Process grease: A possible feedstock for biodiesel production

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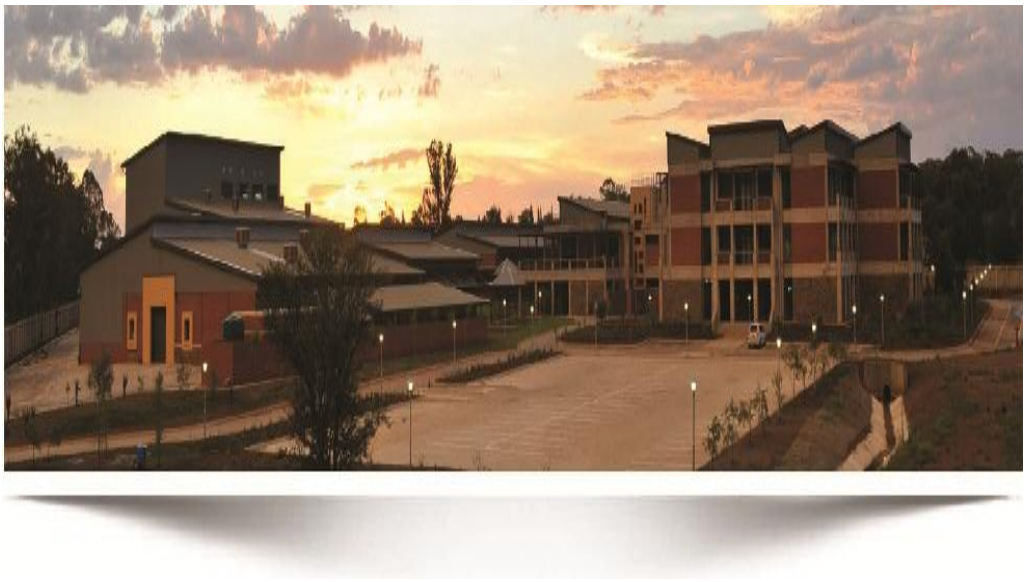

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Abstract

The utilisation of waste process grease (WPG) as feedstock for biodiesel production was investigated in this study. WPG is a lubrication oil used in the metalworking industry and is considered a hazardous waste material. WPG contains vegetable oil and animal fat which are used as base oils in the lubricant formulation.

Three different production routes were followed to produce biodiesel using WPG as feedstock. The first production route involved the conventional two-step production process comprising the acid esterification of the free fatty acids, followed by alkaline transesterification. The second production route involved the extraction of free fatty acids in the WPG by means of liquid-liquid extraction and the production of biodiesel from the extracted free fatty acids through acid esterification. The produced biodiesel was purified by means of chromatography. A third process route was the saponification of the WPG using aqueous sodium hydroxide followed by acidulation with hydrochloric acid. The resulting acid oil was purified by means of column chromatography, using a hydrophobic resin as the stationary phase prior to esterification through acid catalysis to produce biodiesel. The crude biodiesel was purified using column chromatography with silica gel as stationary phase.

The optimum reaction conditions for the reduction of the free fatty acid content of WPG in route 1 to 0.5% were a methanol to oil ratio of 8:1 and a reaction temperature of 65 °C with a catalyst loading of 4 wt%. Acetonitrile was found to be the most effective extraction solvent for the reduction of sulphur compounds in the free fatty acid feedstock in route 2. A reverse phase chromatographic system with a hydrophobic stationary phase and methanol as the mobile phase was found to be an effective system to reduce the sulphur to below 10 ppm as specified by the SANS 1935 biodiesel standard in route 3.

Both the conventional two-step process (route 1) and the liquid-liquid extraction process (route 2) were found not to be suitable for the production of biodiesel from WPG as the sulphur content of the produced biodiesel for routes 1 and 2 was 8 141 ppm and 4 888 ppm, respectively. The sulphur content of the produced biodiesel following route 3 was 9 ppm. The latter approach reduced the sulphur

content of the biodiesel to acceptable levels that conform to the SANS 1935 standard to be used in a B10 biodiesel blend. A biodiesel yield of 45%, calculated as the mass of biodiesel produced as a percentage of the total mass of dried WPG used, was achieved with route 3. The biodiesel conformed to most of the specifications in the SANS1935 standard for biodiesel. The presence of a relatively high concentration of saturated fatty acids reflected in the higher cetane number of 74.7, the high cold filter plugging point of +10 and the oxidative stability of > 6 hours. A comparative cost analysis for route 3 indicated that the production cost of biodiesel, compared to the cost of petroleum diesel is marginally higher at the current Brent crude oil price of $102.41 per barrel. The production of biodiesel from WPG will be economically viable once the crude oil price has risen to about $113 per barrel.

Key words: Process grease, biodiesel feedstock, feedstock purification, conventional two-step process, chromatography, sulphur content.

Uittreksel

Die aanwending van afval-prosesghries as voerstof vir die produksie van biodiesel is geëvalueer in die studie. Prosesghries is 'n smeerolie wat in die metaalvervormingsindustrie gebruik word en die gebruikte ghries word as gevaarlike afval geklassifiseer. Afval-prosesghries bevat plantolies en diervette wat as basis-olies in smeerolie-formulering aangewend word.

Drie verskillende produksieroetes vir die produksie van biodiesel uit afval-prosesghries is geëvalueer. Die eerste roete het die konvensionele biodiesel produksiemetode, wat bestaan uit suuresterifikasie van die vry vetsure gevolg deur alkaliese transesterifikasie, behels. Die tweede produksieroete het die ekstraksie van die vry vetsure uit die afval-prosesghries met behulp van vloeistof-vloeistof-ekstraksie en die produksie van biodiesel deur middel van suuresterifikasie met die vry vetsure as voerstof, behels. Chromatografie is gebruik om die geproduseerde biodiesel te suiwer. Die derde roete het die saponifikasie van die afval-prosesghries deur middel van waterige natriumhidroksied, gevolg deur asidulasie met soutsuur, behels. Die gevormde suurolie is gesuiwer deur middel van kolomchromatografie met 'n hidrofobe hars, waarna biodiesel deur suuresterifikasie geproduseer is. Die ru-biodiesel is vervolgens gesuiwer deur middel van kolomchromatografie met silikajel as stasionêre fase.

Die optimale reaksiekondisies vir die verlaging van die vry-vetsuurinhoud in afval-prosesghries tot 0.5% vir roete 1 was by 'n metanol tot ghries molare verhouding van 8 tot 1, 'n reaksietemperatuur van 65 °C en 'n katalislading van 4 massa%. Met roete 2 is gevind dat asetonitriel die effektiefste ekstraheermiddel was vir die vermindering van swaelverbindings tydens die ekstraksie van vry vetsure uit afval-prosesghries. 'n Omgekeerde fase chromatografie-sisteem met 'n hidrofobe stasionêre fase en metanol as mobiele fase was geskik om met roete 3 die swaelinhoud te verminder tot onder 10 dele per mljoen, soos gespesifiseer deur die SANS 1935-biodieselstandaard.

Beide die konvensionele 2-stapproses (roete 1) en die vloeistof-vloeistof-ekstraksieproses (roete2) was nie geskik vir die produksie van biodiesel uit afval-prosesghries nie, aangesien die swaelinhoud in die biodiesel 8 114 en 4 888 dele

per miljoen, respektiewelik, vir roetes 1 en 2 was. Die swaelinhoud van die biodiesel geproduseer volgens roete 3, was 9 dele per miljoen wat tot gevolg het dat die biodiesel voldoen aan die spesifikasie vir 'n B10-biodiesel-mengsel. ‘n Biodiesel opbrengs van 45%, bereken as massa biodiesel geproduseer uitgedruk as persentasie van die totale massa gedroogte afval-prosesghries gebruik, was behaal met roete 3. Die biodiesel het voldoen aan meeste van die spesifikasies vir biodiesel soos uiteengesit in die SANS 1935 biodiesel standaard. Die teenwoordigheid van ‘n relatiewe hoë konsentrasie versadigde vetsure in die voerstof word gereflekteer in die hoër setaangetal van 74.4, die hoër kouefilter-stolpunt van +10 en die hoër oksidatiewe stabiliteit van > 6 ure. 'n Vergelykende koste-analise het getoon dat die produksiekoste van biodiesel volgens roete 3 marginaal hoër is as die prys vir petroleumdiesel. Die produksie van biodiesel volgens roete 3 sal ekonomies lewensvatbaar wees teen 'n Brent-ru-olieprys van $113 per vat en hoër.

Sleutelwoorde: Prosesghries, biodiesel-voerstof, voerstofsuiwering, konvesionele twee-stapproses, chromatografie, swaelinhoud.

Declaration

I, Roelof Jacobus Venter, hereby declare that I am the sole author of this thesis entitled:

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Nomenclature

Symbol	Description
M_{fa}	Mass of fatty acids
M_{wpg}	Mass of waste process grease
$\bar{x}$	Average
x_i	Data point
δ	Standard deviation
N	Number of data points
ET(30)	Solvent polarity
m_i	Mass of ester
$m_{c9:0}$	Mass of internal standard
k_{ci}	Calibration constant
A_i	Peak area of ester
$A_{c9:0}$	Peak area of interrnal standard
cP	Centipoise

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Chapter 1. - Introduction

An overview of the contents of the study is provided in this chapter. The background and motivation are discussed in section 1.1, section 1.2 contains the problem statement, section 1.3 lists the aim and objectives and section 1.4 provides the scope of the investigation.

1.1 Background and motivation

A fast-growing human population striving for improved living conditions is the driving force behind an ever-increasing pace of industrialisation. Figure 1.1 shows that the global population is estimated at 8.9 billion in 2050, growing from about 7 billion in 2013.

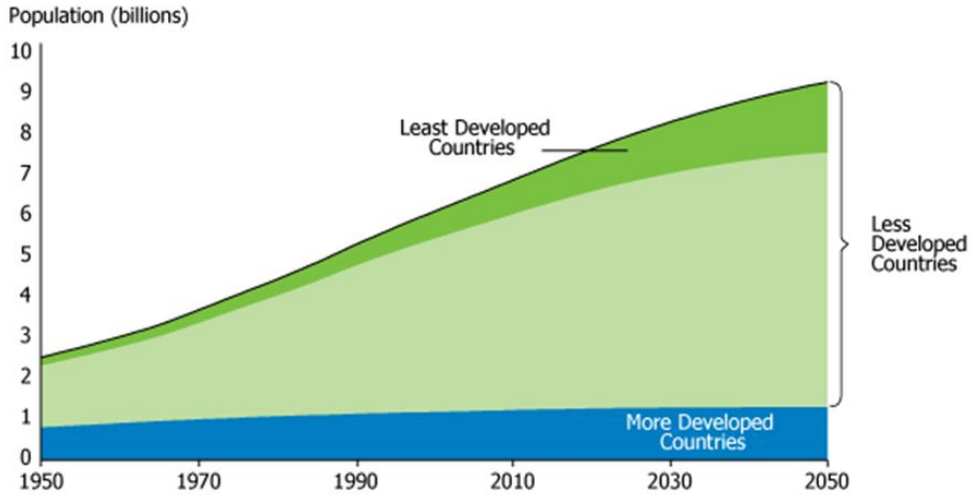

Figure 1.1: Word population (United Nations, 2011)

Industrialisation inspired the development of new technologies which delivered technical progress which is considered to be the main driving force behind economic growth and rising living standards.

Industrialisation and economic growth across the globe also causes a rapid increase in waste generation. More than 2500 million tons of waste is generated in Europe each year and in many countries the generation of waste is linked to economic growth (Eurostat, 2012). Waste creates an environmental burden as it contributes to the degradation of air quality, soil and water. The incineration of waste as well as landfill sites contributes to greenhouse gases through the generation of carbon dioxide and methane, respectively. Waste represents a loss of energy and materials and an economic burden is created by the collection, treatment and disposal thereof. Waste management systems need to adapt with the growing volumes of waste to ensure that no harm is done to the environment. The growth in consumption for renewable energy is slower, compared to the total energy consumption which is a focus area for various governments to increase the utilisation of renewable energy.

Figure 1.2.shows the increase in energy consumption between 1990 and 2011 with the estimated increase until 2030 (BP Energy Outlook, 2013).

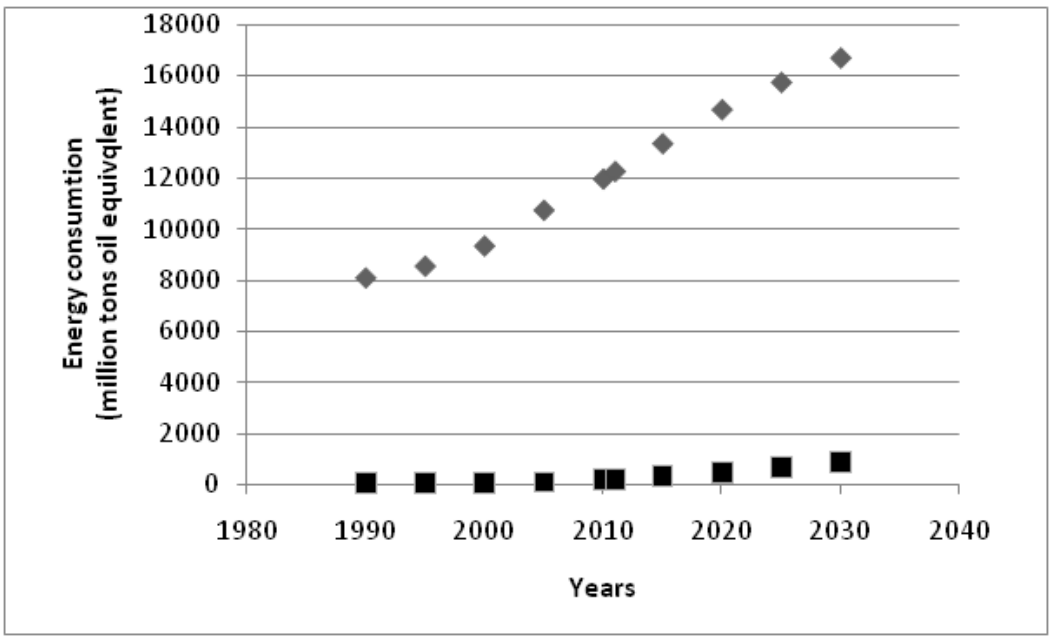

Figure 1.2: Total and renewable energy consumption (BP Energy Outlook, 2013) (♦) Total energy consumption (■) Renewable energy consumption

Most of the world's energy supply is from fossil origin, which is non-renewable and the world is presently confronted with the depletion of fossil fuel resources.

Fossil fuel resources are also confined to certain countries in the world, which results in ever increasing resource prices as it becomes more and more expensive to recover the resources from deposits while demand is also increasing exponentially. The availability of fuel is of strategic importance to those countries without natural oil resources. In addition, the combustion of petroleum-based fuels in engines such as diesel engines results in the formation of pollutants such as carbon monoxide, carbon dioxide, sulphur dioxides, nitrogen oxides and particulate matter (Oner & Altun, 2009:2114). The ever-increasing demand for energy and the problems resulting from the burning of fossil fuel encourage researchers to find alternative energy sources. Research on biomass-based fuels such as biodiesel and ethanol has increased exponentially in the last decade. Biodiesel has several advantages compared to petroleum diesel, such as biodegradability, it is a cleaner burning fuel due to the presence of oxygen in the ester molecule, it is renewable, contains low or no sulphur, has a 90% reduction in cancer risks due to the lower reactive hydrocarbon species and emissions of poly aromatic hydrocarbons, and it is a non-toxic fuel (Murugesan *et al*., 2009:658).

The utilisation of waste as energy source offers opportunities to address various problems faced by a fast-growing human population. This concept of waste- to-energy has the potential to be an effective waste management option, as well as a source of energy (Jamasb & Nepal, 2010:1352). The biomass content of the waste can contribute to renewable energy resulting in less waste being sent to landfill sites. The utilisation of waste oils and fats as feedstock for biodiesel production supports the effort of generating energy from waste. Most waste oils and fats contain impurities which makes it unfit for human or animal consumption, which poses a threat to the environment in the event of improper disposal or use. The utilisation of waste oils as feedstock for biodiesel production is therefore not implicated in the food versus fuel debate.

The growing shortage of feedstock for biodiesel production drives the search for alternative feedstock sources. Examples of bio-oil sources from waste which require further research to result in viable renewable energy options include oil from municipal sewage sludge (Siddiquee *et al*., 2011:1067) bio-oil produced by

microorganisms such as yeast feeding on rotten fruit or food waste (Sankh *et al.*, 2013:1), and oil from insects feeding on farm waste (Li, 2011: 1545).

Eurostat (2013) reports that waste generated from economic activities and households in the European Union amounts to 2 569 850 thousand tons in 2010 of which 94 460 thousand tons or 3.68% is hazardous. Waste from industrial processes is often hazardous waste that could have a detrimental effect on the environment if not handled properly. One such waste material is lubricants. In all sectors of industry, lubricants are being utilised to lubricate machines and materials, and continuing industrialisation worldwide results in a growing demand for lubricants. Petroleum-based oil represents approximately 85% of lubricants used all over the world (Shashidhara & Jayaram, 2010:1073). Lubricants could have a negative effect on the environment in the event of inappropriate use and disposal. Al-Omari (2008:3648) states that used lubrication oil is a significant energy source and supplementary fuel for furnaces. By co-firing even small quantities of this oil with gaseous fuels such as LP gas can result in a significant enhancement of the radiation from the gaseous fuel. However, Al-Omari (2008: 3648) emphasises the concern about the impact of lubricating oil combustion on the environment. Lubricating oil could contain unwanted additives such as sulphur and phosphorous which will end up in the environment when combusted.

Vegetable oils and animal fats are promising alternatives to mineral-based oils in lubricant formulations. Environmental acceptability of lubricants has become increasingly important as lubricants are used in many diverse applications (Lawal *et al*., 2012:2). Significant advantages from an environmental point of view with respect to resource renewability, biodegradability and adequate performance in a variety of applications are offered by vegetable oils and animal fats (Gawrilow, 2003:3). Advantages of vegetable oils as lubricants are high biodegradability, low pollution of the environment, low volatility, compatibility with lubricants, low production cost, high viscosity indices, high flashpoints and low toxicity (Shashidara & Jayaram, 2010:1076). However, vegetable oils lack certain key oil properties or characteristics needed to withstand harsh conditions when used in lubrication applications. Oxidative stability, hydrolytic stability and low temperature properties, thermal stability and corrosion protection are key oil properties in lubrication applications.

The shortcomings of vegetable oils used in lubrication applications could be addressed by the modification of these oils using additives (Erhan & Asadauskas, 2000:278; Gawrilow, 2003:6). Base fluids which usually comprise more than 80% of the lubricant are enhanced by additives such as antioxidants, detergents, dispersants, viscosity modifiers, pour point depressants, anti-wear agents, rust and corrosion inhibitors, demulsifiers, foam inhibitors, thickeners, friction modifiers, dyes and biocides. The lubrication performance of a vegetable oil-based lubricant is also influenced by the fatty acid composition of the base oil, specifically by the ratio and position of the carbon- to- carbon double bonds. Physical properties such as oxidative stability, low temperature properties and hydrolytic stability are influenced by the presence of saturated fatty acids and mono- and poly-unsaturated fatty acids. The markets that hold potential for vegetable-based lubricating oils are two-cycle engine base oils, anti-wear hydraulic fluids, chain bar lubricants, gear oils, metalworking fluids, food machinery lubricants, textile lubricants and grease-base fluids (Gawrilow, 2003:14).

Waste process grease was identified as a potential feedstock for biodiesel production that had not been evaluated previously. Waste process grease is defined as a spent metalworking fluid generated by the metal rolling industry. Rolling is a metal forming process where metal is passed through rolls. The lubricating grease or oil facilitates the feed of the metal between the work roles. Process grease contains base oils consisting of vegetable oil or animal fat modified by the addition of various additives. These additives are supplied by lubrication specialists and the nature and composition of the additive packages are proprietary information.

The utilisation of waste process grease as feedstock for biodiesel production appears to be promising, as this will result in the transformation of a hazardous waste material into energy, reducing the risk of the contamination of the environment, either by releasing it into the soil or water, or by releasing it into the atmosphere by using it as a furnace fuel. When biodiesel is produced from the WPG, there will also be some contribution to the shortage of feedstock for biodiesel production.

A metalworking plant in Gauteng South Africa has been identified as a source of WPG for biodiesel production. Most of the waste process grease generated in

South Africa is generated by four metalworking plants. The addressable market size for metalworking fluids in the USA was estimated at 540 thousand metric tons per annum in 2003 (Gawrilow, 2003:16). The annual use of rolling oil in South Africa using vegetable oil or animal fat as base oil, was estimated at 2 400 metric tons in 2011 (Lubrisol, 2011).

The outcome of this study will be applicable to waste process grease from the metalworking industry as other vegetable-based lubricants have not been evaluated. WPG from the metalworking industry was chosen, based on its availability in larger quantities which makes collection easy as apposed to the generation of smaller quantities such as gear oils which requires a major collection effort.

1.2 Problem statement

Waste process grease is generated by a metalworking facility in the Gauteng province, South Africa. WPG is regarded as a hazardous waste as it contains impurities which are harmful to the environment when released into the soil, water or atmosphere. The disposal cost of WPG is at least USD 500 per ton which makes it attractive to blend the WPG with other waste oils and use it as a low-value furnace fuel. The burning of WPG will result in unwanted impurities released into the atmosphere.

WPG has not yet been evaluated as feedstock for biodiesel production. Existing production methods utilising WPG as feedstock have not been evaluated yet to determine whether the impurities present in the WPG will have a negative effect on the production process and on the quality of biodiesel produced. A production method utilising WPG as feedstock addressing the unusual impurities present in WPG, resulting in biodiesel conforming to the SANS 1935 biodiesel standard has not been developed.

1.3 Aims and objectives

The aim of this study is to evaluate different methods for the production of biodiesel from waste process grease and to determine the suitability of WPG to be used as feedstock for biodiesel production.

Objectives:

1 The optimisation of the pre-treatment reaction parameters (reaction temperature, methanol to oil ratio, catalyst loading) for conventional alkali-catalysed transesterification process as production method to produce biodiesel from waste process grease.
2 The evaluation of the conventional alkali-catalysed process to produce biodiesel that conforms to the SANS 1935 biodiesel standard with specific reference to sulphur.
3 Determine the effect of different feedstock pre-treatment options on the reduction of sulphur in the biodiesel produced and the comparison of the biodiesel with the SANS 1935 specification.
4 The development of a modified biodiesel production process to produce biodiesel that conforms to the SANS 1935 biodiesel standard.

1.4 Scope of the investigation

To fulfil the aims and objectives set out in section 1.3, the following is required:

- Chapter 2 - Literature study
    - A literature study on biodiesel feedstock evaluating the current feedstock situation and the research done on alternative feedstock options.
    - A study on the conventional biodiesel production technologies used and the work done on feedstock preparation and pre-treatment and technology innovations to reduce production costs to ensure biodiesel quality that conforms to the standards set for biodiesel.
    - A study on biodiesel quality, the parameters measured to ensure biodiesel quality and the factors affecting the biodiesel quality.

- Chapter 3 – Experimental
    - A brief description of the origin of the feedstock used, the characteristics of the feedstock and the materials used for the experiments.
    - The experimental set-up for the optimisation of the reaction parameters for feedstock pre-treatment for the conventional alkaline transesterification process.
    - The experimental set-up for the extraction and esterification of the free fatty acids from the WPG feedstock.
    - The experimental set-up for the modification, pre-treatment and esterification of the WPG feedstock.
    - The analytical methods used.

- Chapter 4 – Results and Discussion
    - Results from the effect of the different reaction conditions on the reduction of free fatty acids in the WPG feedstock.
    - Results from the effect of different solvent combinations on the extraction of FFA from the WPG feedstock and the selectivity of the solvent combinations on the selectivity towards sulphur extraction.
    - Results from the different chromatographic parameters on the extraction of sulphur compounds from the modified WPG feedstock.
    - Comparison of the biodiesel produced via the three different routes with the biodiesel standard.
    - Comparison of biodiesel production cost.

- Chapter 5 - Conclusions and Recommendations

1.5 References

Al-Omari, S. B. 2008. Used engine lubrication oil as a renewable supplementary fuel for furnaces. Energy Conversion and Management, 49: 3648- 3653.

BP. 2013. BP Energy outlook 2030: 2013. Energy consumption by fuel. http://www.bp.com/liveassets/bp_internet/globalbp/globalbp_uk_english/reports_and_publications/statistical_energy_review_2011/STAGING/local_assets/sreadsheets/energy-outlook_summary_tables_2013.xlsx-'Consumption Date of access: Feb. 2013.

Erhan, S. Z., Asadauskas, S. 2000. Lubricant basestock from vegetable oils. Industrial Crops and Products, 11: 277- 282.

Eurostat. 2012. File: Waste generation, 2010 (1000tonnes). Png. Statistics Explained. http://epp.eurostat.ec.europa.eu/statistics_explained/index.php?title=File:Waste_generation,_2010_(1_000_tonnes).png&filetimestamp=20121022151900 Date of access: 24 Apr. 2013.

Eurostat. 2013. Waste statistics. Generation of waste by economic activity. Tonnes. All NACE plus households. Total amount of waste generated by households and businesses by economic activity. http://epp.eurostat.ec.europa.eu/tgm/table.do?tab=table&init=1&language=en&pcode=ten00106&plugin=1 Date of access: 24 Apr. 2013.

Gawrilow, L. 2003. Palm oil usage in lubricants. 3rd Global Oil and Fats business Forum USA "Interfacing with the Global Oils and Fats Business". http://americanpalmoil.com/pdf/Ilija%20Gawrilow.pdf.(2003) date of access: January 2012.

Jamasb, T., Nepal, R. 2010. Issues and options in waste management: A social cost-benefit analysis of waste-to-energy in the UK. Resources, Conservation and Recycling, 54: 1314 - 1352.

Lawal, S. A., Choudhury, I. A., Nukman, Y. 2012. Application of vegetable oil based metal working fluids in machining ferrous metals – A review. International Journal of Machine Tools & Manufacturing, 52: 1 - 12.

Li, Q., Zheng, L., Cai, H., Garza, E., Yu, Z., Zhou, S. 2011. From organic waste to biodiesel: Black soldier fly, Hermetia illucens, makes it feasible. Fuel, 90: 1545- 1548.

Lubrison. 2011. Sales representative, Lubrisol South Africa, estimation February 2011).

Murugesan, A., Umarani, C., Subramanian, R., Nedunchezhian, N. 2009. Bio-diesel as an alternative fuel for diesel engines – A review. Renewable and Sustainable Energy Reviews, 13:653- 662.

Oner, C., Altun, S. 2009. Biodiesel production from inedible animal tallow and experimental investigation of its use as alternative fuel in a direct injection diesel engine. Applied Energy, 86: 2114- 2120.

Sankh, S., Thiru, M., Saran, S., Rangaswamy, V. 2013. Biodiesel production from newly isolated Pichia kudtiavzevii strain. Fuel, Article in press.

Shashidhara, Y. M., Jayaram, S. R. 2010. Vegetable oils as a potential cutting fluid- an evolution. Tribology International, 43: 1073- 1081.

Siddiquee, M. N., Rohani, S. 2011. Lipid extraction and biodiesel production from sewage sludges: A review. Renewable and sustainable Energy reviews, 15: 1067- 1072.

United Nations. Department of Economic and Social Affairs. Population Division. 2011. Total population (both sexes combined) by major area, region and country, annually for 1950- 2100 (thousands). http://esa.un.org/unpd/wpp/Excel-Data/DB02_Stock_Indicators/WPP2010_DB2_F01_TOTAL_POPULATION_BOTH_SEXES.XLS Date of access: 24 Apr. 2013

Chapter 2. - Literature Study

Aspects with regard to biodiesel feedstock, biodiesel production technology and biodiesel quality are discussed in this chapter. The current biodiesel feedstock situation and alternative feedstock for biodiesel production is evaluated in section 2.1. In section 2.2 feedstock pre-treatment, transesterification processes and biodiesel refining is discussed and quality assurance with regard to biodiesel and the factors affecting the quality of biodiesel is discussed in section 2.3.

2.1 Biodiesel feedstock

ASTM defines biodiesel as a fuel comprising mono-alkyl esters of long-chain fatty acids derived from vegetable oils or animal fats (ASTM, 2012). These oils and fats contain mainly triacylglycerides or triglycerides and free fatty acids. The reaction of triglycerides with an alcohol such as methanol in the presence of a catalyst is known as transesterification and results in the formation of the mono-alkyl esters. Any material containing fatty acids, whether they are linked to other molecules such as glycerol or present as free fatty acids can be used as biodiesel feedstock. Biodiesel is similar to conventional diesel in terms of its main characteristics and is therefore compatible with it and can be blended in any ratio with petroleum diesel.

2.1.1 Current biodiesel feedstock situation

More than 95% of the biodiesel produced in the world at present uses edible oil as feedstock (Borugadda *et al*., 2012:4764). The typical feedstock sources are rapeseed oil, canola oil (a specific cultivar of rapeseed), soybean oil, sunflower oil and palm oil. Animal oils and fats such as beef tallow, sheep tallow and poultry oil as well as waste oils such as used cooking oil are also used as feedstock for biodiesel production. Other sources include jatropha oil, coconut oil, fish oil, karanja oil, groundnut oil, rice bran oil, cotton oil, sesame oil, sorghum and wheat.

Different regions worldwide are focusing their efforts on different oils depending on climate, availability, local soil conditions and agricultural practices. Lin and co-workers (2011:1023) indicate the major raw material source for biodiesel production in the different regions in the world. Soybean oil is a major biodiesel feedstock in the USA. In Europe rapeseed oil is the major source with sunflower oil in countries such as Spain. In tropical countries such as Malaysia, India, Thailand as well as Brazil, palm oil is an important source. Waste oil as feedstock plays an important role in Japan, Korea, China, Australia, New Zealand, Mexico, as well as in the USA and UK. Animal fat is an important source in Australia, New Zealand and Mexico.

Edible vegetable oil is available from the agricultural industry in large quantities. Approximately 157 million metric tons of vegetable oil will be produced for the 2012/2013 year of which approximately 70% is accounted for soybean oil (26%), palm oil (18%), rape seed oil (12%) and sunflower oil (13%) (USDA, 2012; Rosillo-Calle, 2009:8).

The consumption of vegetable oil falls mainly into three categories, namely food use, industrial use and biofuel use (Rosillo-Calle *et al*., 2009:3). Vegetable oil is predominantly used in the food industry which makes up over 80% of the market. The major growth in the vegetable oil market is driven by growth in the demand for food rather than biofuels (Rosillo-Calle *et al*., 2009:10). As the demand for vegetable oil from a fuel point of view is also expanding rapidly, given increasing environmental concerns, fears exist for future competition between the food and the fuel markets which could lead to starvation in some developing countries.

The production of biodiesel from food-grade oils, which is known as first generation biodiesel, enjoys the benefits of a high purity feedstock which is easily available from the agricultural industry on a large scale and makes the production process less complicated compared to processes where low quality feedstock is used. The majority of biodiesel plants operated today are using conventional acid or alkali catalysed transesterification. Alkaline catalysis is more often used than acid catalysis as the process is much faster and alkali catalysts such as sodium hydroxide, sodium methoxide and potassium hydroxide are less corrosive compared to sulphuric acid (Oh *et al.,* 2012:5134).

The total production of biodiesel for 2007/09 for the main producers was 13.3 million tons per annum and the forecast for 2019 is 36 million tons (Gunstone, 2013:3). The countries considered as main producers are the USA, EU countries such as Germany, France, Italy, Spain and the UK, and countries like Argentina, Brazil, India, Thailand, Malaysia and Indonesia.

Continuing production of biodiesel from first generation feedstock and the further commercialisation of biodiesel face serious drawbacks in terms of the production cost of the biodiesel, availability of feedstock and the food versus fuel debate. The contribution of feedstock cost is more than 75% of the overall biodiesel production cost (Ahmad *et al*., 2011:585).

Using edible oils as feedstock makes biodiesel too expensive compared to petroleum diesel and the situation is getting worse, given the rise in edible oil prices. The price for sunflower oil increased from just above 500 USD per ton in 2001 to USD 1 1254 per ton in 2012 (Gunstone, 2013:1). Soybean oil increased from approximately USD 400 per ton in 2001 to USD 1 241 in 2012. A growing population expected to increase to 9 – 10 billion by 2050 with a growth in urbanisation and wealth pushing up the demand for vegetable oil by 4 to 5 million tons more per annum. (Gunstone, 2013:2). Not all biodiesel feedstock comes from edible oils. This increasing demand for oil together with the increasing cost of agricultural production, as well as unfavourable climate conditions such as recent droughts in many countries will put pressure on the oil prices for the foreseeable future. Therefore, rising vegetable oil prices will result in biodiesel to become less competitive with petroleum diesel in future, affecting the further commercialisation of biodiesel negatively.

Feedstock availability for the production of biodiesel should be placed in perspective, given the energy demand in the world compared to the total production of vegetable oils and animal fats. If all soybean production in the USA is dedicated to biodiesel production, only 6% of the diesel demand would be met (Chapagain *et al*., 2009:1222). This perspective suggests that biodiesel should form a small part of the energy supply in future, making a contribution in replacing fossil fuel in the long term. Although the contribution of biodiesel could be relatively small, it is considered a very important one given its environmental benefits.

The debate on food versus fuel, as well as the environmental concerns with regard to the use of edible oil as biodiesel feedstock has a negative effect on feedstock availability. Extensive use of edible oils may cause significant problems such as starvation in developing countries with almost 60% of humans in the world malnourished (Balat 2011:1480). As the demand for palm oil grows, oil-palm plantations are expanding, posing a major threat to natural forests and biodiversity of the ecosystem (Lim *et al*., 2010:942).

The biodiesel industry will experience increasing pressure in future to move away from using edible oils as feedstock. With the objective in mind to increase the role of biodiesel in the energy supply of the future given its benefits, scientists are focusing on the development of new generation biofuels attempting to reduce the concerns for first generation biofuels. This resulted in the development of second generation and third generation biodiesel.

2.1.2 Alternative feedstock for biodiesel production

Alternative options to edible oils as feedstock are non-edible oils and waste oils. These oils are not part of the food versus fuel debate, they contains toxic substances that are not suitable to be used in food. The lower demand for non-edible oils compared to edible oils is the reason for the lower value of these oils.

2.1.2.1 Non-edible oil and fat

Non-edible oil crops as feedstock for biodiesel production have been investigated and discussed extensively the last couple of years (Bankovic-Ilic *et al*., 2012:3622, Wang *et al*., 2011: 1194). Non-edible oils include oils from non-edible oil plants, waste cooking oils, restaurant grease, animal fats such as pork lard and beef tallow, oils from insect larvae, oil from lignocellulosic biomass and microalgae.

Jatropha, karanja, mahua and castor oils are non-edible oils, most often used as feedstock for biodiesel (Bankovic-Ilic *et al*., 2012:3623). Other non-edible crops

also used as feedstock sources which have potential to be feedstock sources are rubber seed, sea mango, neem, kusum, jojoba, tobacco, rice bran, linseed, soapnut, cotton, moringa, tall oil, coffee ground, butter tree, polanga, lucky bean tree, syringa, yellow pleander, kokum, Mexican prickly poppy, nahor, simaroba and tumba (Gui *et al*., 2008:1647; Demibas, 2009:15; Karmakar *et al*., 2010:7205; Kumar & Sharma, 2011:1793, Balat & Balat, 2010:1820, Leung *et al*., 2010:1084, Borugadda & Goud, 2012:4765).

The use of non-edible oils for the production of fuel could be a way to improve the economy of biodiesel on a large scale. Non-edible oils are potentially cheaper than edible oils, because these oils are not suitable for human consumption and therefore the demand for them is lower. Jatropha is considered the most promising feedstock for biodiesel all over the world. The seeds contain 30 – 35% oil. Jatropha oil contains toxins such as phorbol ester, trypsin inhibitors lectins and phytates and is therefore not suitable for human consumption (Koh & Ghazi, 2011:2242, Borugadda & Goud, 2012:4767).

Non-edible oil plants can more easily be cultivated on barren land not suitable for edible oils, resulting in lower production cost and lesser competition with edible oil plants for agricultural land. A disadvantage of non-edible oils as biodiesel feedstock is that many contain high free fatty acids which require additional pre-treatment in the biodiesel production process. The process of oil collection, expelling and storage conditions result in high concentrations of free fatty acids in the oil. Naik and co-workers (2008:354) produced biodiesel from high free fatty acid Karanja oil using a two-step acid-alkali-catalysed process as the traditional alkali-catalysed process was not suitable, due to unwanted soap formation during the reaction. A biodiesel yield of 96.6 – 97% was achieved.

The production of biodiesel from lipids produced by insects has been described by several researchers. Manzano-Agugliaro and co-workers (2012:3746) reviewed the utilisation of fats from insects as feedstock for biodiesel production and found that the fat content of insects varied widely between orders, species and stages of development.

Zheng and co-workers (2013:620) evaluated the grease from yellow mealworm beetle as a biodiesel feedstock where decayed vegetables were used as feedstock.

The biodiesel yield obtained was 34.2 g from 234.8 g dried larval biomass. The biodiesel consisted of linolenic acid methyl esters (19.7%), palmitic acid methyl esters (17.6%), linoleic acid methyl esters (16.3%, and stearic acid methyl esters (11.4%). Most of the properties of the biodiesel conformed to the EN 14214 biodiesel specification. It is difficult to cost the intensive production of insects on industrial scale as they have only been produced on experimental scale. Given the shortage of agricultural space in highly populated countries where available agricultural land is used for the production of food crops, the production of insects is attractive for the production of lipids and protein where the insect breeding takes place in a warehouse.

Lignocellulosic biomass, the most abundant biomass resource, has been considered as feedstock for the production of biodiesel. This biomass has been suggested as nutritional source for oleaginous microorganisms producing microbial oil or single cell oils as promising alternative to vegetable oils and animal fats as feedstock for biodiesel production. A list of biomass sources is shown in Table 2.1 (Yousuf, 2012:2062).

Table 2.1: Lignocellulosic biomass sources considered as feedstock for biodiesel production (Yousuf 2012:2062).

Food crops	Non-food/energy crops	Forest residues	Industrial process residues
Rice straw	Cardoon	Tree residues (twigs, leaves, bark and roots	Rice husk
Wheat straw	Giant reed	Wood processing residues (sawmill off-cuts and saw dust	Rice bran
Sugarcane tops	Salix	Recycled wood	Sugarcane bagasse
Maize stalks millet	Jute stalks		Coconut shells
Groundnut stalks	Willow		Coconut husks
Corn straw	Poplar		Maize cob
Soybean residue	Eucalyptus		Maize husks
Residues from vegetables	Miscanthus		Groundnut husks
Residue from pulses	Reed canary grass		
	Switch grass		
	Hemp		

The lignocellulosic parts of food crop such as rice, sugar cane, vegetables, wheat, pulses, coconuts, maize, millet and groundnut are easy to collect and only small quantities are used for heat energy in rural areas. Energy crops such as switch grass and woody crops such as poplar require little soil disturbance and they also show higher productivity levels per square area compared to food crops (Panoutsou, 2007:6047). Forest residues are also a significant source of biomass, given the amount of residue produced such as saw mill off- cuts and saw dust, as

well as the large number of trees rejected by the wood processing industry (Hossain & Badr, 2007:1639). Besides agro-residues, forest residues are the second largest source of lignocellulosic biomass (Yousuf, 2012:2063). Residues from industrial processes such as rice bran, maize cobs and sugar cane bagasse are 100% collectable and abundant. These residues have different physical properties, cellulose content and fermentable pentosans and require different processing technologies when used to produce microbial oil (Yousuf, 2012:2063). The crystalline structure of lignocellulosic material offers some challenges for the biological transformation to fermentable sugars.

Researchers focus on oleaginous microorganisms that are capable of producing oil in the form of triacylglycerols of more than 20% of its weight. Meng and co-workers (2009:2) compared some organisms in terms of their oil content and found that microalgae organisms such as *Botryococus braunii* could produce oil at 25 – 75% of its dry weigh and *Schizochytrium* sp. 50 – 77% of its dry weight. Yeast organisms such as *Rhodotorula glutinis* and *Mortierella isabellina* could produce oil of 72 and 86% of their dry weight, respectively. The lipid composition of these microorganisms is similar to the oil composition of other feedstock used for biodiesel production (Meng *et al*. 2009:2). Bio-lipids from micro-organisms consist mainly of C16 to C18 fatty acids. Microbial oils are typically high in unsaturated fatty acids and show high potential as alternative oil sources for biodiesel production. Further research is needed to take this technology closer to the industrialisation phase.

Non-edible oil plants as feedstock source could also be in competition with other crops for agricultural land, resulting in deforestation of natural habitats resulting in a threat to biodiversity. Large scale growth of non-edible oil plant could also have a negative effect on water resources in dryer areas.

Biodiesel produced from microalgae is known as third generation biodiesel and has several advantages compared to first and second generation biodiesel. Microalgae have much higher growth rates and productivity compared to agricultural crops, forestry and other aquatic plants. They have all year round production and an oil content of 20 – 50% dry weight of biomass (Christi, 2007:296). Microalgae can be cultivated in brackish water on non-arable land. Algae can effectively improve bio-fixation of waste carbon dioxide and its ability

to remove CO_2 as biological treatment has been reported numerous times (Cantrell *et al*., 2008:7948; Sanchez *et al*., 2011:211). Microalgae can also be used to treat effluent from the agricultural industry. After oil extraction microalgae residue can also be used as fertilizer, thereby improving the economics of the process. Other useful co-products or by-products are biopolymers, proteins and carbohydrates. (Ahmad *et al.*, 2011:587). The cultivation of microalgae does not require herbicides and pesticides and does not directly affect the human food supply chain and is therefore excluded from the fuel versus food debate.

Lam (2013:6) successfully converted crude microalgae oil from *C. vulgaris* to biodiesel where sulphuric acid was used as catalyst. The crude oil contained high free fatty acid and a high viscosity. Miao and Wu (2006:845) found that the physical and chemical properties of biodiesel from microalgae were comparable in general with those of petroleum diesel. The biodiesel showed a much lower cold filter plugging point compared to those of diesel fuel. Christi (2007:300) emphasized that micro-algal oils differ from most other bio-oils intended for biodiesel production in terms of the presence of a relatively high concentration of polyunsaturated fatty acids which makes the algal oil susceptible to oxidation.

The commercialisation of biodiesel production from microalgae oil still faces a few challenges, such as reduction in production cost with respect to harvesting and oil extraction, attaining high photosynthetic efficiencies, optimum species selection to balance requirements for biofuel production and extraction of other valuable products (Brennan & Owende, 2010:559).

2.1.2.2 Waste oils and fat

Large amounts of waste oils and fats are produced all over the world on a continuous basis from food processing plants, restaurants, industrial processes and households. If not handled properly, these oils could become an environmental and health problem. Incorrect disposal of waste cooking oil is a threat to the environment in terms of possible contamination of water resources and soil. With the rise in vegetable oil prices, health threats are posed by some catering businesses where vegetable oil is used far beyond the time it should be discarded

(Mkentane, 2008:2). Waste fats and oils from slaughterhouses are often used as animal feed which could lead to the spread of diseases (Dias *et al.*, 2009:6355). The use of waste oils and fats as biodiesel feedstock could benefit the environment from being contaminated (Issara *et al*., 2011:269), and contribute to a lower production cost of biodiesel. Phan and co-workers (2008:3490) state that the price of waste cooking oil is 2 – 3 times lower than the price of virgin oil which could significantly reduce the manufacturing cost of biodiesel. The utilisation of waste fats and oils as feedstock does not compete with food production and contributes to the issue of converting waste into energy (Dias *et al*., 2009:6355).

Waste oil and fat originate from various industries such as meat processing facilities, by-products from industrial processes and as a waste product where vegetable oil or animal fat is used in a specific production process such as food processing.

Slaughterhouses produce waste lipids which contain high free fatty acids due to the hydrolysis of the triglycerides in the presence of water if not processed or utilised immediately. This high free fatty acid waste is not suitable for human or animal consumption and could be used as a biodiesel feedstock. Dias and co-workers (2009:6355) produced biodiesel from acid waste lard collected from a local butchery. The lard was blended with soybean oil to improve the biodiesel quality.

Lin and co-workers (2009:134) compared the fuel properties of biodiesel produced from crude fish oil of marine fish with that of commercial biodiesel produced from waste cooking oil. It was found that palmitic acid (C16:0) and oleic acid (C18:1) were the two most prominent fatty acids present in the fish oil. Saturated fatty acids made up 37.06% of the fatty acids. Fish oil differs from normal vegetable oil in that it contains long chain fatty acids in the range of C20 – C22 (Behcet, 2011:1189). The fish oil from marine fish also contains a larger amount of polyunsaturated fatty acids with more than three double bonds. The kinetic viscosity, cetane index, carbon residue and heating value are comparable to that of waste cooking oil.

Bhatti and co-workers (2008:2961) used acid-catalysed esterification with sulphuric acid to produce biodiesel from waste tallow. It was suggested that biodiesel from chicken and mutton fat is suitable as low cost feedstock for biodiesel production and that the physical and chemical properties of the biodiesel are comparable with the recommended properties.

Industries such as the vegetable oil refining industry, the tobacco industry, the wine industry and the paper and pulp industry are examples of potential sources of lipids for biodiesel production.

Most of the biodiesel produced in the world uses feedstock from the vegetable oil refining industry as edible oils. This industry also produces substantial amounts of waste lipids that need to be managed in order to protect the environment. At least three steps in the refining process generate waste lipids which are:

- Neutralisation generates vegetable oil soap stock
- Bleaching generates spent bleach earth which contains oil
- Deodorisation generates fatty acid distillate

Vegetable oil soap stock is an alkaline aqueous emulsion containing about 50% water, free fatty acids, phosphoacyl glycerols, triacyl glycerols, pigments and other non-polar compounds. Haas (2005:1088) discussed multiple approaches to produce fatty acid methyl esters which included enzymatic catalysis and non-enzymatic catalysis. Complete saponification of the soap stock followed by acidulation and esterification using acid catalysis was found to be the most effective method. Biodiesel produced from soap stock was found to be comparable to that produced from refined soybean oil.

Spent bleaching earth is generated in the discolouration (bleaching) of crude vegetable oil containing 30- 50% oil by weight (Pandey *et al*., 2003:115). The recovery of fatty acids includes methods such as organic solvent extraction and supercritical fluid extraction (Du Mont & Narine 2007:967). Huang and co-workers (2010:269) produced biodiesel via a transesterification process with its quality in reasonable agreement with the specification.

Fatty acid distillate is produced from the deodorising process removing the volatile components from the vegetable oil using steam distillation. This process

is done at temperatures up to 200 °C under vacuum (Pandey *et al.,* 2003:105). Cho and co-workers (2012:271) produced biodiesel from palm fatty acid distillate using a non-catalytic esterification process with methanol. The biodiesel was purified by means of distillation and conformed to the European standard for biodiesel (EN14214). A biodiesel yield of 91.2% was obtained.

Tobacco seeds, a by-product in the production of tobacco leaves, contain oil which is not suitable for the food industry. Giannelos and co-workers (2002:5) evaluated tobacco oil seed as a potential source of energy. It was found that the seeds contained almost 38% oil with major constituents comprising linoleic acid, oleic acid and palmitic acid which makes it a potential candidate as feedstock for biodiesel production. Usta and co-workers (2011:2034) evaluated the properties of biodiesel from tobacco seed oil and found that most of the specifications set out in the EN 14214 standard were met. The oxidation stability was a little lower than specification and the iodine number was higher. Blending with biodiesel produced from waste cooking oil was done, as well as the addition of antioxidants and cold flow improvers.

Tall oil is an odourless black and yellow oily liquid which is a by-product of coniferous wood recovered from the paper and pulp industry (Demirbas 2011:2274). It consists of resin acids, terpenoids, fatty acid and triglyceride oils and unsaponifiables. Altiparmak and co-workers (2007:246) studied the engine performance of new and fuel blends from tall oil fatty acid methyl esters. Tall oil methyl ester –diesel fuel blends showed promise in terms of CO emissions, low sulphur content, higher cetane number and low aromatic contents.

The rubber seed production potential from rubber tree plantations in India is about 150 kg per hectare (Ramadhas *et al*., 2005:336). The oil from rubber seeds contain high levels of free fatty acids (about 17%) which is unsuitable for human consumption and is therefore mainly used to manufacture low cost soaps. Morshed and co-workers (2011:2986) produced biodiesel from rubber seed oil by sequential saponification, acidulation and esterification. Properties such as specific gravity (0.85 at 30 °C), viscosity (4.5 mm^2/s at 40 °C), flashpoint (120 °C), cloud point (3 °C), pour point (-5°C) and calorific value (32MJ/kg) were comparable to the values for petroleum diesel.

The production of biodiesel from grape seed oil was studied by Fernandez and co-workers (2010:7019). During winemaking grape seeds are left behind when the juice is pressed from the grapes and grape seed oil is obtained from the seeds which contain 10 – 20% oil. Fernandez and co-workers (2010:7019) used solvent extraction with the Soxhlet method to extract the grape seed oil with different solvents. Oxidation stability and cold filter plugging point were two of the critical parameters for grape seed methyl esters. Extraction procedure where anti-oxidants from the oil were removed, resulted in the decrease of the oxidation stability.

Waste from the leather industry as feedstock for biodiesel production was studied by Ozgunay and co-workers (2007:1897). Environmental problems are created by waste from the processing of hides and skins which contain a considerable amount of fat. It was recommended that biodiesel from waste fats from the leather industry should be used as an additive to petroleum diesel, given the high pour point value of the biodiesel.

Waste vegetable oils and fats are the major feedstock for the production of biodiesel in countries such as Australia, New Zealand, Mexico and China (Lin *et al*., 2011:1023). Waste vegetable oils are not directly implicated in the food versus fuel debate, land requirement issues and biodiversity problems.

Waste cooking oil or waste fryer grease is generated by the food preparation industry mostly using vegetable oil from the edible oil industry. It is the most widely generated waste oil, which is growing in volume as a result of the increase in fast food consumption of working parents and changing habits of young people (Diya'uddeen *et al*., 2012:168). Used cooking oil is easily available and also 2 – 3 times cheaper than edible oil (Zhang *et al*., 2003:2; Phan & Phan 2008:3490).

Significant quantities of waste cooking oil are generated in many countries. China generates approximately 4.5 million tons per year and the USA 10 million tons (Diya'uddeen *et al.*, 2012:168; Gui *et al*., 2008:1650). The use of waste cooking oil as feedstock for biodiesel production supports the concept of transforming waste to energy and solves a serious problem of waste oils being dumped into sewers and on municipal dumping sites. The release of vegetable oils into the environment could lead to the accumulation of this waste in rivers, dams and in the soil. Mudge (1995:188) reviewed the effects of vegetable oil spillages on the

marine environment and rivers. Vegetable oil could become pollutants by smothering, as oil floats on water and causes oxygen depletion underneath. These oils are also toxic to certain animals and the polymerisation of the oils could lead to reduction in biodegradability. The saturation level of the oil increases during cooking by the increase of the saturated fatty acids as well as the increase of the mono-saturated fatty acids relative to the di-unsaturated and tri-unsaturated fatty acids. Knothe and Steidly (2009:5796) found that the acid value increased by 4.02% on average and the dynamic viscosity an average change of 7.46 cP. The tendency of frying oils to undergo an increase in viscosity, acidity and saturation levels with exposure to heating and cooling during the frying process suggests that the cetane number and the oxidative stability of the produced biodiesel will increase as a result of the higher degree of saturation.

Sewage sludge is produced in huge amounts in municipal wastewater treatment plants and is available to be used as feedstock for biodiesel production at no cost. Siddiquee & Rohani (2011: 2247) evaluated methanol and hexane as solvents to extract lipids from primary and secondary sludge sources. Methanol extracted 14.46 weight% lipids from the primary sludge and 10.04% from the secondary sludge, based on the dried sludge. Hexane extracted 11.6 weight% and 3.04 % from the primary and secondary sludge respectively. Acid-catalysed esterification and transesterification using sulphuric acid as catalyst produced 41.25% based on the lipid mass from the primary sludge and 38.94% from the secondary sludge. The biodiesel contained fatty acid methyl esters of myristic acid (C14:0), palmitic acid (C16:0), palmitoleic acid (C16:1), stearic acid (C18:0), oleic acid (C18:1) and linoleic acid (C18:2). Palmitic acid methyl esters were present in the largest amount. Mondala and co-workers (2009:1209) produced biodiesel from primary and secondary sewage sludge using an acid-catalysed in situ transesterification process. Maximum yields of primary sludge (14.5%) and secondary sludge (2.5%) were obtained at 75 °C, 5% (v/v) sulphuric acid and a 12:1 methanol to sludge mass ratio. The cost of this biodiesel was estimated to be lower than the cost of petroleum diesel, as well as soybean biodiesel. Mondala and co-workers (2009:1209) concluded that municipal waste water sludge has a high potential as biodiesel feedstock and is abundant and cost-competitive.

With the growing shortage of biodiesel feedstock, driven by the growing demand for edible oil, the search for alternative feedstock will continue. Waste materials containing fatty acids are considered an important source of biodiesel feedstock. The evaluation of various waste materials showed that impurities in such waste materials needed to be considered in the choice of production technology to ensure the quality of the product produced from that waste material. The critical impurities in biodiesel feedstock are free fatty acids and water. Pre-treatment strategies of low quality feedstock are built to minimise the effect of FFA and water on the production process and quality of the biodiesel.

Waste grease as feedstock for biodiesel production has been evaluated by several researchers. This waste grease refers to a waste material generated by the food preparation industry. Waste process grease (WPG) is a waste matrial generated through an industrial process.

Waste materials generated by industrial processes which contain fatty acids have not been considered a potential feedstock for biodiesel production up to now. These waste materials include lubricating oils such as vegetable oil-based hydraulic fluids, gear oil lubricants, two cycle engine oils, food machinery lubricants, metalworking fluids, textile lubricants, grease base fluids and chain bar lubricants (Gawrilow, 2003:5). The major difference between this fatty acid containing industrial waste products and the traditional waste oils such as used cooking oil is that the industrial oils contain not only the traditional impurities but also other unusual impurities introduced via the incorporation of additives packages in the industrial products to manipulate the properties of the oil.

Industrial waste oils offer even more challenges for biodiesel production technologies to produce biodiesel that conforms to the specification for biodiesel at an acceptable cost.

2.2 The production of biodiesel

The two most prominent factors having a negative effect on the large scale commercialisation of biodiesel are feedstock availability and the production cost of biodiesel. As discussed in the previous section, the use of low quality, low

value feedstock could make a substantial contribution to the reduction of biodiesel production cost. The use of low quality feedstock has opened up many challenges for the development of improved biodiesel production technology to ensure the quality of the final product produced from low quality feedstock, and also to reduce the production cost.

2.2.1 Chemical reactions in biodiesel production

Biodiesel can be produced by two chemical reactions, namely esterification and transesterification. Both reactions yield the same product which is fatty acid alkyl esters. In esterification the reactants are free fatty acids and an alcohol and in the case of transesterification triglycerides and an alcohol.

Esterification is a reaction between a free fatty acid and an alcohol as illustrated in Figure 2.1. The reaction products are esters and water. The reaction is reversible and an access of alcohol is needed to shift the reaction equilibrium to the right for the reaction to approach completion. As the reaction progresses, the water content of the reaction mixture increases, driving the reaction to the left up to a point where the reaction is stopped.

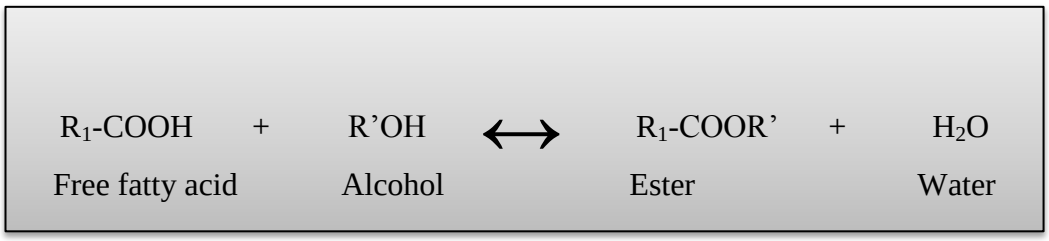

Figure 2.1: The esterification reaction (Boffito *et al*., 2013: 613)

Transesterification is a reaction between fats or oils and alcohol. As shown in figure 2.2, triglycerides are converted in this reaction stepwise to diglycerides, monoglycerides and eventually glycerol and the alkyl esters. Transesterification

consists of these consecutive reactions which are reversible (Atabani *et. al*., 2012:2079). Usually the fatty acid methyl esters float on top of the polar glycerol phase which is at the bottom of the reactor.

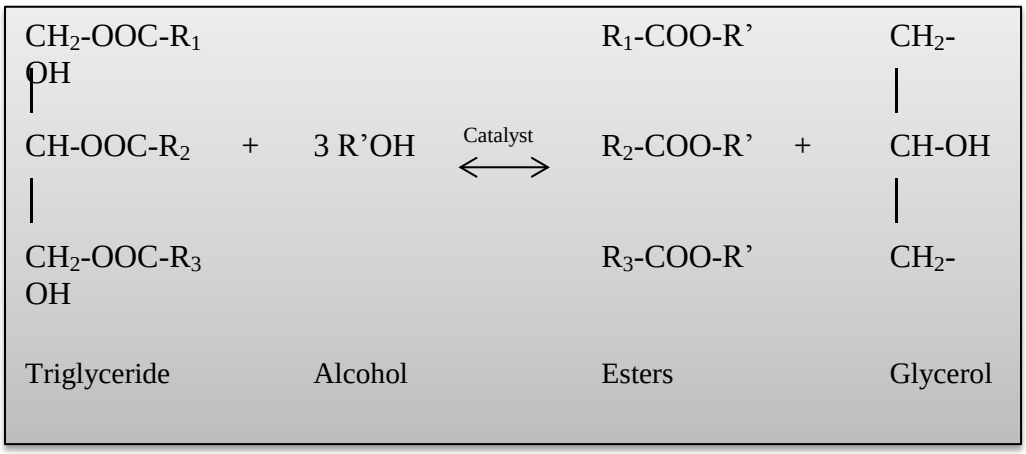

Figure 2.2: The transesterification reaction (Van Gerpen 2005:1099)

Transesterification is most often used for biodiesel production and can be further categorised as shown in Figure 2.3. (Atabani *et. al*., 2012:2080)

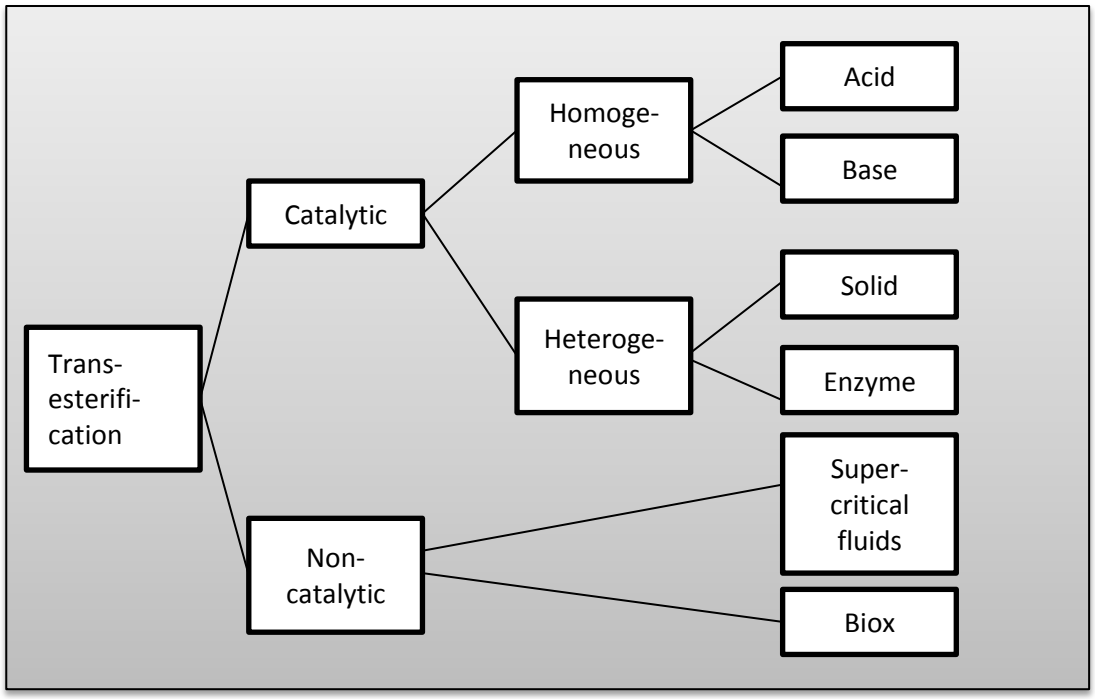

Figure 2.3: Categorisation of transesterification processes (Atabani *et al.*, 2012: 2080)

2.2.2 Catalytic transesterification

Catalysts are used in transesterification to increase the reaction rate as fats and oils are only slightly soluble in alcohol (Karmakar *et. al*., 2010:7202). Two main types of catalytic transesterification are used namely homogeneous and heterogeneous catalysis.

Alkaline homogeneous catalysis is the conventional process most often used in industry. The catalysts often used are sodium hydroxide, potassium hydroxide and sodium methoxide. Singh and co-workers (2006:597) compared four catalysts (potassium hydroxide, sodium hydroxide, potassium methoxide and sodium methoxide) on a molar basis. It was found that the methoxide catalysts gave better yields than the hydroxide catalysts. The methoxide catalysts accelerated the reaction and also elevated the conversion equilibrium.

Advantages of the alkaline catalysts are that the transesterification reaction is relatively fast, executable at mild temperature and pressure and the catalysts are relatively cheap and easily available. Disadvantages are that high quality oil is needed for alkaline transesterification, as the presence of water and free fatty acids has a negative effect on the reaction. Water enhances the formation of free fatty acids and the free fatty acids react with the alkaline catalyst to form soap. This leads to the deactivation of the catalyst and emulsification problems when it comes to glycerol separation and purification by water washing of the methyl esters. Downstream problems also exist where large quantities of water are needed in the biodiesel purification process that needs to be neutralised and treated to conform to environmental regulations.

Acid catalysis solves the problem of low quality feedstock containing high levels of free fatty acids. Typical acids used as acid catalysts are sulphuric acid, hydrochloric acid, phosphoric acid and boron trifluoride (Borugadda & Goud, 2012:4774). Sulphuric acid is the most commonly used acid catalyst. Acid catalysts have slower reaction rates compared to alkaline catalysts and a higher alcohol to oil ratio is needed for the reaction to run to completion. Acid catalysts are also used as a pre-treatment step to esterify the free fatty acids in low quality feedstock reducing the free fatty acids to low enough levels for alkaline

transesterification. A disadvantage of the acid-catalysed process as pre-treatment step is the formation of water in the esterification reaction between the free fatty acid and the alcohol which reduces the reaction rate up to a point where the reaction is stopped.

Atabani and co-workers (2012:2080) grouped heterogeneous catalysts into solid catalysts and enzymes.

Solid catalysts catalyse reactions in a different phase which shows promise in terms of separating the catalyst from the reaction medium after the reaction so that the catalyst could be reused. Research work focusing on heterogeneous catalysis in biodiesel production has increased recently as the potential exists to reduce or eliminate some of the disadvantages associated with homogeneous catalysis. The potential benefits of using heterogeneous catalyst are as follows:

- The catalyst can be separated from the reaction medium after use
- The catalyst can be reused
- Continuous fixed bed operation is possible
- The catalysts are not sensitive towards water and free fatty acids

The potential drawbacks of heterogeneous catalysts are:

- Energy intensive reaction conditions are required to achieve high yield and conversions
- The preparation of solid catalysts can be comprehensive and costly
- Leaching of the catalyst into the reaction medium often causes the deactivation of the catalyst when reused.

Factors influencing the reactivity of solid catalysts should be understood and be quantifiable (Helwani *et al*., 2009:5). The catalyst must have a porous system with interconnecting pores to increase the surface area and therefore the activity of the catalyst for transesterification. The challenge is to obtain uniform pores with control over pore size, geometry and radius as well as the stability of the solid material in the system. A hydrophobic surface should also be created to promote adsorption of hydrophobic species such as oil on the catalyst surface. Adsorption of hydrophilic species such as water and glycerol on the catalyst surface will result in the deactivation of the catalyst. The reliable quantification and

manipulation of these factors will result in a better understanding of the reactivity of solid catalysts.

Enzymatic transesterification has received much attention from researchers because of the advantages as opposed to conventional alkaline catalysis that could benefit the biodiesel production process. (Robles-Medina *et al*., 2009:398).

The presence of free fatty acids in the feedstock poses no problem when enzymatic catalysis is used as the free fatty acids are transformed to biodiesel. The presence of water has no serious effect on lipase catalysis, while in the case of alkaline transesterification water and FFA promote the formation of soap and hydrolysis of the oil resulting in the formation of more soap. Glycerol recovery is easy for lipase catalysis delivering high grade glycerol, while glycerol recovery for the conventional process is complex, delivering low quality glycerol. The environmental impact of enzymatic catalysis is low as the treatment of waste water is not necessary compared to high costs for waste water treatment for the conventional process where alkaline effluents need to be neutralised. Lipase catalysis is also less energy intensive compared to the conventional process. The major disadvantages of the enzymatic process are the high costs of enzymes and the transesterification reaction takes much longer compared to the conventional process.

2.2.3 Non-catalytic transesterification

Attempts by researchers to overcome the poor miscibility of triglycerides and methanol which contributes to slow reaction rates include approaches such as supercritical fluid technology and the inclusion of co-solvents in the reaction mixture.

Tetrahydrofuran (THF) with a boiling point of 67 °C is a commonly used secondary solvent, soluble in both reactants (tryglycerides and alcohol) creating a homogeneous phase resulting in the reduction of mass transfer resistance. This approach is used by the patented Biox process which converts both free fatty acids and triglycerides with a feedstock conversion of greater than 99% (Enweremadu & Mbarawa, 2009:2216).

A fluid is in a supercritical state when the fluid's temperature is above its critical temperature and its pressure is above its critical pressure. In this state, the densities of the two phases are identical with no distinction between the two phases. This homogeneous environment offers a number of unique advantages such as better mixing of the different species, improved heat and mass transfer, fast reaction rates, good scalability, it is more environmentally friendly and biodiesel is easily separated from the reaction mixture. No catalyst is needed and therefore no washing and neutralisation are required (Shahid *et al*., 2011:4741). The presence of water in the feedstock has no negative effect on the reaction. The synthesis of biodiesel by supercritical fluid techniques implies severe reaction, conditions and therefore high operational cost reducing the chances for utilisation on an industrial scale. For this reason researchers focussed the last couple of years on decreasing the severity of the reaction conditions. Co-solvents such as carbon dioxide, hexane and propane are used with the supercritical methanol, allowing the reaction to be completed at lower temperatures and pressures (Borugadda & Goud, 2012:4778). The operating conditions can also be decreased by using subcritical conditions with small amounts of catalysts. Tan and co-workers (2010:88) produced biodiesel from palm oil by using supercritical methanol. It was found that the presence of water in the reaction mixture increased the biodiesel yield and that the presence of a non-polar solvent such as heptane had the potential to decrease the reaction temperature required to achieve high yields.

2.2.4 Feedstock pre-treatment

Processes for the refining of vegetable oils in the agricultural industry to produce edible oil with a high purity for the food industry have been well established. The cost of such high purity oil when used as biodiesel feedstock places an additional burden on the already expensive biodiesel product. The pre-treatment of crude edible oil to be used as biodiesel feedstock involves a less complex and cheaper process (Santori *et al*., 2012:115). The heated oil is mixed with a strong mineral acid to clarify the oil. This step removes the hydrophobic gums from the oil. Next the oil is mixed with 0.1 – 0.2 % sodium hydroxide to neutralise the oil to remove the free fatty acids. Centrifugation is then used to remove the soaps from the

neutralisation step. A water washing step follows with centrifugation to remove trace impurities. The oil is then dried with a vacuum flash–vaporization step at 116 °C and 0.8 – 0.9 bar absolute.

The most significant impurities interfering with the biodiesel production process are water and free fatty acids. The effect of water and free fatty acids has been addressed in numerous studies (Atadashi *et al*., 2011; Kusdiana *et al*., 2004:290). The presence of water retards the transesterification reaction, as the triglycerides are hydrolysed to form diglycerides and free fatty acids. The hydrolysis reaction is intensified at elevated temperatures. Kusdiana and Saka (2004:290) compared the effect of water content in the reaction mixture on alkaline and acid-catalysed transesterification. Water had a significant effect on the conversion for both reactions. For the acid-catalysed reaction the addition of 5% water resulted in a reduction in conversion to 6%. The addition of as little as 0.1% water to the reaction mixture already led to some reduction in the methyl ester conversion. Haas (2005:1093) reported that after the esterification of acid oil using sulphuric acid, between 5 and 10% of the free fatty acids remained unreacted as a consequence of the formation of water in the esterification reaction as shown in Figure 2.4.

Figure 2.4: Formation of water during the esterification reaction

The concentration of water builds up during the reaction, driving the reaction to the left to a point where the reaction is stopped.

The presence of free fatty acids in the oil leads to the formation of soap. The alkaline catalyst reacts with the free fatty acids to form a saponified product as shown in Figure 2.5 (Atadashi *et al*., 2011:440)

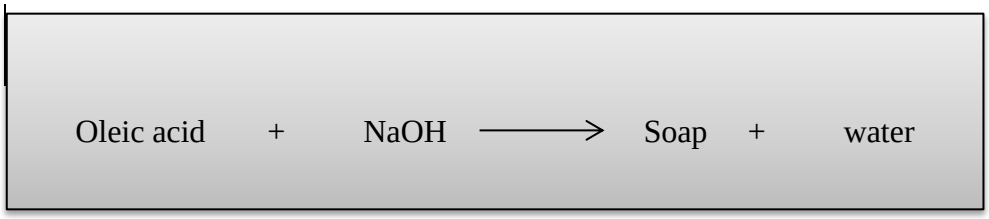

Figure 2.5: The saponification reaction

The formation of soap results in the consumption of the catalyst, the reduction in catalyst effectiveness, emulsification effects which make the separation of glycerol difficult, as well as the separation of water during the washing process. Biodiesel yields are affected due to the loss of methyl esters during washing. The presence of soaps in the final product also causes the biodiesel not to comply with the specification. Minimum concentrations of free fatty acids are recommended by different researchers. Some authors suggested that the FFA level should be below 1% (Tiwari *et. al*., 2007:574). Ramadhas (2005:336) and Sahoo and co-workers (2007:450) suggested that the level should be below 2%.

Different production routes have been investigated to reduce or to accommodate high free fatty acids in feedstock for biodiesel production:

1 Separation of FFA from triglycerides
2 Pre-esterification
3 Hydrolysis of triacylglycerols followed by esterification

2.2.4.1 Separation of FFA from triglycerides

Several processes for the de-acidification of oils and fats exist. The different solubility of neutral glycerides and fatty acids in different organic solvents forms the basis of selective extraction of free fatty acids from tryglycerides. The high difference in boiling points of the solvents and fatty acid compounds makes solvent stripping from refined oil and solvent recovery easy (Bhosle &

Subramanian 2005:487). Qian and co-workers (2010:7028) produced biodiesel from *Jatropha curcas L*. oil after a two-phase extraction of the oil. A methanol/n-hexane solvent mixture at a ratio of 60:40 was used at an extraction temperature of 35 °C and an extraction time of 30 minutes. The polarity of the methanol was increased by the addition of 10% (v/v) water. After separation the hexane phase contained the Jatropha oil and the polar phase contained the methanol, water, free fatty acids and colloid and colouring matter. The free fatty acid concentration of the oil was reduced to 0.42% and the water content to 0.048%. The hexane/oil phase was transesterified to reach a conversion of 98%.

Supercritical fluid extraction with carbon dioxide was evaluated by Taher and co-workers (2011:23) for the extraction of fat from lamb meat to be used as feedstock for biodiesel production. It was possible to extract 87.4% of the total fat from frozen dried meat at optimum conditions of 45 °C, 500 bar and 3 ml per minute flow rate. The advantages of supercritical fluid extraction such as carbon dioxide over conventional solvent extraction are that inert solvents are used, which are pollution free, easily available at low cost and can be separated easily (Bhosle *et al*., 2005:489). It could also be made highly selective by controlling temperature and pressure.

Various approaches have been made using membrane technology for the de-acidification of vegetable oil. The use of membranes in processes for oil refining have several advantages, such as low-energy consumption, operation at ambient temperature, no addition of chemicals and the retention of other desirable components (Bhosle *et al*., 2005:490). Limitations of membrane de-acidification are that the molecular weight differences between triglycerides and free fatty acid are small which makes separation difficult. Low permeate flux causes problems and suitable membranes with high selectivity are not available. Approaches followed by researchers include direct de-acidification, pre-treatment of oil, followed by de-acidification and extraction with a solvent followed by membrane separation. Boshle and co-workers (2005: 490) pointed out that a combination of solvent extraction and membrane separation could be technically feasible with solvent resistant membranes. Further studies on these new approaches are needed.

2.2.4.2 Pre-esterification

The most commonly accepted route to reduce free fatty acids in feedstock is esterification to reduce the acid value of the feedstock to levels suitable for alkaline transesterification. One advantage of this route is that no fatty acids are lost as in the case of other methods such as neutralisation.

For both esterification and transesterification, catalysts are being used to increase reaction rates and to improve yields of triglycerides and free fatty acids being converted to biodiesel.

In homogeneous acid-catalysed esterification, catalysts such as sulphuric acid, phosphoric acid and hydrochloric acid are predominantly used (Atadashi *et al.*, 2012:3277). Heterogeneous catalysts such as solid phase catalysts and enzymes are popular research topics due to the potential to have benefits such as easier separation of the catalyst from the reaction medium, more simplified purification and the reusability of the catalyst.

Many researchers have evaluated the effect of reaction parameters such as alcohol to oil ratio, catalyst concentration, temperature and duration of the reaction on the reduction of free fatty acids prior to alkaline transesterification. Zhang and Jiang (2008:8997) used a methanol to oil ratio of 24:1 at a sulphuric acid concentration of 2%, a temperature of 60 °C and a reaction time of 80 minutes as optimum conditions to reduce the FFA of *Zanthoxylum bungeanum* seed oil from 22.75% to 0.58% in a single step. Zullaikah and co-workers (2005:1895) reduced the free fatty acid concentration of rice bran oil from more than 20% to 3.2% using a reaction time of 120 minutes at a sulphuric acid concentration of 2 weight% and a methanol to oil ratio of 5:1. Ramadhas and co-workers (2005:337) reduced the free fatty acids of rubber seed oil from 17% to less than 1% in 30 minutes using a catalyst concentration (sulphuric acid) of 0.5 % , a methanol to oil ratio of 6:1 at a reaction temperature of 45 °C. Lin and co-workers (2009:688) reduced the free fatty acids of crude rice bran oil from 20% to 0.45% in a two- step acid esterification process at a catalyst concentration (sulphuric acid) of 1 weight% for each step, a methanol to oil ratio of 6:1 for the first step and 7:1 for the second step. For both steps a reaction time of 60 minutes was used. An important

procedure between the two esterification steps is the removal of water formed during the esterification reaction which retards the reaction (Ghadge & Raheman, 2005:604).

The removal of the acid catalyst after the pre-esterification step is one of the main drawbacks of the homogeneous catalysis process. Heterogeneous catalysis is a promising technology to solve this problem. (Di Serio *et al.*, 2005:112).The catalyst does not dissolve in the reaction medium and can be filtered out easily and reused. Peng and co-workers (2008:441) studied the production of biodiesel from low cost feedstock with high free fatty acids using a SO_4^{2-}-TiO_2-SiO_2 solid acid catalyst. Refined cotton seed oil containing different quantities of oleic acid was made up to study the effect of free fatty acids on the ester yield. The reaction was carried out at 200 °C with a catalyst concentration of 3 weight% and a methanol: oil ratio of 9:1. An increase in the free fatty acid content resulted in an increase in the methyl ester content which possibly could be as a result of a higher solubility of the free fatty acids in methanol compared to the cotton seed oil. It was also found that the rate of esterification of oleic acid was much faster than the rate of transesterification of cottonseed oil.

Yan and co-workers (2012:332) developed a novel process to produce biodiesel from waste grease with high free fatty acids using tandem lipases. They solved the current problem of using one enzyme to catalyse both esterification and transesterification reactions by using tandem enzymes, as the one prefers esterification and the other transesterification. Either dry or wet cells of the recombinant *E. coli* co-expressing *C. Antarctica* lipase B (CALB) and *T. lanuginose lipase* (TLL) were used in a solvent-free system. A biodiesel yield of 95% was achieved.

2.2.4.3 Hydrolysis of triacylglycerols followed by esterification

The process route followed by this approach involves the hydrolysis of the triglycerides in the feedstock that contains both triglycerides and FFA, followed by the acid esterification of the hydrolysate.

Haas (2005:1087) evaluated the use of vegetable oil soapstock as low value lipid to improve the economics of biodiesel production. An alternative method than the traditional two-step acid-alkaline-catalysed process, to deal with feedstock that contained both free and glycerol-linked fatty acids was to hydrolyse the lipids, converting all the acylglycerol fatty acids to free fatty acids and then to esterify the resulting free fatty acids. Saponification (alkali-catalysed hydrolysis) using sodium hydroxide was conducted to convert all the acylglycerols to free fatty acids. An amount of 4.2 weight% of sodium hydroxide was added to the already alkaline reaction mixture followed by incubation at 100 °C for 2 to 4 hours. According to Haas (2005:1087) complete hydrolysis was achieved of both acylglycerols and phosphoacylglycerol, using these reaction conditions. Lower concentrations of alkali at lower temperatures failed to achieve complete saponification.

Hydrolysis of soybean oil containing high FFA, using a binary immobilised lipase was done by Ting and co-workers (2008:203) for the production of biodiesel from soybean oil. The hydrolysate was esterified at a sulphuric acid concentration of 2.5%, a methanol to oil molar ratio of 1:15 for 12 hours at 50 °C. A biodiesel yield of 99% was obtained.

2.2.5 Effect of reaction parameters on the transesterification reaction

The reaction parameters that influence the conversion rate, duration of the reaction and the completeness of the reaction include temperature, alcohol to oil ratio, catalyst loading, mixing intensity and the purity of the reactants (Srivastava and Prasad, 2000:126). The catalyst changes the rate by lowering the activation energy of the reaction, thereby increasing the rate of the reaction, but is not part of the reaction itself. The transesterification reaction is reversible and stoichiometrically 3 moles of alcohol are needed for every mole of triglycerides (Demirbas, 2009:20). Excess alcohol is needed to drive the reaction to completion (Ma & Hanna, 1999:7).The optimum reaction temperature depends on the type of alcohol used. A temperature close to the boiling point of the alcohol is often selected as the optimum temperature for the transesterification reaction. Mixing rate also has an important influence on the reaction as the alcohol and the oil are not miscible. Fast enough mixing is needed to ensure mass transfer.

2.2.6 Biodiesel separation

In the conventional process where sodium hydroxide and potassium hydroxide is often used as catalysts, the glycerol is separated from the crude alkyl esters by means of gravity separation or centrifugation. The difference between the densities of glycerol (1050kg/m^3) and biodiesel (880kg/m^3) is enough to have sufficient gravitational settling (Atadashi *et al*., 2011:438). Factors such as concentration of methanol in the reaction mixture, free fatty acids and water in the initial feedstock and type of catalyst used in the process could have a negative effect on the separation of the glycerol phase from the biodiesel. A too high methanol concentration interferes with the separation of glycerol from the biodiesel as solubility of the glycerol in the ester phase increases (Mehr *et al.*, 2006:256). Berios and Skelton, (2008:464) concluded that it was vital to remove as much glycerol as possible in the settling and centrifugation stage. This would

reduce the load on the purification step and result in a much more cost effective process.

2.2.7 Biodiesel refining

Crude biodiesel contains various impurities such as free glycerol, soap, metals, methanol, free fatty acids, catalyst, water and glycerides. The purification of crude biodiesel is necessary to remove unwanted substances from the biodiesel which will have negative effects such as corrosion, bacteriological growth in the engine, deposits in the injectors filter blockages and engine weakening on diesel engines if present in the fuel (Berrios & Skelton, 2008: 460).

Atadashi and co-workers (2011:4239) grouped the biodiesel refining technologies as follows:

- Wet washing technologies
- Dry washing technologies
- Membrane refining technologies

2.2.7.1 Water washing

Water washing and dry washing technologies are generally excepted methods to purify biodiesel (Berios & Skelton, 2008:460). Wet washing is considered as the traditional method and is very effective in the removal of glycerol and methanol as a result of their high solubility in water. Soap, salts, remaining catalyst and free glycerol are being removed with the water washing step (Van Gerpen, 2005:1103). Berios and Skelton (2008:464) compared water washing, ion exchange resins and adsorbents such as magnesol to purify biodiesel and concluded that water washing was the only technique that could successfully purify the biodiesel to the EN 14214 specification directly from the glycerol separation step. Wet washing is performed using deionised water, water containing acids such as phosphoric acid, sulphuric acid and hydrochloric acid and water with organic solvents (Atadashi *et al*., 2011:4242). Haas and co-

workers (2006:675) incorporated water washing in their process model to estimate the production cost of biodiesel. The crude methyl ester stream is washed with acidic water at a pH of 4.5. The acidity neutralises the catalyst and any soaps are being converted to free fatty acids and salts. This step reduces any emulsification tendencies. The water is then separated from the esters using centrifugation. Vacuum drying is further used to dry the biodiesel to a content of 0.045%. Cayli and co-worker (2008:120) treated the crude ester layer with hot water at 70°C followed by 5% phosphoric acid. Organic solvent washing technology involves the extraction of crude biodiesel with a non-polar solvent such as petroleum ether or hexane. Soriano and co-workers (2009:561) synthesised biodiesel via homogeneous Lewis acids in the presence of tetrahydrofuran. The excess methanol and THF were removed after the reaction with vacuum distillation followed by extraction with petroleum ether. Vacuum distillation was used to remove the solvent in obtaining the final product.

Water washing has some disadvantages such as the generation of large quantities of effluent water which needs to be treated as well as the loss of product during the purification stage.

2.2.7.2 Dry washing

Dry washing technologies have been developed to overcome the disadvantages of the wet washing technologies. Dry washing is done by using silicates, ion exchange resins, celluloses, activated carbon, activated clay and activated fibre (Atadashi *et al.*, 2011:4244). The use of magnesium silicate (magnesol) is being promoted by Hydrotechnik in the UK and by The Dallas Corporation in the USA as one of the commercial processes. The acidic and basic adsorption sites of these compounds, which are strongly polar, have an affinity for hydrophilic substances such as methanol, glycerol, mono- and di-glycerides, soap and metals (Atadashi *et al*., 2011:441). Dry washing techniques offer benefits such as lower cost of production, its waste has alternative uses, it saves space, no waste water is generated, it is easily incorporated into existing plants and it is waterless. Two resin manufacturers Rohm and Haas with a product BD 10 Dry and Purolite with

a product PD 206 promote the use of ion exchange resins as biodiesel purification methods.

Ion exchange resins and magnesol were compared with water washing techniques to purify crude biodiesel (Berios & Skelton, 2008:462). It was found that the resins had little effect on the methanol with no effect on the glycerides. The capacity of methanol removal was low, about 20 L biodiesel per kilogram resin. The soap content was reduced by about a factor of 10 and the acid value increase slightly. This could be due to the acidity of the resin. Berios and Skelton (2008:462) concluded that the main advantage of ion exchange resins was to reduce the free glycerol levels to an acceptable standard. Magnesol did not have much effect on the glycerides but its effect on methanol was larger than that of the resins. The methanol content could not be decreased to the levels allowed by the biodiesel specification. Glycerol was removed in a satisfactory way to meet the biodiesel specification and soap levels and acidity were also slightly reduced. None of the three processes evaluated by Berios and co-workers (2008:462) had any significant effect on the water content, glycerides, oxidative stability index and acidity. Sabudak and Yildiz (2010:802) compared water washing with distilled water, and dry washing with magnesol and ion exchange resin as purification methods in biodiesel production from waste frying oils. Biodiesel was produced following three methods of production, one-step basic, two-step basic and two-step acid-basic transesterification. The effect of a co-solvent THF was also evaluated. Ion exchange resin as purification method was determined to be more effective than magnesol, although comparable results were achieved.

Toeneboebn and co-workers (1994) found that in fatty materials, substances such as amorphous silica hydro gels showed promise to remove sulphur containing compounds exhibiting excellent adsorption capacity for these sulphur compounds.

Jalalpoor and co-workers (2009) found that adsorbent particles such as silica hydrogel reduced the phosphorous content of degummed triglycerides to 10 ppm.

2.2.7.3 Membrane purification

Membrane separation processes offer stable effluent quality, occupy small areas, are relativly simple to scale up and uses no chemicals (Sarmento *et al*., 2004:71). Transesterification using a membrane reactor and separative membranes in the refining of crude biodiesel made a huge impact on the production and separation difficulties for biodiesel (Atadashi *et al*., 2010:2002). The passage of unreacted oil to the biodiesel product mixture was restricted by the use of the membrane reactor. The application of membrane technology for the separation and purification of biodiesel was found to be superior compared to the conventional technologies. Membrane processes showed promise in the removal of contaminants such as triglycerides, catalyst, soap and methanol (Cao *et al*., 2008:1028; Wang *et al*., 2009:422; Gomes *et al*., 2010:271). Atadashi and co-workers (2011:5060) concluded that more research was needed to study the fouling effects and stability of membranes for the production and refining of biodiesel.

2.3 Biodiesel quality

2.3.1 Quality assurance

The assurance of in-use biodiesel fuel quality is of critical importance to guarantee that engine parts are not at risk and therefore engine warrantees permit the use of biodiesel. Hoekman and co-workers (2012:161) state that overall product quality is ensured by establishing fuel specifications, the establishment and enforcement of quality assurance programs, the defining of in-use handling guidelines and the applying of in-use surveys.

The establishment of biodiesel specifications forms the basis for quality control of biodiesel fuel. The ASTM D6751 specification of the United States and the EN 14214 specification of the European Union have an important influence on biodiesel specifications worldwide. Numerous countries have defined their own specifications and many of them derived their biodiesel specifications from the

ASTM D6751 or the EN 14214 specifications. In many countries the specifications are modified frequently as the standards evolve. Standards are also developed as national standards for specific countries taking into account circumstances such as climate related requirements in the specific countries. ASTM International also published a standard for biodiesel and diesel fuel oil blends which is the ASTM D7467 – 08 standard. The blending of biodiesel with petroleum diesel provides an opportunity to manage quality deficiencies of the biodiesel resulting from the unfavourable fatty acid composition of the primary feedstock.

2.3.2 Factors affecting the quality of biodiesel.

Van Gerpen (2003:6) listed four factors affecting the quality of biodiesel:

- Quality of the feedstock
- Production process
- Post production parameters
- Fatty acid composition of the primary feedstock

Feedstock quality in biodiesel production is considered one of the critical factors impacting the quality of biodiesel. Economic viability of biodiesel is being promoted by the employment of low quality fats and oils not fit for human and animal consumption as the feedstock cost of biodiesel makes up about 75% of the total production cost. The value of the low quality feedstock is less compared to edible oils as it contains impurities such as water and free fatty acids. Modified processes are needed to produce biodiesel that conforms to the biodiesel specifications.

The effectiveness of the biodiesel production process impacts directly on the biodiesel quality. Low quality feedstock brings about many challenges for the transesterification and purification processes to produce a product conforming to the specification.

Certain impurities present in the initial feedstock could be transferred to the final product when they are not removed during the separation stage and purification

step in biodiesel production. Mendow and co-workers (2011:864) studied the phosphorous balance through the production process where non-degummed vegetable oils were used. The phosphorous originates from the phospholipids and is removed by a process called degumming. Degumming is done by treating the oil with acids such as sulphuric or phosphoric acid. It was found that the largest amount of the original phosphorous was removed from the biodiesel through transfer to the glycerol phase, during the esterification step, done under acidic conditions and during the neutralisation and washing step when finally purified. When the production and purification parameters are not optimised, some of the phosphorous could end up in the final product. Sulphur originating from soaps used in cleaning applications where waste oils are collected and used as feedstock for biodiesel production could be carried through to the final biodiesel product.

The two most important impurities affecting the conventional homogeneous catalysed transesterification process, as reported by numerous researchers (Kusdiana *et al*., 2004; Tiwari *et al*., 2007; Sahoo *et al*., 2007; Ma *et al*., 1999) are water and free fatty acids. The formation of soap when the alkali catalyst reacts with the free fatty acids could weaken the catalytic activity, resulting in incomplete conversion and thus lower yields.

Incomplete conversion of triglycerides leaves mono- di- and triglycerides behind in the biodiesel. The specification for mono- di- and triglycerides in the SANS 1935 standard are 0.8, 0.2 and 0.2 weight% maximum, respectively. The value for total glycerol is made up of the mono- di- and triglycerides minus the free glycerol which should not exceed 0.25 mass fraction.

The formation of soap in the transesterification step causes emulsification during water washing resulting in impurities left behind in the biodiesel after purification. This could include impurities such as the soap itself, residual catalyst and free glycerine.

When hard water is used in the washing step, impurities such as calcium and magnesium are introduced in the biodiesel. The SANS 1935 specification for the group II metals (total Ca and Mg) is specified to be not higher than 5 mg/kg. Unreacted triglycerides in the presence of alkaline catalyst forms soap during the washing step that is carried forward to the final product.

During storage biodiesel could be exposed to oxygen, moisture and rust, causing peroxides to form. Further reactions result in polymerisation that could settle out and form of sludge-like deposits in the engine and filters.

The composition of the feedstock used for biodiesel production is considered the most important factor affecting the quality of biodiesel production. Parameters in the biodiesel standard such as oxidative stability, cloud point, iodine value, cold soak filterability, linoleic acid methyl ester content and polyunsaturated methyl ester content are directly influenced by the composition of the feedstock. Other parameters such as density, viscosity, acid value and distillation properties are influenced by reaction conditions, extent of the reaction, as well as the feedstock composition. The physico-chemical properties of biodiesel produced from the most used feedstock are shown in Table 2.2 (Sarin *et al*. 2007:1368; Berman *et al*. 2011:2864).

Table 2.2: Physico-chemical properties of biodiesel produced from the most used feedstock (Sarin *et al*., 2007: 1368, Berman *et al*., 2011: 2864).

Oil	Density	Viscosity	Cetane number	Flashpoint	Oxidative stability	Cloud point
Non-edible oil						
Jatropha	880	4.4	57.1	163	3.23	4
Karanja		4.16	55.1	141	2.35	4
Mahua	850	3.98	56.61	208		
Castor	899	15.17	48.9		1.1	-14
Edible oil						
Soy	884	4.0	58.1	160	3.8	4.0
Palm	876	4.5	54.6	135	13.37	16
Rapeseed	882	4.44	54.4	170	7.6	-3.3
Sunflower	880	4.1	55.6	180	1.73	4

Biodiesel produced from edible and non-edible vegetable oils used as feedstock for biodiesel production has very similar physico-chemical properties. The typical fatty acids are palmitic acid, stearic acid, oleic acid, linoleic acid and linolenic acid. Castor oil biodiesel is an exception with a viscosity of 15.17. The high viscosity is largely due to the hydrogen bonding of the hydroxyl groups in ricinoleic acid (Berman *et al*., 2011:2864). Castor oil contains approximately 83% ricinoleic acid. Palm oil shows a high cloud point attributed to the high percentage of saturated fatty acids and sunflower oil biodiesel is an example of a fuel with a low oxidative stability due to the high concentration of unsaturated fatty acid methyl esters. Table 2.3 and 2.4 shows some physico-chemical properties for biodiesel produced from different waste materials and by-products as published in the literature.

Table 2.3: Physico-chemical properties of biodiesel produced from waste materials and by-products

	Acid number (mg KOH/g, max)	Viscosity (mm^2/s)	Iodine Value (g I/g, max)	Density (kg/m^3)	Cloud point (°C)	Pour point (°C)	CFPP (°C max)	Reference
SANS 1935	0.5	3.5 – 5.0	140	860 – 900	-	-	-4 winter +3summer	
Acid waste lard	0.03-0.25	4.64-7.73	77	N	N	N	N	Dias *et al*., 2009:6358
Crude fish oil	1.17	7.2	N	860	N	N	N	Lin & Li, 2009:134
Chicken fat	0.25	6.25	130	867	-5	-6	N	Bhatti *et al*., 2008:2965
Mutton fat	0.65	5.98	126	856	-4	-5	N	Bhatti *et al*., 2008:2965
Vegetable oil soap stock	0.05	4.302	129	885	6	N	N	Haas, 2005:1092
Spent bleach earth	0.5	5.0	27	890	N	N	N	Huang & Chang, 2010:273
Palm fatty acid distillate	0.32	N	43	N	N	N	+15	Cho *et al*., 2012:278
Tobacco seed oil	0.3	4.23	136	888.5	N	N	-5	Usta *et al*., 2011:2034
Tall oil	N	7.1	N	922	1	N	-3	Altiparmak *et al*, 2007:243
Rubber seed oil	0.12	4.5	N	850	3	-5	N	Ramadhas *et al*., 2005:339
Leather industry waste	0.27	4.5	50.6	871.4	N	N	N	Ozgunay *et al*., 2007:1899
WCO	0.43	4.89	N	880	3	0	N	Phan&Phan, 2008:3495

Table 2.4: Physic-chemical properties of biodiesel produced from waste materials and by-products

	Flashpoint (°C min)	Cetane number (min)	Carbon residue (M fraction, max)	Sulphur (ppm max,)	Oxidative stability (Hours, min)	References (N = not reported)
SANS 1935	120	51	0.3	10	6	
Acid waste lard	N	N	N	N	N	Dias *et al*., 2009:6358
Crude fish oil	103	50.9 (index)	0.76	N	N	Lin & Li, 2009:134
Chicken fat	N	61	N	N	N	Bhatti *et al*., 2008:2965
Mutton fat	N	59.0	N	N	N	Bhatti *et al*., 20082965
Vegetable oil soap stock	169	51.3	0.01	0.0015%	N	Haas, 2005:1092
Spent bleach earth	168	61 (index)	0.05	<0.01	N	Huang &Chang, 2010:273
Palm fatty acid distillate	N	N	N	N	8.7	Cho *et al*., 2012:278
Tobacco seed oil	165.4	51.6	0.029	8	0.8	Usta *et al*., 2011:2034
Tall oil	89	54	N	5	N	Altiparmak *et al*., 2007:243
Rubber seed oil	120	N	N	N	N	Ramadhas *et al*., 2005:339
Leather industry waste	130	N	N	0.008	N	Ozgunay *et al*, 2007:1899
WCO	120	>47	N	N	>3	Phan & Phan, 2008:3495;Uzun *et al*., 2012:350

Acid waste lard was evaluated by Dias and co-workers (2009:6355) as feedstock for biodiesel production. The biodiesel quality was evaluated according to the EN 14214 standard. The blending of lard with soybean oil increased the iodine value from 77 to 115 g iodine per 100 g of FAME.

Huang and co-workers (2010:269) evaluated the production of biodiesel from residual oil recovered from spent bleaching earth. The properties of the clear yellow liquid were in reasonable agreement with the EN 14214 and ASTM D 6751 standards. The residual oil contained much more saturated fatty acids such as palmitic acid compared to refined vegetable oils which resulted in a higher cetane number and lower iodine number.

Crude fish oil from the soapstock of marine fish contains significantly more poly unsaturated fatty acids compared to waste cooking oil (Lin & Li, 2009:130). The oil contained 37.07% saturated fatty acids and 37.3 % long chain fatty acids in the range of C20 – C22. Polyunsaturated fatty acids with more than three double bonds have low oxidation stability causing precipitation of biodiesel components in the fuel system. Marine-fish oil was found to have higher acid numbers. The presence of water in the feedstock causes the fuel to have higher acid numbers and also the presence of a relatively high unsaturated fatty acid percentage. Lin and co-worker (2009:130) reported a viscosity of 7.2 mm^2/s at 40 °C which is consistent with other findings (Knothe 2005:1067). Longer chain lengths and increasing degree of saturation give higher viscosities. Marine fish soapstock also has a higher cetane index, higher carbon residue and lower flashpoint compared to that of waste cooking oil.

Bhatti and co-workers (2008:2964) concluded that mutton fat contains more saturated fatty acids compared to chicken fat, and a lower percentage of unsaturated fatty acids. The physical and fuel properties of the biodiesel were found to be comparable in general with the recommended properties.

Low value vegetable oil soapstock was used to improve the economics of biodiesel production by Haas (2005:1091). The soapstock was first saponified, followed by acidulation and esterification to obtain methyl esters. It was concluded that the higher cetane number of the palmitate ester in the soap-stock was counteracted by the cetane lowering impact of the slightly elevated

polyunsaturated fatty ester content. Haas (2005:1091) regarded the esterification route to be effective as shown by the low total glycerine value of 0.123 weight% indicating the high efficiency of the saponification reaction with very few acylglycerols remaining. The low acid number was an indication of the effectiveness of the wash protocol where the remaining free fatty acids had been removed.

Usta and co-workers (2011:2034) have evaluated tobacco seed oil as a feedstock for biodiesel production. The oxidation stability of the tobacco seed oil biodiesel was found to be lower than the specification of the EN 14214 standard and the iodine value was higher than the maximum limit of the standard. The low oxidation stability was expected given the higher concentration of unsaturated fatty acids whichwas also responsible for the high iodine value. Usta and co-workers (2011:2034) evaluated the effect of antioxidants to change the oxidation stability of the fuel. Pyrogallol was found to be the most effective antioxidant. Iodine value also depended directly on the fatty acid composition. Waste cooking oil containing more saturated fatty acids was used in blending with tobacco seed oil to decrease the iodine value to just below 120 g iodine per 100 g ester. The other parameters which were within the specification were ester content, carbon residue, sulphated ash content, water content, total contamination, acid value, methanol content, mono- di- and triglyceride content, free glycerol and total glycerol. The cold filter plugging point was increased by the addition of the waste cooking oil containing more saturated fatty acids, and therefore cold flow improvers such as octadecene–1-maleic anhydride copolymer at 0.5 weight% was added. It is also important to consider the effect of additives on any of the other parameters.

Altiparmak and co-workers (2007:246) altered the fuel properties of tall oil fatty acid methyl esters by blending it with diesel fuel. The effect of the fuel blends on the engine performance and exhaust emissions was evaluated. Advantages of tall oil fatty acid methyl ester–diesel fuel blends are that they contain low sulphur and aromatic contents. The cetane number is higher with a decrease in CO emissions.

Ramadhas and co-workers (2005:339) evaluated rubber seed oil containing high free fatty acids as feedstock for biodiesel production. Rubber seed oil contains a high percentage of unsaturated fatty acids with linolenic acid (C18:3) to be 16.3%.

Most of the fuel properties of rubber seed oil methyl esters were found to be comparable with those of petroleum diesel.

Ozgunay and co-workers (2007:1901) did a performance and emission study on biodiesel from leather industry pre-fleshings. Better emission values were expected as the biodiesel from pre-fleshings contained lower unsaturated fatty acids compared to biodiesel from rapeseed, canola and soybean oil. The properties of this biodiesel were comparable to biodiesel produced from tallow with higher cetane numbers.

Phan and Phan (2008:3495) produced biodiesel from waste cooking oil. The chemical and physical properties of the diesel using different sources for feedstock showed little difference. Most of the physical properties of the biodiesel were within specification, but the carbon residue was 4.0 weight%. The volatility of the biodiesel was reduced by mixing with petroleum diesel which also decreased carbon deposits. Pahn and Pahn (2008:3495) concluded that a B20 blend could be used in engines without major modification.

2.4 Conclusion

The biodiesel industry will experience increasing pressure to move away from using edible oil as feedstock fuelled by the rising price for edible oils and the food versus fuel debate.

- The increasing shortage of feedstock for biodiesel production will drive the search for more alternative feedstock sources in the decades to come.
- Alternative feedstock for biodiesel needs more research to better understand the effect of feedstock composition and the presence of certain substances in the feedstock on the production process and the quality of the biodiesel produced.
- Promising innovations on alternative feedstock sources such as bio-oil from microalgae and bio-oil from lignocelluloses biomass as feedstock

for oliganeous micro-organisms still need substantial research work to make them commercially viable.

- Hazardous industrial waste that contains lipids has not been evaluated yet as biodiesel feedstock.
- Multi-feedstock production of biodiesel offers a number of advantages including the reduction in dependency on a single feedstock which avoids price volatility.

2.5 References:

Ahmad, A.L., Yasin, N.H., Derek, C.J.C., Lim, J.K. 2011. Microalgae as a sustainable energy source for biodiesel production: A review. Renewable and sustainable Energy Reviews, 15: 584- 593.

Altiparmak, D., keskin, A., Koca, A., Guru, M. 2007. Alternative fuel properties of tall oil fatty acid methyl ester-diesel fuel blends. Bioresource Technology, 98: 241- 246.

ASTM, 2012. ASTM D6751-11b Standard Specification for Biodiesel Fuel blend Stock (B100) for Middle Distillate Fuels. http://www.astm.org/Standards/D6751.htm Date of access: Oct.2012.

Atadashi, I.M., Aroua, M.K., Aziz, A.A. 2011. Biodiesel separation and purification: A review. Renewable Energy, 36: 437- 443.

Atadashi, I.M., Aroua, M.K., Aziz, A.R.R., Sulaiman, N.M.n. 2011. Refining technologies for the purification of crude biodiesel. Applied Energy, 88: 4239- 4251.

Atadashi, I.M., Aroua, M.K., Aziz, A.A. 2010. High quality biodiesel and its diesel engine application: A review. Renewable and Sustainable Energy Reviews, 14: 1999- 2008.

Atadashi, I.M., Aroura, M.K., Aziz, A.R., Sulaiman, N.M.N. 2011. Membrane biodiesel production and refining technology: A critical review. Renewable and Sustainable Energy Reviews, 15: 5051- 5065.

Atadashi, I.M., Aroua, M.K., Aziz, A.R.A., Sulaiman, N.M.N. 2012. Production of biodiesel using high free fatty acid feedstock. Renewable and Sustainable Energy Reviews, 16: 3275- 3285.

Atabani, A.E., Silitonga, A.S., Badruddin, I.A., Mahlia, T.M.I., Masjuki, H.H., Mekhilef, S. 2012. A comprehensive review on biodiesel as an alternative energy resource and its characteristics. Renewable and Sustainable Energy Reviews, 16: 2070- 2093.

Balat, M. 2011. Potential alternatives to edible oils for biodiesel production- a review of current work. Energy Conversion and Management, 52: 1479- 1492.

Balat, M. Balat H. 2010. Progress in biodiesel processing. Applied Energy, 87: 1815- 1835.

Bankovic-Illic, I. B., Stamenkovic, O.S., Veljkovic, V.B. 2012. Biodiesel production from non-edible plant oils, Renewable and Sustainable Energy Reviews, 16: 3621- 3647.

Behcet, R. 2011. Performance and emission study of waste anchovy fish biodiesel in a diesel engine. Fuel Processing Technology, 92: 1187- 1194.

Berman, P., Nizri, S., Wiesman, Z. 2011. Castor oil biodiesel and its blends as alternative fuel. Biomass and Bioenergy, 35: 2861- 2866.

Berrios, M., Skelton, R.L. 2008. Comparison of purification methods for biodiesel. Chemical Engineering Journal, 144: 459- 465.

Bhatti, H.N., Hanif, M.A., Qasim, M., Rehman, A. 2008. Biodiesel production from waste tallow. Fuel, 87: 2961- 2966.

Boffito, D.C., Pirola, C., Galli, F., Di Michele, A., Bianchi, C.L. 2013. Free fatty acids esterification of waste cooking oil and its mixtures with rapeseed oil and diesel. Fuel, 108: 612- 619.

Borugadda, V.B., Goud, V.V. 2012. Biodiesel production from renewable feedstocks: Status and opportunities. Renewable and Sustainable Energy Reviews, 16: 4763- 4784.

Bhosle, B.M., Subramanian, R. 2005. New approaches in deacidification of edible oils- a review. Journal of Food Engineering, 69: 481- 494.

Brennan, L. Owende, P. 2010. Biofuels from micro-algae- a review of technologies for production, processing, and extractions of biofuels and co-products. Renewable and Sustainable Energy Reviews, 14: 557- 577.

Cao, P., Dube, M.A., Tremblay, A.Y. 2008. High purity fatty acid methyl ester production from canola, soybean, palm, and yellow grease lipids by means of a membrane reactor. Biomass and Bioenergy, 32: 1028- 1036.

Cayli, G., Kusefoglu, S. 2008. Increased yields in biodiesel production from used cooking oils a by two-step process: Comparison with one step process using TGA. Fuel Processing Technology, 89: 118- 122.

Chapagain, B.P., Yehoshua, Y., Wiesman, Z. 2009. Desert date (Balanites aegyptiaca) as an arid lands sustainable bioresource for biodiesel. Bioresource Technology, 100: 1221- 1226.

Cho, H.J., Kim, J., Hong, S.W., Yeo, Y. 2012. Development of a novel process for biodiesel production from palm fatty6 acid distillate (PFAD). Fuel processing Technology, 104: 271- 280.

Christi, Y. 2007. Biodiesel from microalgae. Biotechnology Advances, 25: 294-306.

Cantrell, K.B., Ducey, T., ro, K.S., Hunt, P.G. 2008. Livestock waste –to-bioenergy generation opportunities. Bioresource Technology, 99: 7941- 7953.

Demirbas, A. 2009. Progress and recent trends in biodiesel fuels. Energy Conversion and Management, 50: 14-34.

Demirbas, A. 2011. Methylation of wood fatty and resin acids for production of biodiesel. Fuel, 90: 2273- 2279.

Dias, J.M., Alvim-Ferraz, M.C.M., Almeida, M.F. 2009. Production of biodiesel from acid waste lard. Bioresource Technology, 100: 6355- 6361.

Di Serio, M., Tesser, R., Dimiccoli, M., Cammarota, F. Nastasi, M., Santacesaria, E. 2005. Synthesis of biodiesel via homogeneous Lewis acid catalyst. Journal of Molecular Catalysis A: Chemical, 239: 111- 115.

Dumont, M. Narine, S.S. 2007. Soapstock and deodoriser distillates from North American vegetable oils: Review on their characterisation, extraction and utilisation. Food Research International, 40: 957- 974.

Diya'uddeen, B.H., Aziz, A.R., Daud, W.M.A.W., Chakrabarti, M.H. 2012. Performance evaluation of biodiesel from used domestic waste oils: A review. Process Safety and Environment Protection, 90: 164- 179.

Enweremadu, C.C., Mbarawa, M.M. 2009. Technical aspects of production and analysis of biodiesel from used cooking oil- A review. Renewable and Sustainable Energy Reviews, 13: 2205- 2224.

Fernandez, C.M., Ramos, M.j., Perez, A., Rodriguez, J.F. 2010. Production of biodiesel from winery waste: extraction, refining and transesterification of grapeseed oil. Bioresource Technology, 101: 7091- 7024.

Gawrilow, L. 2003. Palm oil usage in lubricants. 3rd Global Oil and fats business Forum USA "Interfacing with the Global Oils and Fats Business" http://americanpalmoil.com/pdf/IIija%20Gawrilow.pdf.(2003) Date of access: Jan. 2012.

Ghadge, S.V., Raheman, H. 2005. Biodiesel production from mahua (Mahua indica) oil having high free fatty acids. Biomass and Bioenergy, 28: 601- 605.

Giannelos, P.N., Zannikos, F., Stouras, S., Lios, E., Anastopoulos, G. 2002. Tobacco seed oil as an alternative diesel fuel: physical and chemical properties. Industrial Crops and Products, 16: 1- 9.

Gomes, M.C.S., Pereira, N.C., de Barros, S.T.D. 2010. Journal of membrane Science, 352: 271- 276.

Gui, M.M., Lee, K.T., Bhatia, S. 2008. Feasibility of edible vs. non-edible vs. waste edible oil as biodiesel feedstock. Energy, 33: 1646- 1653.

Gunstone, F.D. 2013.Oils and fats in the market place: Biodiesel. http://lippidlibrary.aocs.org/market/biodiesel.htm Date of access: Apr. 2013.

Gunstone, F.D. 2013. Oils and fats in the market place: Prices of commodity oils http://lipidlibrary.aocs.org/market/prices.htm Date of access: Apr. 2013.

Haas, M.J. 2005. Improving the economics of biodiesel production through the use of low value lipids as feedstocks: vegetable oil soapstock. Fuel; Processing Technology, 86:1087- 1096.

Haas, M.J., McAloon, A.J., Yee, W.C., Foglia, T.A. 2006. A process model to estimate biodiesel production cost. Bioresource Technology, 97: 671- 678.

Helwani, Z., Othman, M.R., Aziz, N., Kim, J., Fernando, W.J.N. 2009. Solid heterogeneous catalysts for transesterification of triglycerides with methanol: A review. Applied Catalysis A: General, 363: 1- 10.

Hoekman, S.K., Broch, A., Robbins, C., Ceniceros, E., Natarajan, M. 2012. Review of biodiesel composition, properties, and specifications. Renewable and Sustainable Energy Reviews, 16: 143- 169.

Hossain, A.K., Badr, O. 2007. Prospects of renewable energy utilisation for electricity generation in Bangladesh. Renewable and Sustainable Energy Reviews, 11: 1617- 1649.

Huang, Y., Chang, J. 2010. Biodiesel production from residual oils recovered from spent bleaching earth. Renewable Energy, 35: 269- 274.

Issara, C., Kanokwan, K., Worapon, K., Navadol, L. 2011. Patent review on "Biodiesel Production Process". Recent Patents on Chemical Engineering,4: 265-279.

Jalapoor, M., Page, I. 2009. Physical refining process using adsorbent particles for the production of biodiesel fuel. (Patent: US 20090300972).

Karmakar, A., Karmakar, S., Mukherjee, S. 2010. Properties of various plants and animal feedstocks for biodiesel production. 101: 7201- 7210.

Knothe, G. 2005. Dependence of biodiesel fuel properties on the structure of fatty aid alkyl esters. Fuel Processing Technology, 86: 1059- 1070.

Knothe, G., Steidley, K.R. 2009. A comparison of used cooking oils: A very heterogeneous feedstock for biodiesel production. Bioresource Technology, 100: 5796- 5801.

Koh, M.Y., Ghazi, T.I.M. 2011. A review of biodiesel production from Jatropha curcas L. oil. Renewable and Sustainable Energy Reviews, 15: 2240- 2251.

Kumar, A. Sharma, S. 2011. Potential non-edible oil sources as biodiesel feedstock: An Indian perspective. Renewable and Sustainable Energy Reviews, 15: 1791- 1800.

Kusdiana, D. Saka, S. 2004. Effects of water on biodiesel fuel production by supercritical methanol treatment. Bioresource Technology, 91: 289- 295.

Lam, M.K., Lee, K.T. 2013. Catalytic transesterification of high viscosity crude microalgae lipid to biodiesel: Effect of co-solvent. Fuel processing Technology. (Article in press).

Leung, D.Y.C., Wu, X., Leung, M.K.H. 2010. A review on biodiesel production using catalysed transesterification. Applied energy, 87: 1083- 1095.

Lim, S., Teong, L.K. 2010. Recent trends, opportunities and challenges of biodiesel in Malaysia: An overview. Renewable and Sustainable energy Reviews, 14: 938- 954.

Lin, C., Li, R. 2009. Fuel properties of biodiesel produced from crude fish oil from soapstock of marine fish. Fuel Processing Technology, 90: 130- 136.

Lin, L. Ying, D., Chaitep, S., Vittayapadung, S. 2009. Biodiesel production from crude rice bran oil and properties as fuel. Applied Energy, 86: 681- 688.

Lin, L., Cunshan, Z., Vittayapadung, S., Xiangqian, S., Mingdong, D. 2011. Opportunities and challenges for biodiesel fuel. Applied Energy, 88: 1020- 1031.

Ma, F., Hanna, M.A. 1999. Biodiesel production: A review. Bioresource Technology, 70: 1- 15.

Manzano-Agugliaro, F., Sanchez-Muros, M.J., Barroso, F.G., Martinez-Sanchez, A., Rojo, S. Perez-Banon, C. 2012. Insects for biodiesel production. Renewable and Sustainable Energy Reviews, 16: 3744- 3753.

Meher, L.C., Sagar, D.V., Naik, S.N. 2006. Technical aspects of biodiesel production by transesterification – A review. Renewable and Sustainable Energy Reviews, 10: 248- 268.

Mendow, G., Monella, F.C., Pisarello, M.L., Querini, C.A. 2011. Biodiesel production from non-degummed vegetable oils: Phosphorous balance through the process. Fuel Processing Technology, 92: 864- 870.

Meng, X. Yang, J., Xu, X., Zhang, L., Nie, Q., Xian, M. 2009. Biodiesel production from oleaginous microorganisms. Renewable Energy, 34: 1- 5.

Miao, X., Wu, Q. 2006. Biodiesel production from heterotrophic microalgal oil. Bioresource Technology, 97: 841- 846.

Mkentane, L. 2008. Reused cooking oil poses health threat. Weekend Post, August 02.

Mondala, A., Liang, K., Toghiani, H., Hernandez, R., French, T. 2009. Biodiesel production by in situ transesterification of municipal primary and secondary sludges. Bioresource Technology, 100: 1203- 1210.

Morshed, M., Ferdous, K., Khan, M.R., Mazumder, M.S.I., Islam, M.A., Uddin, M.T. 2011. Rubber seed oil as a potential source for biodiesel production in Bangladesh, Fuel, 90: 2981- 2986.

Mudge, S.M. 1995. Deleterious effects from accidental spillages of vegetable oil. SpillScience& Technology Bulletin, 2: 187- 191.

Naik, M., Meher, L.C., Das, L.M. 2008. Production of biodiesel from high free fatty acid Karanja (Pongamia pinnata) oil. Biomass and Bioenergy, 32: 354- 357.

Oh, P.P., Lau, H.L.N., Chen, J., Chong, M.F., Choo, Y.M. 2012. A review on conventional technologies and emerging process intensification (PI) methodsfor biodiesel production. Renewable and Sustainable Energy Reviews, 16:5131-5145.

Ozgunay, H., Colak, S., Zengin, G., Sari, O., Sarikahya, H., Yuceer, L. 2007. Performance and emission study of biodiesel from leather industry pre-fleshings. Waste Management, 27: 1897- 1901.

Pandey, R.A., Sanyal, N., Chattopadhyay, N., Kaul, S.N. 2003. Treatment and reuse of wastes of vegetable oil refinery. Resources, Conservation and Recycling, 37: 101- 117.

Panoutsou, C. 2007. Socio-economic impacts of energy crops for heat generation in Northern Greece.Energy Policy 35:6046- 6059.

Peng, B., Shu, Q., Wang, J. Wang, G., Wang, D., Han, M. 2008. Biodiesel production from waste oil feedstocks by solid acid catalysis. Process Safety and Environment Protection, 86: 441- 447.

Phan, A.N., Phan, T.M. 2008. Biodiesel production from waste cooking oils. Fuel, 87: 3490- 3496.

Qian, J., Shi,H., Yun, Z. 2010. Preparation of biodiesel from Jatropha curcas L. oil produced by two-phase solvent extraction. Bioresource Technology, 101: 7025- 7031.

Ramadhas, A.S., Jayaraj, S., Muraleedharan, C. 2005. Biodiesel production from high FFA rubber seed oil. Fuel, 84: 335- 340.

Robles-Medina, A., Gonzalez-Moreno, P.A., Esteban-Cerdan, L., Molina-Grima, E. 2009. Biocatalysis: Towards ever greener biodiesel production. Biotechnology Advances, 27: 398- 408.

Rosillo-Calle, F., Pelkmans, L., Walter, A. 2009. A global overview of vegetable oils, with reference to biodiesel. A report for the IEA Bioenergy Task 40. http://www.bioenergytrade.org/downloads/vegetableoilstudyfinaljune18.pdf Date of access: Oct. 2012.

Sabudak, T., Yildiz, M. 2010. Biodiesel production from waste frying oils and its quality control. Waste Management, 30: 799- 803.

Sahoo, P.K., Das, L.M., Babu, M.K.G., Naik, S.N. 2007. Biodiesel development from high acid value polanga seed oil and performance evaluation in a CI engine. Fuel, 86: 448- 454.

Sanchez, E., Ojeda, K., El-Halwagi, M., Kafarov, V. 2011. Biodiesel from microalgae oil production in two sequential esterification/transesterification reactors: Pinch analysis of heat integration. Chemical Engineering Journal, 176-177: 211- 216.

Santori, G., Nicola, G.D., Moglie, M., Polonara, F. 2012. A review analysing the industrial biodiesel production practice starting from vegetable oil refining. Applied Energy, 92: 109- 132.

Sarin, R., Sharma, M., Sinharay, S., Malhotra, R.K. 2007. Jatropha-Palm biodiesel blends: An optimum mix for Asia. Fuel, 86: 1365- 1371.

Sarmento, L.A.V., Spricigo, C.B., Petrus, J.C.C., Carlson, L.H.C., Machado, R.A.F. 2004. Performance of reverse osmosis membranes in the separation of supercritical CO2 and essential oils. Journal of Membrane Science, 237: 71- 76.

Shahid, E.M., Jamal, Y. 2011. Production of biodiesel: A technical review. Renewable and Sustainable Energy Reviews, 15: 4732- 4745.

Siddiquee, M.N., Rohani, S. 2011. Experimental analysis of lipid extraction and biodiesel production from waste water sludge. Fuel Processing Technology, 92: 2241- 2251.

Singh, A. He, B., Thompson, J., Van Gerpen, J. 2006. Process optimizationof biodiesel productionusing alkaline catalysts. Applied Engineering in Agriculture, 22: 597- 600.

Soriano, N.U., Vendetti, R.., Argyropoulos, D.S. 2009. Biodiesel synthesis via homogeneous Lewis acid-catalyzed transesterification. Fuel, 88 560- 565.

Srivastava, A., Prasad, R. 2000. Triglyceride-based diesel fuels. Renewable and Sustainable Energy Reviews, 4: 111- 133.

Taher, H., Al-Zuhair, S., AlMarzouqui, A., Hashim, I. 2011. Extracted fat from lamb meat by super critical CO_2 as feedstock for biodiesel production. Biochemical Engineering Journal, 55: 23- 31.

Tan, K.T., Lee, K.T., Mohamed, A.R. 2010. Effects of free fatty acids, water content and co-solvent on biodiesel production by supercritical methanol reaction. The Journal of Supercritical Fluids, 53: 88- 91.

Ting, W., Huang, C., Giridhar, N., Wu, W. 2008. An enzymatic/acid-catalyzed hybrid process for biodiesel production from soybean oil. Journal of the Chinese Institute of Chemical Engineers, 39: 203- 210.

Tiwari, A.K., Kumar, A., Raheman, H. 2007. Biodiesel production from jatropha oil (jatropha curcas) with high free fatty acids: An optimized process. Biomass and Bioenergy, 31: 569- 575.

Toeneboebn, G.J., Welsh, N.A. 1994. Adsorptive removal of sulphur compounds from fatty materials. (Patent: US 5298638).

USDA. 2012. Foreign Agricultural Services. Table 06: Major Vegetable Oils: World Supply and Distribution (Country View). http://www.fas.usda.gov/psdonline /circulars/oilseeds.pdf Date of access: 25 Apr. 2013.

Usta, N., Aydogan, B., Con, A.H., Uguzdogan, E., Ozkal, S,G. 2011. Properties and quality verification of biodiesel produced from tobacco seed oil. Energy Conversion and Management, 52: 2031- 2039.

Van Gerpen, J. 2003. Biodiesel Production and Fuel quality. Journal of American Oil Chemists Society, 1: 97- 102.

Van Gerpen, J. 2005. Biodiesel processing and production. Fuel Processing Technology, 86: 1097- 1107.

Wang, R., Hanna, M.A., Zhou, W., Bhadury, P.S., Chen, Q., Song, B. 2011. Production and selected fuel properties from promising non-edible oils: Euphorbia lathyris L., Sapium sebiferum L., and Jatropha curcas L., Bioresource Technology, 102: 1194- 1199.

Wang, Y., Wang, X., Liu, Y., Ou, S., Tan, Y., Tang, S. 2009. Refining of biodiesel by ceramic membrane separation. Fuel Processing Technology, 90: 422- 427.

Yan, J., Li, A., Xu, Y., Ngo, T.P.N., Phua, S., Li, Z. 2012. Efficient production of biodiesel from waste grease: One-pot esterification and transesterification with tandem lipases. Bioresource Technology, 123: 332- 337.

Yousuf, A. 2012. Biodiesel from lignocellulosic biomass – Prospects and challenges. Waste Management, 32: 2061- 2067.

Zhang, Y. Dube, M.A., McLean, D.D., Kates, M. 2003. Biodiesel production from waste cooking oil: 1. Process design and technological assessment. Bioresource Technology, 89: 1- 16.

Zhang, J., Jiang, L. 2008. Acid-catalyzed esterification of Zanthoxylum bungeanum seed oil with high free fatty acids for biodiesel production. Bioresource Technology, 99: 8995- 8998.

Zheng, L., Hou, Y., Li, W., Yang, S., Li, Q., Yu, Z. 2013. Exploring the potential of grease from yellow mealworm beetle (Tenebrio molitor) as a novel biodiesel feedstock. Applied Energy, 101:618- 621.

Zullaikah, S., Lai, C., Vali, S.R., Ju, Y. 2005. A two-step acid catalysed process for the production of biodiesel from rice bran oil. Bioresource Technology, 96: 1889- 1896.

Chapter 3 – Experimental

The experimental work conducted in this study is described in this chapter.

Section 3.1 describes the materials used. Section 3.1.1 provides a list of chemicals used and section 3.1.2 describes the origin of the feedstock used in this study. In section 3.1.2.1 the sampling methods and sample preparation are described and in section 3.1.2.2 the characteristics of the feedstock are provided and discussed. The analytical methods followed and analytical equipment used are described in section 3.2. The process routes to produce biodiesel from the feedstock are explained in section 3.3, including the experimental set-up and the optimisation of the experimental conditions.

3.1 Materials

3.1.1 Chemicals used

Table 3.1 provides a list of all the chemicals used in this study.

3.1.2 Feedstock

Used cold rolling lubricant from a steel mill in the Gauteng Province of South Africa was identified as a potential feedstock for biodiesel production. This used lubricant is referred to as waste process grease (WPG) in this study. The lubricant is only used once in the rolling process after which it is disposed of by professional waste treatment companies or by selling it to oil collectors from where it is further utilised in industry. Further utilisation could involve blending with other used lubrication oils and applied as low grade furnace fuel.

Table 3.1: Chemicals used for feedstock preparation, biodiesel production, purification and analysis

Component	Purity	Supplier	Purpose
Sulphuric acid	98%	Merck Chemicals	Catalyst
Hydrochloric acid	32%	ACE*	Acidulation agent
Sodium hydroxide	98%	ACE*	Saponification agent
Sodium sulphate	99% min	ACE*	Drying agent
Methanol	99.5%	Rochelle Chemicals	Reagent/mobile phase/extractant
Ethanol	99.9%	Rochelle Chemicals	Extractant
Iso-propanol	99.5%	Rochelle Chemicals	Extractant
Hexane	95% min	ACE*	Mobile phase
Acetonitrile	➢ 99.9%	Merck Chemicals	Extractant
Sodium methylate	30% in methanol	BASF	Catalyst
Purolite PD-206	-	Purolite	Cation exchange resin
Silica gel	-	Merck Chemicals	Column Stationary phase
Purolite A860	-	Purolite	Anion exchange resin
Purolite MN 200		Purolite	Hydrophobic resin
Potassium hydroxide	1 M	Sigma Aldrich	Acid value analysis
BF_3/methanol	10% in methanol		Catalyst
Methyl nonanoate	97%	Sigma Aldrich	Internal GC standard
Dichloromethane	99%	Sigma Aldrich	Solvent – GC
3Hydranal	-	Sigma Aldrich	Water analysis (KFC)

*ACE: Associated Chemical Enterprises

3.1.2.1 Sampling and preparation of waste process grease

After usage, waste process grease is circulated in a drying plant at the metalworking facility to reduce moisture levels to approximately 4 to 8% prior to disposal. A 200 L sample of waste process grease was drawn from the drying plant and while still warm divided into 10 by 20 L containers after proper mixing each time. The waste process grease was dried to a moisture content of 0.3% prior to experimental work by slowly heating the grease to 110 °C while stirring. The removal of water from WPG should be done very carefully. The reduction of the moisture content of the WPG from about 8% to 0.3% by slowly heating the grease on a hot plate with vigorous stirring was achieved in about 30 to 45 minutes. The temperature was controlled not to go higher than 110 °C in order to prevent any unwanted reactions. The reduction of moisture levels is critical for those experiments where conventional esterification and transesterification reactions are done (Ma & Hanna 1999:1; Kusdiana *et al*., 2004: 289).

3.1.2.2 Characterisation of waste process grease

A cold rolling lubricant or process grease consists of base oil containing various additives. The base oil is usually palm oil or tallow or mixtures thereof. The additive packages are proprietary information and belong to the companies supplying these lubricants. Rolling oils contain high molecular weight non-ionic surfactants incorporated with the cold rolling oil as emulsifying and dispersing agents (Shiraishi et al., 1992). The different non-ionic surfactants have different HLB (hydrophilic/lipophilic balance) values indicating the difference in solubility of such a surfactant molecule in either the polar or non-polar part of a solvent system.

Process grease is exposed to extreme conditions in a metalworking plant which results in changes in the composition of the grease. The grease is mixed with large quantities of water and during the rolling process free fatty acids are formed where the shearing, high forces and high temperatures breaks down the triglycerides. The free fatty acids also bind the free iron to form iron soaps during

the rolling process which act as a lubrication agent. It is expected that waste process grease will contain large quantities of metals, free fatty acids and moisture, as well as various foreign compounds introduced via the addition of additive packages to the base oil during formulation. In this study waste process grease was analysed for free fatty acids, moisture content, fatty acid distribution, viscosity at 40 °C, oil content, metal content and other elements such as sulphur and phosphorous. An average compositional analysis and the standard deviation of each property analysed of the waste process grease is presented in Table 3.2. Analysis for FFA, viscosity, moisture, metals, sulphur and phosphorous were done in triplicate and the GC analysis of fatty acids were done in duplicate.

Table 3.2: Compositional analysis of WPG

Property	Value ± δ	Units	Method
FFA	43.2 ±0.28	wt%	SANS 54104
Viscosity at 40 °C	33.0 ± 0.36	mm^2/s	ISO 3104
Moisture content	0.3 ± 0.004	wt%	ISO 12937
Fe	3232 ± 40.43	ppm	ASTM 5185
Ca	1124 ± 3.61	ppm	ASTM5185
Mn	64 ± 1.73	ppm	ASTM 5185
Mg	88 ± 2.08	ppm	ASTM 5185
S	8956 ± 300.88	ppm	ASTM 5185
P	721 ± 13.85	ppm	ASTM5185
Fatty acids			
Lauric acid (C12:0)	0.73 ± 0.37	wt %	EN 14103
Myristic acid (C14:0)	1.97 ± 0.32	wt %	EN 14103
Palmitic acid (C16:0)	33.50 ± 1.12	wt%	EN 14103
Stearic acid (C18:0)	7.77 ± 0.30	wt %	EN 14103
Total saturated	43.97 ± 2.11	wt%	EN 14103
Palmitoleic acid (C16:1)	0.83 ± 0.08	wt%	EN 14103
Oleic acid (C18:1)	49.23 ± 0.73	wt %	EN 14103
Linoleic acid (C18:2)	5.99 ± 0.42	wt %	EN 14103
Total unsaturated	56.05 ± 1.23	wt %	EN 14103
Total fatty acids, (Oil content) *	64.04 ± 4.24	wt %	EN 14103

*Total fatty acids as mass percentage of the dried WPG

Extreme process conditions and the presence of water in the metalworking process result in many of the triglycerides of the base oil being hydrolysed to form free fatty acids, explaining the high FFA value of 43.2%. The oil content was calculated as the total mass of fatty acids quantified with gas chromatography, as a percentage of the total mass of dried waste process grease. The oil content was measured as 64.04 % with the balance made up of impurities. WPG contains a relatively high percentage of saturated fatty acids, predicting less favourable cold flow properties with good oxidative stability. The metal impurities are being introduced into the waste grease through the rolling process and the sulphur and phosphorous compounds through the additive package as part of the formulation of the process grease.

3.2 Analyses

3.2.1 Fatty acid analysis

Fatty acid analyses were based on the test method EN 14103 and performed using an Agilent 7890A gas chromatograph fitted with a flame ionisation detector. In the sample preparation, the fatty acids were derivatised and the weight percentage of the methyl esters was determined. The gas chromatograph was calibrated using fatty acid methyl ester standards acquired from Sigma Aldrich. Methyl nonanoate was used as the internal standard.

The calibration procedure for the gas chromatograph is discussed in Appendix A. The gas chromatographic method used, is summarised in Table 3.3.

Table 3.3: Method for gas chromatography analysis

Parameter	Item/value
Column	Agilent HP-88 (100 m length, 0.25 mm inside diameter, 0.2 µm film thickness)
Carrier gas	Hydrogen
Linear velocity	35 cm/s
Inlet	Split/splitless
Split ratio	1/10
Injection volume	1µL
Inlet temperature	250 °C
Detector temperature	350 °C
Head pressure	447.12 kPa
Oven temperature program	120 °C for one minute Ramp at 10 ° C/minute to 175 °C, hold for 10 minutes Ramp at 5 °C/minute to 210 °C, hold for 5 minutes Ramp at 5 °C/ minute to 230 °C, hold for 5 minutes
Detector	Flame Ionisation detector
Detector gas flows	Hydrogen: 40 mL/minute; Air: 400 mL/minute
Solvent for needle wash	Dichloromethane
Internal standard	Methyl nonanoate

The preparation of dried waste process grease for fatty acid analysis was based on the method described by Abdulkadir and Tsuchiya (2008:4). A 300 mg dry sample of oil was weighed into a 25 mL reaction vessel and 24 mg of BF_3-methanol complex was added followed by 2 mL of 2,2-dimethoxypropane as water scavenger. The water scavenger removes the water forming during the reaction to drive the reaction to the right and obtain a high ester yield. The reaction mixture was heated to 60 °C for 120 minutes after which 2 mL of water and 2 mL of hexane was added. The reaction vessel was then shaken to transfer all the esters into the non-polar solvent. The upper organic layer was then removed and dried over sodium sulphate, filtered and taken for analysis. This sample was diluted with dichloromethane and consisted of 150 µl of derivatised sample, 40 µl of methyl nonanoate as internal standard and 800 µl of dichloromethane.

To calculate the oil content of the process grease, the total mass of fatty acid methyl esters was determined by the gas chromatographic method. The total mass of the fatty acids present in the sample was calculated, taking into account the molecular mass, and expressed as a percentage of the total mass of dried WPG. During derivatisation the free fatty acids were esterified to methyl esters and the remaining triglycerides were transesterified to methyl esters to be measured as total methyl esters. The total saturated fatty acid were calculated as mass percentage of total mass of fatty acids and the total unsaturated fatty acids as mass percentage of total mass of fatty acids. Fatty acid analysis were done in dublicate.

3.2.2 Elemental analysis

The metals, sulphur and phosphorous in the WPG were analysed by Wearcheck Africa which is an ISO 9001 and ISO 14000 registered company. The analyses were done in triplicate by means of ICP-AES (Inductively Coupled Plasma Atomic Emission Spectroscopy) based on the ASTM 5185 test method. Fluiden 1520, which is also known as a white spirits, supplied by ENGEN, was used to dilute and feed the sample. The dilution ratio of oil to solvent was 1:10. This analysis method can tolerate particles sizes up to 6 micrometres.

3.2.3 Other analyses

The moisture content of the WPG was determined in triplicate using Karl Fischer coulometry according to the ISO 12973 test method and the free fatty acids were measured in triplicate with a 828 Titrino Plus Methrom titrator, using potassium hydroxide as titrate (SANS 54104). The viscosity of the WPG was measured by Wearcheck, according to the ISO3104 test method.

3.2.4 Biodiesel analysis

Analysis of ester content and linoleic acid mass fraction was based on test method EN 14103 as discussed in section 3.2.1. Methanol content was determined using gas chromatography based on the EN 14110 test method. Iodine value (SANS 54111), sulphated ash (ISO 3987), flashpoint (ISO 2917), cold filter plugging point (SANS 50116) and total and free glycerol (AOCS Ca 14-56) were tested by Bioservices cc, South Africa, using the test methods mentioned. Copper strip corrosion was measured following the ISO 2160 test method and oxidative stability according to the SANS 54112 test method using the 837 Biodiesel Rancimat from Methrom. Density (ISO 12185) and cetane number (ISO 5165) were measured with an Eraspec infrared instrument from Euralytics.

Experiments were done in triplicate and the overall experimental error associated with these experiments was calculated to be 12.5% at a 95% confidence level.

3.3 Biodiesel production

In the selection of the technology to produce biodiesel, the quality of the intended feedstock plays an important role. Impurities in the feedstock could have a detrimental effect on the quality of the biodiesel by interfering with the production process or being carried forward to the final product.

In this study, three production routes were investigated, utilising WPG for biodiesel production:

a) The conventional two-step process

b) Free fatty acid extraction and biodiesel production

c) Modified process with solid phase extraction

The conventional biodiesel production process was selected in this study, as this process is the most widely used process all over the world for the production of biodiesel. The conventional process is uncomplicated and is performed at relatively low temperatures and at atmospheric pressure. Catalysts like sulphuric

acid and sodium methoxide are also commonly used, as these catalysts are relatively inexpensive. Results from the conventional process using WPG as feedstock provided critical information regarding further action needed to produce acceptable quality biodiesel from WPG.

The unit operations of the different routes are shown in Figure 3.1.

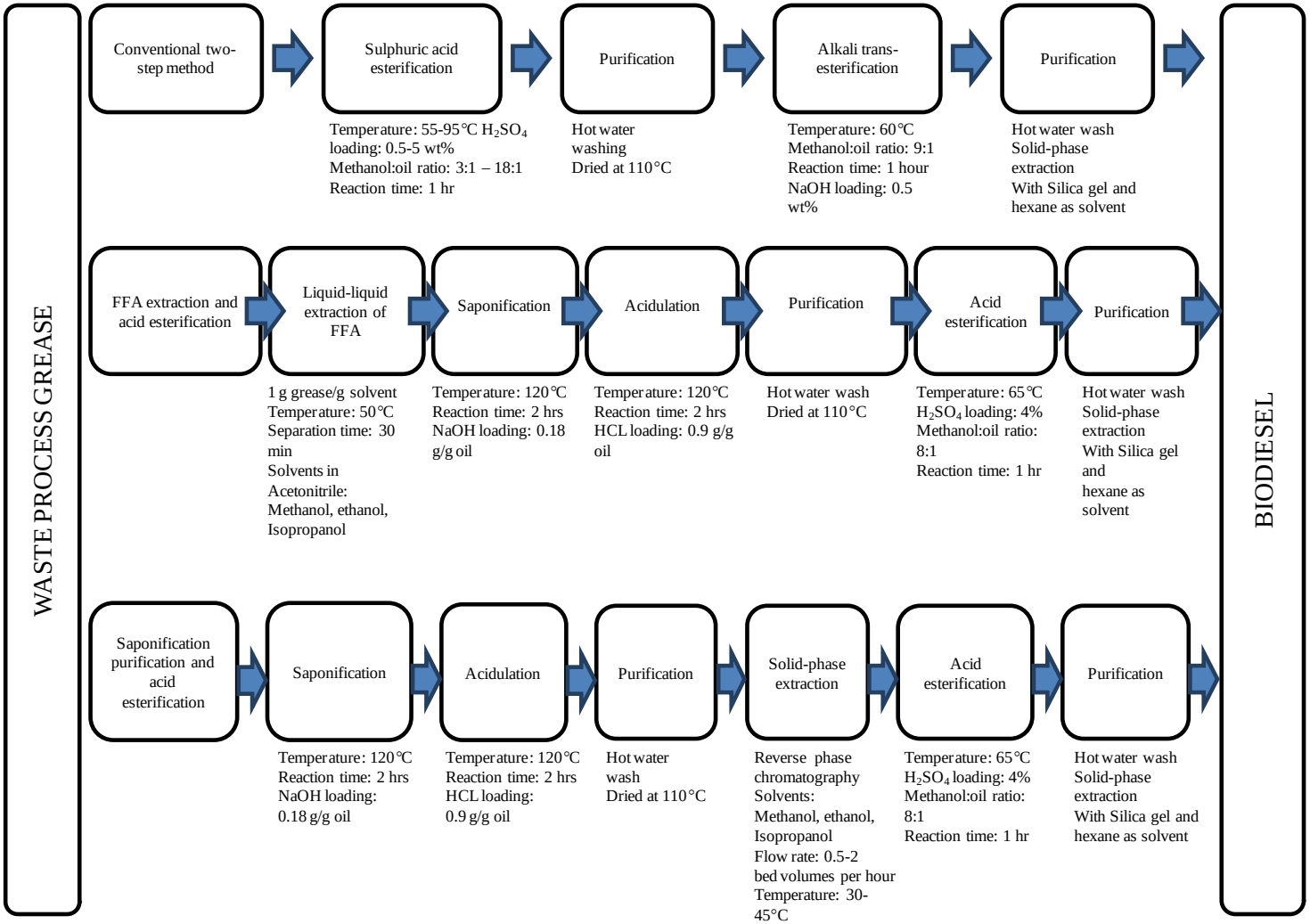

Figure 3.1: Unit operations in production routes for biodiesel production used in this study

3.3.1 Conventional two-step process (route 1)

The objective with route 1 was to evaluate the alkali catalysed transesterification process for biodiesel production using waste process grease as feedstock.

As shown in Figure 3.1, the first unit operation of the two-step process, pre-treatment, involves acid esterification where the free fatty acids were reduced to less than 1% using sulphuric acid as catalyst as suggested by Tiwari and co-workers (2007:569) who optimised the process of producing biodiesel from jatropha oil containing high free fatty acids. The acid esterification step was followed by a purification step where the reaction mixture was washed with hot distilled water and dried at 110 °C on a hot plate. The dried neutralised grease was next transesterified using sodium methoxide as alkaline catalyst. The crude biodiesel was then purified by means of water washing with hot distilled water followed by a further purification step using solid phase extraction with silica gel as stationary phase and hexane as the mobile phase.

3.3.1.1 Experimental set-up

The WPG was filtered using a Buchner funnel prior to drying on a hot plate at 110 °C. A three-neck 500 mL round-bottom flask fitted with a reflux condenser and thermometer was used as a batch reactor. An oil bath heated on a hot plate was used for heating the reactor contents, that was stirred using a magnetic stirrer. Stirring was done fast enough to overcome mass transfer limitations during biodiesel production. The temperature of the reaction was controlled by controlling the temperature of the oil bath.

Samples were drawn from the batch reactor at different time intervals, using a 25 mL pipette after which the sample was transferred to a 50 mL separation funnel. Samples were washed with 150 mL hot water, transferred to a 50 mL beaker, heated in an oven and cooled to allow the water to separate from the oil. After cooling the process grease was dried over sodium sulphate and stored for analysis at 2 °C in a refrigerator.

3.3.1.2 Optimisation of reaction conditions for pre-treatment of WPG

Srivastava and Prasad (2000:126) emphasized that reaction temperature, alcohol to oil ratio, catalyst type, mixing intensity and purity of reactants are the most important variables effecting the conversion and reaction time of the transesterification reaction. Similar variables are also important for the acid esterification reaction as shown by the optimisation of the acid pre-treatment experiments done by various researchers. Table 3.4 shows the results obtained by some of the researchers. The results in Table 3.4 were used as background to choose the range to be investigated for the different parameters in this study.

Table 3.4: Optimal reaction conditions for the pre-treatment of FFA in non-edible feedstock for biodiesel production as determined by various researchers

Feedstock	Acid esterification reaction conditions				FFA reduction (from – to)	Reference
	Catalyst/ Concentration	Temperature (°C)	Duration (min)	MeOH:Oil Molar ratio		
Zanthoxylum bungeanum seed oil	H_2SO_4 2 wt %	60	80	24:1	22.75% - 0.58%	Zhang and Jiang, 2008:8995
Rubber seed oil	H_2SO_4 0.5 v %	45±5	30	6:1	17% - <2%	Ramadhas *et al.*, 2005:335
Kusum	H_2SO_4 1 v %	50±0.5	60	10:1	10.65% - 0.47%	Sharma Y.C., 2010:1470
Jatropha curcas	H_2SO_4 1.43 v %	60	88	0.28 v/v	14% - 1%	Tiwari *et al.*, 2007:569
Tobacco seed oil		60	25	18:1	17% - <2%	Veljkovic *et al.*, 2006:2671
Salmon oil	H_2SO_4 1 wt %	52±2	60	9.1:1	6% - 1.5%	El-Mashad *et al.*, 2008:220
Karanja/Mahua seed oil	H_2SO_4 1.5 v %	55±0.5	60	6:1	10.8% - 1.035%	Sharma and Singh, 2010:1267
Crude Jatropha seed oil	H_2SO_4 1 wt %	50	60	0.6 w/w	15% - <1%	Berchmans and Hirata, 2008:1716

In this study the optimum conditions for the acid-catalysed esterification of WPG were determined by studying the reduction in FFA at different temperatures, molar ratios of methanol to oil and catalyst concentrations during a one hour reaction.

The effect of temperature on the reduction of FFA was studied from 55 to 65 °C for the reaction mixture with a methanol to oil ratio of 8:1 and a sulphuric acid concentration of 3 weight%.

The effect of molar ratio of methanol to oil on the reduction of FFA was studied at ratios from 3:1 to 18:1 at an at a reaction temperature of 65 °C with a sulphuric acid concentration of 3 weight%.

The effect of catalyst concentration on the reduction of FFA was studied at concentrations of 0.5 to 5 weight% at a reaction temperature of 65 °C and a molar ratio of methanol to oil of 8:1.

All experiments were done in triplicate and the experimental data are shown in tables B1, B2, B3 and B4 in Appendix B.

3.3.1.3 Selection of alkaline transesterification reaction conditions

Optimisation of reaction conditions for alkaline transesterification as conducted by several researchers is shown in Table 3.5. Reaction temperature has a strong influence on the rate of the reaction but if allowed enough time, the reaction will proceed to near completion at lower temperatures. Most often, the reaction is conducted at 60 to 70 °C which is close to the boiling point of methanol at atmospheric pressure. The maximum product yield is expected at temperatures between 45 and 65 °C. Both esterification and transesterification are equilibrium reactions. To shift the reaction to the right, a molar ratio of alcohol to oil of 3:1 that is the stoichiometric ratio, should be exceeded. Thus, either a large excess of methanol should be used or one of the reaction products should be removed from the reaction medium. The molar ratios used by the researchers varied between 5:1 and 9:1. Molar ratios that were too high interfered with the separation of glycerol from the esters. The most effective alkaline catalysts are alkali metal alkoxides, compared to acid catalysts. Alkaline catalysts are used in the concentration range of 0.5 to 1.4% by weight achieving a yield of esters of 90 to 99%. Too high a concentration of catalyst would not increase the yield more, but adds to the cost to remove the catalyst from the crude product. The mixing rate during the reaction is

important to ensure mass transfer. Mixing is more important in the beginning of the reaction as the methanol and oil form a two phase system, preventing proper mixing. As more and more methyl esters are formed, they act as a mutual solvent, creating more of a single phase system that results in mixing becoming less critical (Srivastava and Prasad, 2000:127).

Table 3.5: Optimal reaction conditions for the transesterification step of non-edible feedstock for biodiesel production as determined by various researchers

Feedstock	Alkaline transesterification reaction conditions				Yield (%)	Reference
	Catalyst/ Concentration	Temperature (°C)	Duration (min)	MeOH:Oil Molar ratio		
Zanthoxylum bungeanum seed oil	KOH 0.9 wt %	60	90	6.5:1	98	Zhang and Jiang, 2008:8995
Rubber seed oil	NaOH 0.5 %	45±5	30	9:1	98	Ramadhas *et al.*, 2005:335
Kusum	KOH 0.7 wt %	50±0.5	60	8:1	95	Sharma, 2010:1470
Jatropha curcas	KOH 0.55 wt %	60	24	5:1	99	Tiwari *et al.*, 2007:569
Tobacco seed oil	KOH 1 wt %	60	30	6:1	91	Veljkovic *et al.*, 2006:2671
Salmon oil	KOH 0.8 wt %	52±2	30	4.7:1	97	El-Mashad *et al.*, 2008:220
Karanja/Mahua seed oil	KOH 0.8 wt %	55±0.5	60	8:1	94	Sharma and Singh, 2010:1267
Crude Jatropha seed oil	NaOH 1.4 wt %	65	120	0.24 w/w	90	Berchmans and Hirata, 2008:1716

In this study the alkaline transesterification of WPG was done at a molar ratio of 9:1, a sodium methoxide loading of 0.5 weight%, and a temperature of 60 °C based on the method proposed by Ramadhas and co-workers (2005:335).

Based on the optimum reaction conditions for the esterification of free fatty acids in WPG determined in this study and the optimum transesterification parameters as described by Ramadhas (2005: 335), biodiesel was produced from WPG

following route 1 and analysed according to the SANS 1935 specification for biodiesel.

In preliminary work it was attempted to reduce the sulphur levels of WPG. Distillation of WPG under vacuum did not result in the separation of sulphur components as sulphur was measured in all fractions after distillation. The treatment of the WPG with activated bentonite clay had only a minor effect on the sulphur content, the sulphur levels were reduced from 9000 ppm to about 8000 ppm. Acid treatment with sulphuric acid and phosphoric acid resulted in the settling of solid materials from the WPG in the bottom of the beaker. The sulphur content of the WPG was not reduced significantly with the acid treatment. In some cases where higher sulphuric acid dosages were used (5%), it even increased the sulphur content of the WPG.

3.3.2 Free fatty acid extraction and biodiesel production (route 2)

The objective with route 2 was to separate the feedstock to be used for biodiesel production from unwanted impurities affecting the production process and the quality of the final product. The possibility to use liquid-liquid extraction to separate the free fatty acids from the WPG to be used as the biodiesel feedstock was evaluated. The FFA content of WPG was relatively high at 43.2% due to the harsh conditions the grease had been exposed to in the metalworking process.

A shown in Figure 3.1, the first step of route 2 is to use liquid-liquid extraction to extract the FFA from the WPG. Solvent extraction is a widely used process to extract vegetable oils from crushed seeds such as sunflower seeds, using a solvent such as hexane. Two-phase solvent extraction has been evaluated by researchers to extract polar compounds such as free fatty acids from crude oil with the objective to purify the oil to be used as feedstock for biodiesel production. Qian and co-workers (2010:7025) used a 60:40 solvent mixture of methanol and n-hexane at an extraction temperature of 35 °C and an extraction time of 30 minutes. Water at 10% (v/v) was added to increase the polarity of the methanol. After separation the hexane phase contained the jatropha oil and the polar phase contained the methanol, free fatty acids and colloid and colouring matter.

Different solvent combinations were tested in this study to extract the acid oil from the WPG in a search for the solvent combination that would extract the acid oil selectively with sulphur compounds remaining in the WPG.

The production of biodiesel in route 2 was based on the esterification of FFA using homogeneous acid catalysis instead of alkaline transesterifcation of triglycerides as described by Haas (2005:1087) who evaluated the use of vegetable oil soapstock as low value lipid to improve the economics of biodiesel production. All triacylglycerols were hydrolysed to free fatty acids to be esterified to methyl esters. The method described by Haas and co-workers (2005: 1087) is an alternative method to the traditional two-step acid-alkali-catalysed process to deal with feedstock that contains both free and glycerol-linked fatty acids.

The second and third step in route 2 involved the saponification and acidulation of the extracted acid oil mixture from WPG ensuring the complete hydrolysis of the triacylglycerols in the WPG after extraction. FFA measurements on the extracted oil showed that the FFA content of this mixture was approximately 63%, indicating that the acid oil mixture contained besides free fatty acids, other compounds such as triacylglycerols and impurities. Alkaline hydrolysis using sodium hydroxide was conducted in step 2 followed by acidulation in step 3 using hydrochloric acid. The resulting acid oil was purified before esterification as indicated by step 4 by washing the acid oil with hot distilled water to remove any water-soluble impurities. Drying of the acid oil was critical for the esterification step. A two-step acid-catalysed esterification reaction was performed on the purified dried acid oil and the crude biodiesel was further purified by solid phase extraction using silica gel as stationary phase and hexane as mobile phase.

3.3.2.1 Experimental set-up

For each extraction, 100 grams of WPG was placed in a 500 mL separating funnel containing100 grams of a specific solvent. This was done in a water bath at a temperature of 50 °C. After 30 minutes the two layers were separated into 400 mL beakers and the beaker containing the solvent phase was placed on a hot plate in a fume cupboard to evaporate the solvent while stirring with a magnetic stirrer. The

remaining crude acid oil mixture after the evaporation of the solvent was saponified by means of alkaline hydrolysis by heating the oil in a closed 1000 mL Schott Durhan bottle at 120 °C for 2 to 4 hours. Saponification was done with 200 grams of the acid oil mixture, 500 mL of distilled water and 36 grams of sodium hydroxide pellets. The reaction mixture was heated while stirred with a magnetic stirrer in an oil bath. After the saponification step, the Schott Durhan bottle was cooled down to room temperature. The saponified acid oil mixture was then acidulated by adding 180 grams 32% hydrochloric acid and further heated and stirred at 120 °C in an oil bath for 2 to 4 hours. After acidulation the acid oil was separated from the water phase by decanting and washed with three times its volume of hot distilled water and dried on a hot plate at 110 °C. The dried acid oil was the starting material for acid esterification which was done using a 500 mL three- neck round-bottom flask fitted with a reflux condenser as reactor at the same reaction conditions used in route 1.

All experiments were done in triplicate and the experimental data are shown in table B5 and B6 in Appendix B.

3.3.2.2 Optimisation of reaction parameters

Optimal combinations of solvents were investigated with the objective of finding a two-phase solvent system consisting of a polar and non-polar phase where the polar free fatty acids could be extracted into the polar phase, separating it from the non-polar substances such as triglycerides and other hydrophobic substances. Different solvent combinations were therefore evaluated in terms of the amount of FFA extracted and their selectivity towards the sulphur compounds in the WPG. The first step was to find a solvent or solvent combination that separated into a two-phase system with the WPG. Pentane, hexane, octane, methanol, ethanol, iso-propanol and acetonitrile were evaluated and only acetonitrile separated into two distinct layers when mixed with the WPG. This could probably be attributed to the relatively low solubility of WPG in acetonitrile. The influence of polar solvents when mixed at different ratios with acetonitrile on the selectivity towards the sulphur compounds and the amount of acid oil extracted, was further investigated.

Methanol, ethanol and iso-propanol were evaluated and the alcohol concentration in the alcohol–acetonitrile solvent mixtures varied between 10 and 50 grams of methanol, ethanol and iso-propanol per 100 grams of solvent mixture. Each extraction combination was done in triplicate and the mass of acid oil mixture extracted was determined, as well as the sulphur content in the extracted oil.

Haas and co-workers (2005:1090) established optimum conditions to achieve complete hydrolysis of vegetable oil soapstock, using sodium hydroxide. The concentration of the alkali and the temperature of the reaction were mentioned. For the saponification of soapstock only additional alkali was needed for complete hydrolysis as the soapstock already contained an alkali. An amount of 4.2 weight% of sodium hydroxide was added to the already alkaline mixture followed by incubation at 100 °C for 2 to 4 hours. According to Haas (2005:1090) complete hydrolysis was achieved of both acylglycerols and phosphoacylglycerol using these conditions. Lower concentrations of alkali at lower temperatures failed to achieve complete saponification.

In this study the amount of alkali needed for complete hydrolysis was evaluated on an excess amount plus the stoichiometric amount needed to completely saponify the fatty acids present in the WPG. Amounts of sodium hydroxide of 10%, 17.8%, 26.8% and 47.6% in excess of the stoichiometric ratio of oil to sodium hydroxide were evaluated. The free fatty acid concentration of the acid oil was measured using the titration method described in section 3.2.3 after the acidulation step, using an excess of hydrochloric acid. Table 3.6 shows the effect of different amounts of sodium hydroxide on the conversion of WPG to free fatty acids.

Table 3.6: The effect of sodium hydroxide concentration on acid oil yield during saponification followed by acidulation of WPG

NaOH excess to stoichiometric ratio (%)	Acid oil yield (measured as % FFA)	Duration (hours)
10	77.6	2
17.8	78	2
26.8	79.1	2
47.6	79.3	2
47.6	79.4	8

It is clear from Table 3.6 that by increasing the excess to the stoichiometric amount from 10 to almost 18% and by increasing the duration of the reaction did not make a significant difference to the acid oil yield.

Based on the optimum experimental conditions in terms of the selection of the solvent in the liquid-liquid extraction step, the hydrolysis of the WPG and the optimised esterification conditions, biodiesel was produced from WPG, following route 2 and analysed according to the SANS1935 specification.

3.3.3 Modified process with solid phase extraction (route 3)

Route 3 exploited the introduction of a purification step of the WPG feedstock prior to esterification attempting to remove unwanted impurities from the WPG which could be carried forward through the production process, ending up in the final product. The WPG was first modified by a hydrolysis step to ensure that all fatty acids were present as free fatty acids followed by a purification step where solid phase extraction was used.

Chromatography as a separation technique employs two phases, a stationary phase and a mobile phase. The mobile phase introduces the solutes into and carries them through the chromatographic system. The solutes spend time in both phases as they are carried through the system by the mobile phase. Those solutes that spend more time in the solid phase than in the mobile phase will be slowly swept forward through the system while the solutes spending more time in the mobile phase will be swept through the system faster.

3.3.3.1 Experimental set-up

The same procedure to hydrolyse the WPG as describe in section 3.3.2.2 was followed. Acid oil was prepared by saponification of 200 grams WPG in a 1 L Schott Durhan bottle for two to four hours at 120 °C with 35 grams of sodium hydroxide and 500 mL distilled water followed by acidulation with 180 grams of 32% hydrochloric acid at 120 °C for another two to four hours. The acid oil was washed with three times its volume of hot distilled water and dried on a hot plate at 110 °C.

Solid phase extraction was evaluated for the removal of sulphur from the acid oil produced. Different resins and other solid phases were placed inside a modified reflux condenser fitted with a frit and tap, which was used as a chromatography column. Water was heated in a water bath to a desired temperature and flowed through the reflux condenser. The flow rate of the mobile phase through the column was controlled using a pump. A schematic representation of the experimental set-up is shown in Figure 3.2.

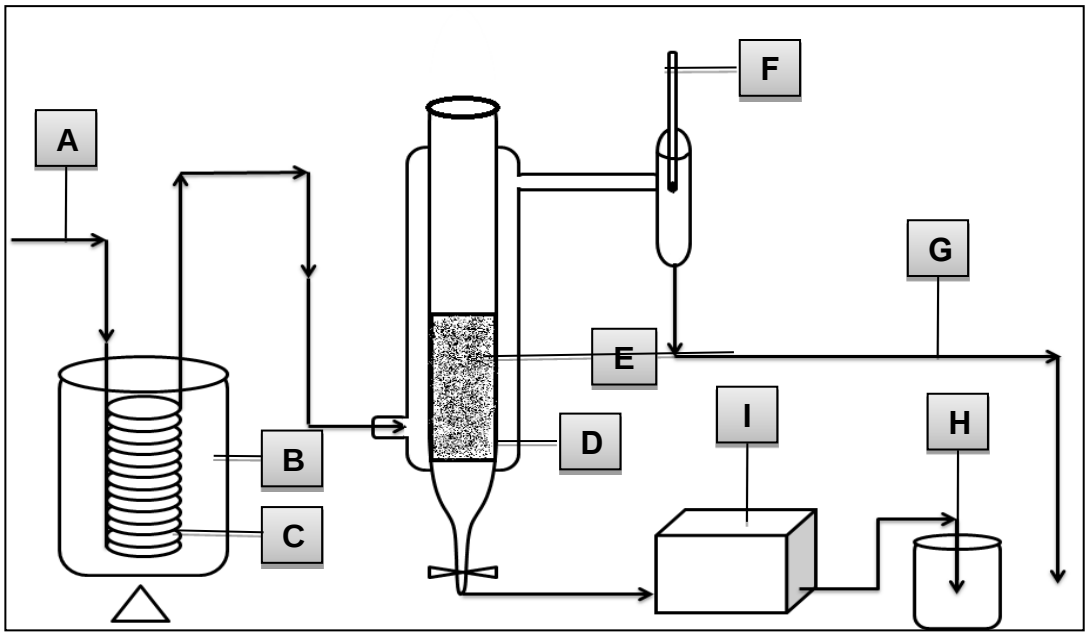

Fig 3.2: A schematic diagram of the experimental set-up for the solid phase extraction of sulphur compounds from WPG: a) water inlet, b) hot water bath, c) coil, d) water jacket, e) inner glass column with resin, f) thermometer, g) water outlet, h) eluate collection, i) flow control pump

Chromatographic separations were done by passing 25 grams of acid oil through 400 mL bed volume of stationary phase. The flow rate was 1 bed volume per hour at 40 °C. The eluate was heated on a hot plate in a fume cupboard to remove the solvent. The purified acid oil was analysed for sulphur by means of Inductively Coupled Plasma Atomic Emission Spectroscopy (ICP-AES).

Acid esterification was performed on the purified acid oil using sulphuric acid as catalyst. The same reaction conditions were used as described in section 3.3.1.2.

3.3.3.2 Optimisation of experimental conditions

Eight different chromatographic systems were compared with regard to their ability to reduce sulphur levels from WPG as well as its ability to remove dark impurities. The different chromatographic systems compared in this study are summarised in Table 3.7.

Table 3.7: Chromatographic systems evaluated for the reduction of FFA in WPG

Solid phase	Commercial name	Mobile phase
Hydrophobic resin	Purolite MN200	Methanol
Hydrophobic resin	Purolite MN200	Hexane
Cation exchange resin	Purolite PD206	Methanol
Cation exchange resin	Purolite PD206	Hexane
Anion exchange resin	Purolite A860	Methanol
Anion exchange resin	Purolite A860	Hexane
Silica gel	Silica gel 60	Methanol
Silica gel	Silica gel 60	Hexane

For each chromatographic system with resins as stationary phase, 25 grams of acid oil was passed through a 400 mL bed volume of resin. For silica gel a bed volume of 200 mL was chosen to reduce the amount of solvent needed to wash the oil through the column. The oil passed much slower through the silica gel column requiring more solvent. The eluate for each chromatographic system was collected in a 400 mL beaker, heated on a hot plate in a fume cupboard while stirred until all the solvent evaporated and the remaining oil was analysed for sulphur using ICP-AES. The chromatographic separations were done in dublicate.

The chromatographic system which separated the most sulphur from the acid oil was selected for further investigation.

Different solvents as mobile phase were evaluated in terms of reducing sulphur from the acid oil using the selected resin as stationary phase. The solvents evaluated were hexane, ethanol, methanol, iso-propanol and methanol with 10% water. For each solvent 25 grams of acid oil was passed through a 400 bed volume

of resin. The eluate was collected in a 400 mL beaker and heated on a hot plate in a fume cupboard while stirred until all the solvent had evaporated. The remaining acid oil was analysed for sulphur using ICP-AES.

The effect of the amount of oil per resin volume on the reduction of sulphur in the acid oil was evaluated by passing 100, 50 and 20 grams of acid oil through a 400 mL bed volume of resin. The eluate was evaporated and the remaining oil analysed for sulphur using ICP-AES.

The effect of passing the same sample of acid oil multiple times through the resin was evaluated by passing 100, 50 and 25 grams of acid oil 5 times through the resin where after the solvent was evaporated and the remaining oil analysed for sulphur using ICP-AES. All experiments were done in triplicate.

Based on the optimised experimental conditions with regard to the hydrolysis of the WPG, the purification of the acid oil, using chromatography and the esterification of the acid oil, biodiesel was produced using WPG as feedstock following route 3 and analysed according to the specification of the SANS 1935 biodiesel standard.

An elution curve for WPG and sulphur components was obtained by plotting the mass of WPG and the concentration of sulphur in successive potions of the eluate against the volume of the eluate as shown in Figure 3.3.

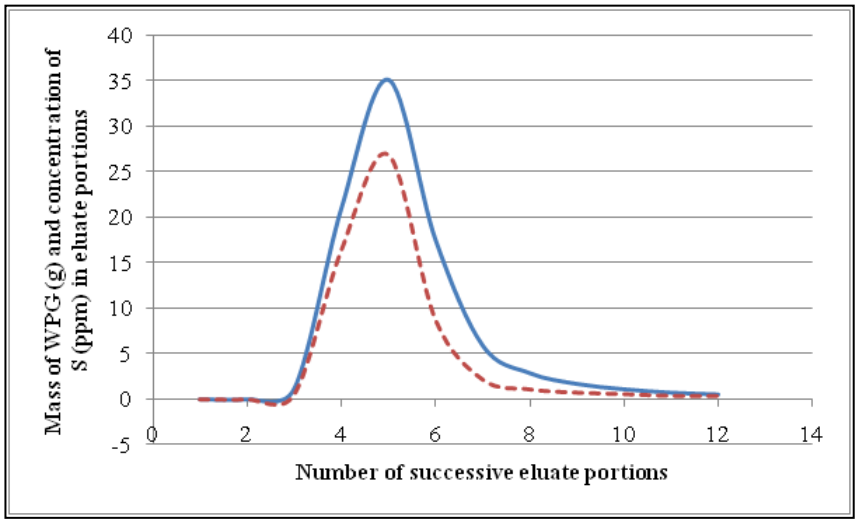

Figure 3.3: Elution curve for WPG and sulphur compounds for a reverse phase chromatographic system. (——) WPG (- - -) sulphur

The elution curve for WPG and sulphur has the same shape and behaviour, indicating that the sulphur components are closely associated with the oil. The sulphur compounds are dissolved in the oil and therefore difficult to separate from the oil.

3.4 References

Abdulkadir, S., Tsuchiya, M. 2008. One-step method for quantitative and qualitative analysis of fatty acids in marine animal samples. Journal of Experimental Marine Biology and Ecology, 352: 1-8.

Berchmans, H. J., Hirata, S. 2008. Biodiesel production from crude Jathropa curcas L. seed oil with a content of free fatty acids. Bioresource Technology, 99: 1716 – 1721.

El-Mashad, H. M., Zhang, R., Avena-Bustillos, R., J.2008. A two-step process for biodiesel production from salmon oil. Biosystems Engineering, 99: 220- 227.

Ma, F., Hanna, M. A., 1999. Biodiesel production: a review. Bioresource Technology, 70: 1- 15.

Haas, M. J. 2005. Improving the economics of biodiesel production through the use of low value lipids as feedstock: vegetable oil soapstock. Fuel Processing Technology, 86: 1087- 1096.

Kusdiana, D., Saka, S. 2004. Effects of water on biodiesel fuel production by supercritical methanol treatment. Bioresource Technology, 91: 289- 295.

Purolite, 201?. Chromatographic Separation of Saccharides, Organic Acids and Amino Acids. http://www.purolite.com/customized/uploads/pdfs/Chromatography%20of%20Saccharides%20%20Organic%20Acids.pdf Date of access: Jul.2012.

Qian, J., Shi, H., Yun, Z. 2010. Preparation of biodiesel from Jatropha curcas L. oil produced by two-phase solvent extraction. Bioresource Technology, 101: 7025- 7031.

Ramadhas, A. S., Jayaray, S., Muraleedharan, C. 2005. Biodiesel production from high FFA rubber seed oil. Fuel, 84: 335- 340.

Sharma, Y. C. 2010. An ideal feedstock, kusum (Schleichera triguga) for preparation of biodiesel: Optimization of parameters. Fuel, 89: 1470 – 1474.

Sharma, Y. C., Singh, B. 2010. A hybrid feedstock for a very efficient preparation of biodiesel. Fuel Processing Technology, 91: 1267 – 1273.

Shiraishi, K., Sharyo, T., Takahashi, R., Watanabe, M., Sonoda, S., Furuyama, O., Kaburagi, K. 1992. Cold rolling oil for steel sheet. (Patent: US5122288).

Srivastava, A., Prasad, R. 2000. Triglyceride-based diesel fuels. Renewable and Sustainable Energy Reviews, 4: 111- 133.

Tiwari, A. K., Kumar, A., Raheman, H. 2007. Biodiesel production from jatropha oil (Jatropha curcas) with high free fatty acids. Biomass and Bioenergy, 31: 569-575.

Veljkovic, V. B., Lakicevic, S. H., Stamenkovic, O. S., Todorovic, Z. B., Lazic, M. L. 2006. Biodiesel production from tobacco (Nicotiana tabacum L.) seed oil with a high content of free fatty acids. Fuel, 85: 2671- 2675.

Zhang, J., Jiang, L. 2008. Acid catalysed esterification of Zanthoxylum bungeanum seed oil with high free fatty acids for biodiesel production. Bioresource Technology, 99: 8995- 8998.

Chapter 4. - Results and Discussion

A detailed description of the results obtained in this study is given in this chapter. The conventional two-step process for the production of biodiesel (route 1) is discussed in section 4.1. The optimisation of the reaction parameters to reduce the free fatty acids to acceptable levels are described, the optimum conditions for the production of biodiesel are summarised, the biodiesel yield was calculated and the characteristics of the biodiesel produced following route 1 is given and discussed.

Section 4.2 describes the results obtained for the extraction of free fatty acids to be used as feedstock for the production of biodiesel. The different extraction parameters for the extraction of the acid oil mixture are discussed, the optimum conditions for the hydrolysis of the remaining triglycerides are described and the characteristics of the biodiesel produced following route 2 are discussed.

The modified process where solid phase extraction is employed to purify the acid oil prior to esterification (route 3) is discussed in section 4.3. The three routes were compared in section 4.4, and the economic viability of the process in route 3 was discussed in section 4.5.

4.1 Conventional two-step process (route 1)

The characterisation of the WPG as described in section 3.1.2.2 showed that the WPG contains various unwanted impurities that could affect the conventional transesterification reaction produce biodiesel and therefore the quality of the biodiesel. The water content of the WPG was reduced from 4% to 0.3% by means of a drying step and the FFA content was measured to be 43.2%. These traditional impurities, water and free fatty acids were to be reduced to acceptable levels preventing the formation of soap allowing the esterification reaction and transesterification reaction to run to completion.

Sulphur and phosphorous content in biodiesel feedstock at concentrations of 8956 ppm for sulphur and 721 ppm for phosphorous as measured and shown in section

3.1.2.2 are exceptionally high. One of the advantages of biodiesel is its low sulphur content due to the low sulphur content of the feedstock and therefore the final product which is indicated by the maximum allowable concentration for sulphur of 10 ppm in the biodiesel standard (SANS 1935).

The sulphur and phosphorous compounds in WPG are artificially introduced through additives to compensate for the shortcomings of vegetable oil and animal fats when used as lubrication oils. Mogwaneng (2004) states that various phosphorous and sulphur containing compounds form part of additives used in cold rolling oils. Formulations for additive packages are proprietary information owned by the lubricating companies.

Optimum reaction conditions for the esterification of free fatty acids in WPG using sulphuric acid as catalyst were investigated.

4.1.1 Effect of reaction temperature on the reduction of FFA content

The experimental data for the esterification of free fatty acids of WPG at different temperatures is shown in Table B1 in Appendix B. Esterification is done to lower the FFA content of the feedstock in preparation for alkaline transesterification. A temperature range of 55 ° C to 65 °C with regard to the reaction mixture was evaluated in this study. The effect of temperature on the reduction of FFA content was evaluated with the molar ratio of methanol to oil at 8:1, the catalyst concentration was 3% sulphuric acid and all reactions were run for 1 hour. The effect of temperature on the FFA content of the WPG is shown in Figure 4.1. The experimental error associated with this set of experiments were calculated to be 7.64% at a 95% confidence level as indicated by the error bars in Figure 4.1. The FFA content at 0°C represents the FFA content of the original WPG and does not represent an experimental point.

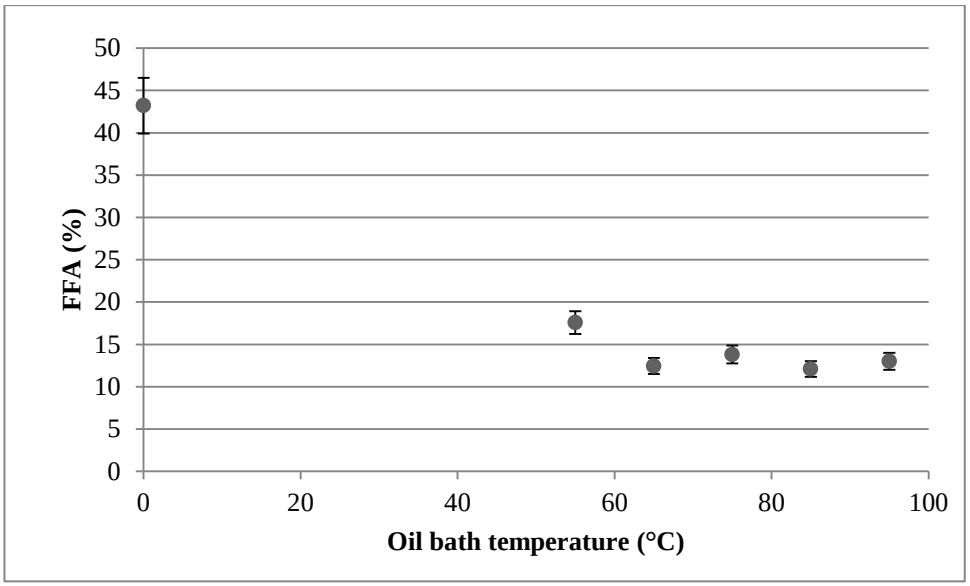

Figure 4.1: Effect of esterification temperature on FFA content of waste process grease

From Figure 4.1 it can be seen that the free fatty acid content of the WPG was reduced to below 13 % from 43.2% in 60 minutes. The viscosity of the reaction mixture increases significantly at temperatures below 55°C due to the saturated fatty acids present in WPG, which makes stirring of the reaction mixture at the same rate as at higher temperatures difficult. The temperature of the reaction mixture reached a maximum as dictated by the boiling point of the reaction mixture and no significant effect of a higher oil bath temperature was noticed on the reduction in acid value of the process grease. Temperature has a significant influence on the reduction of free fatty acids as the viscosity of the oil is reduced with increasing temperatures. At lower viscosities, the miscibility of the oil with the methanol improves which results in improved mass transfer. The results in Figure 4.1 show that the optimum temperature at which the esterification reaction should be conducted is at the boiling point of the reaction mixture (65°C). No significant reduction in free fatty acids was observed with higher oil bath temperatures. Higher oil bath temperatures could result in the evaporation of methanol from the reactor.

Given the reaction parameters selected for esterification, the FFA content could not be reduced to lower than 12 %. This is probably due to the formation of water

during the esterification reaction that slows down the reaction until it is eventually stopped.

4.1.2 Effect of alcohol to oil ratio on the reduction of FFA content

The experimental data for the esterification of FFA in WPG at different molar ratios of methanol to oil is shown in Table B2 in Appendix B. The molar ratio was varied from 3:1 to 18:1 at a constant temperature of 65°C and a catalyst loading of 3wt% sulphuric acid.

Figure 4.2 shows the effect of molar ratio of methanol to oil on the reduction of the free fatty acid content of WPG during the esterification reaction. The experimental error associated with this set of experiments were calculated to be 8.25% at a 95% confidence level as indicated by the error bars in Figure 4.2. The FFA content at the zero intercept represents the original FFA content of the WPG and is not an experimental result.

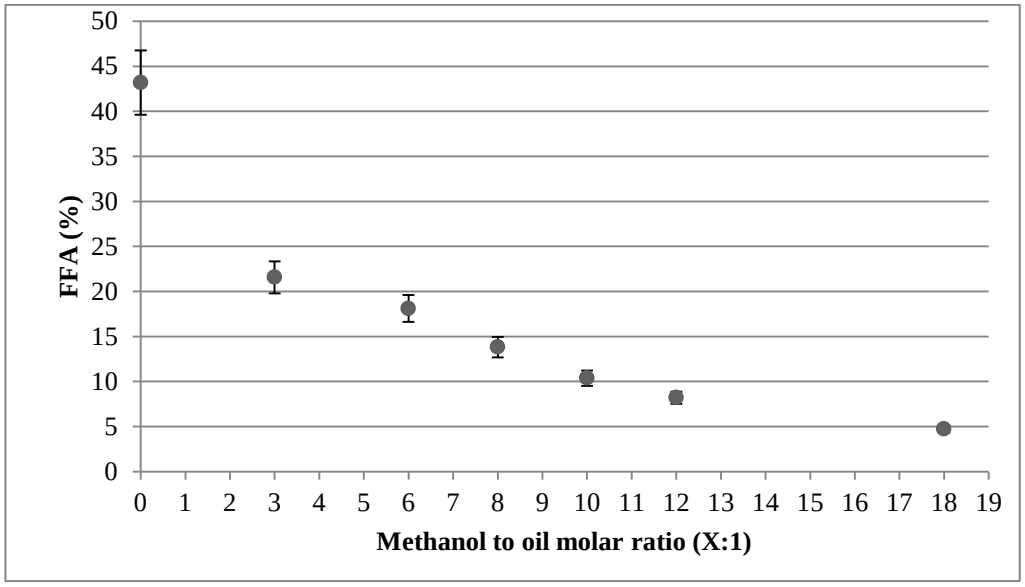

Figure 4.2: Effect of molar ratio of methanol to oil during esterification on FFA content of grease

From Figure 4.2 it can be seen that the amount of alcohol has a significant effect on the reduction of the FFA content with the FFA content gradually decreasing as the methanol to grease ratio is increasing. The free fatty acid content was reduced from 43.2% to 21% at a methanol to oil ratio of 10:1.

The free fatty acid content was reduced to below 10% at a methanol to grease ratio of 12:1 and at a ratio of 18:1, it was reduced to below 5%. Stoichiometrically, 1 mole of methanol is needed to esterify 1 mole of free fatty acid. Higher concentrations of methanol shift the reaction equilibrium to the right increasing the amount of FFA being esterified. The free fatty acid content is still too high for successful alkaline transesterification at these levels, which necessitates a second acid esterification step. A methanol to grease ratio of 8:1 is recommended as higher methanol to grease ratios have a negative impact on the separation of the oil from the water during the washing step and lower methanol usage contributes to a lower biodiesel production cost.

4.1.3 Effect of catalyst concentration on the reduction of FFA content

The experimental data for the esterification of free fatty acids in WPG at different catalyst loadings is shown in Table B.3 in Appendix B. The catalyst loading was varied from 0.5 wt% to 5 wt% with a constant temperature of 65°C and a molar methanol to grease ratio of 8:1.

Figure 4.3 shows the effect of catalyst concentration during esterification on FFA content of WPG. The experimental error associated with this set of experiments was calculated to be 8. 22% for a 95% confidence level as indicated by the error bars in Figure 4.3. The FFA at 0 wt% catalyst represents the original FFA in the WPG and is not an experimental result.

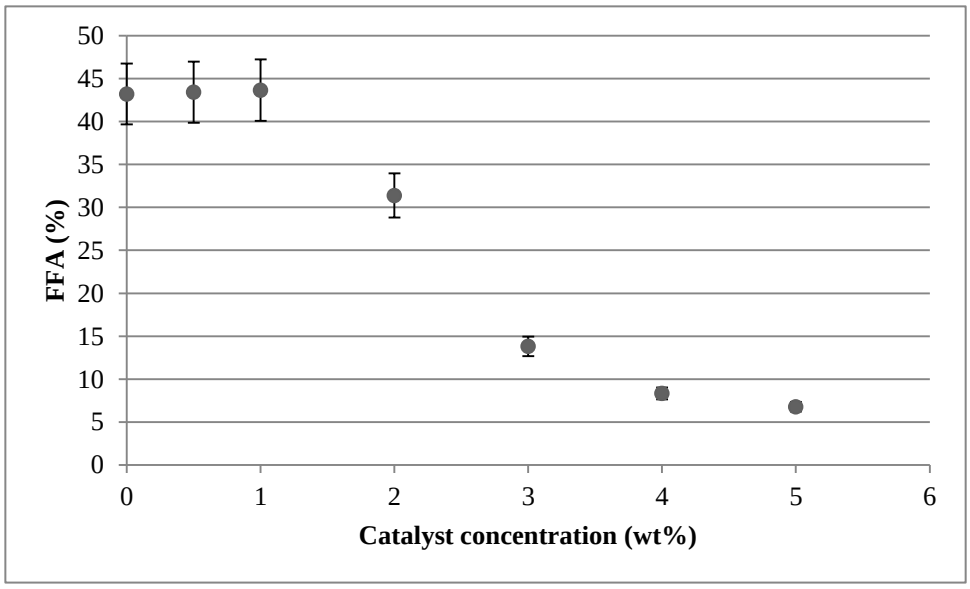

Figure 4.3: Effect of sulphuric acid concentration during esterification on the FFA content of WPG

From Figure 4.3 it can be seen that the FFA content was not reduced for a catalyst loading of 0.5% and 1%. Esterification did not occur at this low catalyst loadings which was further reduced by the depletion of the catalyst through the reaction of the sulphuric acid with the metals and metal ions present in the WPG to form metal salts that separates out at the bottom of the flask.

The FFA content is reduced to just below 10% at a catalyst concentration of 4%. The reason for this high concentration compared to literature [El-Mashad *et al*., 2008: 220; Hayyan *et al*., 2011: 920) is to compensate for the depletion of catalyst by reacting with impurities such as metals and metal ions in the WPG. The optimum reaction conditions selected from the results for this study are a molar ratio of 8:1, a reaction temperature of 65 ° C and a catalyst concentration of 4 wt %. A molar ratio of 8:1 was selected as higher ratios have a significant effect on the costs. The esterification of feedstock containing high FFA offers a challenge to reduce the FFA content to 1% in a single step. Even at molar ratios of methanol to grease of 18:1, the FFA content of the WPG could only be reduced to approximately 5%.

Alptekin and Canakci (2010:4037) used sulphuric acid as catalyst in the pre-treatment of chicken fat containing 13.45% FFA. The optimum reaction conditions selected for the pre-treatment were 20% sulphuric acid at a molar ratio of methanol to oil of 40:1 with a reaction time of 80 minutes at 60 ° C. It was attempted to reduce the FFA content of the chicken fat to 1% in a single step by using an unusually high catalyst concentration and molar ratios.

Stoichiometrically, 1 mole of alcohol is needed for 1 mole of free fatty acid to produce one mole of methyl ester as shown in equation 4.1:

$$R\text{-}COOH + CH_3OH \xleftrightarrow[Catalyst]{} R\text{-}COOCH_3 + H_2O \qquad \text{Equation 4.1}$$

The formation of water as the reaction proceeds inhibits the esterification reaction. An excess amount of alcohol drives the reaction to the right to a point where the reaction is stopped due to an excess amount of water present. By using a high catalyst loading of 20% sulphuric acid, Alptekin and Canakci (2010:4037) reduced the effect of the formation of water that drives the reaction to the left to a point where the reaction is stopped by the affinity of the sulphuric acid for water to act as a water scavenger. An unusually high molar ratio of methanol to oil of 40:1 was combined with the high catalyst loading to drive the reaction to the right to reduce the FFA content to a low enough level for the transesterification reaction. High catalyst concentrations where sulphuric acid is used could have a negative effect on equipment where corrosion problems could occur and too high sulphuric acid concentration could damage the oil. Given the problems associated with high sulphuric acid loadings, it is more advantageous to follow a two-step esterification process with lower sulphuric acid loadings than a single step esterification process with high sulphuric acid high loadings.

Various authors used a double acid esterification step to reduce the FFA content of the feedstock to low enough levels followed by alkaline transesterification (Gadge *et al*., 2005:602, Sahoo *et al*., 2007:448, Lin *et al*., 2009:682, Sabudak and Yildiz, 2010:799). Gadge and co-workers (2005:604) allowed for a settling time

of minimum 1 hour for the removal of the methanol-water mixture to be able to reach low enough FFA levels in the feedstock before alkaline transesterification.

In this study the separation of the methanol-water mixture was made difficult by the emulsification effect of the additives present, and the oil was heated and stirred to reduce the moisture levels to an acceptable level before the next esterification step. A second esterification step using the same optimum conditions determined for the first step was performed on the dried grease from the first esterification step.

The experimental data for the second esterification step is shown in Table B4 in Appendix B. The effect of the selected optimum reaction conditions are shown in Figure 4.4. The FFA content dropped to below 0.5% within the first 2 to 3 minutes. The experimental error associated with this set of experiments were calculated to be 22.2% for a 95 confidence level as indicated by the error bars in Figure 4.4. The FFA at 0 wt% catalyst represents the original FFA in the WPG and is not an experimental result.

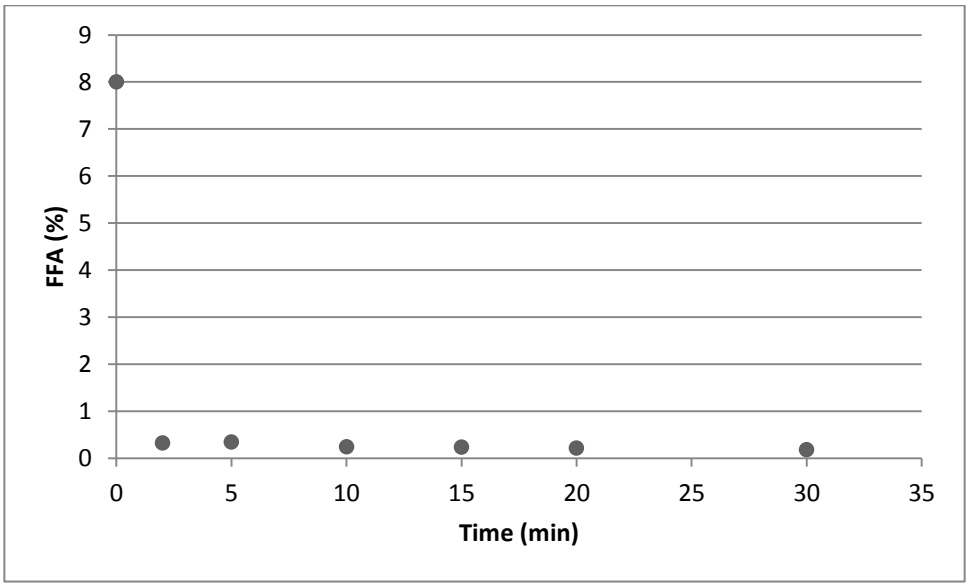

Figure 4.4: Second esterification step at a catalyst concentration of 4%, methanol to oil molar ratio 8:1 and a temperature of 65 ° C.

The sudden drop in FFA content of the oil at the optimum conditions for the first esterification step could allow for a lower methanol to oil ratio and a lower catalyst concentration for the second esterification step and still reach a FFA value of 1% with a more cost effective process in mind.

The optimized reaction conditions needed to achieve a FFA content of 0.5% are summarized in Table 4.1.

Table 4.1: Optimized reaction conditions to reduce the FFA content of WPG to 0.5 wt%

	Esterification stage 1	**Esterification stage 2**
Methanol: oil	8:1	8:1
Catalyst concentration (%)	4	4
Temperature °C	65	65
Duration (minutes)	60	10
FFA content (wt %)	8.3	0.5

4.1.4 Biodiesel production

Biodiesel was produced from WPG following the optimised reaction parameters for the reduction of FFA to 0.5% as shown in Table 4.1. After water washing with hot distilled water and drying, the neutralised oil was used as starting material for the alkaline transesterification at the reaction conditions suggested by Ramadhas and co-workers (2005:335). The molar ratio of methanol to oil was 9:1 with a sodium methoxide loading of 0.5% as catalyst at a temperature of 60 ° C.

The process steps at the optimum reaction conditions and the oil/diesel yields for the steps are shown in Figure 4.5.

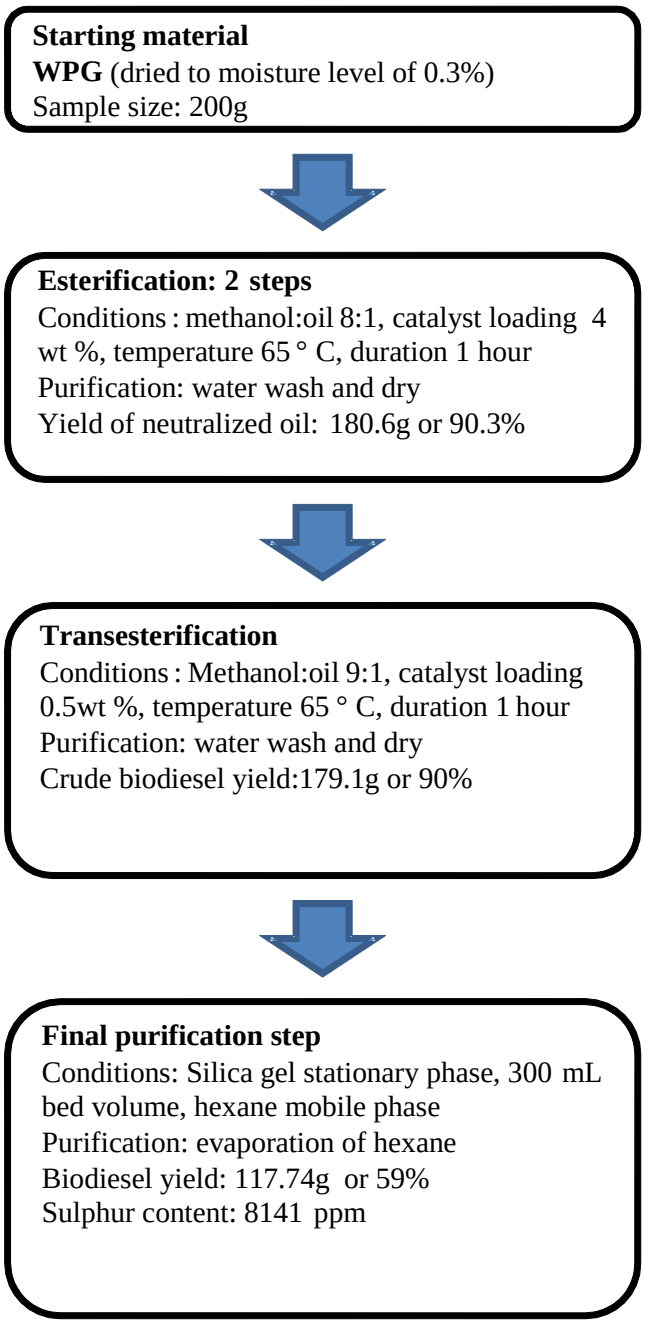

Figure 4.5: Process steps and oil/diesel yields at the optimum conditions for the conventional biodiesel production process (route 1)

The final biodiesel yield was calculated as the mass of biodiesel produced as a percentage of the original mass of dried WPG using equation 4.2.

$$\text{Biodiesel yield} = \frac{\text{Mass of biodiesel produced}}{\text{Mass of dried WPG used}} \times 100 \qquad \textbf{Equation 4.2}$$

A biodiesel yield of 59% was achieved with the conventional biodiesel production process (route 1). The yield of the neutralised oil after two esterification steps was calculated as the mass of neutralised oil as a percentage of the original mass of dried WPG. The yield of the biodiesel after the transesterification step was calculated as the mass of biodiesel produced as a percentage of the original mass of dried WPG. The mass of neutralised oil and crude biodiesel form the transesterification step is much higher than expected. The high mass could be explained by the presence of impurities emulsified by the additives present in the oil and diesel.

4.1.5 Characterisation of biodiesel

The biodiesel was analysed according to the specification of the SANS 1935 standard and the results are shown in Table 4.2. The sulphur content of the biodiesel produced with route 1 is 8141 ppm which disqualifies route 1 as a method for the production biodiesel.

Table 4.2: Comparison of biodiesel produced by route 1 in this study with the SANS 1935 biodiesel specification

Property	Route1	SANS1935	Property	Route1	SANS1935
Ester content, (%mass fraction, min)	93.6	96.5	Acid number (mg KOH/g,max)	0.09	0.5
Cetane number (min)	72	51	Group 1 metals (mg/kg, max)	0	5.0
Density (kg/m^3)	872	860-900	Group 11 metals (mg/kg, max)	0	5.0
Viscosity (mm^2/s)	5.7	3.5-5.0	Linoleic acid (% mass fraction, max)	0	12
S (ppm ,max)	8141	10	Sulfated ash (% mass fraction,max)	0	0.02
P (ppm, max)	12	10	Flashpoint (°C, min)	32	120
Methanol (% mass fraction, max)	ND	0.2	Carbon residue (mass fraction, max)	0.09	0.3
Iodine value (g I/g FAME, max)	25.2	140	Cold filter plugging point (°C, max)	+8	+3 summer -4 winter
Copper strip corrosion (max)	Class 1	Class 1	Total glycerol (% mass fraction, max)	0.42	0.25
Oxidative stability (hrs, min)	>6 hours	6	Free glycerol (% mass fraction, max)	0.02	0.02
Water (%mass fraction, max)	0.01	0.05			

Sulphur compounds are carried through the production process from the feedstock to the final product. The purification steps applied in route 1 which was water washing followed by drying and direct phase chromatography with silica gel as stationary phase and hexane as mobile phase did not make a significant difference

to the sulphur content of the biodiesel compared to the initial sulphur content of the WPG. The sulphur was reduced with approximately 800 ppm from 9112 ppm in the initial WPG feedstock to about 8300 ppm in the final biodiesel. The silica gel chromatographic system removes polar species from the biodiesel. The fact that so little of the sulphur was removed in route 1, leads to the conclusion that a large proportion of the sulphur compounds in the WPG is therefore of a non-polar nature.

The low flashpoint of the diesel (32 °C) is caused by the presence of a volatile compound. No methanol was detected by GC, but some hexane that was used in the purification step could still be present. The presence of emulsifiers as indicated by the high sulphur content, make it more difficult to evaporate the volatile compounds. Even small residual volatile substances can cause the flashpoint to drop significantly. It is expected that methanol is removed by the polar silica gel column.

The specification for cold filter plugging point is not met which could be attributed to the presence of a relatively high percentage of saturated fatty acids in the WPG. High saturated fatty acids also promote the cetane number which shows a relatively high value of 72 for this biodiesel.

The oxidative stability for this biodiesel of > 6 hours is high which could be attributed to the relatively high percentage of saturated fatty acids but also to the presence of additives in the biodiesel originating from the feedstock.

The mass fraction of total glycerol of 0.42% compared to a maximum of 0.25% as per specification could be an indication of the incompleteness of the transesterification reaction. Transesterification reduces the viscosity of triglycerides by the formation of alkyl esters which have lower viscosities. Higher viscosities of the methyl esters than specified by the biodiesel standard could be an indication of incompleteness of the reaction. The kinematic viscosity of 5.7 mm^2/s of the biodiesel from route 1 is slightly higher than allowed for in the specification. The presence of additives such as emulsifiers in the feedstock through- out the biodiesel production process has a negative impact on the unit operations of the process. These emulsifiers make it more difficult to remove

unwanted substances such as water and other impurities in the feedstock that are affecting the process.

4.2 Free fatty acid extraction and biodiesel production (route 2)

4.2.1 Extraction of acid oil to be used as biodiesel feedstock

The results obtained with route 1 in Section 4.1 showed that sulphur compounds in WPG are carried through the production process to the final biodiesel product. The sulphur content of the biodiesel produced following route 1 was 8 141 ppm as shown in Table 4.2.

With route 2 it was attempted to extract the feedstock for biodiesel production from the WPG. Liquid-liquid extraction as separation technique was evaluated to extract free fatty acids from the WPG. The application of solvent extraction in the literature focussed on the utilisation of a polar solvent phase to remove the polar impurities such as free fatty acids from the non-polar oil which was used as the feedstock (Qian *et al*., 2010:7028).

In this study the free fatty acids were extracted as feedstock from the WPG with a polar solvent mixture. The emulsifiers in the WPG made it difficult to find a two-phased solvent system. Different solvents such as acetonitrile, pentane, hexane, heptane, octane, methanol, ethanol and iso-propanol were tested. None of these solvents accept for acetonitrile formed a two-phased solvent system with the WPG. This is probably due to the low solubility of the WPG in acetonitrile. The polarity of the acetonitrile was modified by the addition of different polar solvents at different concentrations. The polar solvents evaluated were methanol, ethanol and iso-propanol.

The experimental data for the mass of acid oil mixture extracted from WPG at different concentrations of methanol, ethanol and iso-propanol in the solvent mixture of alcohol and acetonitrile is shown in Table B5 in Appendix B. Figure 4.6 shows the effect of the different polar solvents at varying concentrations in the

acetonitrile alcohol mixture on the amount of acid oil mixture extracted from the WPG at a 1:1 ratio of solvent mixture to WPG at 50 °C for an extraction time of 30 minutes. The experimental error associated with this set of experiments were calculated to be 15.22% at a confidence level of 95% as indicated by the error bars in Figure 4.6.

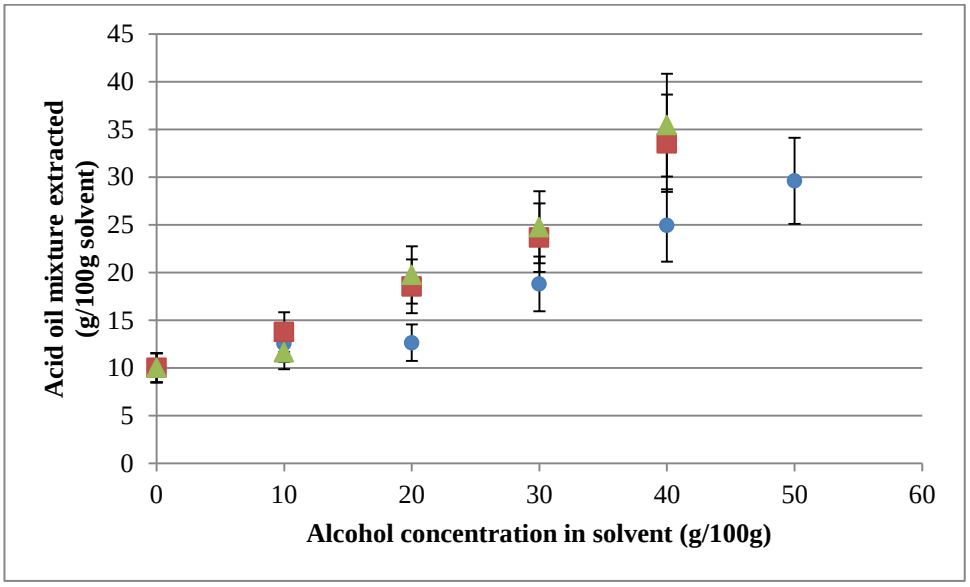

Figure 4.6: Mass of acid oil mixture extracted per grease used at different concentrations of methanol (●), ethanol (■) and iso-propanol (▲) in the solvent mixture

The amount of acid oil mixture extracted increased with an increase in the alcohol concentration of the solvent mixture and was the highest for iso-propanol. The lowest amount extracted was with only acetonitrile as the solvent. Acetonitrile as a solvent resulted in separation of the two phases due to the limited solubility of the acid oil mixture in acetonitrile. The addition of alcohol to the solvent in different ratios with acetonitrile resulted in changes in the solubility of the grease in the solvent. Figure 4.6 shows the increase in solubility of the grease in the solvent mixture with a higher ratio of methanol, ethanol and iso-propanol in the acetonitrile- alcohol mixture. The higher polarity of methanol in the solvent

mixture compared to that of ethanol and iso-propanol did not result in more free fatty acids extracted with methanol in the solvent mixture but instead, the higher partial solubility of the triacylglycerol in the grease in ethanol and isopropanol resulted in more acid oil mixture extracted into the solvent phase. The measurement of the FFA content showed that FFA concentration in the extracted acid oil mixture was higher when only acetonitrile and acetonitrile/methanol was used compared to ethanol and iso-propanol in the solvent mixture.

The second more important objective in the evaluation of liquid-liquid extraction as a technique to reduce certain impurities in the feedstock is to investigate the selectivity of the extraction technique towards the extraction of sulphur compounds. The experimental data for the effect of solvent composition on the extraction of sulphur compounds from the WPG is shown in Table B6 in Appendix B.

Figure 4.7 shows the effect of solvent composition on sulphur concentration in the extracted acid oil mixture. The experimental error associated with this set of experiments was calculated to be 6.16% at a 95% confidence level as indicated by the error bars in Figure 4.7. Acetonitrile without any alcohol extracts the lowest amount of sulphur. A possible explanation could be that the hydrophilic/lipophilic balance of the sulphur compound causes less of the sulphur compound to dissolve in the more polar solvent. The solvent mixtures are to some extent selective in terms of sulphur extraction reducing the sulphur content in the intended feedstock to approximately 4 800 ppm at its best, which is too high given the specification for sulphur in the final biodiesel.

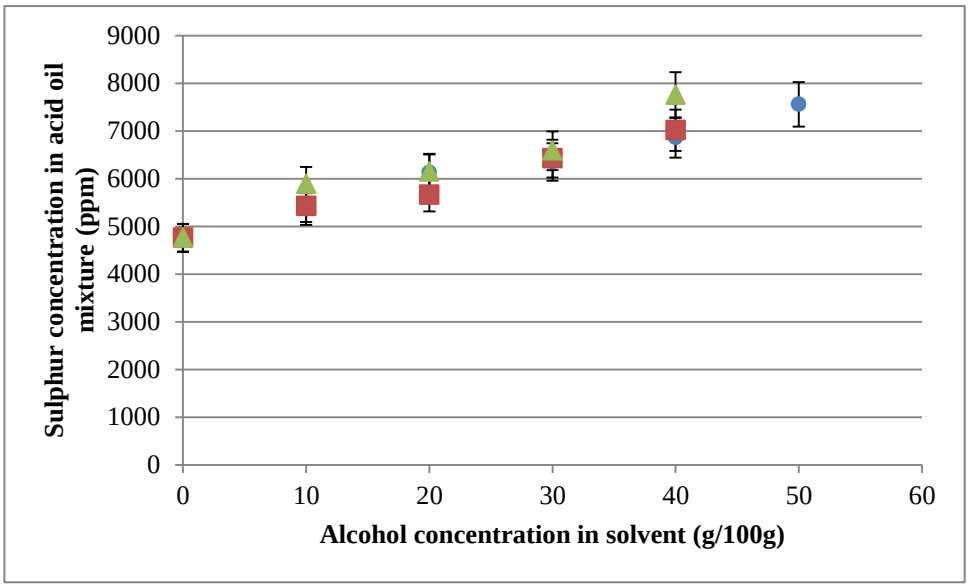

Figure 4.7 Effect of solvent composition on the selectivity towards sulphur extraction from the acid oil mixture (● - methanol; ■ - ethanol, ▲ - iso-propanol)

The evaluation of liquid-liquid extraction as a method to extract free fatty acids from WPG to be used as feedstock for biodiesel production and to reduce the sulphur compounds in the extracted feedstock shows that this route is not feasible as the sulphur content of the acid oil mixture were not reduced to acceptable levels and the biodiesel produced contained 4 800 ppm sulphur which is higher than the requirements of the biodiesel standard. Given the high sulphur content of the treated feedstock, further optimisation of extraction parameters was not performed.

Although the outcome of the evaluation of liquid-liquid extraction showed that this route does not provide a solution for the reduction of sulphur compounds in WPG, the results indicated that some extent of selectivity does exist and that the sulphur compounds tend to associate more with the non-polar phase of the extraction system which corresponds with the effect of the polar silica gel purification step where the sulphur content of the biodiesel was not reduced significantly with the silica gel purification. The findings of route 2 suggested that efforts to separate sulphur compounds from WPG should be focussed on chemical

equilibrium differences rather than solubility differences in these chemical systems. Further investigation was therefore focussed on chromatography as a separation technique in an effort to separate the sulphur compounds from the WPG.

4.2.2 Biodiesel production

Acetonitrile as extraction solvent resulted in the lowest sulphur content for the acid oil mixture and was used to extract the oil to be used as feedstock. Free fatty acid analysis showed that the FFA content of the acid oil was 63%. The saponification and acidulation step described in section 3.3.2.2., hydrolysed the remaining triglycerides in the acid oil mixture which is shown by the increase in the FFA content to 79% confirming the presence of triglycerides in the acid oil extracted. Biodiesel was produced via acid esterification following the same reaction conditions used in route 1.

The process steps and biodiesel yields at the optimum reaction conditions for route 2 are shown in Figure 4.8.

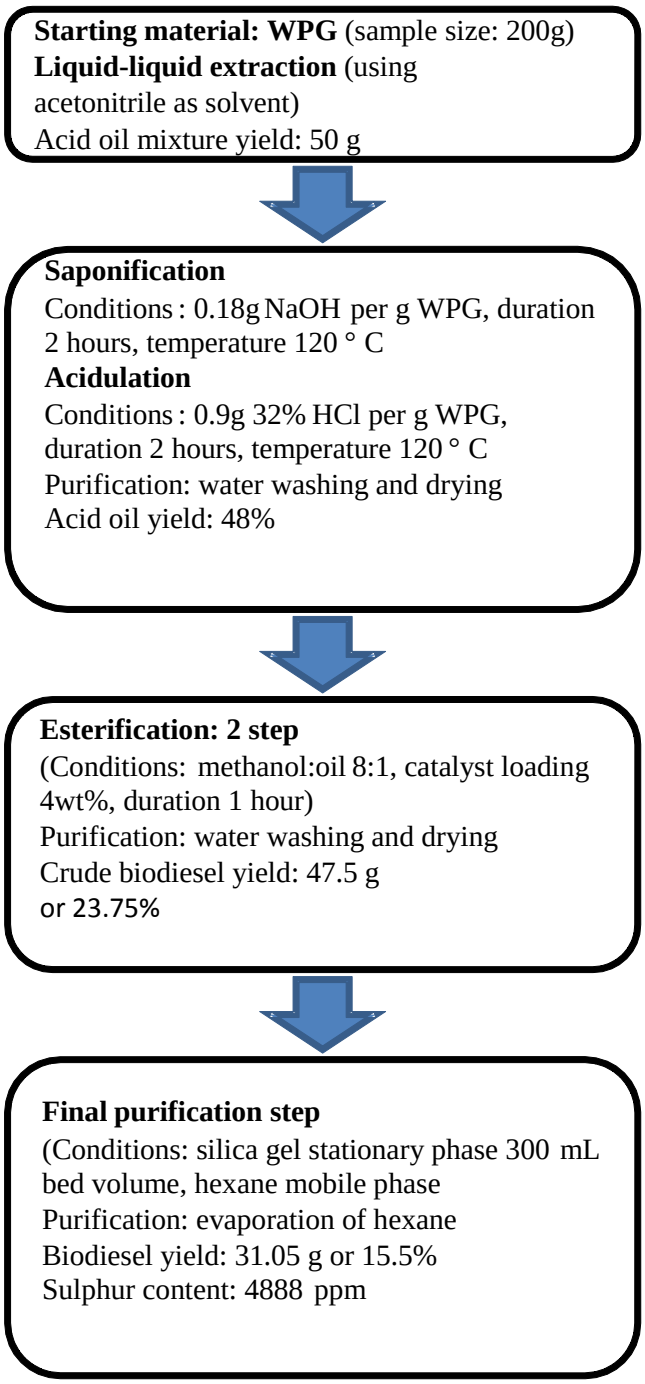

Figure 4.8: Process steps and diesel yields at the optimum reaction conditions for route 2

The final biodiesel yield for route 2 was calculated as the mass of biodiesel produced as a percentage of the mass of WPG used in the experiment. During the extraction of the acid oil mixture, single extractions were performed as multiple extractions increased the final sulphur content of the acid oil mixture. A biodiesel yield of 15.5% was achieved with route 2, based on the original mass of WPG used. The yield of crude biodiesel produced by means of esterification prior to final purification was weighed to be 47.5g. The crude biodiesel mass was reduced from 47.5 g to 31.05 g during the final purification step, indicating that a relatively high percentage of impurities were present in the crude biodiesel. These impurities are of a polar nature which could be removed with the direct phase silica gel chromatographic purification step.

4.2.3 Characterisation of biodiesel

The characteristics of the biodiesel produced with route 2 compared to the requirements of the biodiesel standard are shown in Table 4.3.

Table 4.3 Comparison of biodiesel produced following route 2 with the SANS 1935 biodiesel specification

Property	Route 2	SANS1935	Property	Route 2	SANS1935
Ester content, (%mass fraction, min)	88.6	96.5	Acid number (mg KOH/g, max)	ND	0.5
Cetane number (min)	76.3	51	Group 1 metals (mg/kg, max)	0	5.0
Density (kg/m^3)	870.1	860-900	Group 11 metals (mg/kg, max)	2	5.0
Viscosity (mm^2/s)	4.7	3.5-5.0	Linoleic acid (% mass fraction, max)	0	12
S (ppm, max)	4888	10	Sulfated ash (%mass fraction, max)	0	0.02
P (ppm, max)	6	10	Flashpoint (°C, min)	175	120
Methanol (mass fraction, max)	ND	0.2	Carbon residue (mass fraction, max)	0.01	0.3
Iodine value (g I/g FAME, max)	17.2	140	Cold filter plugging point (°C, max)	+8	+3 summer -4 winter
Copper strip corrosion (max)	Class 1	Class 1	Total glycerol (% mass fraction, max)	0.46	0.25
Oxidative stability (hrs, min)	> 6 hours	6	Free glycerol (% mass fraction, max)	0.01	0.02
Water (% mass fraction, max	0.01	0.05			

The sulphur content of the biodiesel produced with route 2 was reduced from the original concentration of 9 112 ppm in the WPG started with, to 4 888 ppm in the biodiesel product. This is an indication that some of the sulphur containing emulsifiers have been removed but still disqualifies route 2 as the sulphur content

of the biodiesel is too high when compared to the SANS 1935 specification for biodiesel, and a significant amount of emulsifier is still present in the biodiesel. The silica gel purification step using hexane as the mobile phase did not make a significant difference to the sulphur content of the biodiesel. This indicates that the sulphur compounds extracted from the WPG into the acid oil mixture to be used as biodiesel feedstock using the liquid-liquid extraction technique are of a non-polar nature.

The flashpoint of the biodiesel conforms to the specification of the SANS 1935 standard for biodiesel at 175 °C which is much higher than the minimum specification of 120 °C. This is an indication that volatile substances such as methanol which was used as reagent in the esterification process and the hexane which was used as the mobile phase during the silica gel purification step was successfully removed from the biodiesel.

The effect of the fatty acid composition of the original feedstock (WPG) is shown in the results. The relatively high concentration of saturated fatty acids in the original feedstock results in a high cetane number of 76.3 compared to the specification of minimum 51. The high oxidative stability of > 6 hours is supported by the relatively high concentration of saturated fatty acids but also possibly to the presence of additives in the biodiesel. The presence of saturated fatty acids also promotes higher cold filter plugging points which is clear from the CFPP of the biodiesel at + 8 °C where the maximum figure in the specification is +3 °C in summer.

The value for total glycerol is 0.46 % mass fraction which is higher than the specification of 0.25. The value for free glycerol of 0.01 which is within specification is an indication of the effective removal of polar glycerol by the silica gel purification step. The higher total glycerol value could be an indication that traces of triacyl glycerol from the WPG feedstock was carried through the process. The low levels of group I and group II elements is an indication of the effectiveness of the silica gel purification step in removing polar species from the biodiesel.

4.3 Modified process with solid phase extraction (route 3)

With route 1 it was attempted to produce biodiesel from WPG without purification of the feedstock prior to production. With route 2 it was attempted to purify the feedstock prior to production by extracting the acid oil mixture from the feedstock source with the objective to minimise the co-extraction of impurities such as sulphur with the acid oil to be used as feedstock.

The objective in route 3 was to purify the feedstock prior to esterification by introducing an additional purification step. The additional purification step involved the modification of the WPG and the introduction of a chromatographic step. The waste process grease was modified to acid oil by means of the saponification and acidulation step to convert the remaining triglycerides in the WPG to free fatty acids so that the total oil content in the WPG as free fatty acids could be esterified to methyl esters using sulphuric acid as catalyst in the presence of methanol.

The saponification and acidulation conditions for the preparation of acid oil from WPG was evaluated and discussed in section 3.3.2.1. As shown in Table 3.6, the free fatty acid content was 79.3 % in the acid oil. The increase in the free fatty acid content of the WPG by saponification followed by acidulation is owed to the hydrolysis of the remaining triglycerides in the WPG. The free fatty acid yield of 79.3% was achieved with an excess amount of sodium hydroxide of 26.8% to the stoichiometric amount needed to convert all the fatty acids to sodium soap. A further increase in the alkalinity or duration of the reaction did not make a significant difference to the yield of free fatty acids.

A variety of separation technologies such as distillation, crystallisation, precipitation, ion exchange, chromatography and extraction are used in the chemical process industries. Physical/chemical property differences of chemicals are used by different separation methods to separate the different compounds as shown in Table 4.4.

Table 4.4: Separation methods and their physical/chemical property differences used in chemical process industries

Separation method	Physical/chemical property
Distillation	Boiling point differences
Crystallization	Solubility differences
Precipitation	Solubility differences
Ion exchange	Chemical equilibria
Chromatography	Chemical equilibria
Extraction	Solubility differences

The use of chromatographic separation in production processes is a relatively new entrant to the range of unit operations used by chemical engineers. As the demand for high purity materials grow, the use of chromatography is increasing (Purolite, 201?).

4.3.1 Optimisation of chromatographic systems

Different chromatographic systems were evaluated to identify a system that removes the sulphur compounds from the feedstock. Reverse phase chromatography, a non-polar chromatographic system using MN200 from Purolite as a hydrophobic resin was tested. Normal phase chromatography was evaluated using silica gel as stationary phase supplied by Merck Chemical Company. Cation and anion exchange chromatography were tested using PD206 as cationic resin and A860 as anionic resin, both supplied by Purolite. The experimental data on the effect of different chromatographic systems on the reduction of sulphur compounds in WPG is shown in Table B.7 in Appendix B.

Table 4.5 shows the effect of the different chromatographic systems on the reduction of sulphur compounds in the acid oil during purification.

Table 4.5: Effect of different chromatographic systems on the reduction of sulphur levels in waste process grease

Stationary phase	Mobile phase	Sulphur level (ppm)
Hydrophobic resin (MN200)	Methanol	1057
Hydrophobic resin (MN200)	Hexane	5834
Cation exchange resin (PD206)	Methanol	7783
Cation exchange resin (PD206)	Hexane	7854
Anion exchange resin (A860)	Methanol	8118
Anion exchange resin (A860)	Hexane	Ad *
Silica gel	Methanol	8565
Silica gel	Hexane	Ad*

*Ad: oil adsorbed on column

The non-polar chromatographic system (reverse phase) showed the best results for sulphur reduction with both the polar solvent methanol and the non-polar solvent hexane. Sulphur was reduced from 9112 ppm in the acid oil to 1057 ppm using methanol as mobile phase for a 25 g oil sample and a bed volume of 400 ml resin. The sulphur was reduced to 5834 ppm using hexane as the mobile phase. Further passes through the hydrophobic column of the same sample using hexane did not reduce the sulphur levels significantly. In liquid chromatography the solutes in a solution are introduced into the system by means of the mobile phase which carries the solutes through the system. The components in the mixture spend time in both the mobile phase and the stationary phase as the mobile phase passes over and through the stationary phase. A solute that spends more time in the stationary phase as a result of the affinity of the stationary phase for that particular solute will be swept forward through the column at a slower rate. The hydrophobic resin has a greater affinity for non-polar sulphur compounds resulting in the adsorption of some of the non-polar sulphur compounds and the slower passing of other non-polar sulphur compounds through the column.

Shiraishi and co-workers (1992) state that two non-ionic surfactants are included in a formula for cold rolling oils for steel sheets. One of the surfactants has a higher HLB value of 12-16 and the other a lower HLB value of 5-9. The HLB

value of a surfactant refers to the hydrophilic/lipophilic balance of such a molecule. A low HLB value indicates a greater affinity of the molecule for more lipophilic substances. It is therefore suggested that the non-polar sulphur compounds represent the surfactant in the rolling oil formulation with the lower HLB value as the hydrophobic (MN200) resin shows an affinity for this non-polar sulphur compounds.

The principal in reverse phase or non-polar chromatography is to extract non-polar or hydrophobic compounds from polar solutions. The modification of the triglycerides in the process grease by means of saponification followed by acidulation increased the polarity of the grease to decrease its affinity for the non-polar / hydrophobic adsorbent. The polar solvent methanol as mobile phase also promotes a reduced affinity of the non-polar sulphur compounds for the more polar mobile phase to be adsorbed by the non-polar stationary phase. The hydrophobic macronet (MN200) resin supplied by Purolite is a macroporous polystyrene polymer cross-linked with divinylbenzene. The resin is particularly suitable for the efficient sorption of high molecular weight organic molecules with lipophilic properties (Purolite, 201?).

Silica gel with hexane as a mobile phase resulted in the fatty acids to adsorb onto the column which is expected as the more polar fatty acids are being attracted to the polar stationary phase The anion exchange resin also adsorbed the acid oil when hexane was used as mobile phase. Both hexane and methanol as mobile phase did not reduce the sulphur content of the acid oil when the cationic resin was used.

4.3.2 The effect of solvent polarity

Different solvents as mobile phase with the hydrophobic column were compared in terms of their ability to reduce sulphur compounds in the WPG. The experimental data is shown in Table B.8 in Appendix B.

Table 4.6 shows the effect of different solvents as mobile phase on the reduction of sulphur levels in WPG using the hydrophobic resin as stationary phase. The more polar solvents removed more sulphur compounds. The ET (30) value is an

empirical parameter of solvent polarity from spectroscopic measurements (Reichardt, 2003: 111).

Table 4.6: Effect of different solvents as mobile phase with the hydrophobic resin on the removal of sulphur from the oil

Mobile phase	Sulphur level (ppm)	ET(30) value
Hexane	5834	31.0
Iso-propanol	5477	48.4
Ethanol	5024	51.9
Methanol	1057	55.4
Methanol/10%water	176	63.1 (water)

Higher ET (30) values indicate a higher polarity for the specific solvent. Hexane has the lowest polarity of the solvents shown, with an ET (30) value of 31 compared to methanol which is a polar solvent with an ET (30) value of 55.4. For the methanol/10% water mixture the ET (30) value of water of 63.1 was indicated as the value for the methanol/water mixture was not available from the reference. It can be seen from table 4.6 that more sulphur compounds were removed using more polar solvents. The higher the polarity of the solvent as mobile phase, the lower the affinity of the sulphur compounds with the lower HLB value for the mobile phase which is for example the more polar methanol. These non-polar sulphur compounds will spend more time in the lipophilic stationary phase as the mobile phase passes through the column while some will be adsorbed by this stationary phase, reducing the sulphur content of the WPG.

Methanol/10% water as a mobile phase resulted in the lowest sulfur content in WPG for a single stage through the column. The solubility of the acid oil in methanol/ 10% water is relatively low causing some of the grease to get stuck on the column which have a negative effect on the yield.

4.3.3 The effect of oil to resin ratio

The effect of oil to resin ratio on the removal of sulfur using methanol as mobile phase after a single stage through the hydrophobic column is shown in Figure 4.9.

The experimental data is shown in table B.9 in Appendix B. The original sulfur concentration was 9112 ppm and a bed volume of 400 ml resin was used. The experimental error associated with this set of experiments was calculated to be 9.24% at a 95% confidence level as indiacted by the error bars in Figure 4.9.

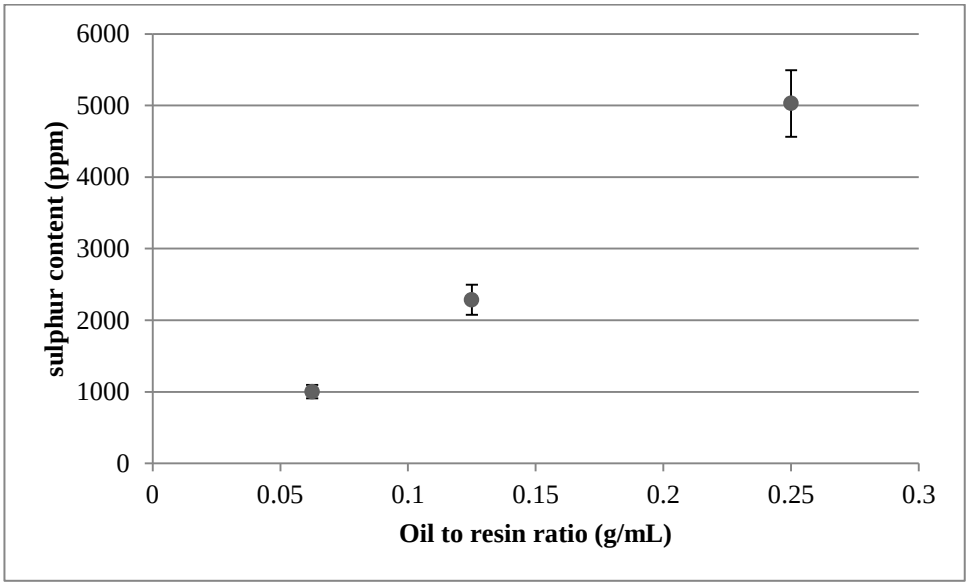

Figure 4.9: Effect of oil to resin ratio on the removal of sulphur from WPG using reverse phase chromatography

The capacity of the chromatographic system is generally expressed in milligrams of sample per grams of stationary phase and corresponds to the amount of sample that can be adsorbed onto a particular stationary phase before overloading occurs (Bauer, *et al.,* 1978: 640). In Figure 4.9 it can be seen that lower oil to resin ratios result in higher sulphur reduction of the acid oil. When WPG is loaded onto the hydrophobic column, an equilibrium of the non-polar sulphur compounds exists where a certain amount of the non-polar sulphur spends time in the hydrophobic stationary phase and a certain amount in the mobile phase. Higher grease loadings where more sulphur compounds are loaded onto the column approach a state where overloading of the column in terms of sulphur could take place where only a certain amount of sulphur is adsorbed and the remaining sulphur stays in the mobile phase.

4.3.4 The effect of multiple passes through the column

The influence of passing the same grease (100g, 50 g and 20 g) multiple times through a hydrophobic column (400 ml bed volume) using methanol as a mobile phase is shown in Figure 4.10. The experimental data is shown in Table B.10 in Appendix B. The experimental error associated with this set of experiments was 15.13% at a 95% confidence level as indicated by the error bars in Figure 4.10.

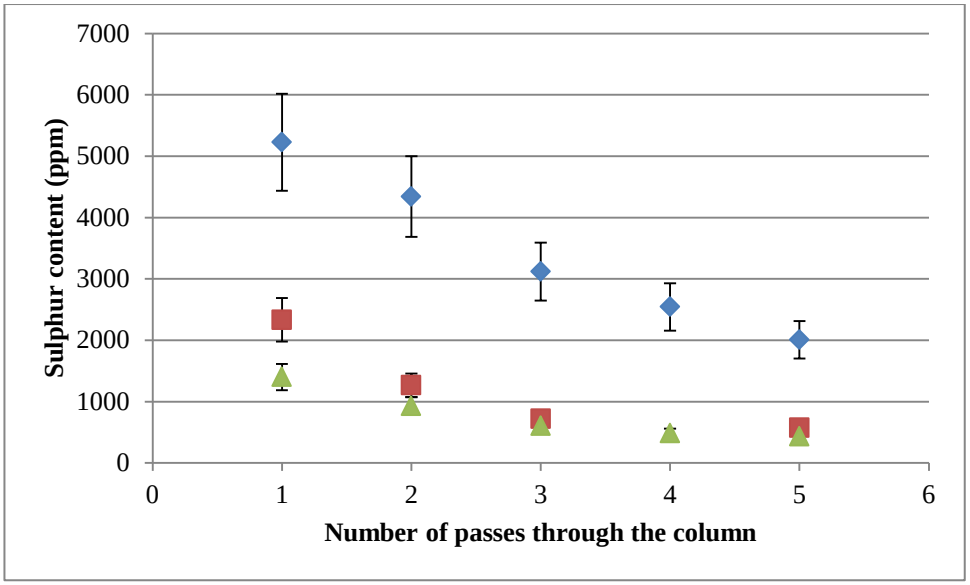

Figure 4.10: Effect of passing the same sample multiple times through the column on sulphur content (●) oil : resin ratio 0.25; (■) oil : resin ratio 0.125 and (▲) oil : resin ratio 0.05

The sulphur content of the acid oil was reduced from 9000 ppm to about 500 ppm by passing the oil 3 times through the hydrophobic column. As more sulphur compounds are removed by each step, the equilibrium of the non-polar sulphur compounds which spends a certain time in the mobile and stationary phase shifts to the lipophilic stationary phase, reducing the sulphur content of the acid oil. More than three passes of the same sample did not have a significant effect on the further reduction of the sulphur compounds in the acid oil. The sulphur content was further reduced to 9 ppm by passing the oil only once through the silica column. By passing the acid oil only once through the hydrophobic column, the

sulphur content of the oil was reduced to approximately 1200 ppm. By further passing the sample through the hydrophilic silica column, the sulphur content of the oil was reduced to 800 ppm. More passes through the silica gel column did not significantly reduce the sulphur content any further. This confirms that the non-polar sulphur is removed from the oil with the hydrophobic resin and the polar sulphur by the silica gel column. Any contribution of the sulphuric acid catalyst to the sulphur content of the biodiesel in route 1, 2 and 3 will be removed by the silica gel purification step.

4.3.5 Biodiesel production

The optimum parameters and process steps for the production of biodiesel from WPG for route 3 are summarized in Figure 4.11.

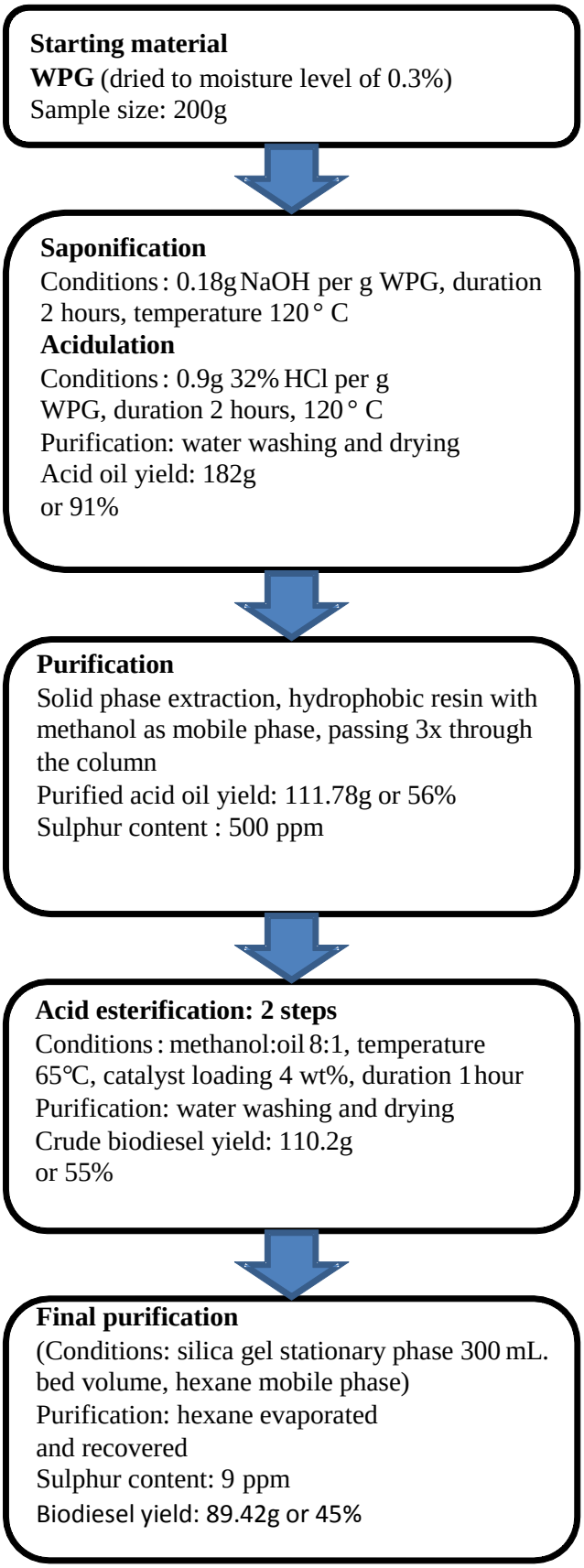

Figure 4.11: Process steps and biodiesel yield at optimum reaction conditions for route 3

A sodium hydroxide concentration of 0.18 gram per gram of WPG was selected. A higher concentration did not make a significant difference to the yield of the acid oil. The concentration of hydrochloric acid in the acidulation step is not critical as long as more than the stoichiometric amount is used. The reaction mixture was stirred during the acidulation step at 120 ° C in a closed Schott Duran bottle until dark clean oil accumulated on top. The system selected to remove the hydrophobic sulphur compounds from the acid oil is the MN200 Purolite resin with methanol as mobile phase. The oil to resin ratio was selected to be 0.05, passing the oil 3 times through the column. The sulphur level in the acid oil was reduced to about 500 ppm by the hydrophobic column. The purified oil was esterified following the two- step process, washed three times with hot distilled water followed by drying and purification with a silica gel column using hexane as mobile phase. The final purification step using silica gel with hexane as mobile phase, removed the polar impurities. The sulphur content is reduced in this step from 500 to 9 ppm.

The biodiesel yield was calculated as the weight of biodiesel produced as a percentage of the weight of WPG used. The lower yield can be attributed to the absorption of fatty acids and FAME on the purification columns.

$$\text{Biodiesel yield} = \frac{\text{Mass of biodiesel produced}}{\text{Mass of dried WPG used}} \times 100$$

A biodiesel yield of 44.71% was achieved following route 3. The acid oil yield was calculated as the mass of acid oil after saponification and acidulation as a percentage of the original mass of WPG used. The yield of the purified acid oil after solid phase extraction of sulphur compounds using the hydrophobic resin was calculated as the mass of purified oil as a percentage of the original WPG used. The yield of crude biodiesel after acid esterification was calculated as the mass of crude biodiesel as a percentage of the original WPG used. The acid oil yield of 182 g from 200 g WPG seems high given the oil content of 64% in the WPG. This could be attributed to the presence of high levels of impurities which are emulsified by the additives present in the oil. The final purification step reduced the mass of biodiesel from 110.2 g to 89.4 g, indicating the presence of a relatively high concentration of hydrophyllic impurities in the crude biodiesel.

4.3.6 Characterisation of biodiesel

The biodiesel produced following route 3 was analysed according to the specification of the SANS1935 biodiesel standard and the results are shown in Table 4.7. The biodiesel conforms to most of the parameters specified in the biodiesel standard.

Table 4.7: Comparison of biodiesel produced following route 3 with the specifications of the SANS 1935 biodiesel standard

Property	Route3	SANS1935	Property	Route3	SANS1935
Ester content, (% mass fraction, min)	93.2	96.5	Acid number (mg KOH/g, max)	0.09	0.5
Cetane number (min)	74.7	51	Group 1 metals (mg/kg, max)	0	5.0
Density (kg/m^3)	865.6	860-900	Group 11 metals (mg/kg, max)	0	5.0
Viscosity (mm^2/s)	4.5	3.5-5.0	Linoleic acid (% mass fraction, max)	0	12
S (ppm, max)	9	10	Sulfated ash (% mass fraction, max)	0	0.02
P (ppm, max)	2	10	Flashpoint (°C min)	170	120
Methanol (% mass fraction, max)	ND	0.2	Carbon residue (mass fraction, max)	0.004	0.3
Iodine value (g I/g FAME, max)	21.3	140	Cold filter plugging point (°C, max)	+10	+3 summer -4 winter
Copper strip corrosion (max)	Class 1	Class 1	Total glycerol (% mass fraction, max)	0.16	0.25
Oxidative stability (hrs, min)	> 6 hours	6	Free glycerol (% mass fraction, max)	0.01	0.02
Water (% mass fraction, max	0.03	0.05			

Sulphur and phosphorous levels were reduced to 9 and 2 ppm respectively. The acid number of 0.09 mg KOH /g biodiesel is within specification which is an indication of a complete esterification reaction and the total glycerol value of 0.16 % confirms that the saponification reaction was also complete.

The presence of a relatively high concentration of saturated fatty acids reflects in the higher cetane number of 74.7, the high cold filter plugging point of +10 and the oxidation stability of > 6 hours. Chain length of the fatty acids also play a role in cold flow properties of the biodiesel produced (Ramos et. al., 2009). Long chain saturated methyl esters in biodiesel contributes to unfavourable cold filter plugging point values. The cold filter plugging point for biodiesel produced by route 3 is one of the parameters out of specification of the biodiesel standard. This problem can be addressed by blending the feedstock with more unsaturated fatty acids prior to biodiesel production reducing the concentration of saturated long chain fatty acids, the addition of additives or by using an on board heating system that warms the fuel when used in a vehicle. No methanol was detected and the high flashpoint of 170 ° C shows that all the hexane was removed from the final product.

4.4 Comparison of the three routes for the production of biodiesel from WPG

In Figure 4.12 the process steps and yields of the 3 routes evaluated to produce biodiesel are compared.

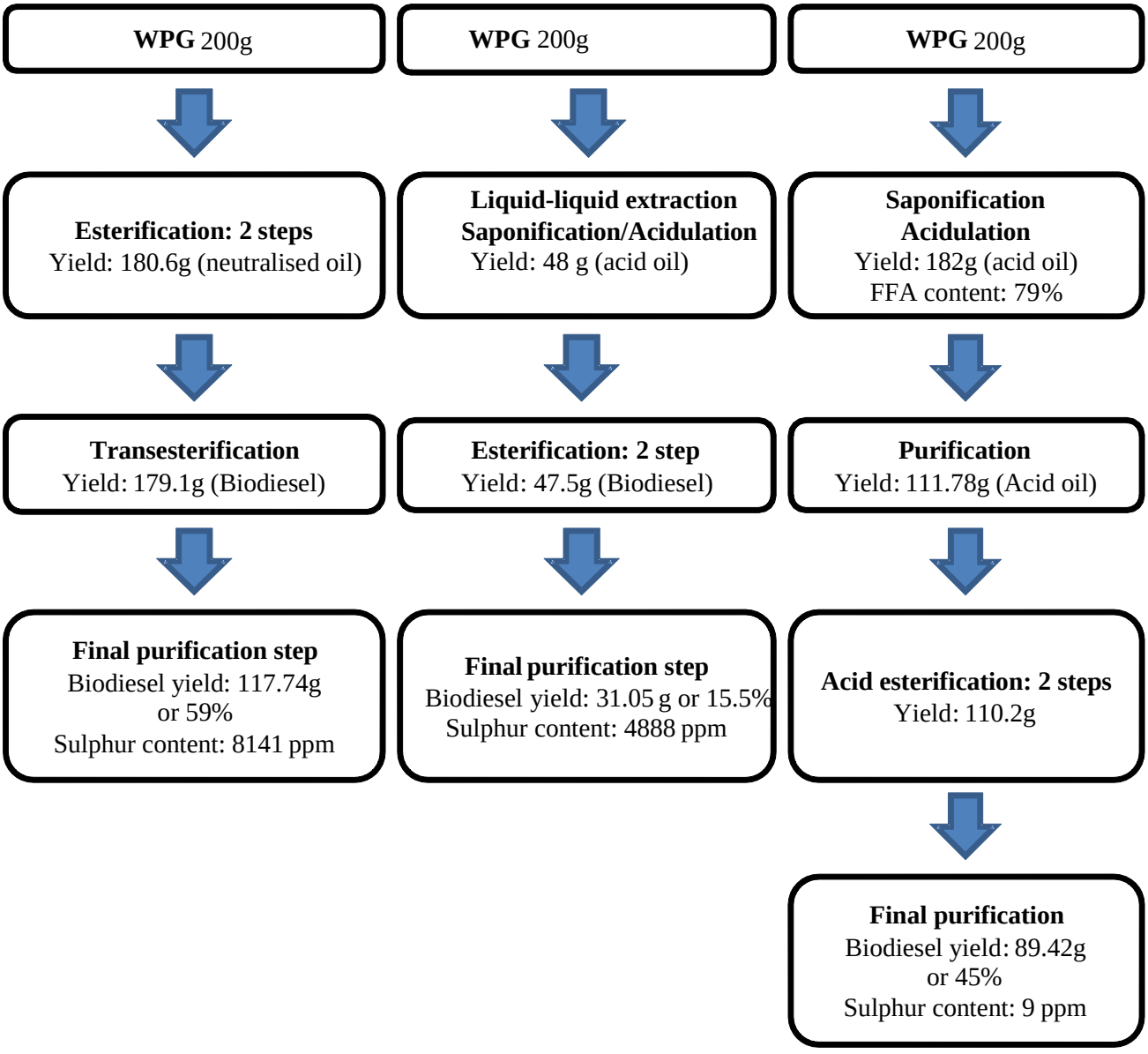

Figure 4.12: Comparison of the process steps and yields of the three production routes followed in this study

In route 1 the conventional biodiesel production process was followed where the traditional impurities (water and FFA) were reduced to acceptable levels prior to transesterification. A final biodiesel yield of 58.87% was obtained. The biodiesel yield was 89.55% before the final purification step using the silica gel column which is an indication that a substantial amount of polar impurities are being removed from the biodiesel by the hydrophilic column. The possibility also exists that some diesel was absorbed on the silica column as a result of the emulsifiers in the biodiesel.

In route 2, liquid-liquid extraction was used to extract the feedstock from the WPG. The biodiesel yield calculated as mass biodiesel as percentage of the WPG started with was 15.5%. A substantial amount of impurities was removed from the biodiesel with the hydrophilic column.

In route 3 an additional purification step was introduced by means of solid phase extraction using a hydrophobic chromatographic column. The acid oil yield reduced from 182 g to 117.8 g by passing it three times through the hydrophobic column. Some of the oil is adsorbed on the column resulting in oil loss.

The comparison of the specification parameters for the biodiesel produced following the 3 routes are shown in Table 4.8. The presence of a relatively high concentration of saturated fatty acids in WPG as shown in Table 3.2 can be seen in the high cetane number, high oxidation stability and high CFPP shown in Table 4.8 for the biodiesel produced by the three routes. The flashpoint for route 2 and route 3 biodiesel are within specification of the SANS 1935 standard with the flashpoint for route 1 much lower as the standard. No methanol was detected in the biodiesel produced by route 1. The presence of small quantities of hexane is responsible for the low flashpoint. High concentrations of emulsifiers as shown by the high sulphur content in the biodiesel produced with route 1 make it more difficult to evaporate the residual hexane. The sulphur content of the biodiesel produced with route 3 indicates that the sulphur containing additives have been removed from the biodiesel.

The final purification step where silica gel as stationary phase with hexane as mobile phase was more effective when used in route 3 compared to route 1 and 2. This is shown in table 4.8 where impurities in the final diesel such as glycerol are

present in low concentrations. The final diesel produced also had a much darker colour compared to the diesel produced by route 3 which was almost colourless as shown in figure 4.13. The presence of sulphur containing additives in the crude biodiesel prior to the silica gel purification step has a negative influence on the effectiveness of the direct phase chromatographic step.

Figure 4.13: Biodiesel produced with the three different routes (routes 1 to 3 from left to right)

The physico-chemical properties of the biodiesel derived from WPG following route 3 was compared with those of biodiesel produced from trap grease (Lu *et al.*, 2010:287) and from waste cooking oil (Wang *et al.*, 2011:1039). The biodiesel specifications for the metioned feedstocks were compared with the SANS1935 standard for biodiesel as shown in table 4.9. Trap grease is suspected to have a relatively high content of saturated fatty acids and as in the case of WPG it is shown by the high cold filter plugging point values as well as the relatively high cetane numbers and high oxidative stability. Trap grease contains 100 ppm sulphur which is much higher than the SANS 1935 specification. Cooking oil

often consists of vegetable oils like sunflower oil, rapeseed oil and soybean oil which contain more volatile fatty acids compared to animal fats like beef tallow, as seen in the lower flashpoint of the biodiesel from waste cooking oil in Table 4.9.

Table 4.8: Comparison of biodiesel specifications produced by the 3 different routes in this study

Property	Route 1	Route 2	Route 3	SANS1935
Ester content, (%mass fraction, min)	93.6	88.6	93.2	96.5
Cetane number (min)	72	76.3	74.7	51
Density (kg/m^3)	872	870.1	865.6	860-900
Viscosity (mm^2/s)	5.7	4.7	4.5	3.5-5.0
S (ppm, max)	8141	4888	9	10
P (ppm, max)	12	6	2	10
Methanol (% mass fraction, max)	ND	ND	ND	0.2
Iodine value (g I/g FAME, max)	25.2	17.2	21.3	140
Copper strip corrosion (max)	Class 1	Class 1	Class 1	Class 1
Oxidative stability (hrs, min)	>6 hours	> 6 hours	> 6 hours	6
Water (% mass fraction, max	0.01	0.01	0.03	0.05
Acid number (mg KOH/g, max)	0.09	ND	0.09	0.5
Group 1 metals (mg/kg, max)	0	0	0	5.0
Group 11 metals (mg/kg, max)	0	2	0	5.0
Linoleic acid (% mass fraction, max)	0	0	0	12
Sulfated ash (% mass fraction, max)	0	0	0	0.02
Flashpoint (°C, min)	32	175	170	120
Carbon residue (mass fraction, max)	0.09	0.01	0.004	0.3
Cold filter plugging point (°C, max)	+8	+8	+10	+3 summer -4 winter
Total glycerol (% mass fraction, max)	0.42	0.46	0.16	0.25
Free glycerol (% mass fraction, max)	0.02	0.01	0.01	0.02

Table 4.9: Comparison of biodiesel specifications produced by route 3 with biodiesel produced from waste trap grease and waste cooking oil as reported in the literature

Property	WTG BD (Lu *et al.*, 2010:287)	WCO BD (Wang *et al.*, 2011:1039)	Route 3 BD (this study)	SANS1935
Ester content, (% mass fraction, min)	N	N	93.2	96.5
Cetane number (min)	60	N	74.7	51
Density (kg/m^3)	890	872.2	865.6	860-900
Viscosity (mm^2/s)	5.28	4.34	4.5	3.5-5.0
S (ppm, max)	100	N	9	10
P (ppm, max)	N	N	2	10
Methanol (% mass fraction, max)	N	N	ND	0.2
Iodine value (g I/g FAME, max)	N	N	21.3	140
Copper strip corrosion (max)	<Class 1	Class 1	Class 1	Class 1
Oxidative stability (hrs, min)	7 hours	5.10	> 6 hours	6
Water (% mass fraction, max	0.04	0.02	0.03	0.05
Acid number (mg KOH/g, max)	0.76	0.11	0.09	0.5
Group 1 metals (mg/kg, max)	N	N	0	5.0
Group 11 metals (mg/kg, max)	N	N	0	5.0
Linoleic acid (% mass fraction, max)	N	N	0	12
Sulfated ash (% mass fraction, max)	0.02	0.0039	0	0.02
Flashpoint (°C, min)	178	135	170	120
Carbon residue (mass fraction, max)	0.84	N	0.004	0.3

Table 4.9: (continue)

Cold filter plugging point (°C, max)	**+15**	**-10**	**+10**	**+3 summer -4 winter**
Total glycerol (% mass fraction, max)	0.22	N	0.16	0.25
Free glycerol (% mass fraction, max)	0.005	N	0.01	0.02

Three routes were evaluated using WPG as feedstock for the production of biodiesel. Route 3 was chosen to be the most suitable route amongst the three routes. Route 3 was the only route where the sulphur content of the biodiesel could be reduced to such levels where it conforms to the SANS1935 standard for biodiesel. The biodiesel produced with route 3 also conforms to most of the specifications outline in the SANS1935 standard.

4.5 Biodiesel economy

It is clear from Table 4.8 that it is technically possible to produce biodiesel from WPG following route 3 that is suitable to be used as an ingredient in biodiesel – petroleum diesel blends. Most of the parameters conform to the biodiesel standard as outlined in the SANS 1935 specification, specifically the values for sulphur and phosphorous which are 9 and 2 ppm respectively.

Based on numerous economic assessments, a generally accepted view is that biodiesel production is not profitable without fiscal support (Bender, 1999:84; Demirbas & Balat, 2006:2379; Balat & Balat, 2008:2738). The high cost of biodiesel is the major obstacle standing in the way of biodiesel commercialisation. Zhang *et al.* (2003: 2) states that biodiesel costs more than US$ 0.5/l which is significantly higher than petroleum diesel at US$ 0.35/l. Comparing the prices mentioned by Zhang and co-workers (2003:2) the price for biodiesel was R 3.05 per litre and R 2.14 per litre for petroleum diesel in 2003.

The feedstock in biodiesel production is a major contributor to the production cost of biodiesel and could be as high as 80% of the total operating cost (Balat & Balat, 2008:2738). The growing demand for edible oil in the food industry, driven by the fast growing human population results in a higher feedstock cost for biodiesel production from edible oil. Given the impact of the feedstock cost, many researchers are focusing on alternative low cost feedstock. Bender (1999:83) compared 12 economic feasibility studies for biodiesel production and concluded that the projected biodiesel cost from oil seed or animal fat fall in a range of US$ 0.30 – 0.69 per litre. Meal and glycerine credits are included in this cost. Rough projections of biodiesel cost from vegetable oil and waste grease are US$ 0.54 – 0.62 and US$ 0.34 – 0.42 per litre respectively (Demirbas, 2006:2275).

The choice of production technology to employ in a production plant has a huge impact on the production cost of the biodiesel and the quality and nature of the feedstock determines the type of technology to be used. Haas and co-workers (2006:671) developed a process model to estimate the production cost of biodiesel. The annual production capacity of the plant was 37,854,118 litres of biodiesel using crude degummed soybean oil as feedstock. The total capital cost

for the plant was estimated at US$ 11348 thousand and the gross operating unit cost for 1 litre of biodiesel was US$ 0.527 based on the time value of dollars of 2006.

A rough estimation of the production cost of biodiesel from WPG using route 3 in this study was made, based on the process model developed by Haas and co-workers (2006:671). Although the annual available quantity of WPG in South Africa is only 2400 tons, the economic evaluation was done for 38000 tons to compare with the evaluation done by Haas and co-workers (2006:671), as the scale down factor for this type of plant is not known. Further more, the total quantity of WPG produced by the metal working industry in the USA was estimated at 540 000 tons per annum in 2003 by Gawrilow (2003: 16). The estimated capital cost for construction and the annual and unit production cost for route 3 is discussed in appendix C. It can be seen in Table C1 and C2 that the capital cost is estimated at a minimum of R324161 thousand and the operating cost per litre diesel produced is estimated at R 7.90. By comparing the estimated operating cost of biodiesel produced from WPG based on the model described by Haas and co-workers (2006: 671) with the basic fuel price (refinery price) for petroleum diesel in South Africa which is R 7.19115 (South Africa. Department of Trade and Industry, 2013), the estimated biodiesel from WPG is about 10% more expensive. When less than 300 000 litres of biodiesel are produced per annum, the biodiesel is exempted from taxes which make it economically viable as the current retail petroleum diesel price in South Africa is above R 12 per litre (South African Department of Trade and Industry, 2013). The feedstock cost has been reduced from R 179634 thousand per annum for a 37.8 thousand ton plant using crude degummed soybean oil to R 97073 thousand for WPG. The initial amount of WPG used was set at 86400 tons as the yield of biodiesel was determined to be 44%. At a current Brent crude oil price of $ 102.41 per barrel the production cost of biodiesel from WPG is marginally higher than the basic fuel price. At a Brent crude oil price of about $ 113 per barrel and higher, the production of biodiesel from WPG will be economically viable.

4.6 Summary

Three process routes to produce biodiesel from WPG were evaluated in this study. In route 1 conventional biodiesel production technology using acid pre-esterification to reduce the FFA and alkaline transesterification using sodium methoxide as catalyst were employed. The strategy in route 2 was to extract the free fatty acids from the WPG as feedstock for acid esterification using sulphuric acid as catalyst to produce fatty acid methyl esters. In route 3 it was attempted to purify the WPG feedstock prior to methyl ester production by means of column chromatography after which the fatty acid methyl esters were produced using acid esterification employing sulphuric acid as catalyst.

In route 1,reaction conditions for esterification was established to be 65°C, a molar methanol to oil ratio of 8:1 and a catalyst concentration of 4% sulphuric acid. Metals ions in the WPG caused the sulphuric acid to form salts during the esterification reaction, and thus more sulphuric acid as what is normally reported in literature was necessary for complete esterification. Biodiesel produced via this route did not conform to the SANS1935 standard. The sulphur and phosphorous levels could not be reduced to below the required levels and the high levels of glycerol indicated that the separation processes employed were not successful in removing all glycerine from the reaction mixture. The low iodine value and high cold filter plugging point is both an indication of the high saturated fatty acid content of the starting material. The latter could be remedied by the addition of suitable additive. An overall biodiesel yield of 59% based on the starting mass of WPG was obtained using route 1.

In route 2, acid oil was extracted from WPG using acetonitrile which resulted in approximately half of the sulphur being removed prior to diesel production. Biodiesel produced via rout 2 did not conform to the SANS1935 biodiesel standard. Most notable of diesel produced via route 2 is the very low iodine value as well as the high flashpoint and high concentration of glycerol still present in the diesel. The latter is an indication that additional purification steps are necessary to remove all glycerine as well as other impurities from the reaction mixture, A biodiesel yield of 15.5%, based on amount of WPG started with was obtained for route 2.

In route 3, reverse phase chromatography was employed to remove all sulphur from the WPG prior to diesel production. Additives and surfactants added to rolling oil are believed to consist of a hydrophilic as well as a lipophilic part (HLB balance) with sulphur contained on both sides. This was confirmed in this study in that not all sulphur could be removed by the hydrophobic resin and polar solvent used in the reverse chromatographic purification step. It is believed that the hydrophobic resin removed the lipophilic part of the additives that included that sulphur. The sulphur bonded to the hydrophilic part of the surfactant could then successfully removed by the silica gel column. The removal of sulphur bonded to the lipophilic part of the surfactant prior to the diesel reaction resulted in the silica gel column being much more effective in removing residual glycerine from the reaction mixture. Biodiesel produced via route 3 conformed for the most part to the SANS1935 biodiesel standard. The high CFPP can be negated with suitable additives to the diesel. A yield of 45% based on the mass of WPG used was obtained for route 3.

This study showed that it is technically feasible to produce biodiesel from WPG that could be used as feedstock for biodiesel blends with petroleum diesel that would conform to the ASTM D6751-10 specification for biodiesel blends. A generic economic comparison of the production cost of biodiesel from WPG with the production of biodiesel from crude degummed soybean oil showed that the economic benefit from the cheap WPG are wiped out by the much higher capital cost to be invested in a 38 million litre per annum biodiesel plant which makes the production of biodiesel from WPG viable only when the crude oil price is higher than $ 113 per barrel.

4.7 References

Alptekin, E., Canakci, M. 2010. Optimization of pretreatment reaction for methyl ester production from chicken fat. Fuel, 89: 4035- 4039.

Balat, M., Balat, H. 2008. A critical review of biodiesel as a vehicular fuel. Energy Conversion and Management , 49:2727-2741.

Bauer, H. H., Christian, G.D., O'Reilly, J.E. 1978. Instrumental Analysis. Allyn and Bacon, Inc. 832 p.

Bender, M., 1999. Economic feasibility review for community-scale farmer cooperatives for biodiesel. Bioresource Technology, 70: 81- 87.

Demirbas, M. F., Balat, M. 2006. Recent advances on the production and utilization trends of bio- fuels: A global perspective. Energy Conversion and Management, 47:2371- 2381.

El-Mashad, H. M., Zhang, R., Avena-Bustillos, R., J. 2008. A two-step process for biodiesel production from salmon oil. Bioresource Technology , 99: 220 – 227.

Gadge, S.V., Raheman, H. 2005. Biodiesel production from mahua (Madhuca indica) oil having high free fatty acids. Biomass and Bioenergy, 28: 601- 605.

Haas, M., J., McAloon, A., J., Yee, W., C., Foglia, T., A. 2006. A process model to estimate biodiesel production costs. Bioresource Technology, 97: 671 -678.

Hayyan, A., Alam, M. Z., Mirghani, M. E. S., Kabbashi, N. A., Hakami, N. I. N. M., Siran, Y. M., Tahiruddin, S. 2011. Reduction of high content free fatty acid in sludge palm oil via acid catalyst for biodiesel production. Fuel, 92: 920- 924.

Lin, L. Ying, D.,Chaitep, S., Vittayapadung, S. 2009. Biodiesel production from crude rice bran oil and properties as fuel. Applied Energy, 86: 681- 688.

Lu, P., Yuan, Z., Li, L., Wang, Z., Luo, W. 2010. Biodiesel from different oil using fixed-bed and plug-flow reactors. Renewable Energy, 35:283- 287.

Mogwaneng, P., J. 2004. Optimization of rolling mill oils evaluation using FT-IR spectroscopy. Pretoria: University of Pretoria (Dissertation - M.Sc).

Purolite, 201?. Chromatographic Separation of Saccharides, Organic Acids and Amino Acids. http://www.purolite.com/customized/uploads/pdfs/Chromatography%20of%20Saccharides%20%20Organic%20Acids.pdf Date of access: Jul.2012.

Qian, J., Shi, h., Yun, Z. 2010. Preparation of biodiesel from Jatropha curcas L. oil produced by two-phase solvent extraction. Bioresource Technology, 101: 7025- 7031.

Ramadhas, A. S., Jayaray, S., Muraleedharan, C. 2005. Biodiesel production from high FFA rubber seed oil. Fuel, 84:335- 340.

Ramos, M.J., Fernandez, C.M., Casas, A., Rodriques, L, Perez, A. 2009. Influence of fatty acid composition of raw materials on biodiesel properties. Bioresource Technology, 100: 261- 268.

Reichardt, C. 2003. Solvents and Solvent Effects in Organic Chemistry. Wiley VCH: Verlag GmbH & Co. KGaA, Weinheim.

Sabudak, T., Yildiz, M. 2010. Biodiesel production from waste frying oils and its quality control. Waste Management, 30:799- 803.

Sahoo, P. K., Das, L.M., Babu, M.K.G., Naik, S.N. 2007. Biodiesel development from high acid value polanga seed oil and performance evaluation in a CI engine. Fuel, 86: 448- 454.

Shiraishi, K., Sharyo, T., Takahashi, R., Watanabe, M., Sonoda, S., Furuyama, O., Kaburagi, K. 1992. Cold rolling oil for steel sheet. (Patent:US 5122288).

South Africa. Department of Trade and Industry. 2013. Star.Business Report: Markets: 22, 26 Apr.

Wang, Y., Ma, S., Zhao, M., Kuang, L., Nie, J., Riley, W.W. 2011. Improving the cold flow properties of biodiesel from waste cooking oil by surfactants and detergent fractionation. Fuel, 90:1036- 1040.

Zhang, Y., Dube, M. A., McLean, D.D., Kates, M. 2003. Biodiesel production Process design and technological assessment. Bioresource Technology, 89: 1-16.

Chapter 5. – Conclusions and Recommendations

The possibility to use waste process grease which is a waste product from the metalworking industry, as feedstock for biodiesel production, was investigated in this study. Three process routes were evaluated to establish the feasibility of producing biodiesel that conforms to the specifications set out in the SANS 1935 biodiesel standard of South Africa.

5.1 Influence of the characteristics of WPG on the conventional alkaline transesterification process (route 1)

The conventional alkaline transesterification technology was not suitable for the production of biodiesel from WPG. Water and free fatty acids in WPG interfered with the transesterification reaction and sulphur containing compounds were carried through the process to end up in the final product.

The pre-treatment of free fatty acids by means of acid-catalysed esterification could be done successfully to reduce the FFA content to acceptable levels for alkaline transesterification. A higher catalyst loading of 4 weight% sulphuric acid than the typical catalyst loadings described in the literature was selected due to the depletion of the catalyst by the reaction of the sulphuric acid with the metals and metal ions in the WPG. The moisture content of WPG could be reduced to low enough levels, although the presence of emulsifiers interfered with the drying step.

The presence of sulphur containing compounds introduced as additives in the process grease interfered with the silica gel purification step, affecting the efficiency of the separation of impurities. The silica gel purification step did not have a significant effect on the reduction of the sulphur content of the biodiesel, indicating that a major part of the sulphur compounds are of a non-polar nature.

A two-step esterification process was necessary to reduce the FFA content to low enough levels for alkaline transesterification.

The conventional alkaline transesterification process (route 1) was not considered as a suitable process for the production of biodiesel from WPG as the sulphur content of the final product was 8 141 ppm which did not conform to the SANS 1935 specification for biodiesel.

5.2 The feasibility of extracting free fatty acids from WPG with liquid-liquid extraction to be used as feedstock for biodiesel production (route 2)

The high concentration of free fatty acids in WPG provided the option to use the free fatty acids as biodiesel feedstock provided that it could be successfully separated from the WPG and separated from the sulphur containing compounds. The presence of emulsifying additives in the WPG feedstock interferes with the separation of the polar free fatty acids from the less polar triglycerides. The reduction in sulphur content of the extracted acid oil mixture recovered by evaporation of the solvent shows that some selectivity exists as far as the sulphur compounds is concerned.

Liquid-liquid extraction of the acid oil mixture from the WPG to be used as biodiesel feedstock resulted in the reduction of the sulphur content from 9112 ppm to 4888 ppm. The sulphur content of the biodiesel is still way too high indicating the presence of sulphur containing additives in the extracted feedstock carried through to the final product. Liquid-liquid extraction of FFA to be used as feedstock could not be considered as a feasible method for the utilisation of WPG as feedstock for biodiesel production.

5.3 The feasibility of feedstock pre-treatment on modified WPG using solid phase extraction on the utilisation of WPG as biodiesel feedstock (route3).

The reduction of sulphur compounds in WPG feedstock and crude biodiesel produced from the modified WPG was successful using process route 3. The

biodiesel produced from WPG following route 3 conforms to most of the parameters specified in the SANS 1935 biodiesel standard.

The reduction of the majority of the sulphur content in WPG by means of a reverse phase chromatographic system confirmed the non-polar nature of these compounds.

The relatively high cold filter plugging point could be attributed to the fatty acid composition of the oil. The relatively high content of saturated fatty acids contributes to the high cetane number and oxidation stability of the biodiesel.

WPG could be used as a feedstock for the production of biodiesel used in biodiesel-petroleum diesel blends judged from the quality of the biodiesel produced by following route 3. The comparison of the biodiesel production cost produced by route 3 shows that the process appears not to be economically viable given the current price for petroleum diesel.

In this study it was shown that WPG could be a used as a feedstock for the production of biodiesel as the specifications of the diesel produced via route 3 conforms to the standard specification for biodiesel production in terms of sulphur content which makes it suitable as a component in biodiesel blending with petroleum diesel. At a Brent crude oil price of $113 per barrel and higher the utilisation of WPG as feedstock would make economic sence.

The production of biodiesel from WPG supports the effort of producing energy from hazardous waste materials. The utilisation of WPG as feedstock will contribute to the protection of the environment by preventing hazardous gaseous compounds from being released into the atmosphere as the WPG is currently used as a low grade furnace fuel. A contribution of about 2400 metric tons of biodiesel feedstock per annum will be made to the biodiesel industry in South Africa which is in short supply. The utilisation of WPG as feedstock will also solve a critical problem for the metalworking industry in terms of their waste management strategy to dispose of WPG in an environmentally acceptable way.

5.4 Recommendations

Further work is needed to utilise industrial waste materials containing biomass such as WPG to generate more environmentally friendly fuel by converting hazardous waste to energy.

WPG contains impurities other than the traditional free fatty acids and water like additives comprising sulphur compounds designed to act as emulsifiers which are difficult to separate from other substances which are not common in traditional biodiesel production. These impurities are difficult and costly to remove from the feedstock or the crude biodiesel.

Further work should focus on the simplification of the production process proposed in route 3, reducing the cost of the process. The approach with route 3 in this study was to find a separation technology to reduce the sulphur content in the biodiesel by removing the excisting sulphur compounds from the diesel and/or the feedstock. Further work could focus on the evaluation of other sorbents to selectively remove specific sulphur compounds from the feedstock. Rheinberg and co-workers (2008:2988) evaluated a Ni/NiO-sorbent for the removal of 4,6-dimethyldibenzothiophene from liquid fuels. For this approach an analytical study is needed to identify the specific sulphur species present in the WPG.

Another approach could be to modify the sulphur compounds in the WPG to molecules that could be removed more easily from the fuel. This could include the evaluation of technologies like hydrodesulfurisation, oxidative desulphurisation and biodesulfurisation. The effect of desulfurisation catalysts on the transformation of sulphur- containing molecules which could more easily be removed from the WPG should be investigated. Pecoraro and Chianelli (1981: 430) showed that transition metal sulphides containing Rhodium, Iridium and Osmium has a high activity as hydrodesulphurisation catalysts.

A further evaluation to determine the feasibility of the purification and reuse of the residual oil, a waste product from route 3 as a supplement rolling oil in the metal working process with the objective to reduce the cost of rolling oil and saving the environment.

The use of WPG as feedstock for the production of renewable diesel should also be investigated. Knothe (2010: 365) compared biodiesel with renewable diesel. When biomass like vegetable oil or animal fat is hydrodeoxygenated with hydrogen and a catalyst, hydrocarbons, propane, water and carbon dioxide is formed. The resulting hydrocarbons are known as renewable diesel. The behaviour of sulphur containing molecules and the ease of removal thereof should be investigated.

5.5 References

Knothe, G. 2010. Biodiesel and renewable diesel: A comparison. Progress in Energy and Combustion Sciences, 36:364- 373.

Pecoraro, T. A., Chianelli, R. R. 1981. Hydrodesulphurisation catalysis by transition metal sulphides. Original research article. Journal of Catalysis, 67: 430-445.

Van Rheinberg, O., Lucka, K., Kohne, H., Schade, T., Andersson, J.T. 2008. Selective removal of sulphur in liquid fuels for fuel cell applications. Fuel, 87:2988- 2996.

Appendix A - Gas chromatography calibration

The calibration of the gas chromatograph is discussed in this section. The gas chromatograph was used to determine the ester content of the feedstock and the biodiesel produced using the 3 routes evaluated in this study.

A1. The identification of fatty acids

Gas chromatography standards as fatty acid methyl esters were obtained from Sigma Aldrich chemical company. The retention times of the different fatty acid methyl esters were determined and are presented in Table A1.

Table A1: Retention times for fatty acid methyl esters separated by the HP-88 column

FAME	Retention time (Minutes)
Dichloromethane (Solvent)	5.553
Methyl hexanoate (C6:0)	7.217
Methyl octanoate (C8:0)	9.611
Methyl nonanoate (C9:0)	11.127
Methyl decanoate (C10:0)	12.629
Methyl laurate (C12:0)	15.567
Methyl myristate (C14:0)	18.569
Methyl myristoleate (C14:1)	19.708 and 20.265 (cis and trans)
Methyl palmitate (C16:0)	22.390
Methyl palmitoleate (C16:1)	23.599
Methyl stearate (C18:0)	27.134
Methyl oleate (C18:1)	27.922 and 28.205 (cis and trans)
Methyl linoleate (C18:2)	29.813
Methyl linoleneate (C18:3)	32.188

A2. Choice of internal standard

The most common fatty acids occurring in oils and fats used as feedstock for biodiesel production are palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1), linoleic acid (C18:2) and linolenic acid (C18:3) (Rezanka et al., 1999:255, Nakpong et al., 2010: 1684, Wang et al., 2012:183, Knothe et al., 2009: 5795). As the fatty acids are mostly even numbered, uneven carbon esters are often used as internal standards. Fernandes and co-workers (2012: 659) used methyl heptadecanoate for the characterisation of methylic and ethylic biodiesel from cottonseed and Jena and co-workers (2010: 1111) used methyl pentadecanoate for the characterisation of biodiesel produced from a mixture of mahua and simarouba oils. In this study, methyl nonanoate was used as the internal standard.

A3. Calibration of gas chromatograph

Calibration curves for fatty acids occurring in WPG were set up to determine the weight percentage values of the esters in the standard samples. A constant mass of the internal standard and a varying mass of the ester was used to prepare the calibration samples. The calibration solution therefore contains a fix known concentration of the methyl nonanoate and a varying concentration of the specific methyl ester.

The chromatogram is obtained for each ester at its specific retention time with the peak area in relation to the abundance of the ester in the sample. The area under each chromatogram peak is integrated by the software of the gas chromatograph (Chem stations ®) and used to quantify the different esters. Each sample is analysed in triplicate. Tables A2-A8 show the calibration data obtained for the methyl esters relevant to this study.

Table A2: Methyl laurate calibration data and mass and area ratios.

Mass C12:0 (g)	Mass C9:0 (g)	$\frac{Mass\ C12:0}{Mass\ C9:0}$	Area C12:0 (pA)	Area C9:0 (pA)	$\frac{Area\ C12:0}{Area\ C9:0}$
0.023	0.018	1.278	5253.00	3845.00	1.37
0.032	0.018	1.778	6630.00	3603.00	1.84
0.044	0.018	2.444	9458.00	3685.00	2.57
0.058	0.019	3.053	13180.00	3895.00	3.38
0.072	0.018	4.000	17110.00	4159.00	4.12

Table A3: Methyl myristate calibration data and mass and area ratios.

Mass C14:0 (g)	Mass C9:0 (g)	$\frac{Mass\ C14:0}{Mass\ C9:0}$	Area C14:0 (pA)	Area C9:0 (pA)	$\frac{Area\ C14:0}{Area\ C9:0}$
0.017	0.019	0.895	5964.00	4399.00	1.36
0.032	0.019	1.684	8135.00	4224.00	1.93
0.045	0.018	2.500	11560.00	4232.00	2.74
0.059	0.019	3.105	14990.00	4262.00	3.51
0.074	0.019	3.895	18620.00	4099.00	4.54

Table A4: Methyl palmitate calibration data and mass and area ratios

Mass C16:0 (g)	Mass C9:0 (g)	$\frac{Mass\ C16:0}{Mass\ C9:0}$	Area C16:0 (pA)	Area C9:0 (pA)	$\frac{Area\ C16:0}{Area\ C9:0}$
0.024	0.018	1.333	6648.00	4525.00	1.48
0.028	0.018	1.556	7748.00	4406.00	1.76
0.074	0.017	4.353	19730.00	4214.00	4.66
0.009	0.017	0.529	2280.00	4267.00	0.53
0.028	0.017	1.647	6799.00	3802.00	1.79

Table A5: Methyl palmitoleate calibration data and mass and area ratios.

Mass C16:1 (g)	Mass C9:0 (g)	$\frac{Mass\ C16:1}{Mass\ C9:0}$	Area C16:1 (pA)	Area C9:0 (pA)	$\frac{Area\ C16:1}{Area\ C9:0}$
0.023	0.018	1.278	5552.00	3834.00	1.44
0.032	0.019	1.684	7948.00	4017.00	1.98
0.046	0.018	2.556	11390.00	3619.00	3.14
0.061	0.019	0.842	14950.00	3820.00	3.92
0.074	0.018	4.111	17670.00	3821.00	4.63

Table A6: Methyl stearate calibration data and mass and area ratios

Mass C18:0 (g)	Mass C9:0 (g)	$\frac{Mass\ C18:0}{Mass\ C9:0}$	Area C18:0 (pA)	Area C9:0 (pA)	$\frac{Area\ C18:0}{Area\ C9:0}$
0.021	0.019	1.105	5542.00	4349.00	1.27
0.033	0.017	1.941	8941.00	3940.00	2.28
0.060	0.018	3.333	15730.00	3651.00	4.30
0.071	0.019	3.737	17620.00	4023.00	4.38
0.083	0.018	4.611	20120.00	3846.00	5.27

Table A7: Methyl oleate calibration data and mass and area ratios

Mass C18:1 (g)	Mass C9:0 (g)	$\frac{Mass\ C18:1}{Mass\ C9:0}$	Area C18:1 (pA)	Area C9:0 (pA)	$\frac{Area\ C18:1}{Area\ C9:0}$
0.024	0.019	1.263	6575.00	3997.00	1.65
0.033	0.019	1.737	8561.00	4058.00	2.10
0.047	0.019	2.474	12310.00	4118.00	2.98
0.061	0.020	3.050	15760.00	3984.00	3.93
0.077	0.019	4.053	19090.00	3913.00	4.92

Table A8: Methyl linoleate calibration data and mass and area ratios

Mass C18:2 (g)	Mass C9:0 (g)	$\frac{Mass\ C18:2}{Mass\ C9:0}$	Area C18:2 (pA)	Area C9:0 (pA)	$\frac{Area\ C18:2}{Area\ C9:0}$
0.023	0.019	1.211	6157.00	4160.00	1.47
0.035	0.018	1.944	8954.00	4103.00	2.18
0.047	0.018	2.611	12120.00	4102.00	2.96
0.064	0.019	3.368	16410.00	4151.00	3.96
0.080	0.018	4.444	19900.00	4027.00	4.96

A straight line is produced when the mass ratio of the ester standards to the internal standard is plotted against the area ratio of the ester standard to the internal standard. The calibration curves for the selected standard ester solutions are shown in Figure A1 to A7.

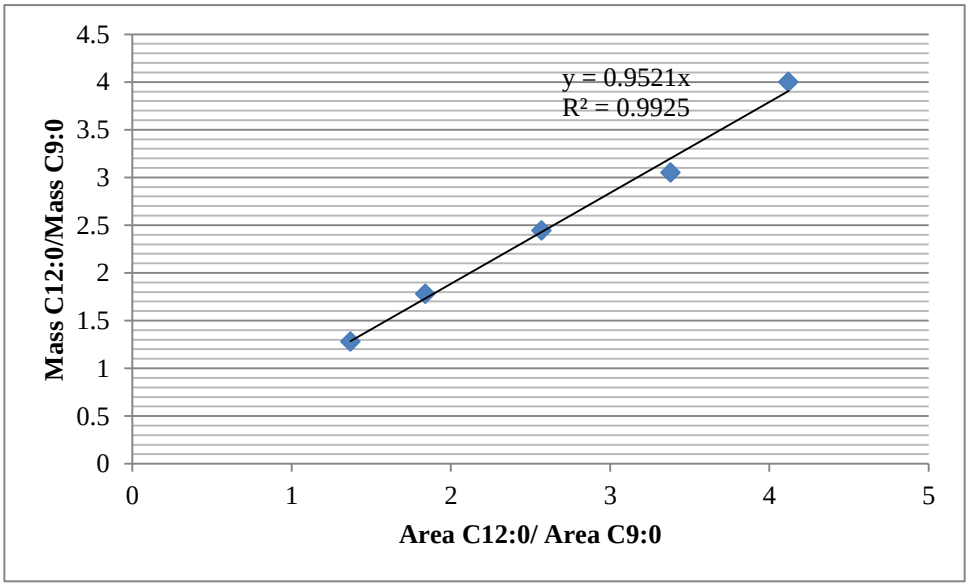

Figure A1: Calibration curve for C12:0

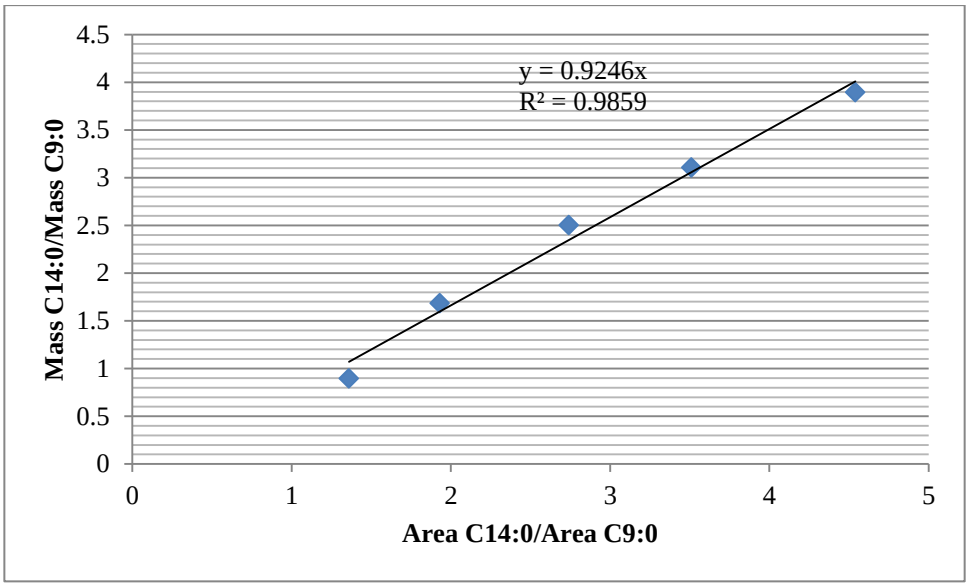

Figure A2: Calibration curve for C14:0

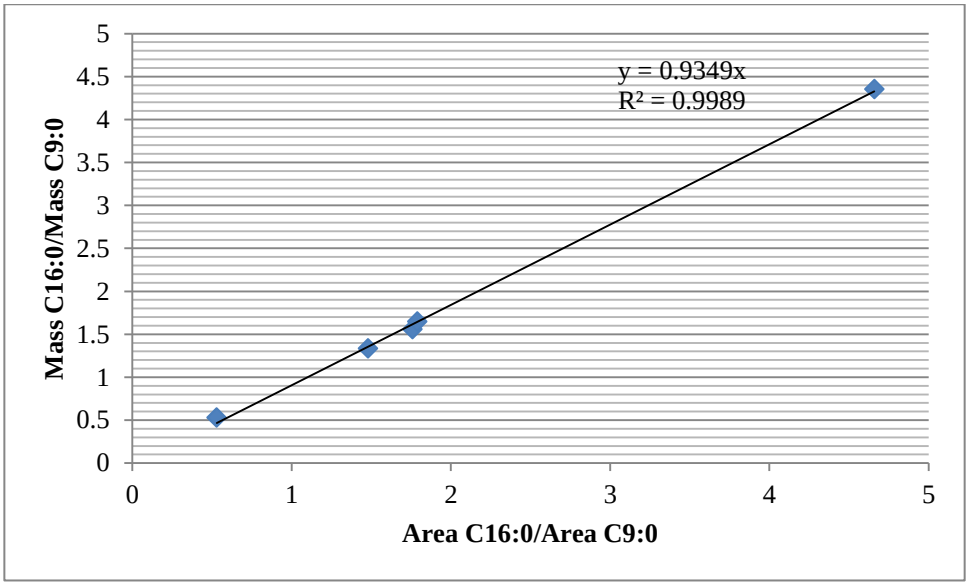

Figure A3: Calibration curve for C16:0

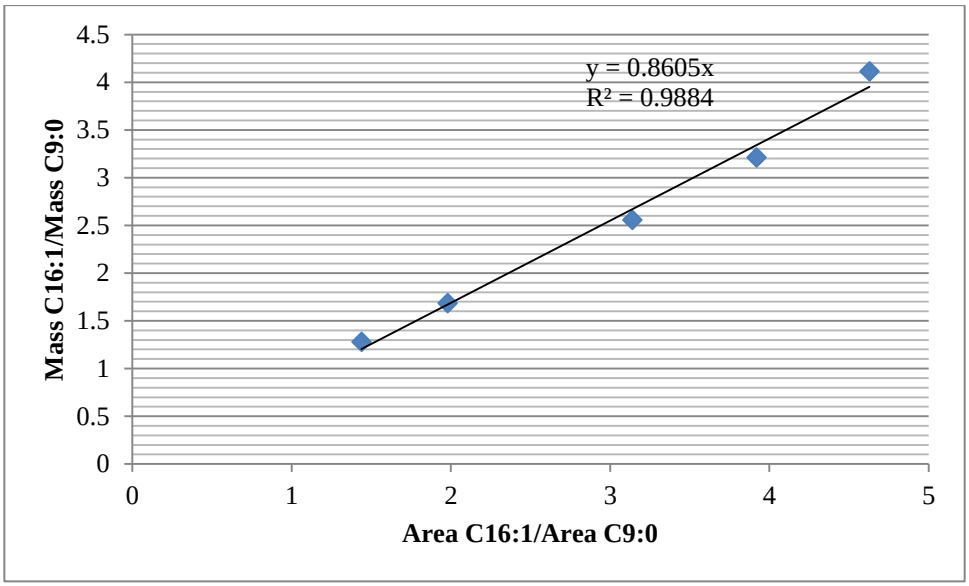

Figure A4: Calibration curve for C16:1

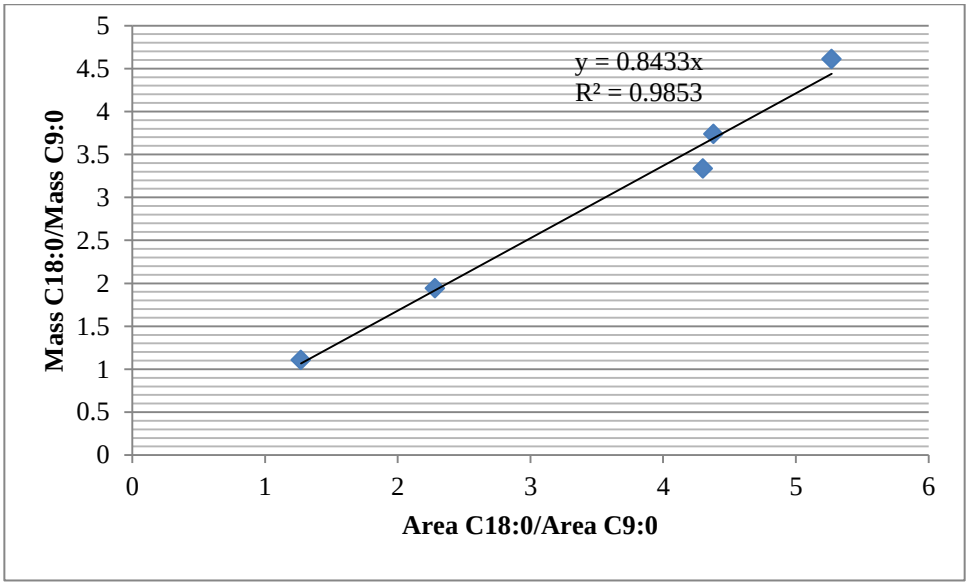

Figure A5: Calibration curve for C18:0

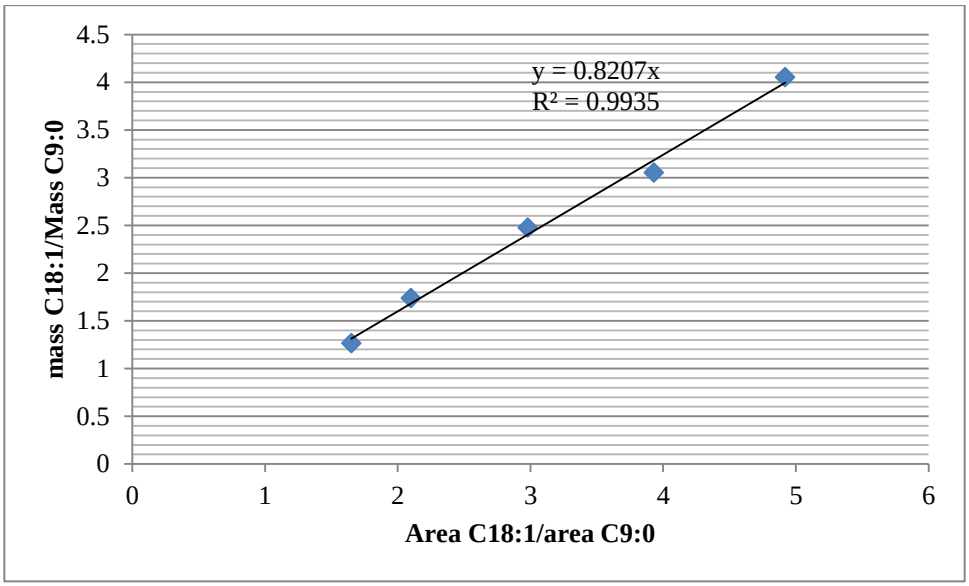

Figure A6: Calibration curve for C18:1

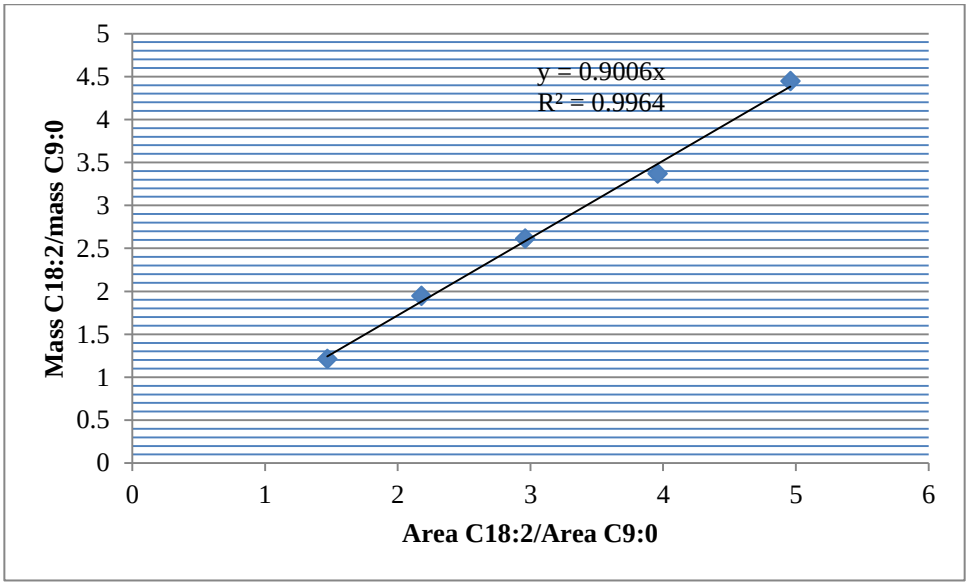

Figure A7: Calibration curve C18:2

Equation A1 was used to calculate the calibration curves for the different esters

$$\frac{m_i}{m_{C9:0}} = k_{ci} \frac{A_i}{A_{C9:0}} \qquad \textbf{Equation A1}$$

Where m_i is the mass of the ester, $m_{C9:0}$ is the mass of the internal standard, k_{ci} is the calibration constant for the ester, A_i is the peak area of the ester and $A_{C9:0}$ is the peak area of the internal standard.The calibration constant is calculated as the slope of equation A1.

The calibration curve constants for the esters are presented in Table A9.

Table A9: calibration constants for the esters.

FAME	K_c-values
Methyl laurate (C12:0)	0.9521
Methyl myristate (C14:0)	0.9246
Methyl palmitate (C16:0)	0.9349
Methyl palmitoleate C16:1)	0.8605
Methyl stearate (C18:0)	0.8433
Methyl oleate (C18:1)	0.8207
Methyl linoleate (C18:2)	0.9006

The quantity of the methyl esters in a sample with an unknown composition can be calculated as this is the only unknown in the equation.

A4 References

Fernandes, D. M., Serqueira, D. S., Portela, F. M., Assuncao, M.N., Munoz, R.A.A., Terrones, G.H. 2012. Preparation and characterisation of methylic and ethylic biodiesel from cottonseed oil and effect of tert-butylhydroquinone on its oxidative stability. Fuel 97: 658- 661.

Jena, P. C., Raheman, H., Kumar, G.V.P., Machavaram, R. 2010. Biodiesel production from mixture of mahua and simarouba oils with high free fatty acids. Biomass and Bioenergy 34: 1108- 1116.

Knothe, G., Steidley, K.R. 2009. A comparison of used cooking oils: a very heterogeneous feedstock for biodiesel. Bioresource Technology 100:5796- 5801.

Nakpong, P., Wootthhikanokkhan, S. 2010. High free fatty acid coconut oil as a potential feedstock for biodiesel production in Thailand. Renewable Energy 35:1682- 1687.

Rezanka, T., Rezankova, H. 1999. Characterisation of fatty acids and triacylglycerols in vegetable oils by gas chromatography and statistical analysis. Analytica Chimica Acta, 398: 253- 261.

Wang, R., Zhou, W.W., Hanna, M.A., Zhang, Y., Bhadury, P. S., Wang, Y., Song, B., Yang, S. 2012. Biodiesel preparation, optimization, and fuel properties from non-edible feedstock, Datura stramonium L. Fuel 91; 182- 186.

Appendix B. - Experimental data

The experimental data used in this study is presented in this Appendix. Section B.1 lists the experimental data for the optimisation of the pre-treatment reaction parameters for the conventional alkaline transesterification method for biodiesel production from WPG. Section B.2 shows the experimental data on the effect of different solvent combinations on the extraction of the acid oil mixture from WPG and the effect of the different solvent combinations on the co-extraction of sulphur compounds into the acid oil mixture. Experimental data for the optimisation of the chromatographic conditions for the reduction of sulphur compounds in the WPG is shown in section B.3.

B1 Optimisation of the pre-treatment parameters for the conventional biodiesel production process

Table B.1 lists the reduction in FFA at different temperatures, Table B.2 the reduction of FFA at different methanol to oil ratios and table B.3 the reduction of FFA at different catalyst loadings. Table B.4 list the experimental data for the reduction of FFA for the second esterification step.

Table B1: Data for the esterification of free fatty acids of WPG at different temperatures.

Run number	Bath temperature ° C	Reactor temperature ° C	Catalyst loading (wt %)	Molar ratio (methanol :oil)	FFA wt %
1	55	53.5	3	8:1	18.43
2	55	53.5	3	8:1	17.72
3	55	53.5	3	8:1	16.61
4	65	63	3	8:1	12.62
5	65	63	3	8:1	11.80
6	65	63	3	8:1	12.91
7	75	64.5	3	8:1	14.01
8	75	64.5	3	8:1	13.40
9	75	64.5	3	8:1	14.03
10	85	65	3	8:1	12.21
11	85	65	3	8:1	12.45
12	85	65	3	8:1	11.61
13	95	65	3	8:1	12.80
14	95	65	3	8:1	13.02
15	95	65	3	8:1	13.14

Table B2: Data for the esterification of free fatty acids of WPG at different molar ratios of methanol to oil.

Run number	Bath temperature °C	Reactor temperature °C	Catalyst loading (wt %)	Molar ratio (methanol :oil)	FFA wt %
16	75	75	3	3:1	21.70
17	75	75	3	3:1	20.91
18	75	75	3	3:1	22.10
19	75	67.5	3	6:1	17.51
20	75	67.5	3	6:1	18.32
21	75	67.5	3	6:1	18.50
22	75	64.5	3	8:1	14.01
23	75	64.5	3	8:1	13.41
24	75	64.5	3	8:1	14.05
25	75	64	3	10:1	10.32
26	75	64	3	10:1	10.01
27	75	64	3	10:1	10.80
28	75	63	3	12:1	8.02
29	75	63	3	12:1	7.91
30	75	63	3	12:1	8.72
31	75	62.5	3	18:1	4.80
32	75	62.5	3	18:1	4.91
33	75	62.5	3	18:1	4.50

Table B3: Data for the esterification of free fatty acids of WPG at different catalyst loadings

Run number	Bath temperature °C	Reactor temperature °C	Catalyst Loading (wt %)	Molar ratio (methanol : oil)	FFA wt %
34	75	62.5	0.5	8:1	42.80
35	75	62.5	0.5	8:1	43.42
36	75	62.5	0.5	8:1	44.01
37	75	63	1	8:1	42.24
38	75	63	1	8:1	45.17
39	75	63	1	8:1	43.53
40	75	64	2	8:1	31.30
41	75	64	2	8:1	30.32
42	75	64	2	8:1	32.51
43	75	64.5	3	8:1	13.42
44	75	64.5	3	8:1	14.01
45	75	64.5	3	8:1	14.04
46	75	65.5	4	8:1	8.40
47	75	65.5	4	8:1	8.32
48	75	65.5	4	8:1	8.26
49	75	66.5	5	8:1	7.10
50	75	66.5	5	8:1	7.22
51	75	66.5	5	8:1	6.01

Table B4: Data for the esterification of free fatty acids of WPG for the second acid esterification step

Run number	Bath temperature °C	Reactor temperature °C	Catalyst loading wt%	Molar ratio (methanol :oil)	Time (min)	FFA (wt %)
1	75	64	4	8:1	2	0.39
2	75	64	4	8:1	5	0.25
3	75	64	4	8:1	10	0.22
4	75	64	4	8:1	15	0.20
5	75	64	4	8:1	20	0.16
6	75	64	4	8:1	30	0.18
7	75	64	4	8:1	2	0.18
8	75	64	4	8:1	5	0.32
9	75	64	4	8:1	10	0.27
10	75	64	4	8:1	15	0.25
11	75	64	4	8:1	20	0.25
12	75	64	4	8:1	30	0.18
13	75	64	4	8:1	2	0.40
14	75	64	4	8:1	5	0.23
15	75	64	4	8:1	10	0.23
16	75	64	4	8:1	15	0.25
17	75	64	4	8:1	20	0.21
18	75	64	4	8:1	30	0.18

B2. The evaluation of different solvent combinations for the extraction of free fatty acids from WPG .

Table B.5 shows the mass of acid oil mixture extracted at different solvent combinations and table B.6 shows the effect of the solvent combination on the reduction of sulphur compounds in the extracted acid oil mixture.

Table B5: Data for the mass of acid oil mixture extracted from WPG at different concentrations of methanol, ethanol and iso-propanol in the solvent mixture.

Run number	Alcohol concentration in solvent (g/100g)	Acid oil mixture extracted (g/100g solvent)		
		Methanol	Ethanol	Iso-propanol
1	0	10.80	10.80	10.80
2	10	12.60	12.88	11.92
3	20	12.65	20.18	19.20
4	30	18.75	23.18	26.87
5	40	26.85	32.28	35.60
6	50	29.45	-	-
7	0	9.80	9.80	9.80
8	10	12.30	13.68	12.12
9	20	12.25	16.72	18.03
10	30	17.65	25.85	25.74
11	40	22.65	36.57	36.79
12	50	28.75	-	-
13	0	9.45	9.45	9.45
14	10	12.80	14.72	10.84
15	20	13.05	18.76	21.98
16	30	20.05	21.95	21.60
17	40	25.30	31.80	33.92
18	50	30.65	-	-

Table B6: Data for the effect of solvent composition on the selectivity towards sulphur extraction from the acid oil mixture.

Run number	Alcohol concentration in solvent (g/100g)	Sulphur concentration in acid oil mixture (ppm)		
		Methanol	Ethanol	Iso-propanol
1	0	4861	4861	4861
2	10	5287	5329	5908
3	20	6185	5530	6201
4	30	6395	6617	6988
5	40	6840	6999	7711
6	50	7559	-	-
7	0	4772	4772	4772
8	10	5892	5427	5971
9	20	6130	5669	6287
10	30	6319	5945	5930
11	40	6886	7063	7762
12	50	7560	-	-
13	0	4657	4657	4657
14	10	4917	5527	5782
15	20	6074	5782	5945
16	30	6337	6708	6847
17	40	6866	6989	7790
18	50	7557	-	-

B3. Optimisation of the chromatographic conditions for the reduction of sulphur in the acid oil

Table B.7 lists the sulphur content of the WPG where different chromatographic systems were used, Table B.8 shows the effect of different solvents as mobile phase on the reduction of sulphur for the hydrophobic column, table B.9 shows the effect of oil to resin ratio on the reduction of sulphur compounds for the hydrophobic column and table B.10 the effect of passing the same sample multiple time through the hydrophobic column with methanol as the mobile phase.

Table B7: Data on the effect of different chromatographic systems on the reduction of sulphur compounds in WPG

Run number	Stationary phase	Mobile phase	Sulphur content (ppm)
	Hydrophobic resin (MN200)		
1		Methanol	1130
2		Methanol	1057
3		Methanol	989
4		Hexane	5834
5		Hexane	5720
6		Hexane	5779
	Cation exchange resin (PD206)		
7		Methanol	7783
8		Methanol	8785
9		Methanol	7653
10		Hexane	7854
11		Hexane	7489
12		Hexane	6999
	Anion exchange resin (A860)		
13		Methanol	8118
14		Methanol	7976
15		Methanol	7675
16		Hexane	Ad
17		Hexane	Ad
18		Hexane	Ad
	Silica gel		
19		Methanol	8565
20		Methanol	8087
21		Methanol	7954
22		Hexane	Ad

Table B7: (Continue)

23	Hexane	Ad
24	Hexane	Ad

Table B8: Data for the effect of different solvents as mobile phase using the hydrophobic resin on the reduction of sulphur compounds from the WPG

Run number	Mobile phase	Sulphur content (ppm)
1	Hexane	5834
2	Hexane	5720
3	Hexane	5779
4	Iso-propanol	5477
5	Iso-propanol	5543
6	Iso-propanol	5590
7	Ethanol	5024
8	Ethanol	4921
9	Ethanol	5235
10	Methanol	1057
11	Methanol	1130
12	Methanol	989
13	Methanol/10% water	176
14	Methanol/10% water	88
15	Methanol/10% water	102

Table B9: Data for the effect of oil to resin ratio on the reduction of sulphur from the acid oil using the hydrophobic chromatography column with methanol as mobile phase

Run number	Oil to resin ratio	Sulphur (ppm)
1	0.0625	1054
2	0.125	2213
3	0.25	5050
4	0.0625	984
5	0.125	2436
6	0.25	5119
7	0.0625	969
8	0.125	2209
9	0.25	4917

Table B10: The effect of passing the same sample at different oil to resin ratios multiple times through the column using the hydrophobic column with methanol as the mobile phase

Run number	Stage	Oil to resin ratio 0.25 Sulphur (ppm)	Oil to resin ratio 0.125 Sulphur (ppm)	Oil to resin ratio 0.0625 Sulphur (ppm)
1	0	9112	9112	9112
2	1	5119	2436	1270
3	2	4315	1400	854
4	3	3127	870	575
5	4	2620	-	501
6	5	2080	570	419
7	0	8996	8996	8996
8	1	5198	2350	1567
9	2	4423	1043	934
19	3	3212	529	570
11	4	2506	-	498
12	5	1978	562	412
13	0	9230	9230	9230
14	1	5362	2213	1362
15	2	4287	1355	999
16	3	3017	752	674
17	4	2505	-	460
18	5	1965	593	459

Appendix C - Biodiesel Costs

Appendix C describes the cost estimation made to give an indication of the economic viability of route 3. The economic estimation is based on the process model developed by Haas and co-workers (2006:671). The purpose of this economic estimation is only to give a rough generic indication of whether the process in route 3 will result in a higher or lower cost compared to the plant capacity, type of feedstock, fixed capital and total manufacturing cost used by Haas and co-workers and should only be used for research purposes.

C1. Biodiesel costing

Based on the capital cost suggested by Haas and co-workers (2006:676), a rough estimation was made for the capital needed to construct a 37,854,118 litre per annum biodiesel plant using WPG as the feedstock based on route 3 as shown in table C1. The US$ values used by Haas and co-workers (2006:671) were converted to South African Rand taking into account the Rand/$ exchange rate for 2006 and the PPI (Production Price Index) for inflation adjustment (Statistics South Africa, 2013).

A block process description of the plant for the production of biodiesel from WPG is shown in figure C1. Results from experimental work for route 3 indicated that a biodiesel yield of 45% based on the mass of dried WPG used was achieved. As shown in table 3.2, the oil content of the WPG was measured to be 64.04% with the difference made up by impurities like solids, metal salts and additives. The crude acid oil yield after saponification and acidulation calculated as the mass of crude acid oil as percentage of the total mass of dried WPG used, was 91% where some of the metal impurities were dissolved in the hydrochloric acid used in the acidulation step resulting in crude acid oil containing additives, some fine solids which did not dissolved during the acidulation step, possibly some glycerine that formed during the saponification step and emulsified by the additives. The yield of purified acid oil after the hydrophobic chromatographic purification step was 55.89%. The yield of crude biodiesel after two esterification steps was 55.1% with a final biodiesel yield of 44.71%.

During saponification and acidulation, the original mass of WPG used in the experiment was reduced by 9% due to the dissolution of metals in the hydrochloric acid and washed out together with other impurities in the water washing step.

During the reverse phase hydrophobic purification step, the original mass of WPG was further reduced by 35.11% where impurities like fine solids and additives as well as some acid oil was retained by the resin columns. After esterification, with the hydrophyllic purification step, the original mass was further reduced by 10.39%.

The waste generated by the route 3 process steps consisted of acid water and residual waste oil. The acidified water containing dissolved metals from the saponification/acidulation steps which require an acid water treatment step. Provision for waste water treatment of 30 cents per liter diesel produced was made in the economic analysis. The regeneration of the hydrophilic and hydrophobic columns resulted in residual oil which contains additives and other imputities. The potential to reuse the recovered additive containing residual oil with neat rolling oil needs further evaluation. The economic benefit of the residual oil and the purification cost of such product were not included in the economic evaluation at this stage.

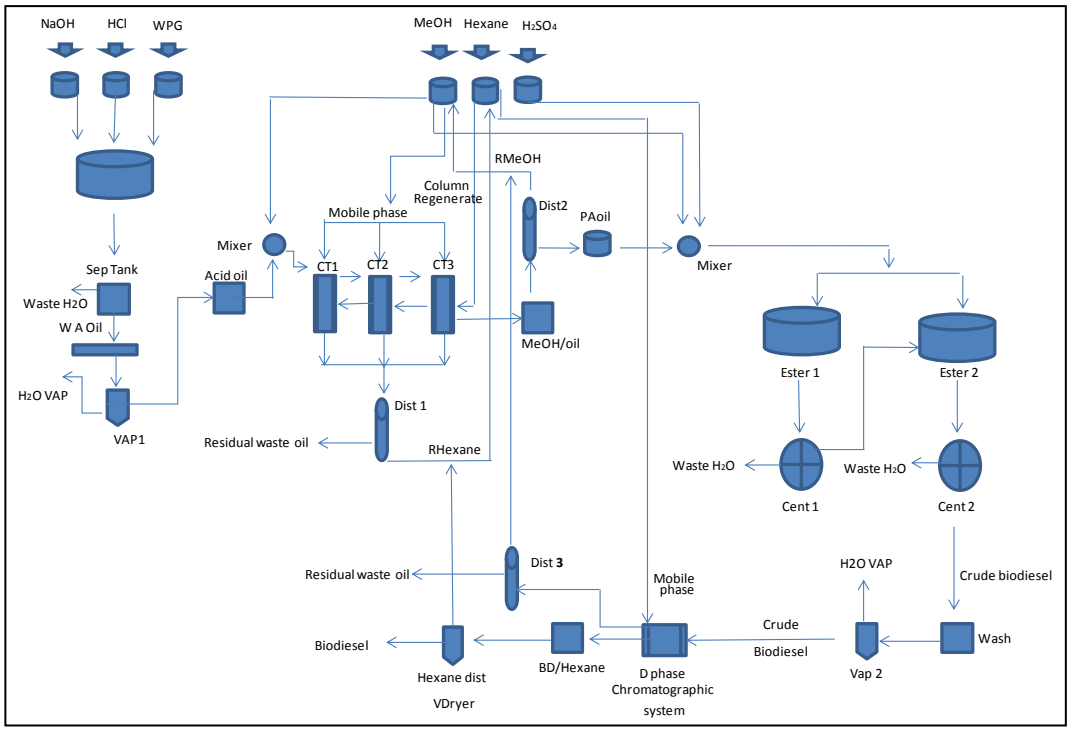

Figure C1: Block process description of the production of biodiesel from WPG. (SEP TANK: separation tank; W A OIL: wet acid oil; H_2O VAP: water vapour; VAP1, VAP2: evaporator 1and 2; MEOH: methanol; RMEOH: recovered methanol; RHEXANE: recovered hexane; DIST1, 2, 3, distillation columns 1,2 and 3; MeOH/OIL: methanol oil mixture; CT1,2,3: hydrophobic chromatographic system; PA OIL: purified acid oil; ESTER 1,2: esterification reactors 1, 2; CENT1,2: centrifugal separators; D PHASE CHROMATOGRAPHIC SYSTEM: direct phase chromatographic system; BD/HEXANE: biodiesel hexane mixture; HEXANE DIST VDRYER: hexane distillation and biodiesel vacuum drier)

Table C1: Capital cost estimation for the construction of a 38 million litre per annum plant Using WPG as feedstock

Item	Cost South African Rand, thousands)
Storage facilities	
WPG storage facility	5146
Biodiesel storage tank	4546
Loading/unloading	508
Pumps to/from storage	206
Subtotal storage facilities	10406
Process equipment	
Sodium hydroxide storage facility	250
Hydrochloric acid storage tank	260
Reactor 1 Saponification/acidulation	712
Reactor 1 pre-heater	31
Water/acid oil separation tank	350
Acid oil/water removal pre-heater	92
Acid oil/water removal heater	20
Acid oil/water removal flash tank	153
Acid oil/methanol mixer	71
Methanol storage tank	244
Hexane storage tank	250
Reverse phase chromatographic system	15813
MN200 resin @ US$ 27777.78/m^2	28239
Methanol/oil tank	350
Methanol distillation tower pre-heater	81
Methanol distillation tower	1932
Distillation reboiler	102
Distillation condenser	264
Hexane distillation tower pre-heater	81
Hexane distillation tower	1932

Table C1: (Continue)

Distillation reboiler	102
Distillation condenser	264
Purified acid oil tank	350
Sulphuric acid storage tank	264
Acid oil/methanol/catalyst mixer	71
Esterification reactor 1 pre-heater	31
Esterification reactor 1	712
Esterification reactor 2 pre-heater	31
Esterification reactor 2	620
Biodiesel/water separator 1	3336
Biodiesel /water separator 2	3336
Biodiesel wash tank	356
Biodiesel water removal pre-heater	92
Biodiesel water removal heater	20
Biodiesel water removal flash tank	153
Direct phase chromatographic system	12203
Silica gel	15000
Biodiesel/hexane tank	350
Biodiesel hexane removal pre-heater	92
Biodiese hexane removal heater	20
Hexane distillation system	364
Biodiesel hexane removal flash tank	153
Biodiesel hexane removal vacuum system	763
Residual waste oil storage tank	250
Subtotal processing	90160
Utility equipment	4098

Table C1: (continue)

Total equipment cost	104664
Other costs	
Installation @ 200% of equipment cost	209328
Miscellaneous	10169
Total capital costs	324161

Table C2 shows the annual and unit costs per litre biodiesel produced from WPG for 37 854 118 litres of biodiesel per annum.

Table C2: Annual and unit cost estimation for the production of 38 million litres per annum of biodiesel from WPG.

Description	Annual use	Annual cost (South African Rand, thousands)	Cost per litre diesel produced (South African Rand)
Raw materials and chemicals			
WPG	86000 ton	97073	2.5
Methanol	3374 ton	9805	0.26
Sodium hydroxide	6840 ton	42913	1.13
Hydrochloric acid	8360 ton	85002	2.25
Sulphuric acid	2064 ton	9233	0.24
Water	2253 ton	8	0.002
Subtotal raw materials		244034	6.38
Utilities		4129	0.11
Labour		5257	0.14
Supplies		1556	0.04
General works		1271	0.03

Table C2: (continue)

Depreciation @ 10% of capital cost per year	32416	0.85
Waste treatment	11400	0.3
Gross operating cost	300063	7.90

C2 References

Haas, M. J., McAloon, A. J., Yee, W.C., Foglia, T. A. 2006. A process model to estimate biodiesel production costs. Bioresource Technology, 97: 671 -678.

Statistics South Africa. 2013. Producer Price Index (PPI) for domestic output of South African industry groups. http://www.stassa.gov.za/keyindicators/ppi.asp Date of access: 2 May 2013.

Appendix D - Calculations

D1 Oil content of WPG

The oil content of the WPG was calculated with equation D1:

$$\text{Oil content } (wt\%) = \frac{M_{fa}}{M_{WPG}} \qquad \textbf{Equation D1}$$

Where M_{fa} is the mass of fatty acids in the WPG as determined by gas chromatography and $M_{WPG}$is the total mass of WPG used in the analysis. The triglycerides and the free fatty acids in the WPG were transesterified and esterified respectively using 10% boron trifluoride as a catalyst based on the method used by Abdulkadir and Tsuchiya (2008:4).

D2 Experimental Error

D2.1 Free fatty acid content

The experimental error for the reduction of FFA in WPG at different methanol to oil ratios is shown below. The experiment was repeated three times and the standard deviation was calculated. The experiment was done at an oil bath temperature of 75 °C, a catalyst loading of 3 wt% and a reaction time of 1 hour.

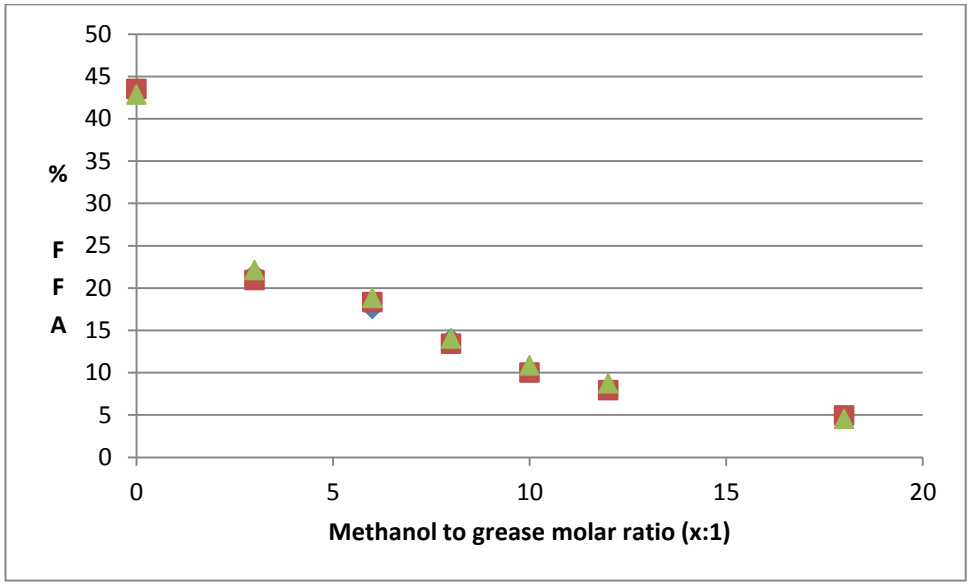

Figure D1: Experimental error for the reduction of FFA at different methanol to grease ratios. Repeat 1 (■) Repeat 2 (▲) Repeat 3 (◆)

Calculation of experimental error

The average of the values for the repeated experiments was calculated using equation D2

$$\bar{x} = \frac{1}{N}\sum_{i=1}^{N} x_i \qquad \textbf{Equation D2}$$

Where $\bar{x}$ is the average, x_i is the individual data points and N is the number of data points.

The standard deviation of the values was determined using equation D3

$$\delta = \sqrt{\frac{\sum_{i=1}^{N}(x_i - \bar{x})2}{n-1}} \qquad \textbf{Equation D3}$$

Where the standard deviation is represented by δ.

The experimental error was calculated using equation D4.

$$Experimental\ error\ = 2 \times \frac{Confidence\ interval}{\bar{x}} \times 100 \qquad \textbf{Equation D4}$$

The experimental error calculations of the FFA content at different methanol to grease ratios are shown in table D1:

Table D1: Experimental error of the FFA content of WPG at different methanol to grease ratios

Molar ratio (x:1)	FFA content Repeat 1	FFA content Repeat 2	FFA content Repeat 3	Average	Standard deviation	Experimental error (%)
3:1	21.70	20.91	22.10	21.57	0.61	6.36
6:1	17.51	18.32	18.50	18.11	0.53	6.59
8:1	14.01	13.41	14.05	13.82	0.36	5.88
10:1	10.32	10.01	10.80	10.38	0.40	8.68
12:1	8.02	7.91	8.72	8.22	0.44	12.09
18:1	4.80	4.91	4.50	4.74	0.21	10.03

The average experimental error for this experiment is 8.27%. The overall experimental error for all the experiments was calculated to be 12.55%.

D2.2. Water content

The moisture content of the dried WPG was measure using Karl Fischer Coulometry. The sample was analysed in triplicate.

Table D2: Experimental error for moisture content of dried WPG measure with Karl Fischer Coulometry

Repeat	Water content (ppm)
1	3188
2	3227
3	3265
Average	3226.667
Standard deviation	38.502
Experimental error	2.700

D2.3 Acid value

The acid value of the dried WPG was measured using a potentiometric titration with KOH as titrant.

Table D3: Experimental error for acid value of WPG measured by potentiometric titration

Repeat	Acid value(mg KOH /g)
1	8.75
2	8.47
3	8.68
Average	8.637
Standard deviation	0.140
Experimental error	3.67

D2.4 Ester content

The ester content of the final biodiesel was determined by gas chromatography based on the EN 14103 standard method for fatty acid analysis. The experimental error reported for GC analysis was based on a measured value which includes the experimental error for the calibration of the gas chromatograph as well as the experimental error for the sample preparation. Therefore the combined experimental error is contained in the final value.

Table D4: Experimental error for ester content of biodiesel produced by route 3

Repeat	Ester content (wt%)
1	93.5
2	93.2
3	93.0
Average	93.2
Standard deviation	0.25
Experimental error	5.12

D3 References

Abdulkadir, S., Tsuchiya, M. 2008. One-step method for quantitative and qualitative analysis of fatty acids in marine animal samples. Journal of Experimental Marine Biology and Ecology, 352: 1-8.