* sp achieved 87.4 %. Addition of solvents affects enzyme and biodiesel yield in reaction mixture. Pollardo et al. [53] examined solvent addition on immobilized *Candida antarctica* lipase B performance on WAFs methanolysis. Authors observed a 50 % increase in biodiesel yield than normal conditions. Similarly, Skoronski et al. [54] reported biodiesel increase on addition of n-hexane to enzyme lipolase 100 L EX. Lee et al. [55] investigated supercritical CO_2 for refined fat and soy bean catalysed by *Candida antarctica* lipase B. The authors concluded that supercritical CO_2 enhanced mass transfer between reactants and enzyme. Further increase of carbon (iv) oxide caused a decreased in methanol-enzyme interaction leading to decrease in biodiesel yield. Enzymatic transesterification has limitation on high cost of enzymes, reactions takes longer period of time and enzymes are inactivated by alcohol at high concentrations.

3.4.5 Supercritical non-catalytic transesterification

A supercritical fluid has no distinct liquid and gas phases above supercritical pressure and temperature hence it can dissolve in liquid with ease and diffusion occurs in solids faster. The main advantage of this process is absence of water formation and soaps as a result of high acid content of animal fats. Sales et al. [56] produced biodiesel from waste animal fat at 240 ℃, 2 h reaction time and pressures ranging from 20 bars to 45 bars. The presence of water and free fatty acids improved conversion in esterification and fatty acid alkyl esters under thermodynamic conditions employed. Niza et al. [57] produced jatropha biodiesel in supercritical methyl acetate at temperature range of 330-420 ℃. Methyl oleate can withstand high temperature than methyl linoleate that degrades at 330 ℃ because hydrogen atoms near methylene group are dislodged easily. Authors observed that for prolonged temperatures biodiesel yield decreased as a result of decomposition of biodiesel formed.

3.5 Assisted transesterification technique (ATT)

Conventional trans-esterification reactions take longer reaction time, consumes high energy and low efficiency in process parameters. These drawbacks can be improved by assisted transesterification techniques categorized as microwave assisted (MA) and ultrasound assisted (UA). Both MA and UA operate in mild reaction conditions with low catalyst concentration and reduced alcohol to oil molar ratio.

3.5.1 Ultrasound assisted technique

Ultrasound waves when propagated through a liquid create acoustic streaming in direction of waves due to pressure gradient of attenuated sound. The US has frequencies ranging from 20 kHz to 100 kHz that creates bubbles in a liquid leading to mass transfer and heat transfer. Application of UA in experimental biodiesel production of waste animal fats has been reported. Maghami et al. [58] compared UA and conventional methods using waste fish oil at 6:1 oil to alcohol molar ratio, 55 ℃ at varying times. Authors reported high yield of 87 % for UA at 30 minutes reaction time and 79.6 % yield of conventional method at 1 h reaction time.

Adewale et al. [32] investigated ultrasonic parameters and lipase *Candica Antarctica* on waste tallow biodiesel yield. The authors concluded that increase in ultrasonic amplitude and enzyme concentration increases biodiesel yield. Constant ultrasonic amplitude rate shows hydrolysis and methanolysis occurs simultaneously.

3.5.2 Microwave assisted technique

Elecromagnetic waves are formed by coupling of magnetic and electric fields in a perpendicular position to each other. Microwaves are electromagnetic waves with frequency between 300 MHz and 300 000 MHz, and wavelength range of 0.01 m to 100 m. High frequency waves absorb more energy with a short wavelength. Methanol is often used in microwave assisted transesterification due to high capability to absorb microwaves and high polarity organic solvents. Da Rós et al. [59] utilized microwave irradiation on beef tallow ethanolysis and 1:6 ethanol to fat molar ratio at 50 ℃ for 8 hrs. The authors reported microwave irradiation has benefits on less enzyme destruction, low alcohol to oil molar ratio and high yield than conventional heating mode.

4. FACTORS AFFECTING TRANSESTERIFICATION REACTIONS OF BEEF TALLOW

4.1 Free fatty acid and water content

Animal cell membranes are build-up of phospholipids containing free fatty acids as a major component. High acid value of beef tallow requires esterification in order to use alkaline catalyst to avoid soap formation and catalyst inactivation. Acid value of beef tallow was reduced from 78.8 % to 2 % using sulphuric acid catalyst [33]. **Table** 1 gives optimal esterification reaction for animal fats.

Table 1 Review of optimum conditions esterification process of various waste animal fats

Feedstock	Catalyst (wt/wt of fat)	Molar ratio	Temp ^{o}C	Time	Reference
Tallow	H_2SO_4(1wt %)	6:1	60	1h	[60]
Mixed WAF	H_2SO_4 (8wt %)	20:1	40	1h	[61]
Mixed WAF	H_2SO_4(10wt %)	30:1	60	1h	[62]
Chicken	H_2SO_4 (20wt %)	40:1	60	1.33h	[62]
Chicken	H_2SO_4 (20wt %)	40:1	60	1.33h	[43]
Pork fat	H_2SO_4 (2wt %)	6:1	65	5h	[63]
Tallow	KOH (1.5wt %)	6:1	55	1h	[64]
Tallow	H_2SO_4	63:1	65	1h	[64]

4.2 Catalyst type and concentration

Beef tallow analysis has shown high concentration of fatty acids > 1 %. This acid value requires pretreatment in order to use alkaline catalyst for transesterification. Chemical reactions and biodiesel yield for beef tallow depends on catalyst concentration. Most catalysts concentration used for beef tallow range from 0.25 % to 2.5 % wt/wt of fat. da Cunha et al. [19] used 1.5 % KOH and Fadhil et al. [50] used 0.5 % KOH for beef tallow biodiesel synthesis. Commonly used catalyst are NaOH and KOH with the latter being more efficient from high solubility in alcohol and easier separation of glycerol.

4.3 Fat-to-alcohol molar ratio

The theoretical stoichiometric ratio for fat to alcohol reaction to occur is normally 1:3. Since the reaction is reversible, high molar ratio than theoretical is used to ensure reaction moves toward formation of products. Excess methanol in products favours formation of glycerol than methyl esters and hence creates problems in gravity separation requiring an additional

separation process like centrifugation as reported by da Cunha et al. [19]. Tippayawong et al. [34] investigated molar ratio of 1:3 to 1:9 on beef tallow and authors observed an increase in biodiesel yield with increase in methanol upto 1:6 molar ratio. High molar ratio decreases yield due to catalyst inactivation by methanol.

4.4 Reaction time and temperature

Animal fats especially beef tallow have high viscosity that slows down reaction at the start of experiment as methanol distributes in the fats. Use of methanol catalyst requires temperature lower than its boiling point of 65 ℃. Yahyaee et al. [65] observed change in colour from dark brown at the start of reaction to light colour after 2 h reaction time for fish biodiesel. Sander et al. [66] reported an increase in efficiency with increase in temperature . Moreover, density and viscosity was lowered at 50 ℃ due to high interface area for mass transfer of reactants.

4.5 Addition of co-solvents

The use of excess methanol in reactants results in insolubility of methanol in fats and forms emulsions that reduces mass transfer. Addition of co-solvents to the reactants in the reactor improves solubility hence reduction in time. Fadhil et al. [48] investigated hexane co-solvent on biodiesel yield of chicken fat. The authors observed an increase in fatty acid ethyl esters and enhanced properties of biodiesel.

5. PROPERTIES AND CHARACTERISTICS OF WASTE BEEF TALLOW BIODIESEL

International regulatory bodies require biodiesel to meet the set standards of ASTM and EN limits (table 5) When it is compared with edible oils, beef tallow biodiesel publications are still scanty. The available articles show beef tallow biodiesel meets acceptable standards for both physical and chemical composition. The physical and chemical composition of beef is shown in Table 3.

Table 2 Review of transesterification parameters for waste beef fat and waste animal fats

Feedstock	Type/volume of reactor, mixer	Catalyst loading (wt/wt of fat)	T °C	Molar ratio Fat:alcohol	Agitation (rpm)	Time (mins)	Production technique	Optimal reaction conditions		Ref.
								Reaction condition	% yield	
Beef tallow	Glass container/250 ml	NAOH/0.25-0.75 wt %	40-60	1:3 to 1:9	Low speed stirrer	10-30 min	Microwave	0.75 wt % at 60 °C, 1:6 molar ratio, 10 mins	98	[34]
Beef tallow	Round bottom flask/500 ml	NaOH/0.5 wt %	32-60	1:6	1100	60 min	Homogeneous	0.5 wt % at 60 °C, 1:6 molar ratio for 60 mins	89	[48]
Beef tallow	Round bottom flask/500 ml	KOH/0.5 wt %	32-60	1:6	1100	60 min	Homogeneous	0.5 wt % at 60 °C, 1:6 molar ratio for 60 mins	91	[48]
Chicken	Flask	KOH	60	1:2.1	600	60 min	Homogeneous base	0.5 wt % at 60 °C, 1:2.1 molar ratio	88	[67]
Beef tallow	flask	KOH	60	1:2.1	600	60 min	Homogeneous base	0.5 wt % at 60 °C, 1:2.1 molar ratio	88	[67]
Commercial fat tallow	Glass flask/2 dm^3	Amberlyst/1-2.7 moles/l	20-65	1:6	-	50-500 min	Heterogeneous	2.2 mo/l at 1:6 molar ratio after 360 min	95	[33]
Poultry fat	Flask/250 ml	Mg-Al hydrocalcite	120	1:30	-	480 min	Heterogeneous base transesteri-fication	1:30 molar ratio, at 120 °C for 6h.	93	[49]
Beef fat	Glass flask/5000 ml	KOH/6 g	60	400g:300ml	Mechani-cal stirrer	60 min	Two-step homogeneous transesteri-fication	0.15 %wt at 60 ℃ for 1h	93	[68]

Table 3 Fatty acid profile of beef tallow and other waste animal fats feedstock

Feedstock	Lauric 12:0	Myristic 14:0	Palmitic 16:0	Palmitoleic 16:1	Stearic 18:0	Oleic 18:1	Linoleic 18:2	Linolenic 18:3	20:0	Ref
Beef tallow	-	2.72	25.3	2.02	34.7	29.87	0.75	-	-	[19]
-	1.43	6.31	28	4.7	18	41	3.3	0.8	-	[67]
-	0.085	3.116	31.376	1.815	25.236	31.091	1.434	0.233	0.77	[66]
-	-	2.72	25.33	2.02	34.7	29.87	0.75	-	0.28	[19]
-	-	1.5	28.1	4.0	12.0	44.6	9.4	0.5	-	[69]
-	-	2.68	26.18	1.90	33.69	30.09	-	-	0.30	[70]
Lard	0.068	1.137	19.19	2.04	11.81	44.6	10.87	0.99	0.234	[66]
Chicken	0.034	0.5	22.055	6.181	5.050	40.314	16.515	0.625	0.106	[66]
Waste fish oil	-	1.19	10.19	14.79	21.76	24.93	11.58	9.31	-	[19]

Table 4 Physico-chemical properties of beef tallow and beef tallow biodiesel

	Waste beef tallow fat			Waste beef tallow methyl ester								
References	[48]	[67]	[71]	[66]	[48]	[67]	[72]	[73]	[74]	[75]	[64]	[76]
Density at 15 °C (kg/m3)	921.6	971	881.5	919	888.9	859	832	873.2	-	872	-	883
Kinematic Viscosity at 40 °C (mm2/s)	37.39	36.39	-	35.5	4.36	5.29	4.89	5.85	5.0	5.3	5.0	4.6
Oxygen %		-	-	-	-	-	-		-	-	-	10.79
Flash point	165	161	-	-	138	161	152	163	150	156.7	>130	160
Cloud point	14	-	-	-	11	-	-	-	11	-	-	3
CFPP		-	-	-		-	15	-	10	-	-	13
cetane number	65.11	-	50.17	-	-	58.2	-	56	-	64.8	-	50
Acid value (mgKOH/g)	4.66	1.07	-	1.076	0.35	0.2	-	-	0.44	-	0.1	0.1
Iodine value (gl2/100 g)	33	44	-	-	31	44	-	-	53.6	-	-	26.4
HHV (MJ/kg)		38.6	-	-	-	41	-	38.35	-	39.932	37.2	36.5
Water content (% mass)				-	-		-	-	0.005	--	-	-
Carbon residue (%)	0.23			-	0.09		-			-	-	76.77

Table 5 Biodiesel standard specification based on ASTM D6751 and EN 14214 [77]

	ASTM D6751		EN 14214	
	Test method	limits	Test method	Limits
Flash point	D 93	130.0 °C min	EN ISO 3679	120.0 °C min
Water	D 2709	0.050 vol.% max	EN ISO 12937	500 mg/kg max
Kinematic viscosity	D 445	1.9 mm2/s to 6.0 mm2/s	EN ISO 3104	3.5 mm2/s 5.0 mm2/s
Density	-	-	EN ISO 3675	860 kg to 900 kg/m3
Ester content	-	-	EN 14103	96.6 mol%
Sulphated ash	D 874	0.020 wt% max	ISO 3987	0.02 mol% max
Sulfur	D 5483	0.0015 % max (S15) 0.05 max (S500)	EN ISO 20846	10.0 mg/kg max
Copper corrosion	D 130	No. 3 max	EN ISO 2160	1 degree of corrosion
Cetane number	D 613	47 min	EN ISO 5165	51 min
Cloud point	D 2500	-	-	-
Carbon residue	D 4530	0.050 wt% max	EN ISO 10370	0.30 mol% max
Acid number	D 664	0.50 mg KOH/ g max	EN 14104	0.50 mg KOH/ g max
Free glycerine	D 6584	0.02 wt% max	EN 14105	0.02 mol% max
Total glycerine	D 6584	0.240 wt% max	EN 14105	0.25 mol% max
Phosphorus content	D 4951	0.001 wt% max	EN 14108	10 mg/kg max
Sodium/potassium	UOP 391	5 ppm max	EN 14107	5 mg/kg max
Distillation temperature	D 1160	360 °C max	-	-
Methanol content	-	-	EN 14110	0.20 mol% max

5.1. Kinematic viscosity

Viscosity is simply the resistance of liquid when measured at time interval to flow through a specified orifice. The experimental test conditions and production technique influence kinematic viscosity. Pure animal fats are impractical to use in diesel engine due to high viscosity that affects flow rate, fuel atomization in combustion chamber and possible soot formation in injectors. Beef tallow has high viscosity than the resultant methyl esters as shown in table 5. Other animal fats show the same trend and more so saturation level is directly proportional to kinematic viscosity. Beef tallow viscosity ranges from 35 to 37.9 mm^2/s [48, 67] while for beef tallow biodiesel ranges from 4.36 to 5.89 mm^2/s [64, 76]

5.2 Density

Density is the amount of mass of a substance acting per unit volume of a liquid or a solid. The SI units of density is kg/m^3. Density has impact on fuel atomization as it indicates the delay period between injection and combustion of fuel in diesel engines. Density is directly proportional to unsaturation and number of carbon atoms in the feedstock. Test methods for density of biodiesel are EN ISO 3675/12185 and ASTM Standard D1298. Density of beef biodiesel ranges from 870 kg/m^3 to 890 kg/m^3 [64, 67].

5.3 Flash Point

Flashpoint is the temperature which biodiesel ignites to liberate heat energy. The double bonds and carbon atoms in beef tallow affects flashpoint for the methyl ester. The flashpoint of biodiesel is tested based on EN ISO 3679 and ASTM D93. In a fuel blend, the flashpoint of petro-diesel can be improved by increasing the concentration of biodiesel. Teixera et al. [72] obtained a linear relation between beef biodiesel concentration and flashpoint in a blend of soy bean, petro-diesel and beef tallow ester. Wyatt et al. [74] reported flashpoint of 150 ℃ for beef tallow biodiesel.

5.4 Cloud Point and Pour Point

Cloud point (CP) and pour point (PP) determine the possibility of biodiesel use when subjected to low temperature conditions like winter seasons. CP is the temperature at which wax crystals appear when biodiesel is cooled under controlled conditions. Pour point is temperature that allows the wax crystals formed to gel biodiesel. Hence, it is the lowest temperature at which biodiesel can still flow. The CP and PP in beef tallow biodiesel is higher than in petroleum diesel. CP and PP are measured based on ASTM D2500 and D97 standards (Table 4). CP and PP indicate amount of saturated fatty acids in biodiesel. Teixera et al. [72] reported CP of 3 ℃ for beef tallow while Wyatt et al. found 3 ℃ cloud point.

5.5 CFPP

Cold filter plugging point (CFPP) is temperature which a known volume of biodiesel completely flows under vacuum through a wire mesh screen filter at time interval of 60 seconds. CFPP indicates crystallization point of biodiesel. CFPP values of biodiesel is based on ASTM D6371 standards. CFPP measures filterability of fuel and better characteristic than CP for biodiesel and diesel. Unsaturated fatty acids have lower melting points than saturated fatty acids thus crystallize at lower temperature than the saturates. Biodiesel fuels that is obtained from saturated fats like beef tallow compounds give a higher CFPP than diesel. Beef tallow biodiesel has CFPP between 10 to 15 [72, 74, 76]

5.6 Iodine Number

Iodine number is a value determining the number of double bonds and degree of unsaturation in biodiesel. Iodine number determines oxidation stability, viscosity, CFPP and cetane number. According to EN 14111 standards, iodine number maximum value is 120 mgI_2/g. Beef tallow-based, which is highly saturated, has the lowest value, while chicken and goose fat-based concentration of biodiesels the have the highest iodine number as reported by Sander et al. [66]. Reported iodine numbers of beef tallow biodiesel are 44 [67], 53.6 [74] and 26.4 [76].

5.7 Calorific Value

Calorific value is an important property for selection beef tallow biodiesel because high calorific value releases high heat during combustion. High calorific value of biodiesel determines performance of engine. The calorific value of biodiesel is lower than diesel because biodiesel has high oxygen content attached to carbon bonds. Mata et al. [78] found that beef tallow and chicken fat methyl esters have higher calorific values of 39.4 MJ/kg and 40.0 MJ/kg respectively, but they are still lower than diesel. Ashraf et al. [67] reported heating value of 41 MJ/kg for beef tallow methyl esters. Other similar data was reported by Teixera et al. [72] , Selvam et al. [73] and Rajak et al. [75]

5.8 Acid Number

Acid number measures the carboxylic group content for chemical compound such as fatty acid or mixtures of compound. Acid number is denoted in mgKOH/g which is the amount needed to neutralize 1 g of free fatty acid in a sample. European standard (EN 14104) and ASTM D 664 has set maximum acid number of biodiesel to be 0.5 mg KOH/g (table 5). A high acid value causes corrosion in the fuel supply system and metallic parts of engine. Acid values of beef tallow are higher than beef tallow fat methyl esters (Table 5). Some acid values of beef tallow biodiesel reported in literature are 0.1 mgKOH/g [76] and 0.44 mgKOH/g [74] which are within acceptable limits.

5.9 Cetane Number

Cetane number which is dimensionless descriptor relates to ignition quality of a fuel. A high cetane number reduces ignition delay of fuel. Thus, the higher the cetane number, the better the ignition quality. Beef tallow with more saturated molecules has more carbon chains which increases cetane number. da Cunha et al. [19] reported a higher cetane index (60.35) of beef tallow biodiesel than diesel fuel (46.52). Sometimes, cetane index is used to estimate the ASTM cetane number small quantity of sample is available for an engine rating or an engine is not available. The high cetane index of biodiesel makes it attractive as an alternative to diesel fuel [19]. Ashraf et al. [67] found cetane number of 58.2 beef tallow biodiesel while Selvam et al. [73] reported a value of 56.

5.10 Fatty Acid Composition

The analysis from gas chromatography of beef tallow for fatty acid profile show high concentration of palmitic and stearic acids. Table 4 indicates the fatty acid profile of waste beef tallow and other animal fats.

6. CONCLUSION

Waste animal fats especially beef tallow are available at low cost for synthesis of quality biodiesel. Beef tallow has high percentage of saturated fats which make it solid at room temperature. Esterification process has been carried out using sulphuric acid to lower the high

acid value prior to transesterification using alkaline catalysts. Transesterification technique has been expansively elaborated due to its wide commercial application as reactions are carried out at mild conditions. Homogeneous catalysts of NaOH and KOH have been used in many experiments. KOH catalyst has advantage of faster reactions and formation of softer glycerols than NaOH. Low chain alcohols of methanol and ethanol are used for high efficiency and high yield. Some solvents like hexane when added onto reactants increases solubility of methanol and increases the yield. Beef tallow biodiesel has high flash point which makes it safe during transportation. Also, high cetane number makes beef tallow methyl esters attractive for blending with diesel fuel for improvement in combustion. The published experimental results show beef tallow methyl esters meet ASTM D 6751 standard requirement making biodiesel a suitable substitute for diesel. However, more research has to be done on heterogeneous catalysts and low cost lipase catalyst to improve productivity.

REFERENCES

[1] Holdren, J. P. Population and the energy problem, *Population and environment,* 12(3), 1991, pp. 231-255. https://doi.org/10.1007/BF01357916

[2] Demirbaş A., Biodegradability of biodiesel and petrodiesel fuels, *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects,* 31(2), 2008, pp. 169-174

[3] Wang, W. C., Bai C.-J., Lin C.-T., and Prakash S., Alternative fuel produced from thermal pyrolysis of waste tires and its use in a DI diesel engine, *Applied Thermal Engineering,* 93, 2016, pp. 330-338.

[4] Bolonio D., García-Martínez M.-J., Ortega M. F., Lapuerta, M. J., Rodríguez-Fernández, and Canoira L., "Fatty acid ethyl esters (FAEEs) obtained from grapeseed oil: A fully renewable biofuel," *Renewable Energy,* 132, 2019, pp. 278-283.

[5] Knothe G., Razon L. F., Madulid D. A., Agoo E. M. G., and de Castro M. E. G., Methyl esters (biodiesel) from Pachyrhizus erosus seed oil, *Biofuels,* 9(4), 2018, pp. 449-454.

[6] Chen H., Xie B., Ma J., and Chen Y., NOx emission of biodiesel compared to diesel: Higher or lower? *Applied Thermal Engineering,* 137, 2018, pp. 584-593.

[7] Shahid E. M. and Jamal Y., Performance evaluation of a diesel engine using biodiesel, *Pakistan Journal of Engineering and Applied Sciences,* 2016.

[8] Canha N., Felizardo P., and Correia M. J. N., "Controlling the oxidative stability of biodiesel using oils or biodiesel blending or antioxidants addition," *Environmental Progress & Sustainable Energy,* 37(3), 2018, pp. 1031-1040.

[9] Knothe G., Krahl J., and Van Gerpen J., *The biodiesel handbook*. Elsevier, 2015.

[10] Koizumi T., Biofuels and food security, *Renewable and Sustainable Energy Reviews,* 52, 2015, pp. 829-841.

[11] Marutani, M., Soria, J. and Martinez M., Characterization of Crude and Biodiesel Oils of Jatropha curcas and Calophyllum inophyllum in Guam, *Micronesica,* 1, 2018 pp. 15.

[12] César, A. d. S., Almeid, F. d. A. a, de Souza, R. P., Silva, G. C., and Atabani, A. E., The prospects of using Acrocomia aculeata (macaúba) a non-edible biodiesel feedstock in Brazil, *Renewable and Sustainable Energy Reviews,* vol. 49, pp. 1213-1220, 2015/09/01/ 2015.

[13] MercyAkaagerger, S., Giwa, S. O., Ibrahim, M., and Giwa, A., Production of biodiesel from desert date seed oil, *International Journal of ChemTech Research,* 9, 2016, pp. 453-463.

[14] Sandouqa, A. and Al-Hamamre, Z., Energy analysis of biodiesel production from jojoba seed oil, *Renewable Energy,* 130, 2019, pp. 831-842.

[15] Islam, M. A., Heimann, K., and Brown, R. J., Microalgae biodiesel: Current status and future needs for engine performance and emissions, *Renewable and Sustainable Energy Reviews,* 79, 2017, pp. 1160-1170.

[16] Kirubakaran, M. and Selvan, V. A. M., A comprehensive review of low cost biodiesel production from waste chicken fat, *Renewable and Sustainable Energy Reviews,* vol. 82, 2018, pp. 390-401.

[17] Stojković, I. J., Miladinović, M. R., Stamenković, O. S., Banković-Ilić, I. B., Povrenović, D. S., and Veljković, V. B., Biodiesel production by methanolysis of waste lard from piglet roasting over quicklime, *Fuel,* 182, 2016, pp. 454-466.

[18] Šánek, L., Pecha, J., Kolomazník, K., and Bařinová, M. Biodiesel production from tannery fleshings: Feedstock pretreatment and process modeling, *Fuel,* 148, 2015, pp. 16-24.

[19] da Cunha, M. E., Beef tallow biodiesel produced in a pilot scale, *Fuel Processing Technology,* vol. 90(4) ,2009, pp. 570-575. https://doi.org/10.1016/j.fuproc.2009.01.001

[20] Lin, C.-Y. and Li, R.-J., Engine performance and emission characteristics of marine fish-oil biodiesel produced from the discarded parts of marine fish, *Fuel Processing Technology,* 90(7-8), 2009, pp. 883-888.

[21] Lin, C.-Y. and Li, R.-J. Engine performance and emission characteristics of marine fish-oil biodiesel produced from the discarded parts of marine fish, *Fuel Processing Technology,* vol. 90(7-8) , 2009, pp. 883-888.

[22] Seffati, K., Honarvar, B., Esmaeili, H., and Esfandiari, N., Enhanced biodiesel production from chicken fat using CaO/CuFe2O4 nanocatalyst and its combination with diesel to improve fuel properties, *Fuel*, vol. 235(1238-1244), 2019. https://doi.org/10.1016/j.fuel.2018.08.118

[23] Keskin, A., Two-step methyl ester production and characterization from the broiler rendering fat: The optimization of the first step, *Renewable energy,* 122, 2018, pp. 216-224. https://doi.org/10.1016/j.renene.2018.01.123

[24] Purandaradas *et al*, A.., Development and quantification of biodiesel production from chicken feather meal as a cost-effective feedstock by using green technology, *Biochemistry and Biophysics Reports*, 14, 2018,pp. 133-139. doi:https://doi.org/10.1016/j.bbrep.2018.04.012

[25] Mikulski, M., Duda, K., and Wierzbicki, S., Performance and emissions of a CRDI diesel engine fuelled with swine lard methyl esters–diesel mixture, *Fuel,* 164, 2016, pp. 206-219. https://doi.org/10.1016/j.fuel.2015.09.083

[26] Duda, K., Wierzbicki, S., Śmieja, M., and Mikulski, M. Comparison of performance and emissions of a CRDI diesel engine fuelled with biodiesel of different origin, *Fuel,* 212, 2018, pp. 202-222. https://doi.org/10.1016/j.fuel.2017.09.112

[27] Lee, J., Tsang, Y. F., Jung, J.-M., Oh, J.-I., Kim, H.-W., and Kwon, E. E., In-situ pyrogenic production of biodiesel from swine fat, *Bioresource Technology*, 220, 2016, pp. 442-447. doi: 10.1016/j.biortech.2016.08.100

[28] de Almeida, V. F., García-Moreno, P. J., Guadix, A., and Guadix, E. M., Biodiesel production from mixtures of waste fish oil, palm oil and waste frying oil: Optimization of fuel properties, *Fuel Processing Technology,* 133, 2015, pp. 152-160. https://doi.org/10.1016/j.fuproc.2015.01.04

[29] Kara, K., Ouanji, F., Lotfi, E. M., Mahi, M. El, Kacimi, M., and Ziyad, M., Biodiesel production from waste fish oil with high free fatty acid content from Moroccan fish-processing industries, *Egyptian Journal of Petroleum,* vol. 27(2) , 2018, pp. 249-255. https://doi.org/10.1016/j.ejpe.2017.07.010.

[30] Hazrat, M. A., Rasul, M. G., Khan, M. M. K., Ashwath, N., and Rufford, T. E., Emission characteristics of waste tallow and waste cooking oil based ternary biodiesel fuels, *Energy Procedia,*. 160, 2019, pp. 842-847. https://doi.org/10.1016/j.egypro.2019.02.149

[31] Ghazavi, M. A., Fallahipanah, M., and Jeshvaghani, H. S., A feasibility study on beef tallow conversion to some esters for biodiesel production, *International Journal Of Recycling of Organic Waste in Agriculture,* 2(1) , 2013, p. 2. https://doi.org/10.1186/2251-7715-2-2

[32] Adewale ,P., Dumont, M.-J., and Ngadi, M. Enzyme-catalyzed synthesis and kinetics of ultrasonic-assisted biodiesel production from waste tallow, *Ultrasonics Sonochemistry,* 27, 2015, pp. 1-9. https://doi.org/10.1016/j.rser.2015.02.039

[33] Vafakish, B. and Barari, M. Biodiesel production by transesterification of tallow fat using heterogeneous catalysis, *Kemija u industriji: Časopis kemičara i kemijskih inženjera Hrvatske,* 66(1-2) , 2017, pp. 47-52.

[34] Tippayawong, N. and Singkham, R. Microwave assisted production of biodiesel from beef tallow, *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects,*. 37(14) , 2015, pp. 1513-1519.https://doi.org/10.1080/15567036.2011.631092

[35] Cunha, A., Feddern, V., De Prá, M. C, Higarashi, M. M., de Abreu, P. G., and Coldebella, A. Synthesis and characterization of ethylic biodiesel from animal fat wastes, *Fuel*, 105, 2013, pp. 228-234.

[36] Kraiem, T., Hassen-Trabelsi, A. B., Naoui, Belayouni, S., H., and Jeguirim, M., Characterization of the liquid products obtained from Tunisian waste fish fats using the pyrolysis process, *Fuel Processing Technology,* 138, 2015, pp. 404-412.

[37] Ben Hassen-Trabelsi, A., Kraiem, T., Naoui, S., and Belayouni, H., Pyrolysis of waste animal fats in a fixed-bed reactor: Production and characterization of bio-oil and bio-char, *Waste Management,* vol. 34(1), 2014,pp. 210-218. https://doi.org/10.1016/j.wasman.2013.09.019

[38] Bora, P., Konwar, L. J., Boro, J., Phukan, M. M., Deka, D., and Konwar, B. K. Hybrid biofuels from non-edible oils: A comparative standpoint with corresponding biodiesel, *Applied Energy,* 135, 2014, pp. 450-460.

[39] Aghbashlo *et al.*, M. A novel emulsion fuel containing aqueous nano cerium oxide additive in diesel–biodiesel blends to improve diesel engines performance and reduce exhaust emissions: Part II – Exergetic analysis, *Fuel*, 205, 2017, pp. 262-271. https://doi.org/10.1016/j.fuel.2017.05.003

[40] Nautiyal P., Subramanian, K. and Dastidar, M. Experimental investigation on performance and emission characteristics of a compression ignition engine fueled with biodiesel from waste tallow, *Clean Technologies and Environmental Policy,* vol. 19(6) , 2017, pp. 1667-1677. https://doi.org/10.1007/s10098-017-1355-8

[41] Shahir, V. K., Jawahar, C. P., Suresh, P. R. and Vinod, V. Experimental Investigation on Performance and Emission Characteristics of a Common Rail Direct InjectionEngine Using Animal Fat Biodiesel Blends, *Energy Procedia,* vol. 117, 2017, pp. 283-290.

[42] Alptekin, E., Canakci, M., Ozsezen, A. N., Turkcan, A., and Sanli, H., Using waste animal fat based biodiesels–bioethanol–diesel fuel blends in a DI diesel engine, *Fuel*, vol. 157, 2015, pp. 245-254. https://doi.org/10.1016/j.fuel.2011.11.014

[43] Behçet, R., Oktay, H., Çakmak, A., and Aydin, H., Comparison of exhaust emissions of biodiesel–diesel fuel blends produced from animal fats, *Renewable and Sustainable Energy Reviews*, vol. 46, 2015, pp. 157-165.

[44] Encinar, J. M., Sánchez, N., Martínez, G., and García, L., Study of biodiesel production from animal fats with high free fat ty acid content, *Bioresource Technology*,102(23), 2011, pp. 10907-10914.

[45] Kwon, E. E., Jeon, E.-C., H. Yi, and Kim, S., Transforming duck tallow into biodiesel via noncatalytic transesterification, *Applied Energy*, 116, 2014, pp. 20-25. https://doi.org/10.1016/j.apenergy.2013.11.043

[46] Motasemi, F. and Ani, F. N., A review on microwave-assisted production of biodiesel, *Renewable and Sustainable Energy Reviews*, 16(7), 2012, pp. 4719-4733.

[47] Fadhil, A. B., Biodiesel production from beef tallow using alkali-catalyzed transesterification, *Arabian Journal for Science and Engineering,* vol. 38(1), 2013, pp. 41-47. https://doi.org/10.1080/15567036.2015.1065299

[48] Liu, Y., Lotero, E., Goodwin, J. G. and Mo, X. Transesterification of poultry fat with methanol using Mg–Al hydrotalcite derived catalysts, *Applied Catalysis A: General*, 331, 2007, pp. 138-148. https://doi.org/10.1016/j.apcata.2007.07.038

[49] Fadhil, A. B., Al-Tikrity, E. T. B., and Khalaf, A. M. Transesterification of non-edible oils over potassium acetate impregnated CaO solid base catalyst, *Fuel*, 234, 2018, pp. 81-93. https://doi.org/10.1016/j.fuel.2018.06.121

[50] Aryee, A. N. A., Simpson, B. K., R. I. Cue, and Phillip,L. E. Enzymatic transesterification of fats and oils from animal discards to fatty acid ethyl esters for potential fuel use, *Biomass and Bioenergy*, vol. 35(10), 2011, pp. 4149-4157. https://doi.org/10.1016/j.biombioe.2011.06.002

[51] Lu, J., Nie, K., Xie, F., Wang, F., and Tan, T., Enzymatic synthesis of fatty acid methyl esters from lard with immobilized Candida sp. 99-125, *Process Biochemistry*, 42(9), 2007, pp. 1367-1370. https://doi.org/10.1016/j.procbio.2007.06.004

[52] Pollardo, A. A., Lee, H.-s., Lee, D., Kim, S., and Kim, J., Solvent effect on the enzymatic production of biodiesel from waste animal fat, *Journal of Cleaner Production*, 185, 2018, pp. 382-388. https://doi.org/10.1016/j.jclepro.2018.02.210

[53] Skoronski, E., de Oliveira, D. C., Fernandes, da Silva, M. G. F., Magalhães, M. d. L. B. and João, J. J. Valorization of agro-industrial by-products: analysis of biodiesel production from porcine fat waste, *Journal of cleaner production*, 112, 2016, pp. 2553-2559.

[54] Lee, H.-s., Lee, D., Kim, S., and Kim, J., Effect of supercritical carbon dioxide on the enzymatic production of biodiesel from waste animal fat using immobilized Candida antarctica lipase B variant, *BMC biotechnology*,17(1), 2017 pp. 70. doi: 10.1186/s12896-017-0390-1.

[55] Sales, E. A., Ghirardi,M. L., and Jorquera, O. Subcritical ethylic biodiesel production from wet animal fat and vegetable oils: A net energy ratio analysis, *Energy Conversion and Management*, 141, 2017, pp. 216-223. https://doi.org/10.1016/j.enconman.2016.08.015

[56] Niza, N. M., Tan, K. TK. Lee, T., and Ahmad, Z. Biodiesel production by non-catalytic supercritical methyl acetate: Thermal stability study, *Applied Energy*, 101, 2013,pp. 198-202. https://doi.org/10.1016/j.apenergy.2012.03.033

[57] Maghami, M., Sadrameli, S. M., and Ghobadian, B., Production of biodiesel from fishmeal plant waste oil using ultrasonic and conventional methods, *Applied Thermal Engineering*, 75, 2015, pp. 575-579. https://doi.org/10.1016/j.applthermaleng.2014.09.062

[58] Da Rós, P. C., de Castro, H. F., Carvalho, A. K., Soares, C. M., de Moraes, F. F. and G. M. Zanin, Microwave-assisted enzymatic synthesis of beef tallow biodiesel, *Journal of industrial microbiology & biotechnology*, 39(4) , 2012, pp. 529-536. https://doi.org/10.1007/s10295-011-1059-8

[59] Panneerselvam, S. I. and Miranda, L. R. Biodiesel production from mutton tallow, in *2011 IEEE Conference on Clean Energy and Technology (CET)*, 2011: IEEE, pp. 83-86.

[60] Gürü, M., Artukoğlu, B. D. A. Keskin, and Koca, A. Biodiesel production from waste animal fat and improvement of its characteristics by synthesized nickel and magnesium additive, *Energy conversion and Management*, 50(3) ,2009, pp. 498-502. https://doi.org/10.1016/j.enconman.2008.11.001

[61] Alptekin, E. and Canakci, M., Optimization of transesterification for methyl ester production from chicken fat, *Fuel*, 90(8), 2011, pp. 2630-2638. https://doi.org/10.1016/j.fuel.2011.03.042

[62] Dias, J. M., Alvim-Ferraz, M. C. M., and Almeida, M. F. Production of biodiesel from acid waste lard, *Bioresource Technology*, vol. 100(24), 2009, pp. 6355-6361. https://doi.org/10.1016/j.biortech.2009.07.025

[63] Fröhlich, A., Rice, B., and Vicente, G. The conversion of low grade tallow into biodiesel-grade methyl ester, *Journal of the American Oil Chemists' Society,* 87(7) , 2010, pp. 825-833. https://doi.org/10.1007/s11746-010-1561-6

[64] Yahyaee, R., Ghobadian, B., and Najafi, G., Waste fish oil biodiesel as a source of renewable fuel in Iran, *Renewable and Sustainable Energy Reviews,* 17, 2013, pp. 312-319,.

[65] Sander, A., Antonije Košćak, M., Kosir, D., Milosavljević,N. Parlov Vuković, J., and Magić, L. The influence of animal fat type and purification conditions on biodiesel quality, *Renewable Energy,* 118, 2018, pp. 752-760.

[66] Ashraf ,S., Hussain, M., Mumtaz, M. W. and Shuaib, M. Biodiesel Production by Alkali Catalyzed Transesterification of chicken and beef fats, *International Journal of Alternative Fuels and Energy,* vol. 1(1) , 2017, pp. 14-20.

[67] Gandure, J., Ketlogetswe, C., and Jonas, M., Production, Composition and Fuel Properties of Tallow Biodiesel: A Case of Botswana, *Energy and Power Engineering,* 9(07) , 2017, p. 355. DOI: 10.4236/epe.2017.97024

[68] Encinar, J., Sánchez, N., Martínez, G., and García, L., Study of biodiesel production from animal fats with high free fatty acid content, *Bioresource Technology,* vol. 102(23) , 2011, pp. 10907-10914. https://doi.org/10.1016/j.biortech.2011.09.068

[69] Moraes *et al.*, M. S. A. Tallow biodiesel: properties evaluation and consumption tests in a diesel engine, *Energy & Fuels,* 22(3) , 2008, pp. 1949-1954.

[70] Alcantara, R., Amores, J., Canoira, L., Fidalgo, E., Franco, M. J. and Navarro, A. Catalytic production of biodiesel from soy-bean oil, used frying oil and tallow, *Biomass and Bioenergy,* 18(6) , 2000, pp. 515-527. https://doi.org/10.1016/S0961-9534(00)00014-3

[71] Teixeira *et al.*, L. S. G. Characterization of beef tallow biodiesel and their mixtures with soybean biodiesel and mineral diesel fuel, *Biomass and Bioenergy,* 34(4) , 2010, pp. 438-441. https://doi.org/10.1016/j.biombioe.2009.12.007

[72] Selvam, D. J. P. and Vadivel, K. Performance and Emission Analysis of DI Diesel Engine Fuelled with Methyl Esters of Beef Tallow and Diesel Blends, *Procedia Engineering,* 38, 2012, pp. 342-358. https://doi.org/10.1016/j.proeng.2012.06.043

[73] Wyatt, V. T. Hess, M. A. Dunn, R. O., Foglia, T. A., Haas, M. J., and Marmer, W. N., Fuel properties and nitrogen oxide emission levels of biodiesel produced from animal fats, *Journal of the American Oil Chemists' Society,* 82(8) , 2005, pp. 585-591. https://doi.org/10.1007/s11746-005-1113-2

[74] Rajak, U. and Verma, T. N. Effect of emission from ethylic biodiesel of edible and non-edible vegetable oil, animal fats, waste oil and alcohol in CI engine, *Energy Conversion and Management,* vol. 166, 2018, pp. 704-718. https://doi.org/10.1016/j.enconman.2018.04.070

[75] Srinivasan, G. R., Shankar, V., and Jambulingam, R.Experimental study on influence of dominant fatty acid esters in engine characteristics of waste beef tallow biodiesel. *Energy Exploration & Exploitation,* 2019, pp. 0144598718821791. https://doi.org/10.1177/0144598718821791

[76] Srinivasan, G. R., Shankar,V. and Jambulingam R., Experimental study on influence of dominant fatty acid esters in engine characteristics of waste beef tallow biodiesel, *Energy Exploration & Exploitation,* 2019, pp. 0144598718821791. https://doi.org/10.1177/0144598718821791

[77] Mata, T. M., Cardoso, N. Ornelas, M., Neves, S., and Caetano, N. S. Evaluation of two purification methods of biodiesel from beef tallow, pork lard, and chicken fat. *Energy & Fuels,* vol. 25(10) , 2011, pp. 4756-4762. DOI: 10.1021/ef2010207

[78] Khammasan, T. and Tippayawong, N., Light liquid fuel from catalytic cracking of beef tallow with ZSM-5, *International Journal of Renewable Energy Research (IJRER),* 8(1), 2018, pp. 407-413.

[79] J. P. Holdren, "Population and the energy problem," *Population and environment,* vol. 12, no. 3, pp. 231-255, 1991.

[80] A. Demirbaş, "Biodegradability of biodiesel and petrodiesel fuels," *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects,* vol. 31, no. 2, pp. 169-174, 2008.

[81] W.-C. Wang, C.-J. Bai, C.-T. Lin, and S. Prakash, "Alternative fuel produced from thermal pyrolysis of waste tires and its use in a DI diesel engine," *Applied Thermal Engineering,* vol. 93, pp. 330-338, 2016/01/25/ 2016.

[82] D. Bolonio, M.-J. García-Martínez, M. F. Ortega, M. Lapuerta, J. Rodríguez-Fernández, and L. Canoira, "Fatty acid ethyl esters (FAEEs) obtained from grapeseed oil: A fully renewable biofuel," *Renewable Energy,* vol. 132, pp. 278-283, 2019/03/01/ 2019.

[83] G. Knothe, L. F. Razon, D. A. Madulid, E. M. G. Agoo, and M. E. G. de Castro, "Methyl esters (biodiesel) from Pachyrhizus erosus seed oil," *Biofuels,* vol. 9, no. 4, pp. 449-454, 2018.

[84] H. Chen, B. Xie, J. Ma, and Y. Chen, "NOx emission of biodiesel compared to diesel: Higher or lower?," *Applied Thermal Engineering,* vol. 137, pp. 584-593, 2018/06/05/ 2018.

[85] E. M. Shahid and Y. Jamal, "Performance evaluation of a diesel engine using biodiesel," *Pakistan Journal of Engineering and Applied Sciences,* 2016.

[86] N. Canha, P. Felizardo, and M. J. N. Correia, "Controlling the oxidative stability of biodiesel using oils or biodiesel blending or antioxidants addition," *Environmental Progress & Sustainable Energy,* vol. 37, no. 3, pp. 1031-1040, 2018.

[87] G. Knothe, J. Krahl, and J. Van Gerpen, *The biodiesel handbook*. Elsevier, 2015.

[88] T. Koizumi, "Biofuels and food security," *Renewable and Sustainable Energy Reviews,* vol. 52, pp. 829-841, 2015/12/01/ 2015.

[89] M. Marutani, J. Soria, and M. Martinez, "Characterization of Crude and Biodiesel Oils of Jatropha curcas and Calophyllum inophyllum in Guam," *Micronesica,* vol. 1, p. 15, 2018.

[90] A. d. S. César, F. d. A. Almeida, R. P. de Souza, G. C. Silva, and A. E. Atabani, "The prospects of using Acrocomia aculeata (macaúba) a non-edible biodiesel feedstock in Brazil," *Renewable and Sustainable Energy Reviews,* vol. 49, pp. 1213-1220, 2015/09/01/ 2015.

[91] S. MercyAkaagerger, S. O. Giwa, M. Ibrahim, and A. Giwa, "Production of biodiesel from desert date seed oil," *International Journal of ChemTech Research,* vol. 9, pp. 453-463, 2016.

[92] A. Sandouqa and Z. Al-Hamamre, "Energy analysis of biodiesel production from jojoba seed oil," *Renewable Energy,* vol. 130, pp. 831-842, 2019/01/01/ 2019.

[93] M. A. Islam, K. Heimann, and R. J. Brown, "Microalgae biodiesel: Current status and future needs for engine performance and emissions," *Renewable and Sustainable Energy Reviews,* vol. 79, pp. 1160-1170, 2017/11/01/ 2017.

[94] M. Kirubakaran and V. A. M. Selvan, "A comprehensive review of low cost biodiesel production from waste chicken fat," *Renewable and Sustainable Energy Reviews,* vol. 82, pp. 390-401, 2018.

[95] I. J. Stojković, M. R. Miladinović, O. S. Stamenković, I. B. Banković-Ilić, D. S. Povrenović, and V. B. Veljković, "Biodiesel production by methanolysis of waste lard from piglet roasting over quicklime," *Fuel,* vol. 182, pp. 454-466, 2016.

[96] L. Šánek, J. Pecha, K. Kolomazník, and M. Bařinová, "Biodiesel production from tannery fleshings: Feedstock pretreatment and process modeling," *Fuel,* vol. 148, pp. 16-24, 2015/05/15/ 2015.

[97] M. E. da Cunha *et al.*, "Beef tallow biodiesel produced in a pilot scale," *Fuel Processing Technology,* vol. 90, no. 4, pp. 570-575, 2009.

[98] C.-Y. Lin and R.-J. Li, "Engine performance and emission characteristics of marine fish-oil biodiesel produced from the discarded parts of marine fish," *Fuel Processing Technology,* vol. 90, no. 7-8, pp. 883-888, 2009.

[99] D. C. Antonio, L. P. Amancio, and I. G. Rosset, "Biocatalytic Ethanolysis of Waste Chicken Fat for Biodiesel Production," *Catalysis Letters,* vol. 148, no. 10, pp. 3214-3222, 2018.

[100] K. Seffati, B. Honarvar, H. Esmaeili, and N. Esfandiari, "Enhanced biodiesel production from chicken fat using CaO/CuFe2O4 nanocatalyst and its combination with diesel to improve fuel properties," *Fuel,* vol. 235, pp. 1238-1244, 2019.

[101] A. Keskin, "Two-step methyl ester production and characterization from the broiler rendering fat: The optimization of the first step," *Renewable energy,* vol. 122, pp. 216-224, 2018.

[102] A. Purandaradas *et al.*, "Development and quantification of biodiesel production from chicken feather meal as a cost-effective feedstock by using green technology," *Biochemistry and Biophysics Reports,* vol. 14, pp. 133-139, 2018/07/01/ 2018.

[103] M. Mikulski, K. Duda, and S. Wierzbicki, "Performance and emissions of a CRDI diesel engine fuelled with swine lard methyl esters–diesel mixture," *Fuel,* vol. 164, pp. 206-219, 2016.

[104] K. Duda, S. Wierzbicki, M. Śmieja, and M. Mikulski, "Comparison of performance and emissions of a CRDI diesel engine fuelled with biodiesel of different origin," *Fuel,* vol. 212, pp. 202-222, 2018.

[105] J. Lee, Y. F. Tsang, J.-M. Jung, J.-I. Oh, H.-W. Kim, and E. E. Kwon, "In-situ pyrogenic production of biodiesel from swine fat," *Bioresource Technology,* vol. 220, pp. 442-447, 2016/11/01/ 2016.

[106] V. F. de Almeida, P. J. García-Moreno, A. Guadix, and E. M. Guadix, "Biodiesel production from mixtures of waste fish oil, palm oil and waste frying oil: Optimization of fuel properties," *Fuel Processing Technology,* vol. 133, pp. 152-160, 2015.

[107] K. Kara, F. Ouanji, E. M. Lotfi, M. El Mahi, M. Kacimi, and M. Ziyad, "Biodiesel production from waste fish oil with high free fatty acid content from Moroccan fish-processing industries," *Egyptian Journal of Petroleum,* vol. 27, no. 2, pp. 249-255, 2018.

[108] M. A. Hazrat, M. G. Rasul, M. M. K. Khan, N. Ashwath, and T. E. Rufford, "Emission characteristics of waste tallow and waste cooking oil based ternary biodiesel fuels," *Energy Procedia,* vol. 160, pp. 842-847, 2019/02/01/ 2019.

[109] M. A. Ghazavi, M. Fallahipanah, and H. S. Jeshvaghani, "A feasibility study on beef tallow conversion to some esters for biodiesel production," *International Journal Of Recycling of Organic Waste in Agriculture,* vol. 2, no. 1, p. 2, 2013.

[110] P. Adewale, M.-J. Dumont, and M. Ngadi, "Enzyme-catalyzed synthesis and kinetics of ultrasonic-assisted biodiesel production from waste tallow," *Ultrasonics Sonochemistry,* vol. 27, pp. 1-9, 2015/11/01/ 2015.

[111] B. Vafakish and M. Barari, "Biodiesel production by transesterification of tallow fat using heterogeneous catalysis," *Kemija u industriji: Časopis kemičara i kemijskih inženjera Hrvatske,* vol. 66, no. 1-2, pp. 47-52, 2017.

[112] N. Tippayawong and R. Singkham, "Microwave assisted production of biodiesel from beef tallow," *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects,* vol. 37, no. 14, pp. 1513-1519, 2015.

[113] A. Cunha, V. Feddern, M. C. De Prá, M. M. Higarashi, P. G. de Abreu, and A. Coldebella, "Synthesis and characterization of ethylic biodiesel from animal fat wastes," *Fuel,* vol. 105, pp. 228-234, 2013/03/01/ 2013.

[114] T. Khammasan and N. Tippayawong, "Light liquid fuel from catalytic cracking of beef tallow with ZSM-5," *International Journal of Renewable Energy Research (IJRER)*, vol. 8, no. 1, pp. 407-413, 2018.

[115] T. Kraiem, A. B. Hassen-Trabelsi, S. Naoui, H. Belayouni, and M. Jeguirim, "Characterization of the liquid products obtained from Tunisian waste fish fats using the pyrolysis process," *Fuel Processing Technology*, vol. 138, pp. 404-412, 2015/10/01/ 2015.

[116] A. Ben Hassen-Trabelsi, T. Kraiem, S. Naoui, and H. Belayouni, "Pyrolysis of waste animal fats in a fixed-bed reactor: Production and characterization of bio-oil and bio-char," *Waste Management*, vol. 34, no. 1, pp. 210-218, 2014/01/01/ 2014.

[117] P. Bora, L. J. Konwar, J. Boro, M. M. Phukan, D. Deka, and B. K. Konwar, "Hybrid biofuels from non-edible oils: A comparative standpoint with corresponding biodiesel," *Applied Energy*, vol. 135, pp. 450-460, 2014/12/15/ 2014.

[118] M. Aghbashlo *et al.*, "A novel emulsion fuel containing aqueous nano cerium oxide additive in diesel–biodiesel blends to improve diesel engines performance and reduce exhaust emissions: Part II – Exergetic analysis," *Fuel*, vol. 205, pp. 262-271, 2017/10/01/ 2017.

[119] P. Nautiyal, K. Subramanian, and M. Dastidar, "Experimental investigation on performance and emission characteristics of a compression ignition engine fueled with biodiesel from waste tallow," *Clean Technologies and Environmental Policy*, vol. 19, no. 6, pp. 1667-1677, 2017.

[120] V. K. Shahir, C. P. Jawahar, P. R. Suresh, and V. Vinod, "Experimental Investigation on Performance and Emission Characteristics of a Common Rail Direct InjectionEngine Using Animal Fat Biodiesel Blends," *Energy Procedia*, vol. 117, pp. 283-290, 2017/06/01/ 2017.

[121] E. Alptekin, M. Canakci, A. N. Ozsezen, A. Turkcan, and H. Sanli, "Using waste animal fat based biodiesels–bioethanol–diesel fuel blends in a DI diesel engine," *Fuel*, vol. 157, pp. 245-254, 2015.

[122] R. Behçet, H. Oktay, A. Çakmak, and H. Aydin, "Comparison of exhaust emissions of biodiesel–diesel fuel blends produced from animal fats," *Renewable and Sustainable Energy Reviews*, vol. 46, pp. 157-165, 2015/06/01/ 2015.

[123] J. M. Encinar, N. Sánchez, G. Martínez, and L. García, "Study of biodiesel production from animal fats with high free fatty acid content," *Bioresource Technology*, vol. 102, no. 23, pp. 10907-10914, 2011/12/01/ 2011.

[124] E. E. Kwon, E.-C. Jeon, H. Yi, and S. Kim, "Transforming duck tallow into biodiesel via noncatalytic transesterification," *Applied Energy*, vol. 116, pp. 20-25, 2014/03/01/ 2014.

[125] F. Motasemi and F. N. Ani, "A review on microwave-assisted production of biodiesel," *Renewable and Sustainable Energy Reviews*, vol. 16, no. 7, pp. 4719-4733, 2012/09/01/ 2012.

[126] A. B. Fadhil, "Biodiesel production from beef tallow using alkali-catalyzed transesterification," *Arabian Journal for Science and Engineering*, vol. 38, no. 1, pp. 41-47, 2013.

[127] Y. Liu, E. Lotero, J. G. Goodwin, and X. Mo, "Transesterification of poultry fat with methanol using Mg–Al hydrotalcite derived catalysts," *Applied Catalysis A: General*, vol. 331, pp. 138-148, 2007/01/01/ 2007.

[128] A. B. Fadhil, E. T. B. Al-Tikrity, and A. M. Khalaf, "Transesterification of non-edible oils over potassium acetate impregnated CaO solid base catalyst," *Fuel*, vol. 234, pp. 81-93, 2018/12/15/ 2018.

[129] A. N. A. Aryee, B. K. Simpson, R. I. Cue, and L. E. Phillip, "Enzymatic transesterification of fats and oils from animal discards to fatty acid ethyl esters for potential fuel use," *Biomass and Bioenergy*, vol. 35, no. 10, pp. 4149-4157, 2011/10/15/ 2011.

[130] J. Lu, K. Nie, F. Xie, F. Wang, and T. Tan, "Enzymatic synthesis of fatty acid methyl esters from lard with immobilized Candida sp. 99-125," *Process Biochemistry,* vol. 42, no. 9, pp. 1367-1370, 2007/09/01/ 2007.

[131] A. A. Pollardo, H.-s. Lee, D. Lee, S. Kim, and J. Kim, "Solvent effect on the enzymatic production of biodiesel from waste animal fat," *Journal of Cleaner Production,* vol. 185, pp. 382-388, 2018/06/01/ 2018.

[132] E. Skoronski, D. C. de Oliveira, M. Fernandes, G. F. da Silva, M. d. L. B. Magalhães, and J. J. João, "Valorization of agro-industrial by-products: analysis of biodiesel production from porcine fat waste," *Journal of cleaner production,* vol. 112, pp. 2553-2559, 2016.

[133] H.-s. Lee, D. Lee, S. Kim, and J. Kim, "Effect of supercritical carbon dioxide on the enzymatic production of biodiesel from waste animal fat using immobilized Candida antarctica lipase B variant," *BMC biotechnology,* vol. 17, no. 1, p. 70, 2017.

[134] E. A. Sales, M. L. Ghirardi, and O. Jorquera, "Subcritical ethylic biodiesel production from wet animal fat and vegetable oils: A net energy ratio analysis," *Energy Conversion and Management,* vol. 141, pp. 216-223, 2017/06/01/ 2017.

[135] N. M. Niza, K. T. Tan, K. T. Lee, and Z. Ahmad, "Biodiesel production by non-catalytic supercritical methyl acetate: Thermal stability study," *Applied Energy,* vol. 101, pp. 198-202, 2013/01/01/ 2013.

[136] M. Maghami, S. M. Sadrameli, and B. Ghobadian, "Production of biodiesel from fishmeal plant waste oil using ultrasonic and conventional methods," *Applied Thermal Engineering,* vol. 75, pp. 575-579, 2015/01/22/ 2015.

[137] P. C. Da Rós, H. F. de Castro, A. K. Carvalho, C. M. Soares, F. F. de Moraes, and G. M. Zanin, "Microwave-assisted enzymatic synthesis of beef tallow biodiesel," *Journal of industrial microbiology & biotechnology,* vol. 39, no. 4, pp. 529-536, 2012.

[138] S. I. Panneerselvam and L. R. Miranda, "Biodiesel production from mutton tallow," in *2011 IEEE Conference on Clean Energy and Technology (CET)*, 2011: IEEE, pp. 83-86.

[139] M. Gürü, B. D. Artukoğlu, A. Keskin, and A. Koca, "Biodiesel production from waste animal fat and improvement of its characteristics by synthesized nickel and magnesium additive," *Energy conversion and Management,* vol. 50, no. 3, pp. 498-502, 2009.

[140] E. Alptekin and M. Canakci, "Optimization of transesterification for methyl ester production from chicken fat," *Fuel,* vol. 90, no. 8, pp. 2630-2638, 2011/08/01/ 2011.

[141] J. M. Dias, M. C. M. Alvim-Ferraz, and M. F. Almeida, "Production of biodiesel from acid waste lard," *Bioresource Technology,* vol. 100, no. 24, pp. 6355-6361, 2009/12/01/ 2009.

[142] A. Fröhlich, B. Rice, and G. Vicente, "The conversion of low grade tallow into biodiesel-grade methyl ester," *Journal of the American Oil Chemists' Society,* vol. 87, no. 7, pp. 825-833, 2010.

[143] R. Yahyaee, B. Ghobadian, and G. Najafi, "Waste fish oil biodiesel as a source of renewable fuel in Iran," *Renewable and Sustainable Energy Reviews,* vol. 17, pp. 312-319, 2013.

[144] A. Sander, M. Antonije Košćak, D. Kosir, N. Milosavljević, J. Parlov Vuković, and L. Magić, "The influence of animal fat type and purification conditions on biodiesel quality," *Renewable Energy,* vol. 118, pp. 752-760, 2018/04/01/ 2018.

[145] S. Ashraf, M. Hussain, M. W. Mumtaz, and M. Shuaib, "Biodiesel Production by Alkali Catalyzed Transesterification of chicken and beef fats," *International Journal of Alternative Fuels and Energy,* vol. 1, no. 1, pp. 14-20, 2017.

[146] J. Gandure, C. Ketlogetswe, and M. Jonas, "Production, Composition and Fuel Properties of Tallow Biodiesel: A Case of Botswana," *Energy and Power Engineering,* vol. 9, no. 07, p. 355, 2017.

[147] J. Encinar, N. Sánchez, G. Martínez, and L. García, "Study of biodiesel production from animal fats with high free fatty acid content," *Bioresource Technology*, vol. 102, no. 23, pp. 10907-10914, 2011.

[148] M. S. A. Moraes *et al.*, "Tallow biodiesel: properties evaluation and consumption tests in a diesel engine," *Energy & Fuels*, vol. 22, no. 3, pp. 1949-1954, 2008.

[149] R. Alcantara, J. Amores, L. Canoira, E. Fidalgo, M. J. Franco, and A. Navarro, "Catalytic production of biodiesel from soy-bean oil, used frying oil and tallow," *Biomass and Bioenergy*, vol. 18, no. 6, pp. 515-527, 2000/06/01/ 2000.

[150] L. S. G. Teixeira *et al.*, "Characterization of beef tallow biodiesel and their mixtures with soybean biodiesel and mineral diesel fuel," *Biomass and Bioenergy*, vol. 34, no. 4, pp. 438-441, 2010/04/01/ 2010.

[151] D. J. P. Selvam and K. Vadivel, "Performance and Emission Analysis of DI Diesel Engine Fuelled with Methyl Esters of Beef Tallow and Diesel Blends," *Procedia Engineering*, vol. 38, pp. 342-358, 2012/01/01/ 2012.

[152] V. T. Wyatt, M. A. Hess, R. O. Dunn, T. A. Foglia, M. J. Haas, and W. N. Marmer, "Fuel properties and nitrogen oxide emission levels of biodiesel produced from animal fats," *Journal of the American Oil Chemists' Society*, vol. 82, no. 8, pp. 585-591, 2005.

[153] U. Rajak and T. N. Verma, "Effect of emission from ethylic biodiesel of edible and non-edible vegetable oil, animal fats, waste oil and alcohol in CI engine," *Energy Conversion and Management*, vol. 166, pp. 704-718, 2018.

[154] G. R. Srinivasan, V. Shankar, and R. Jambulingam, "Experimental study on influence of dominant fatty acid esters in engine characteristics of waste beef tallow biodiesel," *Energy Exploration & Exploitation*, p. 0144598718821791, 2019.

[155] M. Tabatabaei and M. Aghbashlo, *Biodiesel: From Production to Combustion*. Springer, 2018.

[156] T. M. Mata, N. Cardoso, M. Ornelas, S. Neves, and N. S. Caetano, "Evaluation of two purification methods of biodiesel from beef tallow, pork lard, and chicken fat," *Energy & Fuels*, vol. 25, no. 10, pp. 4756-4762, 2011.

CHAPTER 3: CURRENT TRENDS IN ENZYMATIC TRANSESTERIFICATION OF WASTE ANIMAL FATS

This chapter outline on the application of enzymes as catalysts in animal fats transesterification. Animal fats are known to have high level of acid value that reacts with alkaline and basic catalyst leading to saponification and ultimately biodiesel yield reduction. The use of immobilization techniques and controlled parameters during production process is explained. Lastly the economic aspect of enzymes as catalysts is elaborated using available commercial scale application industries.

Cite this Article: Kosgei, C. and Inambao F. L. Current trends in enzymatic transesterification of waste animal fats *International Journal of Mechanical Engineering and Technology, 10(12), 2019, pp. 541-558.*

(This chapter is part of work that was published in *International Journal of Mechanical Engineering and Technology).*

Link to the article:

http://www.iaeme.com/ijmet/issues.asp?JType=IJMET&VType=10&IType=12

International Journal of Mechanical Engineering and Technology (IJMET)
Volume 10, Issue 12, December 2019, pp. 541-558, Article ID: IJMET_10_12_051
Available online at http://www.iaeme.com/ijmet/issues.asp?JType=IJMET&VType=10&IType=12
ISSN Print: 0976-6340 and ISSN Online: 0976-6359

CURRENT TRENDS IN ENZYMATIC TRANSESTERIFICATION OF WASTE ANIMAL FATS

Cheruiyot Kosgei* and Prof. Freddie L. Inambao
Department of Mechanical Engineering
University of Kwazulu-Natal, Durban, South Africa
https://orcid.org/0000-0001-9922-5434
*Corresponding Author Email: inambaof@ukzn.ac.za

ABSTRACT

Biodiesel is an environmentally friendly fuel produced mostly by the transesterification method. Chemical catalysis when used on low quality feedstock with a high acid value leads to soap formation and reduction in biodiesel yield. This has led to lipases gaining popularity for industrial applications. This paper discusses various lipases used in waste animal fats biodiesel synthesis as an emerging feedstock with no competition with food crops. Various immobilization techniques are described in detail as well as the advantages of whole cell catalysts compared to extracellular lipases. Lipases are affected by a number of factors such as solvents, water content, oil to fat molar ratio and temperature. Lastly, the economic aspect of using lipases is discussed in relation to its technical feasibility on an industrial scale.

Keywords: Enzymes, Lipases, Animal Fats, Transesterification.

Cite this Article: Cheruiyot Kosgei and Freddie L. Inambao, Current Trends in Enzymatic Transesterification of Waste Animal Fats. *International Journal of Mechanical Engineering and Technology* 10(12), 2020, pp. 541-558.
http://www.iaeme.com/IJMET/issues.asp?JType=IJMET&VType=10&IType=12

1. INTRODUCTION

Fossil fuels are the backbone of the transport sector globally for powering engines. The increase in the number of vehicles on the roads has led to more fuel consumption and depletion of finite fossil fuel. According to [1], transport sector energy consumption increases at a rate of 1.4 % per year with a slight drop in projected diesel use from 96 % in 2012 to 88 % in 2040. Moreover, countries relying on imported crude oil products have experienced soaring prices. Combustion of fossil fuels leads to emission of nitrogen and carbon oxides and other toxic gases into the environment. Engine emissions contribute to global warming as a result of the buildup of gases in the lower layer of atmosphere causing temperature rise, climate change and disruption of normal human activities. These challenges have led to a search for alternative fuel sources for use in engines. Biodiesel, bioethanol, hydrogen and methane are some of the acceptable clean fuels.

Biodiesel consists of long chain esters of fatty acids derived from natural renewable feed-stocks (animal fats and vegetable oils). Biodiesel merits when compared to diesel are many: it is renewable, biodegradable, sulphur free, environmentally friendly and has properties close to petroleum diesel fuel. Other advantages over petroleum diesel are low carbon and nitrogen oxides [2], high cetane number, low smoke emissions and absence of aromatics.

Feedstock for biodiesel accounts for 75 % of the overall cost of production. Feedstock should possess good triglyceride content, be easily available, be low cost and not cause 'food vs fuel' conflict. Vegetable oils like palm, soybean and sunflower are raw materials with high unsaturated fatty acids commonly used in some countries which have a surplus of those products. However, the high cost of these raw materials increases the overall cost of production, increases the pump price of biodiesel fuel and causes competition with food. This has necessitated a search for low cost feedstock such as waste animal fats (WAFs). WAFs such as lard, tallow, chicken oil, and fish waste are easily available and help solve the environmental problems of landfill dumping. Compared to edible oils, WAFs have not received much attention for commercial biodiesel production. WAFs have high saturated fatty acid content, acid value and are more viscous hence remain as solid at room temperature [3].

WAFs when used directly on engines cause negative effects such as fuel injector blockage, formation of carbon lumps, distortion of oil rings and reduced spray atomization due to high fuel mass transfer. High mass transfer is caused by high viscosity of unmodified base fuel. WAFs can be converted to biodiesel to reduce viscosity by the transesterification method. Transesterification involves the reaction of alcohol and fats accelerated by either acidic, enzymatic or alkaline catalysis. Alkali catalysts and short chain alcohols yield high productivity within a short time. However, use of alkali catalysts with high acid value in WAFs results in poor biodiesel yield and formation of soaps instead of esters. Acid catalysts can tolerate high acid values in WAFs but have limitations regarding slower reaction rates. Both acid and alkali catalysts use in transesterification leads to problems during the separation and purification stage of glycerol and biodiesel.

Biocatalysis has recently gained popularity compared to inorganic catalysts in transesterification. Enzymatic catalysts have low sensitivity to feedstock quality, simple production processes, lower energy consumption, absence of soap formation, easy separation and reuse and produce high purity and quality of biodiesel [4][5][6]. Enzyme catalysts have some drawbacks related to the high cost of enzymes due to new technologies for preparation, slower reaction rates than KOH/NaOH, and alcohols (especially methanol) inhibit enzymes. Lipases are more widely used than other enzymes for most reactions. They can be used in the presence of solvents [7], supercritical conditions or solvent free systems. The use of solvents improves diffusion and improves reaction rates [8].

This paper reviews biodiesel production methods using WAFs with a focus on enzymatic transesterification. A comprehensive analysis has been conducted on enzymatic technologies, factors affecting enzymes in WAF biodiesel production, and the economics of utilizing enzymes as catalyst for low quality feedstock WAFs.

2. WASTE ANIMAL FATS FEED-STOCKS

Feedstock for biodiesel production is generally categorized into four groups, namely, vegetable oils (edible and nonedible), waste cooking oil, microalgae and waste animal fats (WAFs). The commonly used WAFs are lard, chicken oil, waste fish oil, beef, mutton tallow and waste tanning fats. The WAFs are basically low quality feedstocks with free fatty acids of more than 0.5 % which causes saponification and requires additional pretreatment costs in chemical catalysis. The use of enzymes require low reaction temperature and less consumption of methanol to oil ratio [9]. Most animal fats contain a high percentage of

saturated fatty acids such as palmitic, linoleic, linolenic and stearic acids [10-12]. These acids make animal fats remain in a semi-solid or totally solid state at room temperature.

2.1. Why Waste Animal Fats?

Edible vegetable oils such as palm when used for biodiesel production directly influence prices of food in the market. Analysts project that global food prices could be disrupted as a result of fuel generation [13]. The retail price of biodiesel has a direct correlation with raw materials and the demand for crude petroleum oil. Companies depending on edible oils experience a drop in profit margins when demand for crude petroleum oil increases because then less biodiesel is bought [13]. Dermibas et al. [14] reported that the use of edible oils causes starvation in some developing countries and raises major nutritional and ethical concerns.

WAFs comprise meat and bone meal classified into categories 1 and 2 (high risk) according to European Union and are therefore not fit for consumption. These wastes end up being disposed of in landfills and as co-combustion in cement kilns [15]. WAFs have been known to increase profitability when converted to fuel in slaughterhouses by various technologies [16]. The Food and Drug Administration in the USA has set out rules prohibiting the use of cattle derived wastes (fat) for any human or animal consumption because it is considered high risk for the spread of bovine spongiform encelophalophaty, but it is suitable for biodiesel, as reported by Fidel et al. [16]. Leather industry waste can be used directly to power compression ignition engines according to a study conducted by Gheorghe et al. [17]. The authors preheated fats from a tanning factory and blended this product with diesel which was applied in a diesel engine for a heat recovery system.

The main constituents of fish waste are triglycerides and has been used as feedstock for biodiesel production [18]. Fish waste fat has mainly (58 %) oleic acid C18:1 [18]. Lard and beef have oleic and palmitic as dominant acids [19]. Triglycerides and fatty acids in fat materials collected in rendering facilities are therefore a favorable feedstock for synthesis of biodiesel [20]. The economic analysis which was performed by Kara et al. [12] showed that the cost of production of biodiesel from waste fish oil is 0.69 US $/l biodiesel compared to soybean biodiesel and diesel fuel at cost of a 0.527 US $/l and 0.91 respectively, therefore waste fish oil as a feedstock makes economic sense.

2.2. Feedstock Pre-Treatment/Esterification

A number of pretreatment steps are applied on waste fats before mixing with other reagents in the reactor. These physical steps include sieving to separate melted fats from solids, degumming and dewatering, as well as reduction of acid values. Cunha et al. [21] reduced FFA of mixed swine and chicken fat through washing with a carbonate aqueous solution. FFA content was reduced from 1.77 % to 6.40 % to less than 0.1 % to avoid unfavorable reaction conditions such as soap formation. The tannery waste fat was pretreated by an alkali tetramethylammonium hydroxide which reduced free fatty acid from 30.4 mgKOH/g and the resulting feedstock was used for transesterification directly [22]. The authors reported that the final biodiesel had qualities comparable to biodiesel of conventional feed-stocks such as vegetable oils and met most of the EN 14214 required standards.

2.3. Characterization of Animal Fats

Fatty acid of waste animal fats (shown in Table 1) is an important property obtained from gas chromatography (GC) because this determines the oxidation properties of biodiesel. Saturated acids such as palmitic and stearic are fatty acids without double bonds. They do not have double or triple bonds since all carbon atoms have hydrogen atoms attached. Unsaturated fatty

acids such as oleic or linoleic have one or more carbon-carbon double bonds [18]. Fatty acids weight composition of base feedstock has been used in a number of studies to formulate a correlation with cetane number, density, kinematic viscosity and heating values [23, 24]. The above mentioned properties are crucial for biodiesel performance and emission levels.

Table 1 Fatty acid profiles of various waste animal fats

Feedstock	Capric C10:0	Lauric C12:0	Myristic C14:0	Palmitic C16:0	Palmitoleic C16:1	Stearic C18:0	Oleic C18:1	Linoleic C18:2	References
Tunisian fish fat	-	-	0.27	2.39	0.19	40.35	4.75	1.77	[18]
Mixed chicken and lard waste	0.05	0.06	0.98	20.19	2.82	7.52	39.42	21.08	[21]
chicken	-	-	-	21.0	7.7	5.5	48.5	17.3	[28]
Beef tallow	-	-	1	28	-	26	44	1	[29]
Abdominal chicken fat	-	-	-	23.68	5.50	4.98	40.40	25.44	[30]
Lard	-	0.068	1.137	19.19	2.04	11.81	44.6	10.87	[31]
chicken	-	0.034	0.5	22.055	6.181	5.050	40.314	16.515	[31]
Waste fish oil	-	1.19	10.19	14.79	21.76	24.93	11.58	9.31	[32]
Beef tallow	-	2.72	25.3	2.02	34.7	29.87	0.75	-	[33]
Beef tallow	0.085	3.116	31.376	1.815	25.236	31.091	1.434	0.233	[31]
Waste beef tallow	-	2.68	26.18	1.9	33.68	30.03	-	-	[34]
Waste lard	-	-	1.24	24.67	3.13	12.61	37.79	16.43	[20]

Table 2 Benefits and limitations of various catalysts

Catalysts	Benefits	Limitations
Homogeneous alkaline	High biodiesel yield. Low-cost, easily available in the market. They are non-corrosive	Forms soap when used for high acid feed-stocks Requires many washing steps to purify biodiesel
Homogeneous acid	Highly effective in mild conditions The reactions takes short time period than heterogeneous catalysts	Low reaction than homogeneous catalyst The acid is very corrosive Waste water generated pollutes environment
Heterogeneous base	The catalyst can be reused. Less water is used for purification of biodiesel.	Leaching may occur in active site It is expensive to synthesize catalyst High affinity to water and oxidation during storage
Heterogeneous acid	The catalyst is separated easily from products It can be reused for more reactions	It is costly to prepare the catalyst. The reaction conditions are normally severe. Deactivation rate of catalyst is high
Enzyme	Lipases are separated easily from products The quality of final products-glycerol and biodiesel is high	The production costs of lipase is high Enzymes are sensitive to methanol, which may inhibit its activity.

The cetane number of biodiesel should be above 47 according to ASTM D 6751 standards, with saturated fats such as animal fats giving a value of 60 [25]. A low cetane number gives a long ignition delay, that is time between fuel injection and combustion. This is more obvious in cold conditions. As the percentage of unsaturated acids like linoleic and linolenic increases in feedstock, the number of double bonds increases which lowers the cetane number according to Peterson et al. [26]. Unlike cetane number, density increases with an increase in number of double bonds of base feedstock oil hence more fuel mass is injected into the combustion chamber. The high unsaturated oils like palm oil and soy bean oil have higher density than animal fats [27]. Viscosity of a fluid measures its resistance to flow when tensile or shear stress is applied. When WAF biodiesel is used in an engine, its high viscosity

(higher than for diesel) affects atomization thus reducing the ignition period. High viscosity is seen as beneficial to injector pumps since high kinematic viscosity reduces fuel leakage losses and ultimately increases mass of fuel injected and injection pressure [23]. The kinematic viscosity of biodiesel is limited to 1.9 mm^2/s minimum and 6.0 mm^2/s maximum according to EN 14214.

3. TRANSESTERIFICATION METHODS

WAFs can be converted to biodiesel to reduce their viscosity through pyrolysis [35, 36], emulsification [17], blending [37] and transesterification [38, 39]. Transesterification is used for commercial production more often than the other methods. Trans/inter/esterification are usually interchangeable terms describing conversion of triglycerides to alkyl esters. Transesterification or alcoholysis is the exchange of the alkoxy group of an ester by an alcohol (acyl acceptor) which results in conversion of triglycerides in fats to methyl/ethyl esters and glycerol (Cavonius et al. [40]). On the other hand, interesterification transforms triglyceride in fats to triacylglycerol and methyl acetate ester by-products, while direct esterification of FFA can be accomplished using alcohol resulting in water and fatty acid alkyl esters. The type of catalyst is a major parameter that influences biodiesel yield from WAFs. Catalysts can be chemical in nature like basic/alkaline, acidic or biocatalysts lipase/enzymes. Depending on the catalyst, a transesterification reaction is categorized as homogeneous, heterogeneous or enzymatic.

The homogeneous alkaline process is a one step or two step process utilizing NaOH or KOH as a catalyst. Low quality feedstock containing free fatty acids negatively affect alkaline catalyzed transesterification. Free fatty acids neutralize base catalysts forming soaps instead of biodiesel. Deactivation of enzymes by FFA reduces the yield of biodiesel. Soaps formed in the final mixture makes separation of glycerol and esters difficult and contributes to emulsion formation during water wash [21]. Ghazavi et al. [41] studied the feasibility of refined beef tallow for biodiesel using KOH as the catalyst and methanol as the acyl acceptor. The authors concluded that low cost beef tallow is a good source of biodiesel from methanol and KOH without pretreatment, but the economic aspects of such processes can be improved by recycling glycerol and methanol. Fadhil [42] performed experiments to compare single and two step NaOH and KOH catalyzed transesterification of beef fat. The use of KOH gave better results than NaOH and the two-step process produced more biodiesel yield (94 %) than the single-step process (91 %). The solubility of KOH in methanol is higher than NaOH so gave better results. Ana Lucia et al. [43] reported higher yields for heterogeneous basic catalysts than homogeneous alkaline catalysts for acidic feed-stocks. Emphasis was laid on composite structures like grafting amine inside porous silica and strong sulfonic acids on mesoporous materials. To avoid soap formation and increase yield, acid catalyst such as sulphuric acid, hydrochloric acids and sulfated titania [44] are used in non-edible oils and animal fats with a high acid content.

Homogeneous acid catalysts combine esterification and transesterification simultaneously, but the process is affected by slow reaction rates, elevated temperatures and pressure, and more fats to alcohol molar ratios which means it is not economical. Moreover, residual acids in biodiesel corrodes engine pipes, and many washing steps are required to improve biodiesel quality which creates an environmental hazard. The commonly used acids are H_2SO_4, HCl, BF_3 and H_3PO_4. To avoid several washing steps and increase the reusability of homogeneous catalysts, heterogeneous catalysts have been developed. In heterogeneous catalysis, catalyst and alcohol are in separate phases. This allows for reuse of the catalyst and a reduction in washing steps of biodiesel and easier separation from glycerol. Heterogeneous catalysts have problems of leaching in active phases, are more expensive than homogeneous catalysts, and

have lower reaction rates than homogeneous ones. Fadhil et al. [45] synthesized biodiesel from bitter almond oil and waste fish oil on calcium oxide impregnated on potassium acetate. Biodiesel yield of 91.22 % for almond and 93.30 % fish oil were obtained respectively, under the same conditions of 9:1 methanol to oil ratio at 60 ℃ and 120 min. The catalyst was reused for 4 cycles with 75 % yield.

In enzymatic catalyzed reactions lipases and microbial enzymes are used as biocatalysts for reactions as shown in Figure 1 [46]. Lipase efficiently converts free fatty acids to form alkyl esters without the pretreatment steps that are required in alkali catalyzed reactions as reported by Marta et al. [47]. Lipases offer a number of other advantages like lower energy consumption as reactions can be performed under mild conditions, easy separation and reuse of the catalyst, and no soap formation in the system. Due to minimal water wash requirements, lipase offers an environmentally friendly substitute for acid and alkaline catalysts and therefore is more economical when used on WAFs feed-stock. However, enzymes have the challenge of low reaction rates, higher costs of enzymes and possible enzyme inhibition. Table 2 gives an overview of the benefits and limitations of homogeneous, heterogeneous and enzyme catalysis.

4. ENZYME AS CATALYST

As mentioned in the previous section, the disadvantages of homogeneous and heterogeneous catalysts are after treatment of waste water, high energy consumption, high temperature reactions, separation and purification of biodiesel and glycerol, and corrosion caused by alcohol. These problems are minimized/eliminated by enzymes (biocatalysts) which are environmentally friendly. The commonly used enzymes for transesterification reaction are lipases. Lipase, also termed triacylglycerol acylhydrolase EC 3.1.1.3 performs hydrolysis of long chain TG into glycerol and alkyl esters. Lipase belongs to the ubiquitous and diverse family produced from bacteria, animals, plants and fungal sources. Animal lipases are obtained from the pancreatic glands of pigs and fore-stomach tissue of calves or lambs. Animal lipase are not suitable for a vegetarian diet. Lipases from pig pancreas have trypsin which exhibits bitter taste amino acids, and viruses [48]. Animal lipases are used in food industries.

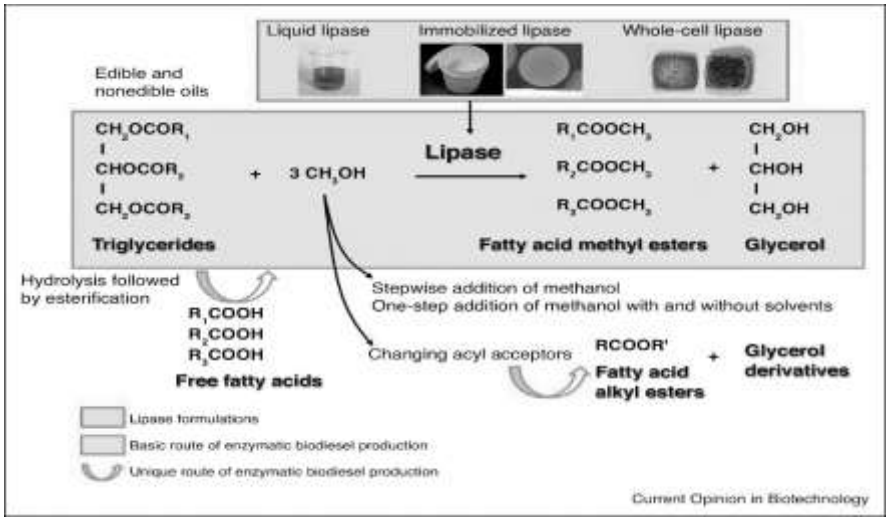


Figure 1 Lipase catalyzed biodiesel transesterification mechanism [46]

Plant lipases are obtained from castor seeds, rapeseed, papaya latex and oats. Lipases from plants can be readily extracted from seeds, bran and latex for synthesis of biodiesel and are less expensive compared to microbial lipases. Rodrigues et al. [49] utilized plant lipase

immobilized *Carica papaya* for synthesis of biodiesel from jatropha where equilibrium reached 64.8 % FAME yield after 4 h reaction time.

Microbial lipases are the most common enzyme sources in commercial applications. The most widely used lipases from microorganisms are from fungi, bacteria and yeast. Microbial lipases are stable, selective and can attack specific parts of the triglyceride molecule making it more common for industrial use compared to other lipases [50]. A large percentage of microbial lipases are cultured in nutrient rich controlled environments. The production of microorganisms depends on the type of strain, cultivation conditions and temperatures. Also, microbial lipases are sensitive to pH, carbon and nitrate sources [51].

4.1. Properties of Microbial Lipase

Microbial lipases are either extracellular or intracellular. Extracellular lipase refers to enzymes that have been extracted from the organism and purified while intracellular lipases are enzymes that are used while they are still in their producing organisms. Extracellular enzymes with a molecular weight of 30 kDa to 50 kDa and a pH range of 7.5-9 are the most common microbial lipases. Some bacterial lipases such as *Pseudomonas gessardii* and *Spirulina platensis* work optimally at an acidic pH [52]. Lipase properties are categorized as mesophilic or thermophilic. Thermophilic lipases such as *Pyrobaculum calidifonti, Pyrococcus furiosus, thermohydrosulfuricus* and *Caldanaerobacter subterraneus* can withstand high heat up to 100 ℃ while mesophilic lipases are stable up to a maximum temperature of 70 ℃. Lipases are stable in their cultured environment and when introduced to high temperatures in a reactor, but the harsh surfaces of reactors inhibit and inactivate enzymes. Short chain alcohols and high alcohol to oil molar ratios have been found to reduce the activity of enzymes. Lipase origin and specific properties determine the mode of action of lipase in terms of specificity and regioselectivity of the glycerol backbone. Lipases catalyse transesterification of fats and oils and alcohol by forming acyl enzymes at an intermediate stage as a donor of acyl moiety to produce alkyl ester and glycerol by-products. Lipases can be 1,3 specific, 2 specific or non-specific depending on the movement of lid during activation. 2 specific enzymes such as *C. rugose* act on the middle and the surface of triglycerides, while 1,3 specific enzymes like *Thermomyces lanuginosus* act primarily on the ester bonds on the extreme positions of the triglyceride molecule and rarely attack the middle ester bond. Non-specific enzymes such as *Candida antarctica*, *Pseudomonas cepacia and Candida cylindracea* show no preference.

4.2. Lipase Immobilization

Immobilization of lipase is the attachment of the enzyme onto a solid support for the substrate to be converted to a product by passing over an enzyme. The aim of immobilization is stability and recycling of expensive enzymes in various conditions [53]. Immobilized enzymes are preferred to free lipases due to elimination of longer reaction time and enantioselectivity [54]. Immobilized lipases exhibit advantages like easy reuse, easy separation of products and enzyme, no contamination of enzyme and product, and stable enzyme due to binding support [55]. Some important enzyme properties are affected positively by immobilization so can operate in harsher environments of pH, temperature and solvents. The main issue for enzyme immobilization is the physical characteristic of support which determines the type of reactor to be used (stirred, fluidized bed or fixed bed) [56]. The cost of lipase is directly proportional to enzymatic production cost and raises the retail price of biodiesel. A choice of low-cost support material that allows sufficient mass transfer is paramount. Other factors that need to be considered for a carrier molecule are mechanical strength, chemical durability, hydrophobic character, loading capacity and microbial strength.

Over 100 immobilization lipase techniques on natural and synthetic supports are available, grouped into four techniques: covalent binding, entrapment/encapsulation, crosslinking and adsorption as shown in Figure 2. Comparison of these techniques are shown in Table 3. The selection of technique to use depends on cost analysis, enzyme activity and desired final output of immobilized enzyme. All techniques can be applied on both intracellular and extracellular lipases.

4.2.1. Adsorption

Adsorption is the simplest and least expensive immobilization method involving weak physical interaction between enzymes, Vanderwaal forces and hydrogen bonds. However, desorption of enzyme from support and sensitivity of enzyme to environmental conditions and ions limits this method. Materials employed as carriers in adsorption range from organic, inorganic, natural and synthetic. Krysztafkiewicz et al. [57] reported that silica is the most common carrier. Silica has a well-developed surface area, is low cost and is more highly available than titanium, zirconium and aluminium and has similar properties to them in terms of thermal, chemical and mechanical strength. Materials from natural origin such as chitin, chitosan and cellulose have recently been used due to biocompatibility and availability. The most frequently used lipases immobilized by adsorption are *C. Antarctica* immobilized on acrylic resin (Novozym 435), Mucormiehei lipase immobilized on ion exchange resin (Lipozyme IM) and Rhizomucor miehei lipase immobilized on macroporous ion exchange (Lipozyme RM IM). Disadvantages are enzymes being stripped off the support, and enzyme loss due to high glycerol levels. Another disadvantage is the low stability of carrier compared to other methods.

4.2.2. Covalent Binding

Covalent binding is used when leaching of enzymes from support is a major concern such as when the absence of enzyme in a product is a strict requirement. Garmroodi et al. [53] grafted octyl/epoxy by GPTMS and OTES support systems to immobilize *Rhizomucor miehei* lipase (RML). The immobilized RML had greater thermal stability and less contamination with higher activity than free lipases. Miao et al. [58] studied immobilized lipase covalently bonded on superparamagnetic Fe_3O_4 effects on biodiesel synthesis for varying temperatures and molar ratios. The immobilized lipase was reused for 5 cycles with the maximum yield of 89 % achieved at 45 ℃, 6:1 molar ratio and 24 h reaction time. The authors concluded that magnetic nanoparticles are good support material for immobilization and the resultant biocatalyst is environmentally friendly for biodiesel production.

Da Rós et al. [59] used covalent binding to immobilize Burkholderia lipase on two different non-commercial supports of hybrid matrix (polysiloxane-polyvinylalcohol SiO_2–PVA), and inorganic matrix (niobiumoxide,Nb_2O_5) for synthesis of beef tallow with ethanol as alcohol. The authors reported that lipase immobilized on hybrid matrix (SiO_2–PVA) produced a yield of 89.70 % while lipase immobilized on inorganic matrix (Nb_2O_5) produced a yield of 40.21 % at the same reaction time for beef tallow.

4.2.3. Cross-Linking

Cross linking involves chemically linking lipase molecules with other molecules using reagents such as glutaraldehyde to form more robust structures. Cross linked immobilized lipase gives pure protein with a high concentration per unit volume, which then is a highly stable and active biocatalyst as reported by Lopez et al. [60]. In cross-linked enzymes, enzymes themselves act as their own carrier instead of fixing enzymes to a carrier. The cross-linked and immobilized enzyme is carrier free which eliminates the disadvantages of using a carrier. Hence, the enzyme has high purity. Alfaro et al. [61] prepared *Candida rugose* lipase

B by cross linking with glutaraldehyde CALB aggregates onto the surface of magnetic nanoparticles carriers. The obtained lipase was applied to synthesize biodiesel from used vegetable oil allowing easy separation and reusability without apparent loss of activity. Other merits of cross-linked lipase are highly concentrated enzymatic activity, cost saving by omitting support material and no enzyme purification requirements.

Table 3 Comparison of various immobilization techniques

	Adsorption	**Covalent binding**	**Entrapment/ Encapsulation**	**Cross-linking**	**Whole cell**
Activity of enzymes	Hydrophobic support increases its activity	Has distorted shape which reduces its activity	Low activity because of strong chemicals and high polymer density	High activity due to large surface area of particles	Has high enzyme activity
Merits	Reactions occur at mild conditions so less damage to enzymes	Has increased activity as a result of more attachment points	Has increased yield from addition of gel beads in industrial setup	Free from carrier hence high purification yield	Fewer procedures followed hence very versatile
Demerits	There is easy desorption and it is favored by proteins that reduces loading efficiency	Support materials used are made from harsh chemical reactions which reduce activity	Enzymes may leach in some cases and harsh environment disrupts enzymes	Reduced activity on high loading of enzymes	Susceptible to methanol poisoning and challenge of aseptic immobilization
Potential for commercial applications	The desorptive properties hinder its reuse efficiency	The method is best for stabilization but distorted shape that reduces enzyme activity hampers on production capacity	Can be used for commercial use but requires further treatment which increases production costs	Is flexible for optimized process hence scaling up production is possible	Due to its high stability, it is attractive for industrial production

4.2.4. Entrapment and Encapsulation

Entrapment of enzymes entails capture of lipase within an inner matrix where the entrapped enzymes are not attached to a polymer but their free diffusion is restrained. Polymeric networks allow substrates and products to pass through and retain enzymes. Entrapment methods of gel and microencapsulation are limited by mass transfer so are effective only for low molecular weight substrates [62]. Entrapment immobilization is simpler than covalent binding and maintains activity of the enzymes. Biodiesel properties as a result of entrapment immobilization needs improvement. I-Ching et al. [63] reported the potential of *P. cepacia* lipase entrapped on biomimetic silica as a catalyst applied for soya bean and waste cooking oil biodiesel synthesis. The authors reported optimum biodiesel conversion similar to silica based supports, but the biodiesel still requires improvement.

Encapsulation is similar to entrapment except encapsulation confines enzymes onto a porous membrane. Encapsulation allows separation of enzymes by providing a cage which prevents enzyme leakage and improves mass transfer. Sirajunnisa et al. [64] investigated encapsulated enzyme mixed cultures of *B. cepacia* and *B. subtilis* on waste cooking oil and methyl acetate acyl acceptor. The immobilized encapsulated lipase showed stability after 20 repeated cycles which contributes to cost saving in the production process.

Because the enzymes are physically attached to the support materials in the process of entrapment and encapsulation, there is the possibility of leaching of enzymes in some cases.

Also, resistance in internal mass transfer is strong resulting in lower reaction rates. To solve these problems, two or more immobilization techniques can be combined to form a hybrid method which is more effective.

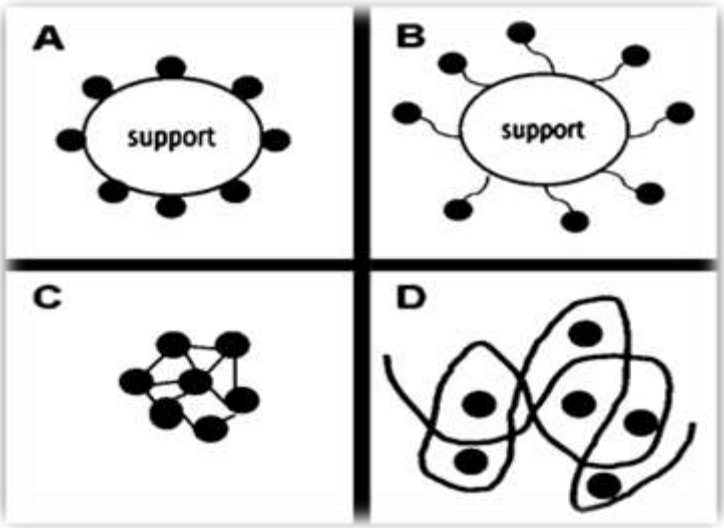


Figure 2 Immobilization; A. adsorption, B. Covalent binding, C. Cross-linking, D. Entrapment

4.3. Whole Cell Immobilization

Over the past few years, whole cell catalysts have attracted attention due to their profit making possibilities. Whole cell immobilization was first developed by Matsumoto et al. [65] on soy bean oil using overproduced *Rhizopus ory-zae* lipase (ROL) in *Saccharomyces cerevisiae* MT8-1. Whole cell immobilization (also known as intracellular lipase) consists of entire microorganisms as biocatalysts and is a less expensive catalyst for biodiesel production. Whole cell immobilization is produced with fewer steps at a low cost from readily available cultures. Phospholipids in oils affect enzymatic transesterification. Phospholipids constitute 30 % of total lipid content of microalgae oil with a significant portion of the C16 and C18 polyunsaturated fatty acids which are required for biodiesel [66]. Jerome et al. [67] produced biodiesel from algal lipid with whole cell *A. oryzae* expressing *Fusarium heterosporum* lipase as biocatalyst. *A. oryzae* whole cell culture was cultured for 6 days at 30 ℃, harvested and reticulated in polyurethane foam then immobilized on DP medium. rFHL showed potential for reuse with activation of active sites being crucial to optimise FAME production.

Whole cell immobilization has disadvantages on immobilization of cell looms and aseptic handling. Another demerit of using whole cell catalysts is the mass limitation from cell membrane transport of the substrate. Research for whole cell immobilization is still in its early stages and hopefully in the near future it will be applied to lower cost for WAF feedstock biodiesel.

4.4. Factors Affecting Enzymatic Transesterification of Animal Fats

There are several factors which affect the rate of enzymatic transesterification and biodiesel yield using WAFs as feedstock. The factors are type of alcohol, solvents, concentration of oil to molar ratio, water content, reaction temperature and lipase pre-treatments. Table 4 gives an overview of the mentioned parameters on waste animal biodiesel synthesis.

4.4.1. Type of Alcohol

Alcohol is a vital property for biodiesel production. Methanol and ethanol are common alcohols used for transesterification with preference for the former in terms of chemical and physical properties as well as low cost. Long chain alcohols (isobutanol, isopropanol and n-butanol) are rarely used while short chain alcohols are used often due to their low cost but these deactivate immobilized lipase. Methanol is the lowest short chain alcohol hence the most deactivating of lipases. Lipases' affinity for long chain alcohols is higher than short chain alcohols. Ethanol, which is a higher chain alcohol than methanol, is a more environmentally friendly and renewable alcohol than deactivating methanol. Methanol is obtained as a by-product of fossil fuel while ethanol is from renewable plant sources. To solve the inhibiting effect of methanol, two strategies have been developed: stepwise addition and the use of solvents [9]. Stepwise addition ensures alcohol is maintained below critical levels to avoid solubility of oil in alcohol and undesirable deactivation of lipase. The use of high volumes of enzymes has been experimented with to compensate for the inhibition, but this is not economical. Some lipases from *pseudomonas* have shown more resistance to inhibition than *Thermomyce lanuginosus.*

4.4.2. Solvents

Solvents added onto substrates increase solubility of alcohol and saves enzymes from being denatured by alcohol. Solvents increase the solubility of glycerol which is advantageous since glycerol can coat enzymes inhibiting its performance. Organic solvents such as hexane, isooctane, n-heptane, tert-butanol, cyclohexane and ionic liquids have been widely used. Pinyanphong et al. [7] reported tert-butanol solvent produced the highest yield compared to other solvents for *Carica papaya* lipase. T-butanol enhanced the solubility of methanol in oil maintaining enzyme activity. Furthemore, t-butanol dissolved glycerol thus eliminated its negative effects. Everton et al. [68] observed that addition of n-hexane in the reaction mixture of porcine fat catalyzed by *Thermomyces lanuginosus* lipase raised the yield of reaction. The effect was observed when n-hexane was raised from 40 ml to 100 ml which raised fat conversion from 30 % to 70 %. Similar results were obtained by Chulalaksananukul et al. [69]. Immobilized lipases show better results than free lipases [69]. *Thermomyces lanuginosus* immobilized on silica gel produced a yield of 88.52 %. Pollardo et al. [70] studied solvent effect on Candida Antarctica lipase B (CALB) utilizing refined animal fats. The authors concluded that organic enzymes are indestructible and pretreatment of enzymes with organic solvents increases yield by 50 %. In the final product mixture solvent must be separated which requires additional processing and some solvents pose health risks since they are hazardous. Moreover, addition of solvents into the reactor increases its volume adding to operational cost.

4.4.3. Alcohol to Oil Molar Ratio

The molar ratio of alcohol to oil in enzymatic transesterification is one of the important variables affecting biodiesel yield and conversion efficiency. The stoichiometric ratio of fat to alcohol has been found to be 1:3 [29] and since the reaction is reversible, more molar ratio is required to improve miscibility and shift the reaction towards formation of products. A higher volume of alcohol to fat gives a higher alkyl ester within a short period of time. Kumar et al. [71] experimented with immobilized lipase catalyst NS88001 (Novozyme) on beef tallow with a fat to alcohol molar ratio of 1:1 to 1:5. Biodiesel yield increased when the oil to alcohol molar ratio was raised from 1:2 to 1:4 followed by a yield decrease of biodiesel when oil to alcohol was increased further from 1:4 to 1:5. No reaction was observed for molar ratios 1:1 and 1:2. Da Silva et al. [30] reported abdominal chicken fat yield of 81 %, 87 % and 69 % at 1:3, 1:4.5 and 1:6 abdominal fat to methanol molar ratio respectively.

Methanol which is a short chain alcohol may cause structural changes to the enzymes causing denaturing [72]. Pinyanphong et al. [7] observed an increase from 49 % to 51 % methyl ester yield when fish oil to fat molar ratio was changed from 1:3 to 1:4. However, reduction occurred when the molar ratio was increased to 1:6 oil to fat molar ratio, indicating enzyme inactivation by methanol which is insoluble at high concentrations.

4.4.4. Reaction Temperature

Reaction temperature depends on alcohol to fat molar ratio, enzyme stability and addition of solvent. Most enzymes have shown stability at temperatures between 30 ℃ to 60 ℃ with some remaining stable as high as 90 ℃. Temperature is a fundamental factor for enzymes due to their proteins which can be denatured at elevated temperatures. The reaction rate is directly proportional to temperature as reported in a number of literature articles [9, 72]. Danielle et al. [73] reported ethanolysis of waste chicken fat with CALB biocatalyst. CALB shows tolerance to the temperature range 30 ℃ to 45 ℃; at 60 ℃ biodiesel efficiency decreased possible due to denaturing or a shift of chemical equilibrium. Kumar et al. [29] reported the effect of reaction time on beef tallow transesterification with Novozyme NS88001 lipase catalyst for temperatures 40 ℃ to 50 ℃. Biodiesel yield increased when temperature was raised after 4 h from 40 ℃ to 45 ℃ after which further temperature increase from 45 ℃ to 50 ℃ decreased biodiesel yield. High temperatures denature specific structures which leads to a decrease in yield. A similar trend was observed on waste cooking oil [74]. Da Silva et al. [30] observed no effect on methyl ester content when temperature was increased from 30 ℃ to 45 ℃ for abdominal chicken fat at 24 h reaction time. The authors emphasized the need to maintain temperature at optimum levels to avoid enzyme denaturing and to save on energy costs. Pinyanphong et al. [7] reported that an increase in temperature from 30 ℃ to 40 ℃ increased fish oil methyl esters and further temperature rises to 60 ℃ lowered methyl ester and raised free fatty acid formation. Higher temperature denatured the enzymes and promoted hydrolysis reactions.

4.4.5. Water Content

Water content affects FAME yield and biodiesel efficiency by minimizing hydrolysis and increasing activity of the lipase biocatalyst. Water affects the structure of an enzyme by maintaining it in a three dimensional form. Antczak et al. [75] found that water content in the reactants is expressed as water concentration for enzyme-water bound molecules. Removal of water disturbs optimal conditions of protein conformation within the enzyme molecular domain. Excess water is attached within enzyme active sites. Absence of water causes no reaction as formation of lipase-fat complexes occurs in the presence of oil-water interface. Addition of water to Ns 40116 (*Thermomycesuss lanugino)* in the course of abdominal chicken fat reaction increased the yield of methyl esters to 82 % while removal of water attained only a 16 % yield as reported by da Silva et al. [30]. The presence of water shielded enzymes from the action of enzymes and kept lipase in its active form. Authors concluded that lipase can be used on low quality feedstock without necessarily undergoing pretreatment.

The percentage of water in the mixture is usually optimized for different types of biocatalysts and characteristics in the reaction medium as well as water content in fats. Pinyanphong et al. [7] investigated addition of water to *Carica papaya* enzymes in a pre-equilibrated saturated salt solution from 0.11-0.53 water activity. Methyl ester yield increased up to 0.23 water activity as minimal water favors active enzyme activity. Higher quantity of water above 0.23 decreased methyl esters as a result of enzyme aggregation. Arumugam and Ponnusami [9] studied water content of 5 % to 15 % for weight of sardine oil and water content of 10 % (v/v), which yielded 92.5 % methyl ester. Addition of water has been shown to increase the interfacial area for lipase action with sardine oil.

5. ECONOMIC BENEFITS OF LIPASE TRANSESTERIFICATION OF WASTE ANIMAL FATS

The results presented in most experiments demonstrated that slaughterhouses can generate extra profit from biodiesel production as the entire project is technically feasible. This represents an important waste recycling strategy for handling WAF. Enzymatic catalysis produces high biodiesel yield, similar to chemical transesterification methods, with more advantages from an environmental point of view, since the process can be accomplished at lower pressures and low temperatures and produce good quality biodiesel. Moreover, when immobilized lipase is successfully used in the process the lipase cost will be low, since enzymes are reused several times without yield loss [68]. Fjerbaek et al. [4] showed that for an acid/alkaline catalyst, productivity is more important than robustness of the production technology. The authors estimated a price of 1 kg of Novozyme 435 is 1000 US $ while 1 kg NaOH is 0.62 US $ but when productivity is factored in, the enzyme price gives 0.14 US $ while NaOH is 0.006 US $ for 1 kg. Hence, enzymes use is more economically feasible than chemical catalysis.

Table 4 Summary of WAFs biodiesel synthesis utilizing enzyme as catalysts

Enzyme	Enzyme loading weight %	Feedstock/substrate	alcohol	Alcohol:oil loading wt%	Temperature (℃)	Reaction time (h)	Additional information	Yield, optimal conditions	Ref.
C. Antarctica lipase B-	4-8	Waste beef tallow	methanol	1:4-1:8	35	8	Ultrasonic assisted,30-50% amplitude	85.6 %, 1:4, 6 % enzyme, 20 min reaction time at 40 % amplitude	
Novozyme Ns 88001		Beef tallow	methanol	1:1- 1:5	45-50	4-16	No solvent was used	94.04 %, 1:4 molar ratio at 45 ℃ and 16 h reaction time	[29]
Thermomyces lanuginosus		Abdominal chicken fat	methanol	1:3-1:6	30-45	24	Solvent free, water was added	0.3 wt% enzyme, 2 wt% water and 3 h reaction time, 250 rpm, 1:4.5 molar ratio	[30]
Commercial lipases	12.5- 50	Residual fish oil	ethanol	1:0.25-1:4	35	1-8	Response surface method was used for optimal yield	76.35 % enzyme loading, 8 h reaction time and 35 ℃	[78]
Carica papaya	5-25	Fish oil	methanol	1:3-1:6	30-60	6-24	Solvent was used for comparison	51 % yield,1:4 oil methanol molar ratio, 40 ℃ temperature	[7]
C. Antarctica lipase B	5	Refined animal fats	methanol	6:50 g/g wt	40	6	Various solvents were tested		
Burkholderia lipase	20	Beef tallow	ethanol	1:12	50	48	Immobilized lipase	89.70 % yield at 50 ℃, 1:12 molar ratio and 48 h time.	[59]
C. Antarctica	12.5-25	Tallow	methanol	1:3	45	4-8	Addition of hexane solvent	95 % yield, 1:3 molar ratio, 5 h reaction time and 200 rpm	[79]
Aspergillus niger lipase	-	Waste sardine oil	methanol	1:6 – 1:12	30-50	4- 72	Water added on mixture	94.55 yield, 9:1 molar ratio, 10 % water content at 30 ℃	[9]
Commercial lipases	-	Lard	methanol	-	50	-	Tert-butanol reaction medium	90.5 % yield	[80]
Lipozyme TL IM	1.25-2.5	Porcine fat	methanol	1:3	35	3-8	n-hexane solvent	1:3 molar ratio, 55 ℃ temperature, 3 h reaction time	[68]

Sotoft et al. [76] conducted economic analysis of two different biodiesel plants using rapeseed feedstock, the first plant using Novozym 435 and Lipozyme TL IM enzymes and

tert-butanol solvent and the second plant Novozym 435 enzyme but solvent free. The solvent-free plant was found to be economically viable, having a payback time of 3 months and a biodiesel production cost of 73 euros per 100 kg. The plant using solvent used high energy to recover the solvent making it uneconomical. The production costs in the two plants studied were contributed to by the price of raw materials and glycerol.

Recently, large scale pilot plants for biodiesel production using enzymes have been built. A company in Israel, Transbiodiesel, invented and developed a new enzyme (TranZyme A) to convert low grade feedstock to biodiesel. The company provides efficient solutions for biodiesel producers using WFs and waste vegetable oils at a low cost. In 2012, Piedmont Biofuels in North Carolina stepped up its production to continuous enzyme catalyst usage from the usual chemical process. A Chinese company, Lvming Co. Ltd started production in 2007 in China using immobilized *Candida Sp.* Lipase as a catalyst with a capacity of 10 000 tons [77].

6. CONCLUSION

The enzymatic transesterification process has more benefits than chemically catalyzed methods. Enzymes are environmentally friendly, there is no pretreatment required for feedstock, and the reaction conditions are mild. Since there is no soap formed in the reaction, any kind of feedstock having enough triglyceride content can be used. Moreover, separation of phases formed is easier permitting quality glycerol and biodiesel. The numerous steps required in chemical reactions are not necessary in lipases which reduces the overall production cost of biodiesel. When the same economic issue of enzymes is applied to low cost WAF, this generates more profit and becomes feasible for long term projects.

The cost of enzymes, sensitivity to methanol, and slow reaction conditions, are the main limiting factors in the enzymatic process. This reduces the number of plants currently in production. Thus, enzymes have to be reused, in most cases when immobilized onto a support. The immobilization methods (adsorption, covalent binding, entrapment/encapsulation and crosslinking) discussed shows that adsorption is most commonly used due to its simplicity in fabrication and low damage of enzymes. It was also noted that enzymes are affected by temperature, water content, solvent and alcohol to oil molar ratio. Amongst these factors, water content has a positive effect on the performance of enzymes as enzymes favor an aqueous medium for action.

REFERENCES

[1] Conti, J., et al., *International Energy Outlook 2016 with Projections to 2040*. 2016, USDOE Energy Information Administration (EIA), Washington, DC, USA

[2] Joy, N., et al. Exhaust Emission Study on Neat Biodiesel and Alcohol Blends Fueled Diesel Engine. *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects*, 40(1), 2018, pp. 115-119. https://doi.org/10.1080/15567036.2017.1405119

[3] Kumar, M.S., et al. Ethanol Animal Fat Emulsions as a Diesel Engine Fuel–Part 2: Engine Test Analysis. *Fuel*, 2006. 85(17-18): p. 2646-2652. https://doi.org/10.1016/j.fuel.2006.05.023

[4] Fjerbaek, L., Christensen, K.V. and Norddahl, B. A Review of the Current State of Biodiesel Production Using Enzymatic Transesterification. *Biotechnology and Bioengineering*, 2009. 102(5): p. 1298-1315. https://doi.org/10.1002/bit.22256.

[5] Guieysse, D., et al. Lipase-Catalyzed Enantioselective Transesterification Toward Esters of 2-Bromo-Tolylacetic Acids. *Tetrahedron: Asymmetry*, 2003. 14(3): p. 317-323. https://doi.org/10.1016/S0957-4166(02)00784-X

[6] Sharma, S. and Kanwar, S.S. Organic Solvent Tolerant Lipases and Applications. *The Scientific World Journal*, 2014. 2014. http://dx.doi.org/10.1155/2014/625258

[7] Pinyaphong, P., Sriburi, P. and Phutrakul, S. Biodiesel Fuel Production by Methanolysis of Fish Oil Derived from the Discarded Parts of Fish Catalyzed by Carica Papaya Lipase. *World Academy of Science, Engineering and Technology*, 2011. 76: p. 466-472.

[8] Tacias-Pascacio, V.G., et al. Comparison of Acid, Basic and Enzymatic Catalysis on the Production of Biodiesel after RSM Optimization. *Renewable Energy*, 2019. 135: p. 1-9. https://doi.org/10.1016/j.renene.2018.11.107

[9] Arumugam, A. and Ponnusami, V. Production of Biodiesel by Enzymatic Transesterification of Waste Sardine Oil and Evaluation of its Engine Performance. *Heliyon*, 2017. 3(12): p. e00486. https://doi.org/10.1016/j.heliyon.2017.e00486

[10] Costa, J.F., et al. Biodiesel Production Using Oil from Fish Canning Industry Wastes. *Energy Conversion and Management*, 2013. 74: p. 17-23. https://doi.org/10.1016/j.enconman.2013.04.032

[11] de Almeida, V.F., et al. Biodiesel Production from Mixtures of Waste Fish Oil, Palm Oil and Waste Frying Oil: Optimization of Fuel Properties. *Fuel Processing Technology*, 2015. 133: p. 152-160. https://doi.org/10.1016/j.fuproc.2015.01.041

[12] Kara, K., et al. Biodiesel Production from Waste Fish Oil with High Free Fatty Acid Content from Moroccan Fish-Processing Industries. *Egyptian Journal of Petroleum*, 2018. 27(2): p. 249-255.

[13] Johari, A., et al. The challenges and Prospects of Palm Oil Based Biodiesel in Malaysia. *Energy*, 2015. 81: p. 255-261.

[14] Demirbas, A., et al. Biodiesel Production from Non-Edible Plant Oils. *Energy Exploration & Exploitation*, 2016. 34(2): p. 290-318.

[15] Leon, M., et al. Thermochemical Conversion of Animal By-Products and Rendering Products. *Waste Management*, 2018. 73: p. 447-463.

[16] Toldrá, F., Mora, L. and Reig, M. New insights Into Meat By-Product Utilization. *Meat Science*, 2016. 120: p. 54-59.

[17] Lazaroiu, G., et al. Solutions for Energy Recovery Of Animal Waste From Leather Industry. *Energy Conversion and Management*, 2017. 149: p. 1085-1095.

[18] Kraiem, T., et al. Characterization of the Liquid Products Obtained From Tunisian Waste Fish Fats using the Pyrolysis Process. *Fuel Processing Technology*, 2015. 138: p. 404-412.

[19] Hong, I.K., Park, J.W. and Lee, S.B. Optimization of Fish-Oil-Based Biodiesel Synthesis. *Journal of Industrial and Engineering Chemistry*, 2013. 19(3): p. 764-768.

[20] Baladincz, P. and Hancsók, J. Fuel from Waste Animal Fats. *Chemical Engineering Journal*, 2015. 282: p. 152-160.

[21] Cunha, A., et al. Synthesis and Characterization of Ethylic Biodiesel from Animal Fat Wastes. *Fuel*, 2013. 105: p. 228-234.

[22] Šánek, L., et al. Biodiesel production from Tannery Fleshings: Feedstock Pretreatment and Process Modeling. *Fuel*, 2015. 148: p. 16-24.

[23] Giakoumis, E.G. and Sarakatsanis, C.K. Estimation of biodiesel Cetane Number, Density, Kinematic Viscosity and Heating Values from Its Fatty Acid Weight Composition. *Fuel*, 2018. 222: p. 574-585.

[24] Gülüm, M. and Bilgin, A. Two-term Power Models for Estimating Kinematic Viscosities of Different Biodiesel-Diesel Fuel Blends. *Fuel Processing Technology*, 2016. 149: p. 121-130.

[25] Giakoumis, E.G., A statistical Investigation of Biodiesel Physical and Chemical Properties, and Their Correlation with the Degree of Unsaturation. *Renewable Energy*, 2013. 50: p. 858-878.

[26] Peterson, C., et al. The Effect of Biodiesel Feedstock on Regulated Emissions in Chassis Dynamometer Tests of a Pickup Truck. *Transactions of the ASAE*, 2000. 43(6): p. 1371.

[27] Caldeira, C., et al. Fatty acid Based Prediction Models for Biodiesel Properties Incorporating Compositional Uncertainty. *Fuel*, 2017. 196: p. 13-20.

[28] Marulanda, V.F., Anitescu, G. and Tavlarides, L.L. Investigations on Supercritical Transesterification of Chicken Fat for Biodiesel Production from Low-Cost Lipid Feedstocks. *The Journal of Supercritical Fluids*, 2010. 54(1): p. 53-60.

[29] Marchetti, J., Miguel, V. and Errazu, A. Techno-Economic Study of Different Alternatives for Biodiesel Production. *Fuel Processing Technology*, 2008. 89(8): p. 740-748.

[30] da Silva, J.R.P., et al. Lipase NS40116 as Catalyst for Enzymatic Transesterification of Abdominal Chicken Fat as Substrate. *Bioresource Technology Reports*, 2018. 4: p. 214-217.

[31] Sander, A., et al. The influence of Animal Fat Type and Purification Conditions on Biodiesel Quality. *Renewable Energy*, 2018. 118: p. 752-760.

[32] Kara, K., et al. Biodiesel production from Waste Fish Oil with High Free Fatty Acid Content from Moroccan Fish-Processing Industries. *Egyptian Journal of Petroleum*, 2018. 27(2): p. 249-255.

[33] da Cunha, M.E., et al. Beef Tallow Biodiesel Produced in a Pilot Scale. *Fuel Processing Technology*, 2009. 90(4): p. 570-575.

[34] Moraes, M.S.A., et al. Tallow biodiesel: Properties Evaluation and Consumption Tests in a Diesel Engine. *Energy & Fuels*, 2008. 22(3): p. 1949-1954.

[35] Wiggers, V., et al. Biofuels from Waste Fish Oil Pyrolysis: Continuous Production in a Pilot Plant. *Fuel*, 2009. 88(11): p. 2135-2141.

[36] Jung, J.-M., et al. Biodiesel Synthesis Using Chicken Manure Biochar and Waste Cooking Oil. *Bioresource Technology*, 2017. 244: p. 810-815.

[37] Nicolici, A., et al. The use of Animal Fats in the Diesel Fuelled Engine. in IOP Conference Series: Materials Science and Engineering. 2018. IOP Publishing.

[38] Tashtoush, G.M., Al-Widyan, M.I. and Al-Jarrah, M.M. Experimental Study on Evaluation and Optimization of Conversion of Waste Animal Fat into Biodiesel. *Energy Conversion and Management*, 2004. 45(17): p. 2697-2711.

[39] Bhatti, H.N., Hanif, M.A. and Qasim, M. Biodiesel Production from Waste Tallow. *Fuel*, 2008. 87(13-14): p. 2961-2966.

[40] Cavonius, L.R., Carlsson, N.-G. and Undeland, I. Quantification of Total Fatty Acids in Microalgae: Comparison of Extraction and Transesterification Methods. *Analytical and Bioanalytical Chemistry*, 2014. 406(28): p. 7313-7322.

[41] Ghazavi, M.A., Fallahipanah, M. and Jeshvaghani, H.S. A Feasibility Study on Beef Tallow Conversion to Some Esters for Biodiesel Production. International Journal Of Recycling of Organic Waste in Agriculture, 2013. 2(1): p. 2.

[42] Fadhil, A.B. Biodiesel Production from Beef Tallow using Alkali-Catalyzed Transesterification. *Arabian Journal for Science and Engineering*, 2013. 38(1): p. 41-47.

[43] de Lima, A.L., Ronconi, C.M. and Mota, C.J. Heterogeneous Basic Catalysts for Biodiesel Production. *Catalysis Science & Technology*, 2016. 6(9): p. 2877-2891.

[44] Lu, H., et al. Production of Biodiesel from Jatropha Curcas L. Oil. *Computers & Chemical Engineering*, 2009. 33(5): p. 1091-1096.

[45] Fadhil, A.B., Al-Tikrity, E.T.B. and Khalaf, A.M. Transesterification of Non-Edible Oils Over Potassium Acetate Impregnated Cao Solid Base Catalyst. *Fuel*, 2018. 234: p. 81-93.

[46] Hama, S., Noda, H. and Kondo, A. How Lipase Technology Contributes to Evolution of Biodiesel Production Using Multiple Feedstocks. *Current Opinion in Biotechnology*, 2018. 50: p. 57-64.

[47] Marín-Suárez, M., et al. Reuse of Immobilized Lipases in the Transesterification of Waste Fish Oil for the Production of Biodiesel. *Renewable Energy*, 2019.

[48] Vakhlu, J. Yeast Lipases: Enzyme Purification, Biochemical Properties and Gene Cloning. *Electronic Journal of Biotechnology*, 2006. 9(1): p. 0-0.

[49] Rodrigues, J., et al. Biodiesel Production From Crude Jatropha Oil Catalyzed by Non-Commercial Immobilized Heterologous Rhizopus Oryzae and Carica Papaya Lipases. *Bioresource Technology*, 2016. 213: p. 88-95.

[50] Dutra, J.C., et al. Lipase Production in Solid-State Fermentation Monitoring Biomass Growth of Aspergillus Niger using Digital Image Processing, in Biotechnology for Fuels and Chemicals. 2007, Springer. p. 431-443.

[51] Souissi, N., et al. Production of Lipase and Biomass By Staphylococcus Simulans Grown on Sardinella (Sardinella Aurita) Hydrolysates and Peptone. A*frican Journal of Biotechnology*, 2009. 8(3).

[52] Demir, B.S. and Tükel, S.S. Purification and Characterization of Lipase from Spirulina Platensis. *Journal of Molecular Catalysis B: Enzymatic*, 2010. 64(3-4): p. 123-128.

[53] Garmroodi, M., et al., Covalent Binding of Hyper-Activated Rhizomucor Miehei Lipase (RML) on Hetero-Functionalized Siliceous Supports. *International Journal of Biological Macromolecules*, 2016. 86: p. 208-215.

[54] Mohamad, N.R., et al. An Overview of Technologies for Immobilization of Enzymes and Surface Analysis Techniques for Immobilized Enzymes. *Biotechnology & Biotechnological Equipment*, 2015. 29(2): p. 205-220.

[55] Anand, A. and Weatherley, L.R. The Performance of Microbial Lipase Immobilized Onto Polyolefin Supports for Hydrolysis of High Oleate Sunflower Oil. Process Biochemistry, 2018. 68: p. 100-107.

[56] Mattiasson, B. *Immobilization Methods*, in *Immobilized Cells and Organelles*. 2018, CRC Press. p. 3-26.

[57] Krysztafkiewicz, A., Binkowski, S. and Jesionowski, T. Adsorption of Dyes on a Silica Surface. *Applied Surface Science*, 2002. 199(1-4): p. 31-39.

[58] Miao, C., et al. Lipase immobilization On Amino-Silane Modified Superparamagnetic Fe3O4 Nanoparticles as Biocatalyst For Biodiesel Production. *Fuel*, 2018. 224: p. 774-782.

[59] Da Rós, P.C., et al. Evaluation of the Catalytic Properties of Burkholderia Cepacia Lipase Immobilized on Non-Commercial Matrices to be used in Biodiesel Synthesis from Different Feedstocks. *Bioresource Technology*, 2010. 101(14): p. 5508-5516.

[60] Lopez-Serrano, P., et al. Cross-Linked Enzyme Aggregates with Enhanced Activity: Application to Lipases. *Biotechnology Letters*, 2002. 24(16): p. 1379-1383.

[61] Cruz-Izquierdo, Á., et al. Magnetic Cross-Linked Enzyme Aggregates (Mcleas) of Candida Antarctica Lipase: an Efficient and Stable Biocatalyst for Biodiesel Synthesis. *PloS One*, 2014. 9(12): p. e115202.

[62] Zhang, B., et al. Enzyme Immobilization for Biodiesel Production. *Applied Microbiology and Biotechnology*, 2012. 93(1): p. 61-70.

[63] Kuan, I.-C., et al. Optimizing the Production of Biodiesel Using Lipase Entrapped in Biomimetic Silica. *Energies*, 2013. 6(4): p. 2052-2064.

[64] Razack, S.A. and S. Duraiarasan, Response Surface Methodology Assisted Biodiesel Production from Waste Cooking Oil using Encapsulated Mixed Enzyme. *Waste Management*, 2016. 47: p. 98-104.

[65] Matsumoto, T., et al. Yeast Whole-Cell Biocatalyst Constructed by Intracellular Overproduction of Rhizopus Oryzae Lipase is Applicable to Biodiesel Fuel Production. *Applied Microbiology and Biotechnology*, 2001. 57(4): p. 515-520.

[66] Amoah, J., et al. Converting oils High in Phospholipids to Biodiesel Using Immobilized Aspergillus Oryzae Whole-Cell Biocatalysts Expressing Fusarium Heterosporum Lipase. *Biochemical Engineering Journal,* 2016. 105: p. 10-15.

[67] Amoah, J., et al. Conversion of Chlamydomonas sp. JSC4 lipids to Biodiesel using Fusarium Heterosporum Lipase-Expressing Aspergillus Oryzae Whole-Cell as Biocatalyst. *Algal Research,* 2017. 28: p. 16-23.

[68] Skoronski, E., et al. Valorization of Agro-Industrial By-Products: Analysis of Biodiesel production from porcine fat waste. *Journal of Cleaner Production,* 2016. 112: p. 2553-2559.

[69] Chulalaksananukul, W., et al. Kinetic study of Esterification by Immobilized Lipase in n-Hexane. *FEBS Letters,* 1990. 276(1): p. 181-184.

[70] Pollardo, A.A., et al. Solvent Effect on the Enzymatic Production of Biodiesel from Waste Animal Fat. *Journal of Cleaner Production,* 2018. 185: p. 382-388.

[71] Kumar, S., Ghaly, A. and Brooks, M. Production of Biodesiel from Animal Tallow via Enzymatic Transesterification using the Enzyme Catalyst Ns88001 with Methanol in a Solvent-Free System. *Journal of Fundamentals of Renewable Energy and Applications,* 2015. 5(2): p. 1-8.

[72] Lopresto, C.G., et al. Enzymatic Transesterification of Waste Vegetable Oil to Produce Biodiesel. *Ecotoxicology and Environmental Safety*, 2015. 121: p. 229-235.

[73] Antonio, D.C., Amancio, L.P. and Rosset, I.G. Biocatalytic Ethanolysis of Waste Chicken Fat for Biodiesel Production. *Catalysis Letters,* 2018. 148(10): p. 3214-3222.

[74] Desikan, S., et al. Application of Immobilized Lipase Enzyme for the Production of Biodiesel from Waste Cooking Oil. *Asian Journal of Biological Sciences*, 2013. 6: p. 322-330.

[75] Antczak, M.S., et al. Enzymatic biodiesel Synthesis–Key Factors Affecting Efficiency of the Process. *Renewable Energy*, 2009. 34(5): p. 1185-1194.

[76] Sotoft, L.F., et al. Process Simulation and Economical Evaluation of Enzymatic Biodiesel Production Plant. *Bioresource Technology,* 2010. 101(14): p. 5266-5274.

[77] Christopher, L.P., K. Hemanathan, and V.P. Zambare, Enzymatic biodiesel: Challenges and Opportunities. *Applied Energy*, 2014. 119: p. 497-520.

[78] Marín-Suárez, M., et al. Reuse of Immobilized Lipases in the Transesterification of Waste Fish Oil for the Production of Biodiesel. *Renewable Energy,* 2019. 140: p. 1-8.

[79] Nelson, L.A., Foglia, T.A. and Marmer, W.N. Lipase-Catalyzed Production of Biodiesel. *Journal of the American Oil Chemists' Society*, 1996. 73(9): p. 1191-1195.

[80] Huang, Y., Zheng, H. and Yan, Y. Optimization of lipase-Catalyzed Transesterification of Lard for Biodiesel Production using Response Surface Methodology. *Applied Biochemistry and Biotechnology*, 2010. 160(2): p. 504-515.

CHAPTER 4: DEVELOPMENT AND CHARACTERIZATION OF WASTE BEEF TALLOW METHYL ESTERS

This chapter gives experimental production of beef tallow biodiesel by transesterification process. Included are beef tallow biodiesel properties and characterization.

Cite this Article: Kosgei, C. and Inambao F. L. Development and characterization of waste beef tallow methyl esters. *International Journal of Mechanical and Technology, 10(11), 2019, pp. 254-258.*

(This chapter is part of work that was published in *International Journal of Mechanical Engineering and Technology*)

Link to the article:

http://www.iaeme.com/ijmet/issues.asp?JType=IJMET&VType=10&IType=11

International Journal of Mechanical Engineering and Technology (IJMET)
Volume 10, Issue 11, November 2019, pp. 245-258, Article ID: IJMET_10_11_022
Available online at http://www.iaeme.com/ijmet/issues.asp?JType=IJMET&VType=10&IType=11
ISSN Print: 0976-6340 and ISSN Online: 0976-6359

DEVELOPMENT AND CHARACTERIZATION OF WASTE BEEF TALLOW METHYL ESTERS

Cheruiyot Kosgei* and Prof. Freddie L. Inambao
Department of Mechanical Engineering
University of Kwazulu-Natal, Durban, South Africa
https://orcid.org/0000-0001-9922-5434
*Corresponding Author Email: inambaof@ukzn.ac.za

ABSTRACT

The current heatwave, sporadic rainfall and longer drought seasons experienced globally are occurring due to climate change which is largely caused by engine emissions. Exhaust emissions from diesel engines increase CO and SO_2 levels in the atmosphere. There is a need for alternative fuels to diesel for diesel engines. The objective of the study was to utilize low cost raw material (waste beef tallow) as an alternate feedstock for biodiesel production. The methanol to fat molar ratio was varied from 3:1 to 9:1, reaction time from 60 min to 120 min and catalyst concentration of 0.5 w/w%. to 2 w/w%. The beef tallow biodiesel was characterized for chemical properties using gas chromatography-mass spectroscopy (GC-MS), thermogravimetric analyses (TGA) and Fourier transform infrared (FT-IR) equipment. According to TGA, one smooth curve for decomposition was formed indicating complete transesterification of triglycerides and therefor high quality biodiesel. The physical properties of beef tallow i.e. methyl esters density, kinematic viscosity, cloud point, pour point, cold filter plugging point (CFPP), acid value, calorific value, and flash point, were within ASTM standards for biodiesel.

Keywords: Waste beef tallow, methyl esters, esterification, transesterification, properties, characterization

Cite this Article: Cheruiyot Kosgei and Prof. Freddie L. Inambao, Development and Characterization of Waste Beef Tallow Methyl Esters. *International Journal of Mechanical Engineering and Technology* 10(11), 2019, pp. 245-258.

http://www.iaeme.com/IJMET/issues.asp?JType=IJMET&VType=10&IType=11

1. INTRODUCTION

The rising global population has put pressure on natural resources as competition intensifies day in and day out. According to the estimated demographic data from UN, the world population in 2015 was 7.35 billion people [1] with the largest percentage from the developing countries. At the same time, these less developed countries have massive problems meeting their energy targets for running their countries and improving their economies [2]. Studies have confirmed that a successful economy is largely due to energy efficiency [3]. The traditional source of energy is fossil fuel, which contributes significantly to

the rise in global temperatures, climate change, increase in emissions of carbon dioxides, nitrogen oxides, smoke emissions and particulate matter. The transport sector in particular is dominated by diesel engine vehicles while moving heavy tons of goods which emit sulphur oxides that are toxic to the environment and contribute to acid rain that has been reported to cause cancer [4]. This has necessitated a search for an alternative fuel that is clean, environmentally friendly and biodegradable. Amongst fuels for engines that have so far been researched, biodiesel has gained wide acceptability as a substitute for diesel in compression ignition engines. Biodiesel is produced from edible and non-edible oils by various production methods discussed in published work [5]. Biodiesel fuel has the possibility of replacing diesel fuel due to it being renewable and biodegradable [6-8].

However, the pump cost of biodiesel is dependent on the purchase value of the feedstock and the manufacturing cost. This impacts on the overall cost of production and the profitability of the biodiesel plant. It is reported that biodiesel feedstock accounts for 75 % of the total production cost of biodiesel [9]. When compared with petroleum diesel production costs, biodiesel is less profitable for commercialization. Thus, a less costly feedstock could mean that biodiesel would be a more cost effective fuel than diesel [10, 11].

Societies where cattle meat is consumed in large quantities generate a lot of waste and South Africa is no exception. Abattoirs produces bio-residues like hoof, skin, horn and tallow. Beef tallow produced in abattoirs has large fat content which is primarily used in soap making. When the soap industry is fully supplied, tallow ends up in landfills polluting the surrounding environment with foul smells and possible disease outbreaks [12]. Beef tallow can be better utilized for fuel generation to power diesel engines in the form of biodiesel [13]. Beef tallow is a low cost fuel but it is a challenge to process due to the high amount of free fatty acids, but this can be overcome by improvements in the production process using a two stage process of esterification and then transesterification employing optimum reaction conditions. Beef tallow and other animal fats have high cetane numbers that can contribute to better engine performance and reduced emissions in their neat form or blended with diesel [14, 15].

Several production methods for biodiesel production are available such as pyrolysis, emulsion, thermochemical, blending and transesterification [5, 16-18]. The most common method is transesterification of oil/fats with an alcohol and alkali or acid catalyst to produce methyl esters and glycerol as by-products. Methanol is a low chain alcohol and is often used due to high productivity within a short time and it is less costly. Ethanol though derived from renewable sources tends to form azeotropes with water which is expensive to purify during the recovery process. Ethanol produces a low ethyl ester yield and difficult glycerol separation. Most studies report that KOH is preferred as catalyst rather than NaOH since it produces biodiesel with better properties and softer glycerol that permits easier separation from methyl esters [19, 20].

The objective of the present work was to develop optimum conditions of methanol to fat molar ratio, reaction time and catalyst concentration for biodiesel production of waste beef tallow using KOH as a catalyst in methanol. The biodiesel was characterized for chemical properties using various instruments and physical properties according to ASTM methods.

2. MATERIALS AND METHODS

2.1. Materials

The waste beef tallow was collected from Buxons slaughterhouse in Durban, South Africa. The main characteristics of the waste beef tallow were acid value =2.4 mgKOH/g, iodine value = 42.1 cg/g, saponification value = 167.8 mgKOH/g and moisture content = 0.164 % m/m.

Methanol, sulphuric acid (99.96 % purity), potassium hydroxide (86.38 % purity), phosphoric acid and anhydrous sodium sulphate (99.0 % purity) were acquired from Radchem Chemicals, Durban, South Africa and used without further purification.

2.2. Rendering of Waste Beef Tallow

The waste beef tallow collected from the slaughterhouse was heated slowly in an open metallic vessel for 60 minutes at 110 °C to avoid degradation in order to melt the fats (Figure 1). The melted fats (Figure 2) were then filtered using a 5 µm strainer, cooled and kept in a tightly sealed container to prevent oxidation.

Figure 1. Dry rendering of beef tallow in metallic vessel

Figure 2. Melted and filtered beef tallow at 60 °C

2.3. Transesterification

Transesterification of the waste beef tallow was performed in a two-step process since it had an acid value of 2.404 mgKOH/g. A 2-step process is performed when free fatty acid is more than 1 % as reported by Sahar et al. [21]. The first step is esterification catalyzed by an acid catalyst while the second step is catalyzed by a basic catalyst.

2.3.1. Acid Catalyzed Transesterification

The mixture of methanol and 1 % w/w sulphuric acid were prepared separately by stirring for 30 min at 50 °C and 300 rpm with a magnetic stirrer. The molar ratio of methanol to beef tallow used was 6:1. The mixture was added into preheated fats at 60 °C, stirred for 60 min at 1000 rpm using an overhead mechanical stirrer and the temperature was kept below 64 °C. The reactants were then placed in a separating funnel for 60 min to separate by gravity as shown in Figure. 3. The upper layer consisted of unreacted methanol and sulphuric acid which

was discarded while the lower phase consisting of unreacted fats was cleaned with warm distilled water at 60 °C three times until the distilled water was clear. The cleaned fats were subjected to heat at 110 °C for 60 min for the remaining water and methanol to evaporate.

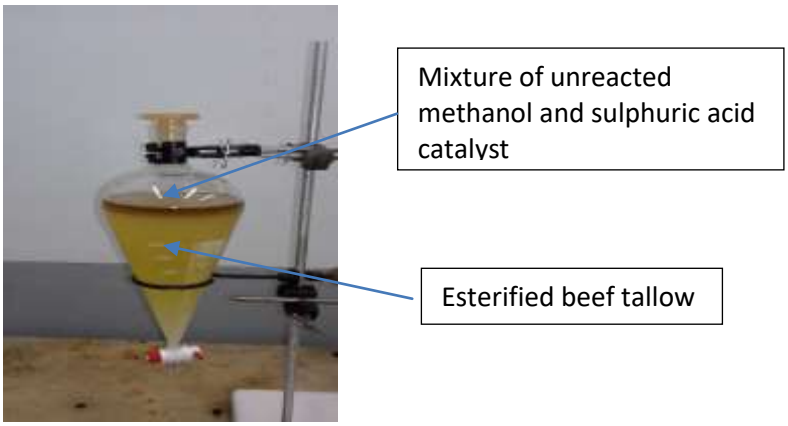


Figure 3. Separation of esterified fat and unreacted methanol

2.3.2. Alkaline Catalyzed Transesterification

The following reaction parameters were controlled: methanol to fat molar ratio 3:1 to 9:1, catalyst loading weight 0.5-2 w/w % of fats and reaction time 60 min to 120 min. The temperature was kept constant at 60 ℃ and the agitation speed at 1000 rpm. A known amount of methanol and KOH were separately prepared by stirring at 300 rpm at 50 °C for 20 min to form potassium methoxide. The mixture was added into esterified fat which was cooled to 60 °C in a 2000 ml round bottom flask. The reactants were stirred at constant speed of 1000 rpm using an overhead mechanical stirrer as shown in Figure 4. The products formed were channeled to a separating funnel and left for 12 hours to settle as shown in Figure 5. The glycerol at the lower layer was discarded. Biodiesel at the upper layer was first splashed with acidified water prepared by adding 3 ml 35 % phosphoric acid to warm distilled water and cleaned further with warm distilled water at 60 °C by rolling the separating funnel gently on a table for 60 sec. The biodiesel was subjected to heat of 110 °C for 60 min to evaporate any excess water and methanol. Lastly, the biodiesel was dried by mixing with sodium sulphate in a beaker for 3 h then filtered.

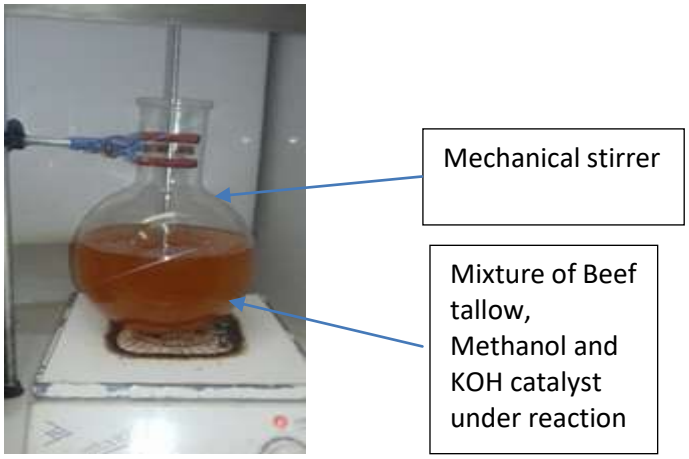


Figure 4. Beef tallow alkali catalyzed transesterification setup

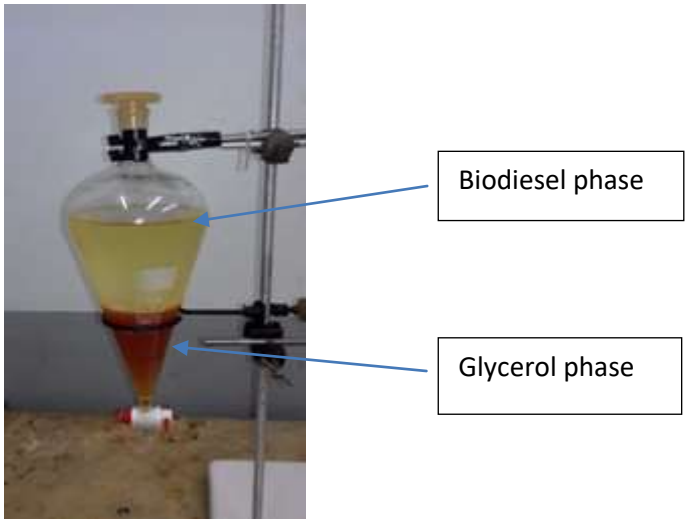


Figure 5. Separation of beef tallow biodiesel and glycerol

2.4. Measurement of Physical Properties of Waste Beef Tallow and Beef Tallow Methyl Esters

Physical properties of waste beef tallow and produced biodiesel were measured based on ASTM standards at Intertek laboratory in Durban, South Africa. The physical properties measured were density, iodine value, acid value, moisture content, saponification value, kinematic viscosity, calorific value, cold filter plugging point, flash point and pour point as shown in Table 1. The last column in the table indicates ASTM requirements for biodiesel.

Table 1 Properties of waste beef tallow and beef tallow methyl esters

Property	Test procedure	Units	Beef tallow	Beef tallow methyl ester	Diesel	ASTM D6751
Density @ 15 °C	ASTM D4052	g/cm^3	0.8949	873.1	0.8303	0.87-0.90
Iodine value	AOCS Cd 1b-87	cg/g	42.1	-	8	-
Acid value	AOCS Ca 5a-40	% m/m	2.404	0.4	0.40	0.50 mgKOH/g max
Moisture content	ASTM D1364	% m/m	0.164	0.035	-	0.050 vol. % max
Saponification value	AOCS Cd 3-25	mgKOH/g	167.8	-	-	-
Kinematic viscosity at 15 °C	ASTM D7042	mm^2/s	-	5.85	2.04	1.9-6.0 mm^2/s
Calorific value	ASTM D240	MJ/kgK	-	39.32	44.90	-
CFPP	IP 309	-	-	13	-	-
Flash point	ASTM D93	°C	-	163	77	130.0 °C min
Pour point (°C)	ASTM D97	°C	-	11	-16	-15- 16
Cloud point	ASTM D97	°C	-	15	-	-

2.5. Characterization

2.5.1. Gas Chromatography and Mass Spectroscopy

The biodiesel was analyzed using gas chromatography mass spectroscopy (GC-MS) (GC QP 2010 ultra) equipment manufactured by Shidmadzu, Japan, to determine the percentage of methyl esters present in beef tallow biodiesel. The GC was equipped with Ultra Alloy capillary column 30 m*0.25 mm*0.25 µm. A volume of 0.25 µl of beef tallow biodiesel was

injected for gas chromatography. Helium was used as the carrier gas at a flow rate of 74.0 ml/min and splitless injection mode of 70.0 split ratio with the program as per Table 2.

Table 2 The Gas chromatography and mass spectroscopy operating program

Rate	Temperature	Hold Time (Min)
-	100.0	1.00
5.00	230.0	2.00
10.00	300.0	15.00

2.5.2. Fourier Transform Infrared

A Fourier transform infrared (FTIR) instrument was used to determine various functional groups present in the waste beef tallow and beef tallow methyl esters. The instrument used for the analysis was Spectrum 100 with Universal ATR manufactured by Perkin Elmer (USA). The wave number range used was 4000 cm^{-1} to 400 cm^{-1}.

2.5.3. Thermogravimetric Analysis

The thermogravimetric (TG) and the derivative thermogravimetric (DTG) curves for beef tallow biodiesel sample were recorded using Simultaneous Thermal Analyser 6000 (STA 6000) manufactured by Perkin Elmer (USA). The sample was heated at 10 °C/min under nitrogen atmosphere at flow rate of 20 ml/min in a temperature range of 25 °C to 1000 °C with a sample mass of 86.25 mg. The loss in the weight was recorded as a function of temperature.

3. RESULTS AND DISCUSSION

3.1. Biodiesel Yield and Parameters Affecting Transesterification Reactions

The beef tallow with acid value of 2.4 mgKOH/g was pretreated by adding 1 w/w % H_2SO_4 and methanol (methanol to fat molar ratio 6:1) at a temperature of 60 ℃ for 1 h reaction time and 1000 rpm which successfully reduced FFA to less than 1 %. Esterification was followed by transesterification using KOH catalyst under varying conditions. The biodiesel yield was calculated using the following equation:

$$Yield = \frac{Weightofbiodiesel}{weightofesterifiedfatused} * 100$$

It was observed that the beef tallow biodiesel product separates quickly from glycerol, which occurs within 20 min when left untouched in a separation funnel. The color of beef tallow was brown which changed to light yellowish color for biodiesel after the transesterification process. A constant 1000 rpm mechanical stirrer was used for all experiments because when a magnetic stirrer was used incomplete agitation occurred which resulted in soap formation and fats solidifying at the upper layer of the reactor.

3.1.2. The influence of methanol to fats molar ratio

Stoichiometrically, the quantity of methanol to fats necessary for transesterification reaction is 3:1 molar ratio. In actual experimental work, this ratio is insufficient to drive the reaction to product formation thus more methanol is needed to increase the reaction rate [22, 23]. The effect of the molar ratio varied from 3:1 to 9:1 with biodiesel yield is shown in Figure 6. It is observed that lower molar ratio produces a low yield. The best yield of 97.83 % was obtained with a molar ratio of 6:1 and addition of more methanol had no effect on yield. It was also noted that soaps form at high molar ratios. The excess methanol for high molar ratio remains in the ester phase which decreases the flash point of biodiesel.

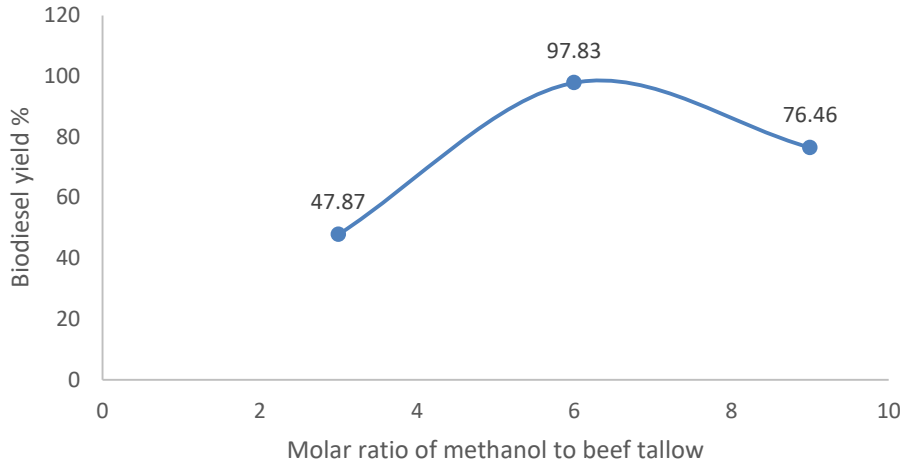


Figure 6. Influence of molar ratio of methanol to beef tallow on yield at 60 ℃ temperature and KOH load of 1.5 wt %

3.1.3. The Influence of Catalyst Concentration

Potassium hydroxide (KOH) catalyst was varied in the range 0.5 w/w % to 2.0 w/w % of fats used for the experimental work. The highest biodiesel yield (98.21 %) was achieved using 1.5 % KOH. It was observed that an excessive amount of catalyst led to reduction of biodiesel yield and formation of an emulsion in the biodiesel. The influence of catalyst concentration and biodiesel yield on beef tallow is shown in Figure 7.

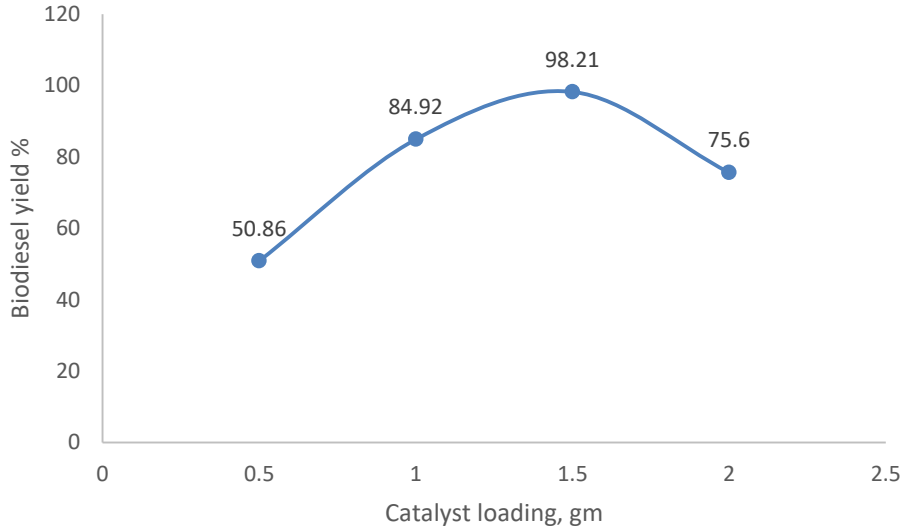


Figure 7. Influence of catalyst concentration on beef tallow biodiesel yield at 60 ℃ temperature, 90 min reaction time and 1000 rpm agitation speed.

3.1.4. The Influence of Reaction Time

Reaction time was varied from 60 min to 120 min while keeping other reaction conditions constant. Results show biodiesel yield increased with increase in reaction time and a maximum yield of 95.8 % was achieved after 90 min. Further increase in reaction time reduced yield to 87.88 %. The reduction in yield after 120 min might be due to the reversible nature of the transesterification process. This is due to biodiesel reverting back to reactants

hence reducing the overall biodiesel yield. The influence of reaction time and biodiesel yield is shown in Figure 8.

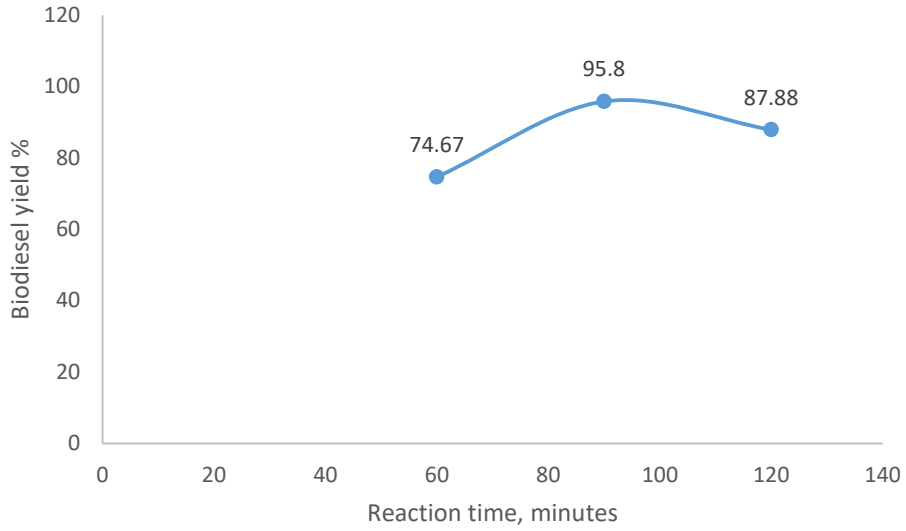


Figure 8. Influence of reaction time on biodiesel yield at reaction temperature 60 ℃ and catalyst concentration of 1.5 w/w % and 1000 rpm agitation speed.

3.2. Characterization of Biodiesel

3.2.1. Physical Properties

The physical properties of beef tallow methyl esters as shown in Table 1 were assessed to determine biodiesel suitability when compared to the standards for diesel fuel. The density of the beef tallow biodiesel after transesterification was reduced from 0.8949 kgm^{-3} to 0.873 kgm^{-3}. The values met the ASTM standard limits of below 900 kg/m^3.

The kinematic viscosity (a measure of resistance to flow of fuel) can be used to select the fatty acid profile of raw materials used for production of biodiesel. Kinematic viscosity plays a vital role in fuel atomization and penetration [13]. It can be observed from Table 1 that viscosity of beef tallow biodiesel is 5.85 mm^2s^{-1} which met the ASTM standards for engine applications.

Beef tallow biodiesel has high cloud and pour points of 15 ℃ and 11 ℃ respectively, which impacts negatively on their use at low temperatures. Unsaturated fatty compounds have significantly lower melting points compared to saturated fatty compounds. Saturated compounds crystallize at higher temperatures thus biodiesel from beef tallow feed-stocks display higher CFPP [24].

Flash point of biodiesel is indicative of the safety of biodiesel during transport, storage and handling [13]. From Table 1 it is evident that the resultant biodiesel had a flashpoint of 138 ℃ which is within the ASTM standard requirements and indicates an acceptable methanol limit.

Acid value (AV) of biodiesel is a measure of the free fatty acids and of the aging of the fuel [25]. It is measured in terms of KOH required to neutralize the fatty acids in 1 g of sample. The AV of beef tallow was 2.406 mgKOH/g. It is clear that the two-step process reduced AV to 0.4 mgKOH/g which is within the recommended ASTM limit of 0.60 max.

The calorific value refers to the energy liberated upon combustion of fuel. Beef tallow biodiesel yielded calorific value of 39.32 MJ/kg which is closely comparable to diesel fuels at

44.90 MJ/kg. The lower energy of beef tallow biodiesel is because of high oxygen present in biodiesel [26].

3.2.2. Chemical Properties of Beef Tallow Methyl Esters

3.2.2.1. Fatty Acids Profile

The GC-MS was used to quantify compounds present in beef tallow biodiesel. The GC-MS chromatogram shows 13 peaks detected as fatty acid methyl esters (Figure 9 and Table 3). From Figure 9 one can see that six peaks of the biodiesel sample had larger areas. The main components shown by these peaks were contributed by methyl tetradecanoate, hexadecenoic acid, hexadecanoic acid, heptadecanoic acid, 9-octadecenoic acid and methyl stearate as tabulated in Table 3.

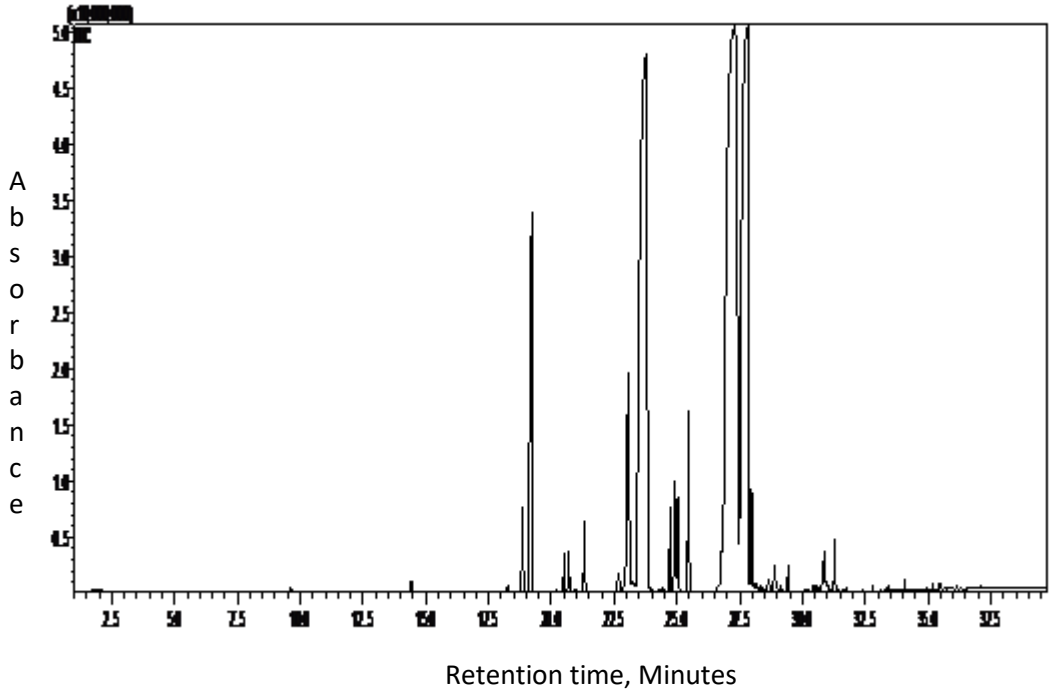


Figure 9. GC-MS chromatogram beef tallow biodiesel sample

Table 3 Fatty acids profiles of beef tallow biodiesel from GC-MS

Peak no.	Identified methyl ester	Formula	Molecular weight	Beef tallow biodiesel	
				Retention time	Area
1	Methyl myristoleate	$C_{15}H_{28}O_2$	240.3816	18.852	0.75
2	methyl tetradecanoate	$C_{15}H_{30}O_2$	242.403	19.222	5.05
3	Pentadecanoic acid	$C_{16}H_{32}O_2$	256.43	20.511	0.29
4	Tridecanoic acid	$C_{15}H_{30}O_2$	242.3975	20.692	0.30
5	Pentadecanoic acid	$C_{16}H_{32}O_2$	256.4241	21.320	0.53
6	9-hexadecenoic acid,methyl ester	$C_{17}H_{32}O_2$	268.434	23.064	3.45
7	hexadecanoic acid,methyl ester	$C_{17}H_{34}O_2$	270.4507	23.797	24.12
8	Heptadecanoic acid	$C_{18}H_{36}O_2$	284.4772	24.739	0.61
9	hexadecanoic acid	$C_{18}H_{36}O_2$	284.4772	4.932	1.59
10	heptadecanoic acid	$C_{18}H_{36}O_2$	284.4772	25.483	1.52
11	9-octadecenoic acid	$C_{19}H_{36}O_2$	296.4879	27.303	37.55
12	methyl stearate	$C_{21}H_{40}O_2$	298.511	27.793	23.56
13	cis-11-Eicosenoic acid, methyl ester	$C_{21}H_{40}O_2$	324.549	30.864	0.67

These six compounds are long chain compounds that that are saturated, constituting 90 %. These characteristics have a direct effect on fuel properties. The high saturated fatty acids of beef tallow biodiesel have high cloud and pour points, hampering its applicability in winter seasons [25]. Moreover, properties like cetane number, oxidation stability and heat of combustion will increase with an increase in the number of carbon atoms but decreases with an increase in the number of double bonds. A fuel with compounds that are fully saturated has better oxidation stability and cetane number but poor cloud and pour points. The low level of unsaturation depicted by double bonds in methyl stearate and 9-octadecenoic acid has significant inhibiting gel formation thus lowering the cloud and pour points of the fuel [27]. A low cloud point is a beneficial property that maintains biodiesel at a liquid state in low temperatures. Waste beef tallow methyl esters have excellent properties in terms of cloud points, pour points, atmospheric oxidation and a high cetane number.

3.2.2.2. Fourier Transform Infrared Spectroscopy Results

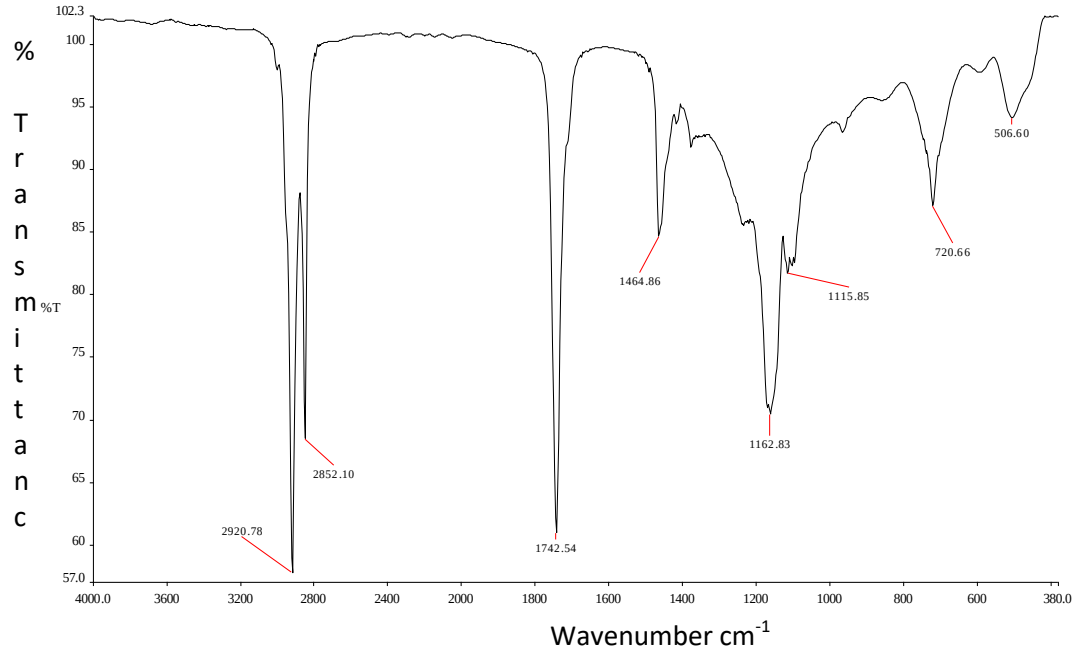


Figure10. FTIR waveform of beef tallow

The Fourier transform infrared spectroscopy (FTIR) spectra of the beef tallow and produced beef tallow methyl esters were carried out in the range 4000 cm^{-1} to 400 cm^{-1} to access the important functional groups related to biodiesel content, loss and formation of linkages, production and loss of OH [28]. The FTIR spectra of beef tallow and beef tallow biodiesel are shown in Figures 10 and 11 respectively. The FTIR of tallow and methyl ester have similar patterns due to similarities that exist among triglyceride and methyl esters in the functional groups. There are changes in (%) absorbance and functional group which indicates that modifications occur in the transesterification process. In Figure 11 several absorption features are observed and when FAME increases, these peaks increase. The strong ester peaks at 1742.16 cm^{-1} (C=O) and at 1100 cm^{-1} to 1200 cm^{-1} (C-O ester) are clearly present in the spectra. The absence of a broad band in the 3100 cm^{-1} to 3500 cm^{-1} region related to axial deformation of OH groups suggest that the biodiesel had a low water content which is verified by the thermogravimetric curve in the next section. The results show the produced biodiesel is of good quality for running a diesel engine.

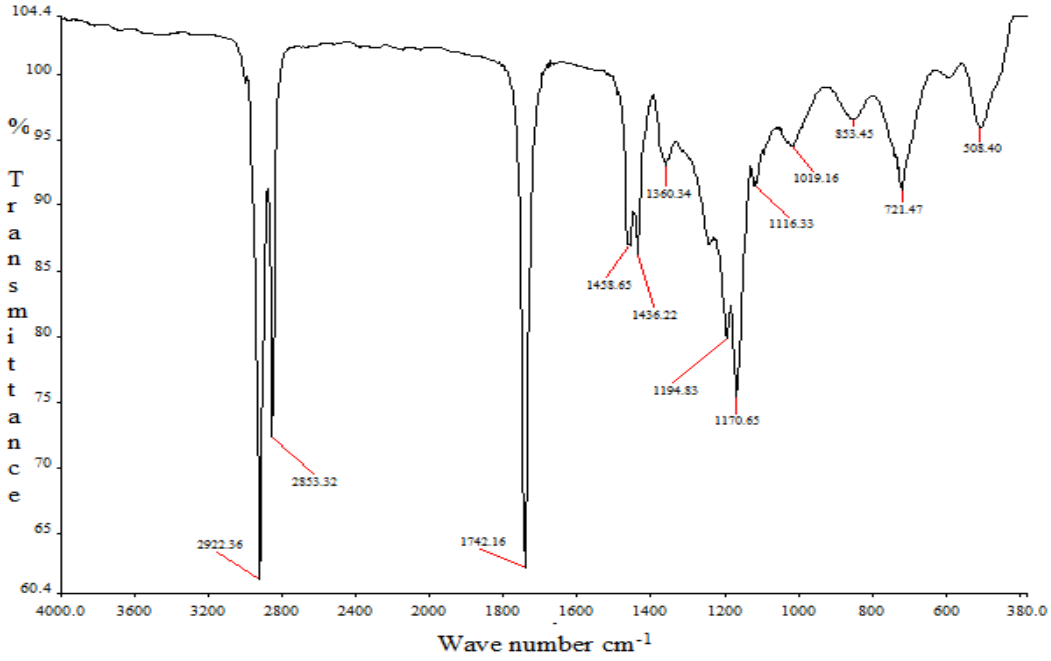


Figure 11. FTIR waveform of biodiesel produced from beef tallow

3.2.2.3. Thermal Analysis

TG and DTG analysis generates thermal decomposition curves that can be used as efficient, low cost methods to determine biodiesel quality. The curves show degradation stages as a function of time or temperature as reported by Silva et al. [29]. Thermal stability and volatility also indicate the ignition quality of biodiesel [7, 30]. The curves in the Figure 12 show decomposition and loss of weight of biodiesel and loss of derivative weight at the corresponding temperatures. The change in weight is a result of combustion of beef tallow methyl esters. Only one heating rate was used in order to show thermal decomposition in a form that is more visually accessible. The biodiesel obtained by KOH revealed only one degradation step in the temperature range of 150 °C to 350 °C with a mass loss of 95 % with degradation of methyl esters. The smooth degradation step indicates an absence of triglyceride and a low water content in biodiesel sample which confirms the high conversion rate of biodiesel.

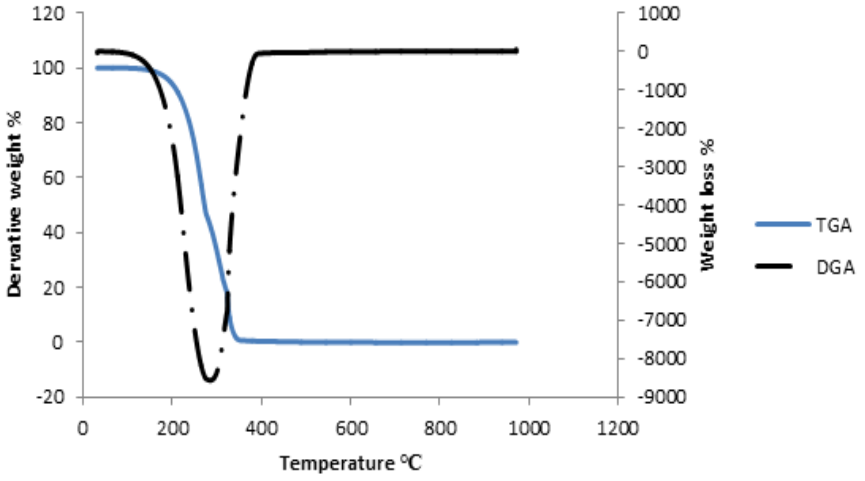


Figure 12. TG/DTG curves of beef tallow biodiesel produced by transesterification

4. CONCLUSION

Waste beef tallow with high acid value can produce quality biodiesel with a high yield when a two-step transesterification process is employed. According to experimental work, the best conditions for methanolysis of beef tallow is 1.5 w/w % catalyst loading, methanol to oil molar ratio 6:1 and reaction time of 90 min. Waste beef tallow biodiesel has a unique short separation period from glycerol of 20 min. Transesterification is an available method employed to reduce the viscosity of beef tallow that is accepted as fuel for diesel engines. The viscosity of beef tallow biodiesel at 40 ℃ is 5.85 mm^2/s which meets ASTM D 6751 international standards and is close to that of diesel fuel.

The chemical composition of beef tallow biodiesel is typically constituted by methyl tetradecanoate, hexadecenoic acid, hexadecanoic acid, heptadecanoic acid, 9-octadecenoic acid and methyl stearate. A smooth TG degradation curve for thermal analysis confirms the absence of triglycerides and therefore quality biodiesel is obtained. Therefore, economically viable and renewable biodiesel could be produced from the low cost feedstock of waste beef tallow which can be blended with diesel and used in existing diesel engines without any modification to meet the ever rising demand for fuel.

REFERENCES

[1] Murdoch, W. W., et al. Improving Wellbeing and Reducing Future World Population. *PloS One*, 13(9), 2018, p. e0202851. https://doi.org/10.1371/journal.pone.0202851

[2] Cantore, N., Calì, M. and te Velde, D. W. Does Energy Efficiency Improve Technological Change and Economic Growth in Developing Countries? *Energy Policy*, 92, 2016, p. 279-285. https://doi.org/10.1016/j.enpol.2016.01.040

[3] Gerarden, T. D., Newell, R. G. and Stavins, R. N. Assessing the Energy-Efficiency Gap. *Journal of Economic Literature*, 55(4), 2017, p. 1486-1525. DOI: 10.3386/w20904

[4] Boffetta, P., et al. Occupational Exposure to Diesel Engine Emissions and Risk of Cancer in Swedish Men and Women. *Cancer Causes & Control*, 12(4), 2001, p. 365-374.

[5] Marchetti, J., Miguel, V. and Errazu, A. Possible Methods for Biodiesel Production. *Renewable and Sustainable Energy Reviews*, 11(6), 2007, p. 1300-1311. https://doi.org/10.1016/j.rser.2005.08.006

[6] Dias, J. M., Alvim-Ferraz, M. C. M. and Almeida, M. F. Production of Biodiesel from Acid Waste Lard. *Bioresource Technology*, 100(24), 2009, p. 6355-6361. https://doi.org/10.1016/j.biortech.2009.07.025

[7] Sbihi, H. M., et al. Production and Characterization of Biodiesel from *Camelus dromedarius* (Hachi) Fat. *Energy Conversion and Management*, 78, 2014, p. 50-57. https://doi.org/10.1016/j.enconman.2013.10.036

[8] Sendzikiene, E. et al. Biodegradability of Biodiesel Fuel of Animal and Vegetable Origin. *European Journal of Lipid Science and Technology*, 109(5), 2007, p. 493-497. https://doi.org/10.1002/ejlt.200600243

[9] Gui, M. M., Lee, K. and Bhatia, S. Feasibility of Edible Oil vs. Non-Edible Oil vs. Waste Edible Oil as Biodiesel Feedstock. *Energy*, 33(11), 2008, p. 1646-1653. https://doi.org/10.1016/j.energy.2008.06.002

[10] Tulashie, S. K., et al. Biodiesel Production from Shea Butter: A Suitable Alternative Fuel to Premix Fuel. *Materialia*, 3, 2018, p. 288-294. https://doi.org/10.1016/j.mtla.2018.08.038

[11] Canakci, M. The Potential of Restaurant Waste Lipids as Biodiesel Feedstocks. *Bioresource Technology*, 98(1), 2007, p. 183-190. https://doi.org/10.1016/j.biortech.2005.11.022

[12] Mujere, N. Water Quality Impacts of Abattoir Activities in Southern Africa In: Impact of Meat Consumption on Health and Environmental Sustainability. IGI Global. 2016, p. 64-74.

[13] da Cunha, M.E. et al. Beef Tallow Biodiesel Produced in a Pilot Scale. *Fuel Processing Technology*, 90(4,) 2009, p. 570-575. https://doi.org/10.1016/j.fuproc.2009.01.001

[14] Shahir, V. K., et al. Experimental Investigation on Performance and Emission Characteristics of a Common Rail Direct InjectionEngine Using Animal Fat Biodiesel Blends. *Energy Procedia*, 117, 2017, p. 283-290. https://doi.org/10.1016/j.egypro.2017.05.133

[15] Demirbaş, A. Chemical and Fuel Properties of Seventeen Vegetable Oils. *Energy Sources*, 25(7), 2003, p. 721-728. https://doi.org/10.1080/00908310390212426

[16] Kraiem, T., et al. Characterization of the Liquid Products Obtained from Tunisian Waste Fish Fats using the Pyrolysis Process. *Fuel Processing Technology*, 138, 2015, p. 404-412. https://doi.org/10.1016/j.fuproc.2015.05.007

[17] Yilmaz, N., Atmanli, A. and Vigil, F. M. Quaternary Blends of Diesel, Biodiesel, Higher Alcohols and Vegetable Oil in a Compression Ignition Engine. *Fuel*, 212, 2018, p. 462-469. https://doi.org/10.1016/j.fuel.2017.10.050

[18] Dias, J. M., Alvim-Ferraz, M. C. and Almeida, M. F. Production of Biodiesel from Acid Waste Lard. *Bioresource Technology*, 100(24), 2009, p. 6355-6361. https://doi.org/10.1016/j.biortech.2009.07.025

[19] Issariyakul, T., et al. Production of Biodiesel from Waste Fryer Grease Using Mixed Methanol/Ethanol System. *Fuel Processing Technology*, 88(5), 2007, p. 429-436. https://doi.org/10.1016/j.fuproc.2006.04.007

[20] Vicente, G., Martınez, M. and Aracil, J. Integrated Biodiesel Production: A Comparison of Different Homogeneous Catalysts Systems. *Bioresource Technology*, 92(3), 2004, p. 297-305. https://doi.org/10.1016/j.biortech.2003.08.014

[21] Sahar, et al. Biodiesel Production from Waste Cooking Oil: An Efficient Technique to Convert Waste into Biodiesel. *Sustainable Cities and Society*, 41, 2018, p. 220-226. https://doi.org/10.1016/j.scs.2018.05.037

[22] Ramadhas, A. S., Jayaraj, S. and Muraleedharan, C. Biodiesel Production from High FFA Rubber Seed Oil. *Fuel*, 84(4), 2005, p. 335-340. https://doi.org/10.1016/j.fuel.2004.09.016

[23] Panneerselvam, S. I. and Miranda. L. R. Biodiesel Production from Mutton Tallow. In: 2011 IEEE Conference on Clean Energy and Technology (CET). IEEE, 2011. DOI: 10.1109/CET.2011.6041441

[24] Yuan, M.-H., et al. Dependence of Cold Filter Plugging Point on Saturated Fatty Acid Profile of Biodiesel Blends Derived from Different Feedstocks. *Fuel*, 195, 2017, p. 59-68. https://doi.org/10.1016/j.fuel.2017.01.054

[25] Mata, T. M., et al. Properties and Sustainability of Biodiesel from Animal Fats and Fish Oil. *Chemical Engineering Transactions*, 38, 2014, p. 175-180. DOI:10.3303/CET1438030

[26] Demirbas, A. Combustion Efficiency Impacts of Biofuels. *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects*, 31(7), 2009, p. 602-609. https://doi.org/10.1080/15567030701743718

[27] Mata, T., et al. Sustainable Production of Biodiesel from Tallow, Lard and Poultry Fat and its Quality Evaluation. *Chemical Engineering Transactions*, 19, 2010. p. 13-18. DOI: 10.3303/CET1019003

[28] Zhou, J., et al. Analysis of the Oxidative Degradation of Biodiesel Blends Using FTIR, UV–Vis, TGA and TD-DES methods. *Fuel*, 202, 2017, p. 23-28. https://doi.org/10.1016/j.fuel.2017.04.032

[29] Silva, G., et al. Physico-Chemical, Spectroscopical and Thermal Characterization of Biodiesel Obtained by Enzymatic Route as a Tool to Select the most Efficient Immobilized Lipase. *Brazilian Journal of Chemical Engineering*, 29(1), 2012, p. 39-47. DOI: 10.1590/S0104-66322012000100005

[30] Vega-Lizama, T., et al. Thermogravimetric Analysis as a Rapid and Simple method to Determine the Degradation Degree of Soy Biodiesel. *Fuel*, 156, 2015, p. 158-162. https://doi.org/10.1016/j.fuel.2015.04.047

CHAPTER 5: EXPERIMENTAL STUDY OF PERFORMANCE AND EMISSION CHARACTERISTICS OF WASTE BEEF TALLOW BIODIESEL

This chapter gives the performance and emission characteristics of beef tallow biodiesel when used in compression ignition engine.

Cite this Article: Kosgei, C. and Inambao F. L. Experimental Performance and Emission Characteristics of Waste Beef Tallow Biodiesel. *International Journal of Mechanical Engineering and Technology*, 10(11), 2019, pp. 371-381.

(This chapter is part of work was published in *International Journal of Mechanical Engineering and Technology*)

Link to the article:
http://www.iaeme.com/ijmet/issues.asp?JType=IJMET&VType=10&IType=11

International Journal of Mechanical Engineering and Technology (IJMET)
Volume 10, Issue 11, November 2019, pp. 371-381, Article ID: IJMET_10_11_031
Available online at http://www.iaeme.com/ijmet/issues.asp?JType=IJMET&VType=10&IType=11
ISSN Print: 0976-6340 and ISSN Online: 0976-6359

EXPERIMENTAL STUDY OF PERFORMANCE AND EMISSION CHARACTERISTICS OF WASTE BEEF TALLOW BIODIESEL

Cheruiyot Kosgei and Prof. Freddie L. Inambao*
Department of Mechanical Engineering,
University of Kwazulu-Natal, Durban, South Africa
https://orcid.org/0000-0001-9922-5434
* Corresponding Author Email: inambaof@ukzn.ac.za

ABSTRACT

Climate change has led to extreme cold and hot seasons and unexpected changes in global weather patterns. This negative effect is attributed to emissions of pollutant gases from industries and vehicles used in the transport sector. Burning diesel fuels in compression ignition (CI) engines *emits poisonous gases such as SO_2, CO, NOx and UHC which together contribute to the rise in global temperatures. It is imperative to utilize an alternate fuel to diesel in compression ignition (CI) engines that is clean and emits non-poisonous gases. Biodiesel fuel can substitute diesel fuel without any modification to engine combustion chamber geometry. Biodiesel feedstock is easily available from plants and animal fats which are renewable in nature. The net carbon contribution of biodiesel is lower than diesel emission. Moreover, biodiesel's sulphur emission (which is extremely poisonous) is negligible compared to diesel fuel. Production of feedstock from edible oil sources has led to food vs fuel conflicts and contributes to malnutrition in developing countries. This research utilized low cost feedstock of waste beef tallow for biodiesel synthesis and its application in a CI engine. The biodiesel and diesel blends B0, B20, B40, B60, B80 and B100 were used in a stationary tractor diesel engine. Performance and emission parameters brake specific fuel consumption (BSFC), brake thermal efficiency (BTE), exhaust gas temperature (EGT), carbon monoxide (CO), oxides of nitrogen (NOx) and hydrocarbon (HC) were tested at a constant speed of 2000 rpm and varying loads. The BSFC, EGT and NOx increased for biodiesel than diesel fuel while BTE, CO and HC were lower for biodiesel fuel. Thus, waste beef tallow biodiesel can substitute diesel in CI engines.*

Keywords: biodiesel, performance, emissions, physical properties.

Cite this Article: Cheruiyot Kosgei and Freddie L. Inambao, Experimental Study of Performance and Emission Characteristics of Waste Beef Tallow Biodiesel. *International Journal of Mechanical Engineering and Technology* 10(11), 2019, pp. 371-381.
http://www.iaeme.com/IJMET/issues.asp?JType=IJMET&VType=10&IType=11

1. INTRODUCTION

The transport sector consumption of energy globally has been projected to increase by 56 % between 2010 to 2040 according to [1, 2]. This growth in energy demand leads to an increase in exhaust emissions of vehicle engines. There are quite a number of fuels sourced from biomass which can power diesel engine. The inventor of the diesel engine, Rudolf Diesel, experimented first prototype on neat vegetable oil [3]. Developments and intensive research on engines with lean fuel engines for agricultural purposes are on the rise. In agricultural vehicles, especially tractors, combustion is quality governed since a rich fuel/air mixture produces smoke while producing less power [4]. The most frequently used biomass based fuel in diesel engines is biodiesel commonly known as fatty acid alkyl ester. European Union and American standards (ASTM) for biodiesel are often used in many countries. Any differences among the standards are related to the dominant feedstocks for a particular country [5].

Studies on biodiesel's life cycle in the environment reveal a positive impact as it is an environmentally friendly fuel [6]. Biodiesel is biodegradable [7], though its quality may deteriorate when exposed to light, oxygen and metal contaminants, but additives are used to mitigate these effects [8]. Before being used in an engine, biodiesel blending can be done in any ratio with diesel and used without any engine modifications due to its characteristics being so close to diesel fuel [9]. Further, biodiesel contains 10 % to 15 % oxygen content attached to carbon atoms [9] which makes it an oxygenated fuel, reducing carbon monoxide (CO) emissions to the atmosphere.

Wood et al. [10] investigated biodiesel for motorsport application in an automotive diesel engine by varying in-cylinder pressure for soybean B100 and B50 beef tallow. The torque achieved for all the injection timings were lower for B100 soybean and B50 beef tallow than fossil diesel. The authors altered injection timing and increased fuelling of biodiesel to achieve the high torque output required for motorsport. Abed et al. [11] examined emissions of biodiesel at different engine loads and constant 1500 rpm speed in a 4-stroke single cylinder engine. The authors found increased emissions of oxides of nitrogen (NO_x) and carbon dioxide (CO_2) for B10 and B20 in comparison to diesel fuel and overall, biodiesel conformed with the required emissions standards. Similar results in emissions conformation with ASTM standards were observed by [12]. Gad et al. [13] found lower thermal efficiency and air-fuel ratio for B20 and B100 biodiesel when compared with diesel fuel for tested engine loads. Similarly, specific fuel consumption and mechanical efficiency decreased while using biodiesel as reported by [14].

Senthilkumar et al. [15] conducted an experiment on a 4-stroke compression ignition engine with B20, B40, B45 and B50 palm biodiesel. Authors reported CO and hydrocarbon (HC) reduction of 46 % and 73 % respectively, while brake thermal efficiency (BTE) for B40 was lower in comparison to diesel for all tested speeds. Most of the mentioned tests and published articles show tests of biodiesel produced from vegetable oils. Biodiesel tests from waste animal fats are still scarce but recently it has gained attention as assessed from available publications. Alptekin et al. [16] conducted experiment for chicken biodiesel and its blends under 0.15 kNm, 0.3 kNm, 0.45 kNm and 0.6 kNm load conditions. Brake specific fuel consumption (BSFC) values was inversely proportional to engine loads for the blends tested. The high proportion of bioethanol raised level of BSFC, CO_2 and NO_x while CO level reduced. Shahir et al. [17] carried out an experiment on a common rail direct injection (CRDI) engine with animal fat biodiesel mixed as B10, B20, B30, B40 and B50 at a constant speed of 2800 rpm. The authors reported B30 biodiesel had better BSFC and emissions than petroleum diesel. This study presents low cost beef tallow biodiesel which was run in a stationary tractor compression ignition engine to evaluate performance and emission parameters.

2. MATERIALS AND METHODS

2.1. Biodiesel Production and Specification

Two feed-stocks named WBT 1 and WBT 2 both from beef tallow were converted to biodiesel in 2 stage of acid and base catalysed process. During the first step catalysed by acid, the quantity sulphuric acid used was 1.5 % w/w oil plus 6:1 methanol to oil molar ratio. A known methanol weight and oil was separately stirred for 15 minutes using a mechanical stirrer. The mixture was added to 850 g of melted fat at 50 ℃ and stirred for 60 minutes at 1000 rpm keeping a temperature of 60 ℃. The product was then separated in a separating funnel and cleaned with warm purified water. The second step was catalysed by KOH and 6:1 methanol to esterified fat molar ratio. 5.92 g of KOH and 190.3 g methanol were stirred to form a homogeneous solution. The solution was added to the esterified fat and stirred at 1000 rpm using a mechanical stirrer for 90 minutes at 60 ℃. Mixture of glycerol and biodiesel was left for 12 h to settle. A water wash was done to clean the biodiesel which was then dried with anhydrous sodium sulphate.

Blending of biodiesel and diesel was performed using the splash blending method. During splash blending, biodiesel was heated to 45 ℃ and mixed with diesel in a beaker while being stirred continuously to ensure uniform mixing. The blending ratio produced B0, B20, B40, B60, B80 and B100 biodiesel. The measured physical properties of beef tallow biodiesel and diesel are indicated in Table 1.

Table 1 Physical properties of obtained beef tallow biodiesel

Property	Waste beef tallow (WBT 1)	Beef tallow biodiesel		Diesel
		WBT 1	WBT 2	
Acid value mg KOH/g	2.404	0.40	0.31	0.40
Density g/cm^3	0.8949	0.8731	0.8649	0.8303
Kinematic viscosity@40cst	-	5.85	4.708	2.04
Flash point ℃	-	163	83	77
Calorific value MJ/kg	-	39.32	42.72	44.04
CFPP ℃	-	13	10	-
Pour point ℃	-	11	18	-16
Cloud point	-	15	-	-
Water ppm	-	-	797	-
Saponification value	167.8	-	-	-
Iodine value	42.1	-	-	-

2.2. Engine Test Procedure

Biodiesel used for the engine test was produced from WBT 1 with properties shown in Table 1. The engine test arrangement consisted of stationary tractor diesel engine (Figure 1), P2000 hydraulic dynamometer, computer and emission gas device. Figure 2 shows a diagrammatic arrangement of the test system. A three cylinder, four-stroke, direct injection tractor engine was used for the test. Specifications of TT55 engine are summarized in Table 2. The fuels used were denoted as B0, B20, B40, B60, B80 and B100 depending on the percentage of biodiesel used in the blend.

The exhaust gases of engine CO, NO_x and HC were measured using an emission gas analyser while a gravimetric fuel gauge measured fuel consumption. A thermocouple inserted at exhaust end recorded exhaust gas temperature (EGT). The data was taken in triplicate and average recorded for analysis.

Table 2 Specifications of TT55 tractor engine

Make and model	New Holland TT 55
Bore (mm)	104
Stroke (mm)	115
Type	four stroke, 3-cylinder, direct injection
Maximum power	41.1 kw
Torque	184.6 Nm at 1500 rpm
Starter volts	12
Rated rpm	2500

Figure 1 Biodiesel blends used during engine test

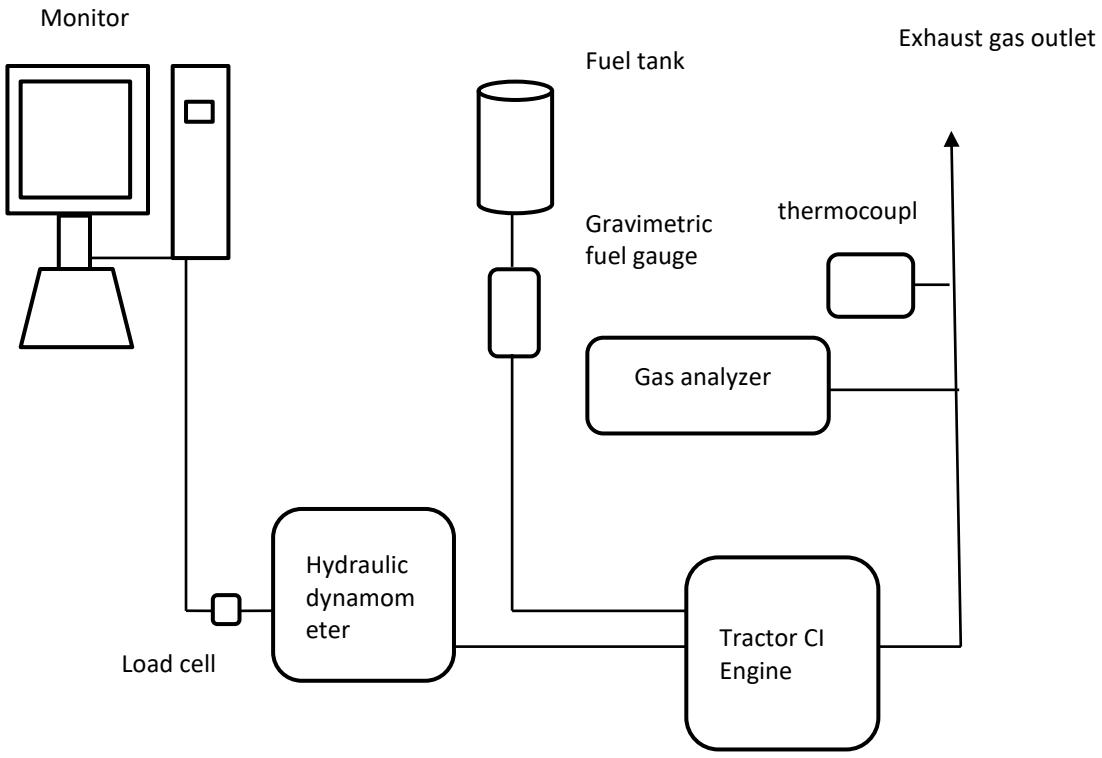


Figure 2 Diagram showing setup of test equipment

3. RESULTS

3.1. Physical Properties Influencing Performance and Emission

Physical properties of biodiesel have a close relationship with performance and emission. Feedstock source and production conditions determine the overall properties of biodiesel. Density is the mass of a material contained in a unit volume. The density of both biodiesel fuels (WBT 1 and WBT 2) conform with international ASTM standards. Density of diesel fuel is 0.8303 kg/m^3. Waste beef tallow has a density of 0.8949 kg/m^3 while WBT 1 has 0.8731 kg/m^3 and WBT 2 has 0.8696 kg/m^3. High density of biodiesel means a higher mass of biodiesel will be injected into the combustion chamber and affect the A/F ratio [18]. Kinematic viscosity influences the ease of flow of fuel in the delivery pipes of the engine. WBT 1 and WBT 2 have KV of 5.85 cst and 4.708 cst which are higher than diesel KV at 2.04 cst. With regard to a compression ignition engine, a low viscosity shortens ignition delay and same time increases ignition pressure [19]. An acid value that is out of the ASTM limits of 0.05 % max hastens the aging of fuel. WBT 1 and WBT 2 fuels have acid values of 0.40 mg and 0.31 mg KOH/g respectively, which are within ASTM standard limits.

Flash point determines the safety of biodiesel during transportation. WBT 1 has a flashpoint of 163 ℃ which conforms to ASTM standards while WBT 2 has a flashpoint of 83 ℃ which is out of ASTM specifications. A high remaining methanol content in WBT 2 is attributed to a lower flashpoint [20] close to diesel fuel at 77 ℃. Flashpoint of biodiesel influences the calorific value as shown in Table 1. WBT 1 with a high flashpoint has a lower calorific value while WBT 2 with a lower flashpoint has a high calorific value closer to diesel fuel calorific value at 44.04 MJ/kg. Pour point (PP) and cloud point (CP) determines biodiesel usability at cold temperatures. PP is always lower than CP. WBT 2 has higher CP at 18 ℃ than WBT 1 which has a CP of 11 ℃. Biodiesel PP and CP is better measured by CFPP which gives an estimate of lowest temperature at which biodiesel will clog in fuel filters in winter [21].

3.2. Performance

3.2.1. Brake Specific Fuel Consumption (BSFC)

BSFC measures the charge efficiency of an engine and how fuel is constructively transformed into work. Figure 3 shows BSFC of blends of biodiesel used. Results indicate that BSFC was inversely proportional to the applied load. The effectiveness of engine increases at high load which requires more fuel consumption. In addition, BSFC was higher when the biodiesel percentage in the blend was higher. Neat biodiesel had the highest BSFC and diesel had the lowest BSFC which is attributed to lower gross calorific value, higher kinematic viscosity and higher density of biodiesel [22]. The higher density of biodiesel implies a larger fuel consumption on a mass basis is required to achieve a power output closer to diesel fuel. Less fuel leakages were observed for biodiesel in the fuel pump as a result of the denser structure of biodiesel. At maximum load, the BSFC of B20, B40, B60, B80 and B100 were 1.05 %, 6.90 %, 12.09 %, 19.57 % and 22.54 % respectively which was less than fossil diesel fuel.

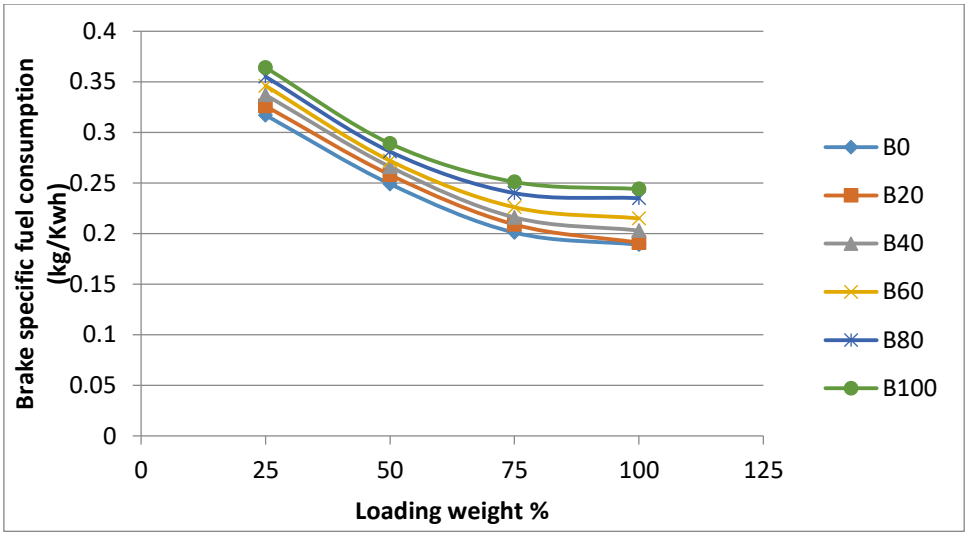


Figure 3 BSFC vs load

3.2.2. Brake Thermal Efficiency (BTE)

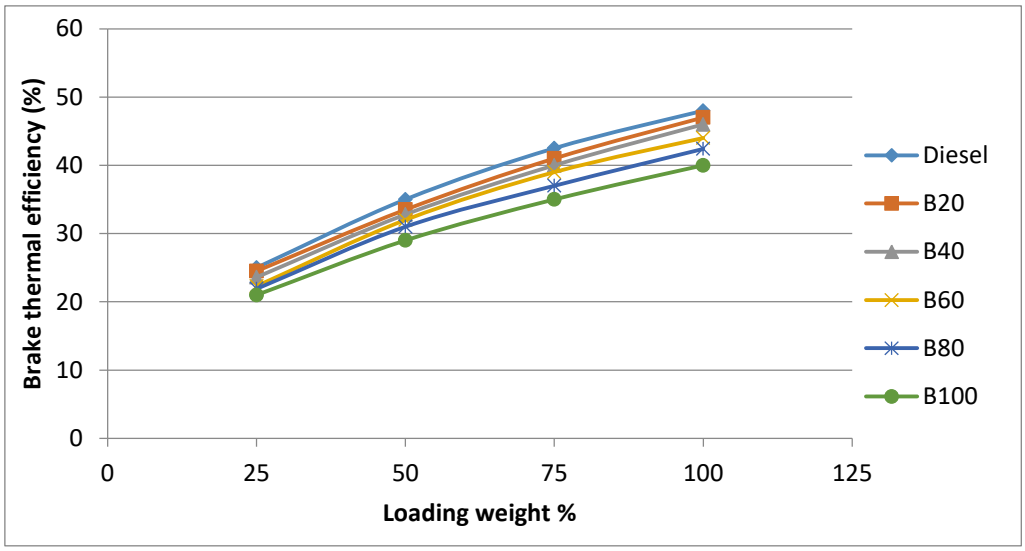


Figure 4 BTE vs load

BTE is the ratio of work output to the thermal energy of the fuel burnt in an engine. BTE is an important property used to analyze how chemical energy of fuel is converted into work done. Figure 4 gives the BTE at measured loads. Brake thermal efficiency was found to decrease with the increase in ratio of biodiesel in the blend. BTE varies directly proportionally to the applied load. Diesel fuel had the highest BTE while neat biodiesel had the lowest BTE for loading conditions. This difference in BTE is attributed to lower density and high calorific value of diesel than biodiesel which causes biodiesel to have poor fuel atomization leading to incomplete combustion [22, 23]. At maximum load condition, the BTE of B20, B40, B60, B80 and B100 biodiesel was lower by 2.08 5, 4.17 %, 8.33 %, 11.67 % and 16.67 % respectively for diesel comparison.

3.2.3. Exhaust Gas Temperature (EGT)

The quality of combustion process in cylinder can be obtained by EGT. EGT depends on the oxygen content in fuel. Available data shows reduction in cetane number reduces length of

time in premixing chamber. Thus, more heat is released due to continuous burning of fuel [24]. The EGT of various proportions of biodiesel blends and diesel fuel are shown in Figure 5. The EGT value rises with an increment in load.

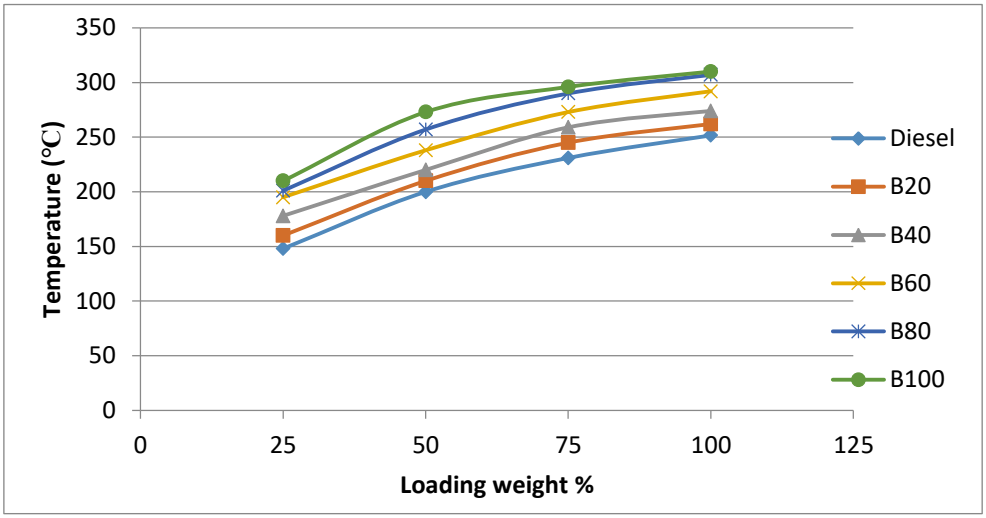


Figure 5 Variation of EGT with load

The highest proportion of biodiesel blend (B100) had increased EGT while diesel had decreased EGT for all test loads. Thermal efficiency decreased for biodiesel blends leads to increased heat loss for biodiesel and increased fuel consumption. The high value of the EGT of the B100 biodiesel blend is as a result of combustion enhancement due to availability of more oxygen content in the biodiesel. At the maximum load of 100 %, the EGT of B20, B40, B60, B80 and B100 are 3.97 %, 8.18 %, 13.84 %, 18.05 % and 18.84 % higher, respectively, compared to diesel fuel.

3.3. Exhaust Emission Characteristics

3.3.1. Oxides of Nitrogen Emissions

Nitrogen oxides comprise a group of gas emissions created due to ignition in the diesel engine. NOx is dependent on temperature and pressure. Figure 6 shows various oxides of nitrogen (NOx) emissions of biodiesel blend and diesel for different load.

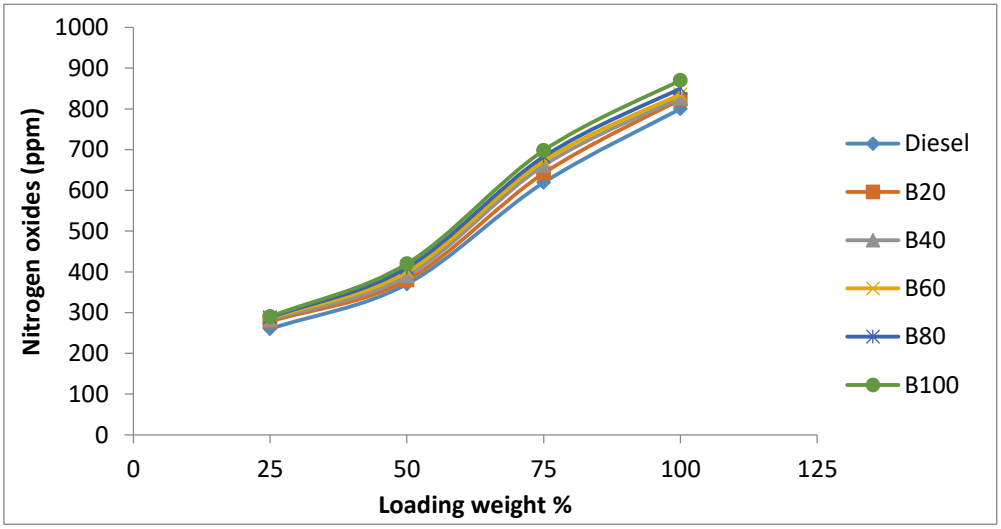


Figure 6 Oxides of nitrogen vs load

The NOx emission of pure beef tallow methyl esters is higher than diesel fuel. Higher NOx emissions are obtained when biodiesel is increased in the fuel blend. EGT and NOx emissions are correlated. Graph shows NOx is dependent on the load because an increase in engine load increases temperature in the combustion chamber and formation of NOx is dependent on temperature [25]. Also at lower load, NOx emission for all blends were closer to diesel fuel. This might have been because of low combustion temperature of biodiesel and their combinations. At maximum load, NOx emission of B20, B40, B60, B80 and B100 were higher than diesel by 0.9 %, 2.3 %, 3.3 %, 4.07 % and 5.5 % respectively.

3.3.2. Carbon Monoxide Emission

Carbon monoxide (CO) from internal combustion engines is associated with insufficient oxygen for total combustion of carbon compounds in fuel. CO is an extremely hazardous gas for the body as it inhibits proper blood flow and results in devastating poisoning. The CO emissions of biodiesel blends at different loads is shown in Figure 7. When the load increased CO emission were found increase by a wider margin.

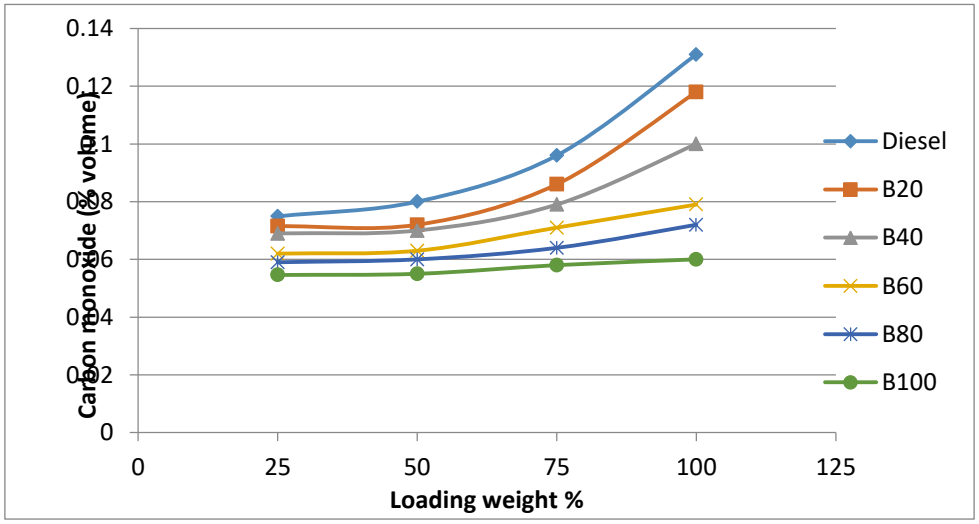


Figure 7 Carbon monoxide vs load

The CO emissions were higher at larger loading condition when compared to lower loading conditions. The reason for this increase is that as the fuel-air ratio becomes greater than the stoichiometric ratio, the level of CO emissions rise. For all loading conditions, diesel had more CO emissions than the diesel-biodiesel blend, while neat biodiesel generated the least CO emissions. Decrease in CO emissions were due to high percentage for oxygen molecules and lower carbon content in the biodiesel blend. At maximum load condition of the engine, CO reduction of B20, B40, B60, B80 and B100 were 1.08 %, 2.644 %, 3.18 %, 3.46 % and 4.06 % lower diesel fuel, respectively.

3.3.3. Hydrocarbon Emissions

Figure 8 presents the influence of hydrocarbons with respect to engine load for beef tallow biodiesel blends. When the biodiesel proportion in the mixture increased the HC emission decreased, with B100 having the lowest value for the entire tested load. The main reason is sufficient carbon combustion contributed by high oxygen content in biodiesel fuel and hence reduced HC emission. Another reason for the low HC is the high cetane number of beef tallow biodiesel. At low loading conditions, HC emission was lower than at high loads which are due to efficiency in dispersion by injector nozzle. At the maximum loading condition, HC

for B20, B40, B60, B80 and B100 were 10.86 %, 19.56 %, 28.26 %, 34.78 % and 43.47 % lower than diesel fuel, respectively.

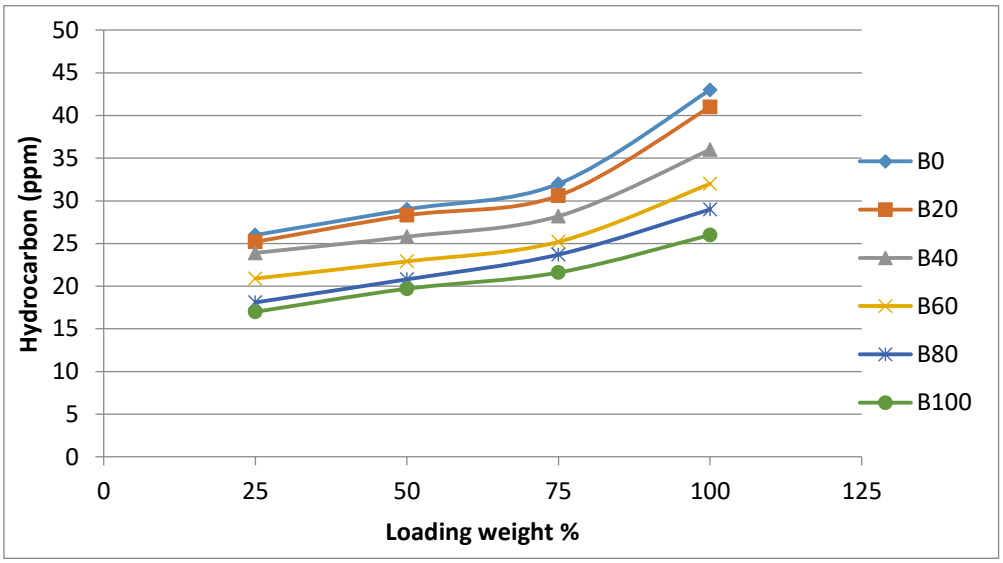


Figure 8 Variation of hydrocarbon emissions with load

4. CONCLUSIONS

A three-cylinder stationary tractor diesel engine was run using beef tallow biodiesel. Performance and exhaust emissions were measured at a constant speed of 2000 rpm and loads applied, measured on a hydraulic dynamometer. BTE, EGT and thermal efficiency performance indicators were measured. Exhaust emission of CO, NOx and HC were benchmarked with diesel as base fuel. Conclusions can be summarized from the experimental work as follows:

- The flashpoint for biodiesel has a close relation with residual methanol which makes biodiesel unsafe during transportation.
- Neat diesel fuel has higher BTE compared to biodiesel. Biodiesel B100 with the highest biodiesel percentage had the lowest BTE.
- At lower loads BSFC is higher than at high loads. The BSFC of biodiesel are higher than diesel fuel for the blends used.
- The oxides of nitrogen increased for biodiesel and biodiesel blends for all the loads. The NOx emissions increased from low loads to higher loads.
- The emissions of CO and HC were lower for biodiesel at all tested loads than neat diesel. For all the tested loads, emissions increased from lower loads to high loads.
- Biodiesel blends comprise high oxygen content fuels and hence the renewability provides a complete combustion process.

Thus, from the mentioned conclusions, biodiesel from beef tallow can be used as an alternate fuel to diesel in order to reduce pollutant emission of gases and overreliance on imported fossil diesel.

REFERENCES

[1] Van der Hoeven, M. World Energy Outlook 2012. Tokyo: International Energy Agency, 2013.

[2] US Energy Information Administration. Briefing, U.S. International Energy Outlook 2013. Washington: US Energy Information Administration, 2013.

[3] Nitske, W.R. Rudolf Diesel: Pioneer of the Age of Power. *Business History Review (pre-1986)*, 39(000002), 1965, p. 280.

[4] Eastop, T.D. and Mc Conkey, A. Applied Thermodynamics for Engineering Technologies. London: Longman, 1986.

[5] Barabás, I. and Todoruţ, I.-A. Biodiesel Quality, Standards and Properties. In: Montero, G and Stoytcheva, M. eds, Biodiesel-Quality, Emissions and By-Products. Rijeka: InTech, 2011, pp. 3-28. http://dx.doi.org/10.5772/25370.

[6] Farrell, S. and Cavanagh, E. An Introduction to Life Cycle Assessment with Hands-On Experiments for Biodiesel Production and Use. *Education for Chemical Engineers*, 9(3), 2014, pp. e67-e76. https://doi.org/10.1016/j.ece.2014.04.003

[7] Demirbaş, A. Biodegradability of Biodiesel and Petrodiesel Fuels. *Energy Sources, Part A: Recovery, Utilization, and Environmental Effects*, 31(2), 2008, pp. 169-174. https://doi.org/10.1080/15567030701521809

[8] Chandrasekaran, V., et al. The Role of Nano Additives for Biodiesel and Diesel Blended Transportation Fuels. *Transportation Research Part D: Transport and Environment*, 46, 2016, pp. 145-156. https://doi.org/10.1016/j.trd.2016.03.015

[9] Emiroğlu, A.O. and Şen, M. Combustion, Performance and Exhaust Emission Characterizations of a Diesel Engine Operating with a Ternary Blend (Alcohol-Biodiesel-Diesel Fuel). *Applied Thermal Engineering*, 133, 2018, pp. 371-380. https://doi.org/10.1016/j.applthermaleng.2018.01.069

[10] Wood, B.M., et al. Study of Combustion Performance of Biodiesel for Potential Application in Motorsport. *Journal of Cleaner Production*, 93, 2015, pp. 167-173. https://doi.org/10.1016/j.jclepro.2014.12.091

[11] Abed, K.A., et al. Effect of Biodiesel Fuels on Diesel Engine Emissions. *Egyptian Journal of Petroleum*, 28(2), 2019, pp. 183-188. https://doi.org/10.1016/j.ejpe.2019.03.001

[12] Khalid, A., et al. Performance and Emissions Characteristics of Diesel Engine Fuelled by Biodiesel Derived from Palm oil. In: Ismail, A. E. et al., Applied Mechanics and Materials. Zurich: Trans Tech Publ. 2013. https://doi.org/10.4028/www.scientific.net/AMM.315.517

[13] Gad, M.S., et al, Performance and Emissions Characteristics of C.I. Engine Fueled with Palm Oil/Palm Oil Methyl Ester Blended with Diesel Fuel. *Egyptian Journal of Petroleum*, 27(2), 2018, pp. 215-219. https://doi.org/10.1016/j.ejpe.2017.05.009

[14] Deepanraj, B., et al. Use of Palm Oil Biodiesel Blends as a Fuel for Compression Ignition Engine. *American Journal of Applied Sciences*, 8(11), 2011, pp. 1154-1158. https://doi.org/10.3844/ajassp.2011.1154.1158

[15] Senthilkumar, S., Sivakumar, G. and Manoharan, S. Investigation of Palm Methyl-Ester Bio-Diesel with Additive on Performance and Emission Characteristics of a Diesel Engine under 8-Mode Testing Cycle. *Alexandria Engineering Journal*, 54(3), 2015, pp. 423-428. https://doi.org/10.1016/j.aej.2015.03.019

[16] Alptekin, E., et al. Using Waste Animal Fat Based Biodiesels–Bioethanol–Diesel Fuel Blends in a DI Diesel Engine. *Fuel*, 157, 2015, pp. 245-254. https://doi.org/10.1016/j.fuel.2015.04.067

[17] Shahir, V.K., et al. Experimental Investigation on Performance and Emission Characteristics of a Common Rail Direct InjectionEngine Using Animal Fat Biodiesel Blends. *Energy Procedia*, 117, 2017, pp. 283-290. https://doi.org/10.1016/j.egypro.2017.05.133

[18] Giakoumis, E.G., et al. Exhaust Emissions of Diesel Engines Operating under Transient Conditions with Biodiesel Fuel Blends. *Progress in Energy and Combustion Science*, 38(5), 2012, pp. 691-715. https://doi.org/10.1016/j.pecs.2012.05.002

[19] Li, B., et al. Combustion and Emission Characteristics of Diesel Engine Fueled with Biodiesel/PODE Blends. *Applied Energy*, 206, 2017, p. 425-431. https://doi.org/10.1016/j.apenergy.2017.08.206

[20] Demirbas, A. Biodiesel from Triglycerides via Transesterification. In: Biodiesel: A Realistic Fuel Alternative for Diesel Engines. London: Springer, 2008, pp. 121-140. https://doi.org/10.1007/978-1-84628-995-8_5

[21] Dwivedi, G. and Sharma, M. Impact of Cold Flow Properties of Biodiesel on Engine Performance. *Renewable and Sustainable Energy Reviews*, 31, 2014, pp. 650-656. https://doi.org/10.1016/j.rser.2013.12.035

[22] Shrivastava, P., Verma, T.N. and Pugazhendhi, A. An Experimental Evaluation of Engine Performance and Emission Characteristics of CI engine operated with Roselle and Karanja Biodiesel. *Fuel*, 25, 2019, p. 115652. https://doi.org/10.1016/j.fuel.2019.115652

[23] Qi, D., et al. Experimental Studies on the Combustion Characteristics and Performance of a Direct Injection Engine Fueled with Biodiesel/Diesel Blends. *Energy Conversion and Management*, 51(12), 2010 pp. 2985-2992. https://doi.org/10.1016/j.enconman.2010.06.042

[24] Rajak, U., et al. Numerical Investigation of Performance, Combustion and Emission Characteristics of Various Biofuels. *Energy Conversion and Management*, 156, 2018 pp. 235-252. https://doi.org/10.1016/j.enconman.2017.11.017

[25] Rajak, U., Nashine, P. and Verma, T.N. Assessment of Diesel Engine Performance Using *Spirulina* Microalgae Biodiesel. *Energy*, 166, 2019, pp. 1025-1036. https://doi.org/10.1016/j.energy.2018.10.098

CHAPTER 6: CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusion

The aims and objectives of this study were to produce WAF biodiesel by the acid-base transesterification method and an experiment regarding its performance and emission characteristics was successfully explored. The goal was to productively utilize a low-cost feedstock that does not compete with edible oil consumed by humans and provide sustainable energy for developing countries. The objectives were two step acid-base transesterification, characterization of physical and chemical properties, and test for suitability in an engine application. The experimental results were successfully achieved as reflected in peer-reviewed journal publications documented in this thesis.

Chapter 2 was a comprehensive review of low cost WAF biodiesel pertaining to various production methods, properties and published work on WAF usage as fuel. This chapter emphasized diversification to newer feed-stock sources to prevent food insecurity especially in developing countries. WAF biodiesel has properties similar to fossil diesel.

Chapter 3 was a review on the application of enzymes during transesterification of animal fats. Animal fats have a high level of free fatty acids that react with chemical catalysts causing saponification and reduction of biodiesel yield. Enzyme catalysts have gained wide acceptability to lower the overall cost of biodiesel. However, the development of lipase catalysts is still hampered by the lack of available technology for production of enzymes. Whole cell catalysts have been shown to be more economical.

Chapter 4 covers experimental work conducted on the acid-base transesterification method during conversion of WAF feedstock to biodiesel. The optimum parameters which resulted in high biodiesel yield were 1.5 w/w % KOH catalyst, 6:1 methanol to fat molar ratio and reaction time 90 min. Both physical and chemical properties characterization showed WAF biodiesel complied with ASTM D 6751 standard requirements.

Chapter 5 described the experiment conducted to test the suitability of WAF biodiesel as applied in a compression ignition engine. The performance parameters indicate that brake specific fuel consumption is higher for biodiesel than diesel fuel. Brake thermal efficiency for WAF biodiesel were lower than diesel fuel at tested loads. Emissions of CO, HC and NO_X

show that WAF biodiesel is a cleaner fuel in the environment due to lower values of CO and HC except for the higher oxides of nitrogen compared to diesel.

This study has shown that waste from abattoirs can be converted to biodiesel to service energy demand and reduce the polluting emissions to the environment caused by burning fossil diesel. Furthermore, transesterification is a less costly production process so can be undertaken at small scale in developing countries.

6.2 Recommendations

On the basis of the challenges encountered while conducting this research, the following recommendations can be made for improvement:

1) The study showed WAF biodiesel has poor cold flow properties which discourages its commercialization in the industry. WAF is solid at room temperature and tends to gel when transesterification is carried out at low rpm agitator speed. This property limitation was found to arise from the long chain saturated fatty acid esters present in biodiesel. Cold flow properties impact negatively on fuel combustion which may cause engine knocking in the combustion chamber. Cold flow improvers like olefin-ester copolymer and Octa-1-maleic anhydride copolymer can be researched to solve this problem.
2) NO_X emission for biodiesel can be reduced by creation of a low temperature zone using exhaust gas recirculation such as used for tractors. In addition, hybridization of biodiesel to reduce ignition delay is proposed for use in direct ignition engines. The formation of NO_X in engines should be simulated using software like ANSYS as shown in various published work in order to further understand the factors involved and search for solutions.

APPENDIX: EQUIPMENT USED

Figure 1 Fume hood chamber

Figure 2 Separator funnel and other chemicals used

Figure 3 Preparation of sample

a b

Figure 5(a)Simultaneous Thermal Analyser-STA6000

(b) STA 6000 top view

Figure 4 Spectrum 100 Instrument