In-situ biodiesel production using liquefaction and supercritical extraction

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Abstract

Biodiesel is an alternative source of energy with the potential to replace diesel fossil fuel. Full scale production however, has been hampered by high production costs since the extraction of oil causes excessive losses of energy, time and money when producing biodiesel. Currently, there is a lack of innovative technologies that combine known techniques to collectively overcome individual shortfalls. The aim of this project is to investigate the feasibility of combining liquefaction, *in -situ* trans-esterification and supercritical carbon dioxide extraction, to develop a biodiesel production process that requires a minimum amount of energy, is fast and efficient in the extraction of oil from biomass, while simultaneously converting extracted oil into biodiesel.

Sunflower seeds were used as feedstock for liquefaction in a methanol/-catalyst solution to produce biodiesel under high temperatures and pressures. Experiments were conducted with three different catalysts, potassium hydroxide (KOH), sulphuric acid (H_2SO_4) and calcium carbonate ($CaCO_3$), at five different temperatures, varying with 10˚C intervals from 320˚C to 360˚C; and with three different biomass loadings of 15wt%, 20wt% and 25wt% of the reaction mixture in the presence of supercritical CO_2. The final product was analysed with gas chromatography, gas chromatography-mass spectrometry, elemental analysis and FT-IR to determine the product quality.

The results from the various temperatures peaked at 340˚C; this temperature delivered yields of 120 $g.kg^{-1}$ bio-oil, 84.7 $g.kg^{-1}$ FAME and 147.9 $g.kg^{-1}$ biochar. The bio-oil had a HHV of 41.12 $MJ.kg^{-1}$ and the biochar 28.12 $MJ.kg^{-1}$. It was evident that temperature had little influence on the elemental composition, but it was a major determinant of bio-oil, FAME, and biochar production.

The results obtained for the different types of catalysts varied. Potassium hydroxide catalysed reactions peaked at 320˚C, delivering yields of 110.55 $g.kg^{-1}$ bio-oil, 71.72 $g.kg^{-1}$ FAME and 108.95 $g.kg^{-1}$ biochar. The bio-oil had a HHV of 40.12 $MJ.kg^{-1}$ and the biochar 25.20 $MJ.kg^{-1}$. The catalyst favoured gas production. Sulphuric acid catalysed reactions peaked at 320˚C; this temperature delivered yields of 61.15 $g.kg^{-1}$ bio-oil, 36.61 $g.kg^{-1}$ FAME and 367.25 $g.kg^{-1}$ biochar. The bio-oil had a HHV of 42.28 $MJ.kg^{-1}$ and the biochar 28.73 $MJ.kg^{-1}$. The catalyst favoured biochar production. Calcium carbonate catalysed reactions peaked at 340˚C,

delivering yields of 120 $g.kg^{-1}$ bio-oil, 84.7 $g.kg^{-1}$ FAME and 147.9 $g.kg^{-1}$ biochar. The bio-oil had a HHV of 41.12 $MJ.kg^{-1}$ and the biochar 28.12 $MJ.kg^{-1}$. The catalyst was superior to the other two and favoured bio-oil production. The three catalysts had varying influences on bio-oil, FAME, biochar production and subsequently, on calorific values.

The results for the biomass loading peaked at 20wt% loading using 20 $g.kg^{-1}$ $CaCO_3$ catalysts at 330°C; this load delivered yields of 120 $g.kg^{-1}$ bio-oil, 84.7 $g.kg^{-1}$ FAME and 147.9 $g.kg^{-1}$ biochar. The bio-oil had a HHV of 41.12 $MJ.kg^{-1}$ and the biochar 28.12 $MJ.kg^{-1}$. Biomass loading illustrated adverse effects on product distribution between biochar, bio-oil and biogas production.

There was an abundance of aromatic compounds, stemming from unsaturated, branched, uneven hydrocarbon chains present in the bio-oil. Analysis of the C/H molar ratios against the C/O content showed the change in oil quality relative to specific fossil fuels. The HHV increased with higher carbon-hydrogen ratios and lower oxygen ratios. The results motioned an apparent threshold limit to de-oxygenation. The bio-oil can therefore be classified as paraffin-like, but a high level of methyl esters means that the bio-oil should be classified as low quality diesel oil.

The products of *in -situ* trans-esterification by liquefaction and supercritical carbon dioxide can compete with fossil fuels regarding the HHV. However, a low quality biodiesel was produced; this requires secondary processing to reduce the cyclic compounds and oxygen content. These findings contribute towards the current growing bed of knowledge to overcome the blatant lack of innovative usages of the existing technology.

Keywords: Biodiesel; Liquefaction; Supercritical carbon dioxide; *In -situ* trans-esterification; Sunflower seed.

Opsomming

Biodiesel is 'n alternatiewe bron van energie wat as kandidaat geïdentifeer is om diesel-fossielbrandstof te vervang. Groot skaalse produksie word egter steeds gekniehalter deur buitensporige produksie koste. Buitensporige hoeveelhede tyd, geld en energie gaan verlore tydens die olie-ekstraksie stappe in produksie. Daar is tans 'n gebrek aan innoverende tegnologieë wat bestaande metodes kombineer om individuele metodes se gebrekke te oorkom. Hierdie navorsings-projek is daarop gerig om ondersoek in te stel na die haalbaarheid van 'n gekombineerde tegniek bestaande uit vervloeiing, *in -situ* trans-esterifikasie en superkrietiese koolstofdioksied ektraksie. Die word gedoen om 'n biodiesel-produksie-proses te ontwikkel wat die minimum hoeveelheid energie verbruik, wat olie vinnig en effektief uit biomassa ekstraheer en terselfde tyd die olie omskakel in biodiesel.

Sonneblom-saad is as olie-bron vir die vervloeiings-operasie, gekombineer met 'n metanol/ katalisator-oplossing, gebruik om biodiesel te vervaardig by hoë temperature en drukke. Eksperimente is gedoen om ondersoek in te stel na die effektiwiteit van drie katalisators, kalium hidroksied (KOH), swaelsuur (H_2SO_4) en kalsium karbonaat ($CaCO_3$). Die temperatuur het gewissel met 10°C intervalle vanaf 320°C to 360°C en die biomassa-ladings is van 15%, 20% en 25% van die massa-oplossing gebruik. Hierdie reaksies van die variasies het almal in die teenwoordigheid van superkrietiese CO_2. Die finale produk is ge-analiseer deur middel van gas-chromatografie, gas-chromatografie gekoppeld aan 'n massa spektrometer, elementale analiese en FTIR om die kwaliteit daarvan te bepaal.

Die resultate vir die verskillende temperature dui daarop dat dar by 340°C die beste opbrengs gelewer is met resultate van 120 $g.kg^{-1}$ bio-olie, 84.7 $g.kg^{-1}$ FAME en 147.9 $g.kg^{-1}$ bio-houtskool. Die bio-olie het 'n HHV van 41.12 $MJ.kg^{-1}$ gehad en die van bio-houtskool was 28.12 $MJ.kg^{-1}$. Temperatuur het geen drastiese effek op die elementale samestelling van die eindprodukte gehad nie, maar was wel die bepalende faktor in die bio-olie-, FAME- en bio-houtskool-produksie.

In die resultate van die eksperimente waar in die katalisators gewissel is het die kalium-hidroksied-katalisator die beste resultate by 320°C gelewer met 'n opbrengs van 110.55 $g.kg^{-1}$ bio-olie, 71.72 $g.kg^{-1}$ FAME en 108.95 $g.kg^{-1}$ biohoutskool. Die katalisator gee voorkeur aan gas- en seep-produksie. Swaelsuur-gekataliseerede reaksies se hoogste opbrengs was by

320°C; wat 61.15 $g.kg^{-1}$ bio-olie, 36.61 $g.kg^{-1}$ FAME en 367.25 $g.kg^{-1}$ bio-houtskool gelewer het. Die HHV van die olie was 42.28 $MJ.kg^{-1}$ en die van bio-houtskool 28.73 $MJ.kg^{-1}$. Die katalisator het voorkeur gegee aan bio-houtskool-produksie. Kalsium karbonaat-gekataliseerde reaksies het die maksimum opbrengste by 340°C gelewer wat uit 120 $g.kg^{-1}$ bio-olie, 84.7 $g.kg^{-1}$ FAME en 147.9 $g.kg^{-1}$ bio-houtskool bestaan het. Die bio-olie se HHV was 41.12 $MJ.kg^{-1}$ en die van die bio-houtskool was 28.12 $MJ.kg^{-1}$. Die katalisator was die effektiefste in die afbreuk van biomassa en bio-olie produksie. Elke katalisator het sy eie unieke effek op produksie gehad met betrekking tot die bio-olie-, FAME-, en bio-houtskool produksie, asook die HHV- en elementale samesteling.

Verwisseling van die biomassa ladings het optimale resultate by die 20wt% lading met die 20 $g.kg^{-1}$ $CaCO_3$ katalisator en by 330°C gelewer met 'n opbrengs van 120 $g.kg^{-1}$ bio-olie, 84.7 $g.kg^{-1}$ FAME en 147.9 $g.kg^{-1}$ bio-houtskool. Die bio-olie het 'n HHV van 41.12 $MJ.kg^{-1}$ gehad en die van bio-houtskool was 28.12 $MJ.kg^{-1}$. Biomassa ladings het 'n nadelige effek op produk-verspreiding en opbrengs gehad indien dit nie optimaal was nie.

Daar was n oorvloed van aromatiese molekules, direk afkomstig van onversadigde, vertakte, ongelyke koolwaterstofkettings wat teenwoordig was in die bio-olie. ONdersoek was ingestel na die C/H molare verhoudings teenoor die C/O inhoud wat n verandering in die qualiteit van die bio-olie teenoor die fossiel brandstof. The energie inhoud het vermeerder met hoër koolstof-waterstof verhoudings en laer suurstof inhoud. Dei resultate dui n moontlike beperking of die tegniek se vermoe om suurstof te verwyder. Die bio-olie kan dus s paraffinagtig geklassifiseer word, maar met die hoë inhoud van metiel esters kan die bio-olie dus onder a lae kwaliteit biodiesel klas val.

Die produkte van *in -situ* trans-esterifikasie deur middel van die gekombineerde vervloeiing en superkritiese koolstof dioksied ekstraksie kan dus geoormerk word as 'n haalbare vervanging vir fossielbrandstof. Indien hierdie proses wel toegepas word, sal die bio-olie-produk eers sekondêre verwerking moet ondergaan om die groot hoeveelhede onsuiwerhede te verwyder. Hierdie resultate dra by tot die algehele wetenskaplike kennis oor biodiesel produksie.

Kernwoorde: Biodiesel; Vervloeiing; Superkrietiese koolstof-dioksied; *In -situ* trans-esterifikasie; Sonneblom saad.

Declaration:

I, Jacobus Hercules de la Rey, hereby declare that I am the sole author of the dissertation entitled:

In -situ biodiesel production using liquefaction and supercritical extraction.

Jacobus Hercules de la Rey

October 2016

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Chapter 1:

This chapter provides a brief introduction of this dissertation. It consists of four subsections, an introduction (1.1), the problem statement (1.2), the aims and objectives (1.3) and the scope of the dissertation (1.4).

1.1) Introduction

Global urbanisation, industrialisation and rumours of dwindling fossil fuel stocks triggered an uncontrollable increase in the demand for fuel, contributing to skyrocketing fuel prices. Combine these factors with an environmentally concerned green revolution and the end result is a global outcry for an alternative fuel source. Thousands of years ago oil seeped through the ground and was collected for coating houses and as fuel for lamps in ancient China and Persia (Rodger, 2010). Today, things look significantly different, the demand for fossil fuels have increased exponentially, so much so, that the oil consumed by humans since 1985 constitutes 50% of all the oil consumed over the course of human history (Rodger, 2010). Despite the sources being finite and located in restricted locations around the world more than 85% of the energy consumed by the world originates from fossil fuels (Gorman, 2009). Diesel is the driving force behind modern industrialisation. Diesel is a mixture of ethers, esters, aromatics, alkanes, alkenes and any other organic substance that has a boiling point between 180°C and 360°C. Biodiesel is a mixture of methyl esters with carbon chain lengths of between 12 and 20 carbons when it is harvested from renewable sources such as vegetable oil (Mondala *et al.*, 2009:1203 ; Sharma & Singh, 2009:1647). Biodiesel is produced from lipids in biomass. Lipids in biomass consist of many different organic molecules that can be used to produce a variety of fuels using different processing methods (Delrue *et al.*, 2013:206).

Investigations into biofuel production increased during World War II when Germany experienced an energy scarcity. Biodiesel can be produced from conventional food crops, such as edible oil (called 1st generation biofuel) or from inedible crops or waste oil (called 2nd generation biofuel). Trans-esterification is one of the processes most often used to produce biodiesel from extracted oils. During trans-esterification, a simple alcohol and oil

(triglycerides) is heated in the presence of a catalyst. Fatty acids are cleaved away from the triglyceride during the reaction to produce a class of diesel like compounds called fatty acid methyl esters (FAME) and glycerol. A variation of this method is *in -situ* trans-esterification. *In -situ*, meaning in place, is a trans-esterification reaction, to extract the oil without pre-treatment. The idea behind this technique is to establish direct contact between the oil bearing material with the catalyst/alcohol medium, without including the excessive energy consumption of prior oil extraction (Georgogianni *et al.*, 2008:504; Wagner & Haas, 2011:1220; Lam & Lee *et al.*, 2012:685).

Liquefaction is used to produce a bio-oil fraction (bio-oil) from biomass at high temperatures (300°C to 500°C) and pressures (4-20 MPa). The oil is composed of a vast array of different liquid fuel like compounds that can be used in a one step *in -situ* trans-esterification reaction to produce biodiesel. However, the lack of prior processing does result in a greater amount of interference from non-oil producing contaminants such as unprocessed plant mass and phosphates that reduce the exposure of the catalyst and alcohol to the oil.

Supercritical extraction makes use of supercritical fluids to extract components of different polarity and/or variable miscibility from a solid matrix. A supercritical state is an in between phase where substances have the flow properties of gasses and the densities of fluids. Carbon dioxide is a gas used in the industry for the supercritical extraction of oils. Supercritical carbon dioxide could be used as a reaction atmosphere during *in -situ* biodiesel production to facilitate the extraction of oils from the biomass for subsequent conversion to biodiesel.

1.2) Problem statement

Oil extraction processes are energy intensive and time consuming. Supercritical carbon dioxide can be utilized for combining the oil extraction and trans-esterification of oils to diesel into a single processing step.

1.3) Aim and objectives

The aim of this study is to determine the influence of various processing parameters on the bio-oil and FAME yields obtained from *in -situ* trans-esterification of extracted oil from sunflower seeds using supercritical carbon dioxide.

The following specific objectives are defined to reach the aim of the study:

- Investigate the effect of the reaction temperature on the bio-oil and biodiesel yields by varying the temperatures from 320°C to 360°C.
- Investigate the effect of the catalyst on the bio-oil and biodiesel yields by utilising three catalysts, namely potassium hydroxide, sulphuric acid and calcium carbonate.
- Investigate the effect of biomass loading on the bio-oil and biodiesel yield by varying the reactor biomass loading from 15wt% to, 20wt% to 25wt% biomass loading.

1.4) Scope

In order to reach the aim and objectives set out for this study in the previous section, the following is required.

Chapter 1: Provides an overview of this dissertation.

- Brief introduction
- A problem statement
- The aim and objectives of this study
- The scope of the dissertation

Chapter 2: Literature review

- Fossil fuels and diesel
- Biodiesel and biodiesel production
- Trans-esterification and *in -situ* trans-esterification
- Liquefaction and how it works
- The cell components during liquefaction
- The Parameters influencing yield during liquefaction

- Supercritical fluid extraction and how it works

Chapter 3: Provides an overview of the techniques used in the experimental procedures.

- Chemicals used
- Biomass and preparation
- Overview of methods
- Sample analysis

Chapter 4: Contains the results and discussion of the study.

- Effect of catalyst
    - On bio-oil, FAME and biochar yields
- Effect of temperature
    - On bio-oil, FAME and biochar yields
        - Effect of catalyst and temperature on elemental composition
- Effect of Biomass loading
    - On bio-oil, FAME and biochar yields
    - On elemental composition
- Compositional analysis

Chapter 5: Concluding chapter

- Conclusion and recommendations.

Chapter 2: Literature review

This chapter provide a brief overview of the available theoretical information on biodiesel production. The first sections cover fossil fuels (2.1) and biodiesel (2.2). In the sections following, focus is placed on the production processes (2.3), trans-esterification (2.4) and liquefaction (2.5) and the extraction method supercritical extraction (2.6).

2.1) Fossil fuels

Fossil fuels were formed in the carboniferous period during which humid and warm conditions ensured that most of the land mass was covered in swamps with lush forests and oceans with massive algal blooms. The detriment of this abundance of organic life, created large organic sediment layers and over time, fossil fuels were formed (Rodger, 2010; Gorman, 2009). Fossil fuels can be found in a solid (coal), liquid (oil) and gas phase (natural gas) (Rodger, 2010; Gorman., 2009). Petroleum gas (1-4 carbons), naphtha (5-9 carbons), gasoline (5-12 carbons), kerosene/paraffin (10-18 carbons), diesel (12-20 carbons), lubricating oil (20-50 carbons), heavy gas oil (20-70 carbons), the residual (70 + carbons) and other petroleum based products can be obtained from hydro treatment and subsequent fractional distillation of crude oil (Rodger, 2010; Gorman, 2009).

2.2) Diesel /Biodiesel

Biofuels are derived from biomass and are produced using different chemical or biological methods which are renewable (Delrue *et al.*, 2013:205 ; Gasparatos *et al.*, 2013:12). Biodiesel is composed of methyl and ethyl esters with hydrocarbon chain lengths similar to that of diesel. Biodiesel is produced from vegetable oils, has more or less the same properties as fossil based diesel and has 88-95% of the energy content of fossil based diesel, but a better lubricity and cetane value (Gude *et al.*, 2013:1; Timilsina & Shrestha, 2010:2056). During the 1980s, the focus in the production of biodiesel in South Africa was on using land grown crops, such as

palm oil, soybean oil, sunflower oil, and rapeseed oil. The European Union is currently the world leader in biodiesel production and they use rapeseed oil.

Biodiesel is classified into different generations; according to the feedstock from which it is produced. First generation biofuels are produced from edible oil food crops such as soybeans, rapeseed and palm oil. Rudolph Diesel, the inventor of the diesel engine, originally tested his engines with peanut oil, a first generation feedstock. Almost 95% of biodiesel today is produced from first generation crops (Leung *et al.*, 2010:1084). Second generation biofuels are produced from lignocellulose and vegetable oils. Third generation biofuel is the production of fuel from micro- and macro alga. Algae, as feedstock for biodiesel production, do not compete with any food, cosmetic or industrial market for oil (Gude *et al.*, 2013:8). The choice of feedstock for biodiesel production (1st, 2nd or 3rd generation) is mostly based on an assessment of the availability and the cost of crops, which vary greatly depending on the location (Sharma & Singh, 2009:1647). The main disadvantages of land grown crops are that the cultivation of these crops takes up arable land, contributes towards deforestation, are slow growing, and require profuse amount of water. The competition between the energy and the food crops (1st generation crops) or crops used in the cosmetic and industrial sector (2nd generation crops), result in an increase in food and commodity prices (Gude *et al.*, 2013:8).

The production of energy and fuel is only economically and environmentally feasible if the production yields a net profit and energy gain. Biodiesel from 1st and 2nd generation feedstocks are only just becoming economically feasible following policy incentives, whereas 3rd generation production currently falls short in terms of energy gain and profitability. Alternative techniques for biofuel production are thus needed to provide economically viable production without the aid of policy incentives (Mondala *et al.*, 2008:1204; Miao *et al.*, 2006:845; Gude *et al.*, 2013:8).

2.3) Biodiesel production processes

Four techniques are used for biofuel production, each with its own technical difficulties. The ability to overcome the small technical difficulties will eventually determine the success of biodiesel production. The four methods of producing biodiesel are:

2.3.1) Blending

Blending involves mixing biofuels and vegetable oils with regular petroleum fuels. The blends are named according to their mixing ratio for example BXX: where the XX denotes the percentage of biofuels in the mixture. B20, for example, contains 20% biodiesel or vegetable oil. The fuel contains ≈80% of the heat content of fossil based fuels, is readily available and renewable (Leung *et al.*, 2010:1086). The main disadvantages of blending are that the fuel has a higher viscosity, a lower volatility and a lower degree of reactivity due to unsaturated hydrocarbon chains. This may cause carbon residues and oil rings sticking that lead to coking of engines (Leung *et al.*, 2010:1086).

2.3.2) Micro-emulsion

During micro-emulsion the viscosity of vegetable oil is reduced by the addition of compounds such as paraffin, methanol and ethanol and then used for fuel. Micro-structures with dimensions of between 1- and 150 nm are formed spontaneously from two immiscible liquids and one or more ionic or non-ionic amphiphiles (Leung *et al.*, 2010:1086). The fuel has good spray properties during combustion and a lower fuel viscosity, but these are at the expense of a lower cetane number and thus, a lower energy content. Micro-emulsions cause irregular needle sticking and incomplete combustion that lead to carbon deposits and increased lubricating oil viscosity (Leung *et al.*, 2010:1086).

2.3.3) Thermochemical processing (cracking)

This is a process in which larger organic hydrocarbon chains are thermo-chemically decomposed to smaller hydrocarbon chains to produce biodiesel. This is done under high temperatures and low oxygen levels (Leung *et al.*, 2010:1086). The fuel product is chemically

very similar to fossil based fuels but the process is energy intensive, hence resulting in high production costs.

2.3.4) Trans-esterification

Trans-esterification is the chemical reaction between a triglyceride and an alcohol in the presence of a catalyst to yield fatty acid methyl esters (biodiesel). In a stoichiometrically accurate situation one mole of triglyceride will react with three moles of alcohol to produce three moles of fatty acid esters and one mole of glycerol.

2.4) Overview of trans-esterification

Trans-esterification, also called alcoholysis, is the chemical reaction between a triglyceride and an alcohol in the presence of a catalyst to yield fatty acid methyl esters (biodiesel). The cleavage attacks the ester bonded hydrocarbon chains one by one, releasing them to produce fatty acid methyl esters, as indicated in Figure 2.1 (Leung, 2010:1086: Singh Chouhan & Sarma, 2011:4380).

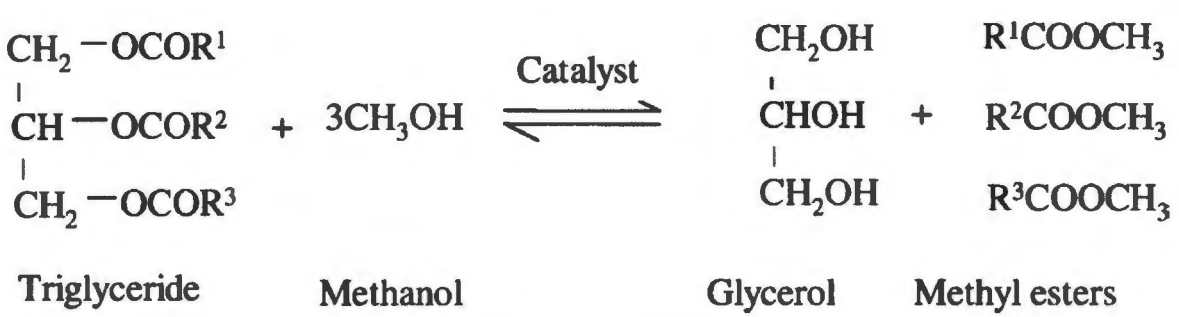

Figure 2.1: Trans-esterification reaction (Leung, 2010:1086: Singh Chouhan & Sarma, 2011:4380).

The feedstock of trans-esterification is completely renewable and delivers biodiesel with a high cetane number, lower emissions and high combustion efficiency. The single drawback is the by-products, glycerol and waste water. Recent progress has overcome these setbacks by producing value added products, such as carbon nanotubes, hydrogen and ethanol, from the glycerol waste, whereas waste water is currently limited by using alternative dry washing

methods, which use an ion exchange resin or magnesium silicate powders to remove impurities (Chatzifragkou *et al.*, 2011:1097; Wu *et al.*, 2013:1).

Trans-esterification has been well explored and current research focus on obtaining the fine balance between the catalyst, biomass, reaction temperature used and the alcohol to oil molar ratio. Research focussed on the catalyst has, and continues to explore the possibilities of different catalyst types. These include, but are not limited to, alkaline earth metal oxides and their derivatives (CaO, MgO), Boron based catalysts, alkali metal oxides and their derivatives, zeolite catalysts, various acid heterogeneous catalysts, sulphated oxides and biocatalysts (Singh Chouhan & Sarma, 2011:4379). Until now, more than 300 different possible biomass types have been identified as potential feedstock for biodiesel production (Shahid & Jamal, 2011:4734). The most current research on trans-esterification, focus on combination virgin biomass oils (FFA <0.5% v/v) and alkaline catalysts (NaOH or KOH) (Ehimen *et al.*, 2010:677). The yields of virgin oils vary between 80-99% FAME at the boiling point of the alcohol used.

2.4.1) In -situ trans-esterification

In -situ meaning in place, trans-esterification is performed through the simultaneous extraction of oil and the reaction with alcohol to oil, to form biodiesel. The aim of this technique is to circumvent the need to process biomass to get the oils beforehand, and it is believed that some financial and production gains can be made through this (Wagner & Haas, 2011:1220; Lam & Lee *et al.*, 2012:685). *In -situ* production brings the oil bearing material directly into contact with the catalyst/alcohol medium, without prior oil extraction (Georgogianni *et al.*, 2008:504).

In the presence of an inorganic acid or base, the *in -situ* trans-esterification reaction can occur at a moderate temperature and atmospheric pressure, without the need for prior processing, therefore, a higher alcohol to oil molar ratio and catalyst loading are needed to overcome interference from contaminating particles in the biomass (Wagner & Haas, 2011:1220). With *in -situ* methods, the presence of water in the feedstock requires an increase in the alcohol to oil molar ratio during production, because of the occurrence of unwanted hydrolysis that

produces soap as a by-product. A feedstock sample with more than 3wt% moisture is regarded unfit for use when a base catalyst is used (Wagner & Haas, 2011:1220). The most current research on trans-esterification, focuses on the combination of virgin biomass oils (FFA <0.5% v/v) and alkaline catalysts (NaOH or KOH). However, this is not feasible for an *in -situ* application due to the high presence of water, FFA and the abundance of different compounds that may lead to saponification (Ehimen *et al.*, 2010:677).

The major disadvantage of *in -situ* biodiesel production is the increased alcohol to oil molar ratio necessary to compensate for contaminating particles that inhibits the diesel reaction. A too high alcohol to oil molar ratio will increase the operating cost of the process, thus nullifying the advantages gained by combining the extraction and reaction processes (Wagner & Haas, 2011:1220).

2.5) Liquefaction

Thermochemical processing of biomass is the treatment of materials at high temperatures and pressures in the presence of a working fluid and reaction gas. It is a method of altering the state of matter with temperature in order to concentrate and harvest its energy. During thermochemical treatment, the carbon and hydrogen content of the material is increased and the oxygen content is decreased by a complex series of dehydrogenating, decarboxylating and hydro-deoxy-carbonylation reactions (Chen *et al.*, 2011:1939).

Biomass breaks down to smaller molecules to form biochar, bio-oil and biogas during hydrothermal liquefaction (Anastasakis & Ross, 2011:4877). The formation of biochar is favoured at the lower end of the liquefaction temperatures when less bio-oil and biogas are formed. Around the median temperatures, conditions favour the production of bio-oil and the highest temperatures support conditions favouring the production of bio-gas (Chen *et al.*, 2011:1935; Anastasakis & Ross, 2011:4879). Liquefaction conditions mostly favour bio-oil production with lesser amounts of biochar and bio-gas produced, and occurs at temperatures of 200˚C to 500˚C and pressures ranging from 4 MPa to 20 MPa (Aresta *et al.*, 2005: 138; Chen *et al.*, 2011:1935; Barnard, 2009:31).

2.5.1) A description of the process of liquefaction

When a substance is heated in a closed vessel under pressure, there is a change in the density and dielectric constant that results in an increase of the apparent miscibility of polar and non-polar substances (Anastasakis & Ross, 2011:4877). This allows components to mix thoroughly, regardless of their polarity. Different solvents behave differently under these adverse conditions for instance a solvent such as water acts as both a hydrogen donor and as a hydrolysing agent for high molecular weight substances. Therefore, the selection of a solvent is important for thermochemical processing.

Liquefaction has a lower production cost, an increased biodiesel yield and an increased total energy production despite the high energy consumption (due to high heat demand). The major advantages of liquefaction are the time and energy savings associated with the elimination of the drying step (Delrue *et al.*, 2013:205). The process can be used to concentrate energy on site, which eases transport and handling. A potential disadvantage of this technique is the high pressure and temperature requirements that contribute to high energy demands and additional health and safety risks.

In biodiesel production, liquefaction is used to produce bio-oil, which can then be converted into biodiesel. Almost any form of lipid bearing biomass can be used for bio-oil production. Liquefaction can be combined with different techniques to produce the desired products for example trans-esterification is combined with liquefaction to directly produce biodiesel from the bio-oil, as the oil is formed.

Most of the groundwork in the field of liquefaction was done by Appel and his co-workers in the 1970s at the Pittsburgh Energy Research Centre (Yang *et al.*, 2004:22). Since then, liquefaction has been studied regularly using different parameters and feedstock (Yang *et al.*, 2004:22). Many studies have analysed the different effects that parameters have on a variety of biomass materials. Oil yields have varied greatly, from 15.60% oil (300°C; 15 MPa for 30 min with 3% dry weight algae) to a 33% maximum oil yield from alga liquefaction (Barnard, 2009:115 ; Yang *et al.*, 2004:26). The liquefaction of sunflower stalks on the other hand was done using supercritical extraction with an organic solvent (methanol, ethanol and acetone) (Erzengin & Kucuk, 1998:1203). It was found that yields increased with the use of a catalyst

(10% NaOH) (Erzengin & Kucuk, 1998:1203). From this initial investigation, it became clear that there are a large number of parameters that influence the liquefaction process and all of them must be examined to understand the influence of the various parameters on product yields.

2.6) Cell components

Each biomass with its unique composition can produce different bio-oils with different characteristics, depending on the lignocellulosic, carbohydrate, protein and lipid content of the biomass (Anastasakis & Ross, 2011:4879). Oil is not only produced from the oil fractions, but also from the carbohydrate and protein fractions of the biomass (Biller *et al.*, 2011: 4841). During liquefaction, oil production from different biomass components tend to achieve higher yields: lipid content (up to 100% conversion), protein content (20% conversion efficiency), and starch content (10% efficiency) (Biller *et al.*, 2011:4845).

Some functional groups found in the feedstock are more difficult to convert to biochar, bio-oil and bio-gas, for instance glycerol structures of triglycerides are harder to remove than hydroxyl groups of carbohydrates and hydroxyl groups of glucose are easier to deoxygenate than the oxygen in starch. Operating parameters thus need to be adjusted for each different type of biomass to obtain the same yield and product distribution (Biller & Ross, 2011: 219).

Fatty acids are rarely found in free form in nature, but they form part of other lipids such as triacylglycerols (TAG), phospholipids, waxes, spingolipids, glycolipids and steroids. Fatty acid molecules are produced in the chloroplast of a cell, catalysed by Acetyl CoA carboxylase (Ac-CoA) and stored in densely packed lipid bodies (Hu *et al.*, 2008:826). Under normal conditions, cells will produce anywhere between 5- and 20% lipids (dry weight percentage), but under adverse conditions, lipid content can increase to between 20- and 50%. TAG forms the largest portion of this increased lipid content (Hu *et al.*, 2008:829).

Table 2.1: Typically, natural occurring fatty acids

Common Name	Hydrocarbon Chain
Lauric Acid	12:0
Myristic Acid	14:0
Palmitic Acid	16:0
Palmitoleic Acid	16:1▲9
Stearic Acid	18:0
Oleic Acid	18:1▲9
Linoleic Acid	18:2▲9,12
Linolenic Acid	18:3▲9,12,15
Arachidic Acid	20:0
Arachidonic Acid	20:4▲5,8,11,14

Lipids are fully decomposed to fatty acids in subcritical water at 260- to 280°C, with full decarboxylation of hydrocarbons also occurring at low levels. Lipids normally end up as hydrocarbons of lengths C5- to C22 that can be methylated or ethylated depending on the alcohol used. In the absence of a catalyst, higher molecular weight saturated fatty acids can be produced suggesting that fatty acids are hydrogenated by the hydrogen produced from hydrolysis. An *in -situ* donor is present after CO has been produced from oxygenated molecules. As the saturation of the lipids increase, so does the oxidative stability of the oil (Biller *et al.*, 2011: 4846). Bio-crudes with lower oxygen numbers are preferred in these reactions since they deteriorate slower (Biller *et al.* 2011: 4846).

During liquefaction, carbohydrates primarily break down to sugars and oligomers at temperatures of about 200°C (Peterson *et al.*, 2008: 42). This causes the attack of glycosidic linkages, which leads to the subsequent cleavage and breakdown of carbohydrates into sugar monomers (Kucuk, 2001:366). Sugar monomers, such as glucose and xylose, are present in all three forms (open chain, pyranose ring and furanose ring), the 6-carbon sugars then become furfurals, lactic acid, formic acid, acetic acid, and 1, 2, 3-benzenetrial, whereas the 5-carbon sugars are transformed to glyceraldehyde, glycolaldehyde, pyruvaldehyde, lactic acids, acetal and various aromatics (especially in acid solutions in excess of 300°C) (Peterson *et al.*, 2008:40). Carbohydrates are the parent molecules of carboxylic acids in bio-oil (Peterson *et al.*, 2008:40). The main mechanisms to break down these molecules are dehydration and decarboxylation of the molecules (Kucuk *et al.*, 2001:366). The highest bio-oil yield from carbohydrates comes from using an alkali catalyst, with approximately 10wt% of the oil yield attributed to carbohydrates.

Lignocellulose is an inedible biopolymer that makes up more than 30% of the worlds' plant matter (Timilsina & Shrestha, 2010:2056). Lignocellulose is a mixture of lignin (15-30wt%), cellulose (30-50wt%) and hemicellulose (23-32wt%). Free radicals produced during the heating process of lignocellulose decompose lignin and cellulose at higher temperatures to stochastically condense macromolecules (Kucuk, 2001:367). Lignin is made of phenyl propane polymers linked by ether bonds that degrade to monomeric derivatives of phenyl propane (p-coumaryl alcohol, conferyl alcohol and sinapyl alcohol) and their derivatives (Peterson *et al.*, 2008:43; Centi *et al.*, 2011: 23). Products from the breakdown of lignin include polyphenolic and polycyclic aromatics. The process of lignin degradation starts with dehydration to form syringols, guaiacols and catechols (Behrendt *et al.*, 2008:670). Further hydrolysis leads to the production of methanol, hydrocarbons, acids, aldehydes, phenols and aromatics. These molecules spontaneously interact and can be separated into an aqueous phase (acids, aldehydes and catechol phenols) and an oil phase (phenolics, hydrocarbons) (Behrendt, 2008: 670). The oil phase is supplemented by depolymerisation of polyaromatic char and a continuous dehydration/hydration reaction that lead to the production of hydrophobic compounds (Behrendt, 2008:670).

Hemicelluloses decompose early in the temperature profile; freeing the hemicellulose bound lignin (Kucuk, 2001:367). Hemicellulose, a highly branched 5-carbon sugar polymer (mainly xylan) is readily degraded by an alkaline catalyst (Centi *et al.*, 2011:24). Hemicellulose has no crystal structure and is easily broken down to monomers of sugar and furfurals at 180°C (Peterson *et al.*, 2008:43).

Cellulose is a linear glucose polymer that is susceptible to acid treatment (Centi *et al.*, 2011:23). It breaks down at elevated temperature and pressure through the disruption of the hydrogen bonding in the crystalline structure and hydrolysis of the ß(1-4) glycosidic linkages, leading to the production of sugar monomers and oligomers (Peterson *et al.*, 2008:42). The breakdown paths of cellulose and hemicellulose occur primarily through dehydration to sugar monomers, which in turn break down to water soluble fragments, these fragments then repolymerise and depolymerise to form biochar and bio-oil (Behrendt *et al.*, 2008:671).

Amino acids are the building blocks of proteins. These amino acids are linked via a peptide bond (C-N) between a carboxyl and an amine group. This is characteristic of all amino acids (Peterson, 2008:45). The amino acids are released by hydrolysis of the peptide bonds (Peterson *et al.*, 2008:45). The contribution of proteins to the production of oil during liquefaction occurs when the C-N peptide bond hydrolyses and breaks the protein down to amino acids. Amino acids start to become unstable at 300°C (particularly positively charged glutamine and arginine) and are then decomposed via deamination into ammonia and organic acids or through decarboxylation to carbonic acids, amines and other water soluble organic compounds (Peterson *et al.*, 2008:45). Catalyst free liquefaction results in the highest portion of proteins being converted to bio-oil, while catalysts convert proteins into solid biochar. Products that are phenolic, indole and pyrole derivatives are likely products of complex and cyclic amino acids, such as tryptophan (Ross *et al.*, 2010: 2238). A disadvantage of direct liquefaction is the distribution of protein into the oil product fraction resulting in higher sulphur and nitrogen levels in the oil.

Repolymerisation of intermediates occurs via a Fischer-Tropsch-type reaction to form hydrocarbons (Peterson *et al.*, 2008:45). The repolymerised bio-oil product has 10-20wt% oxygen content with a HHV of 30-36MJ.kg^{-1}. The oxygen exits as gas via the small organic molecules; CO, CO_2, or H_2O (Peterson *et al.*, 2008:46).

2.7) Parameters that influence product yield and distribution

During liquefaction different operating parameters (internal and/or external) have varying effects on the rate of reaction, yield and quality of product obtained.

2.7.1) Temperature

Researchers in general accept that temperature is the main parameter influencing yield and type of product (Shuping *et al.*, 2010: 5410; Ross *et al.*, 2010:2242; Kucuk, 2001:366; Chen *et al.*, 2011:1940). The oil yield and conversion to biodiesel increase in direct proportions to an increase in temperature (Shuping *et al.*, 2010:5480; Ross *et al.*, 2010:2242; Kucuk, 2001:366;

Chen *et al.*, 2011:1935). Controlling the temperature determines the ratios of the formation of solids, liquids and gasses. As biomass breaks down at elevated conditions, the higher temperatures generally resulting in an increased H/C molar ratio, a decreased O/C molar ratio, and formation of smaller molecular weight products (Chen *et al.*, 2011:1935-1940). There is an inverse relationship between temperature and product molecular weight (Chen *et al.*, 2011:1935). More severe reaction conditions should increase the carbon, hydrogen, and nitrogen content, and decrease the oxygen content. This means that the reaction temperature should be as high as possible to trigger a fast effective reaction, yet not so high that it breaks down the product (Sharma & Singh, 2009:1650; Leung *et al.*, 2010:1091; Jazrawi *et al.*, 2013:267).

Anastasakis and Ross (2011:4479) found a maximum yield of 19.3 $g.kg^{-1}$ at different temperatures for different types of biomass, for instance green macro alga (300˚C), cattle manure (310˚C), woody biomass (320˚C), micro algae (340˚C) and brown macro algae (350˚C). This shows that the biomass composition is the major determining factor of reaction temperature, due to the extent of the free radical production and potential shielding against free radical production. If the biomass has a high level of hydrogen content, the onset of thermal degradation can be delayed when the hydrogen acts as a reducing agent to stabilise free radicals (Barnard, 2009:51).

Barnard (2009:ii) found that the optimum hydrothermal processing parameters of microalgae are 300˚C, 15 MPa with a holding time of 30 min and a biomass dosage 30 $g.kg^{-1}$ to obtain a 156 $g.kg^{-1}$ bio-oil yield. Shuping *et al.* (2010:5406) used algae as raw material and a bio-oil yield of 250 $g.kg^{-1}$ using Na_2CO_3 at 360˚C was achieved. Yu *et al.* (2011:239) produced refined oil using liquefaction with yield of between 240 $g.kg^{-1}$ and 354 $g.kg^{-1}$ at temperatures between 200- and 300˚C. Yang *et al.* (2004:32) achieved a 395 $g.kg^{-1}$ maximum oil yield from alga liquefaction at 340˚C with a 30 min holding time and a 5wt% Na_2CO_3 catalyst loading. Ross *et al.* (2010:2234) investigated conditions for the production of high quality low molecular weight bio-oil via hydrothermal processing, obtaining bio-oil yields of between 700- and 750 $g.kg^{-1}$ at 300˚C and 350˚C.

Table 2.2: Elemental composition of different bio-oils from literature sources

Biomass	Cwt%	Hwt%	Owt%	Nwt%	HHV(MJ.kg^{-1})	Reference
May chang oil (*Litsea cubeba*)	76.2	11.9	10.4	1.6	40.8	(Wang *et al.*, 2013:509)
Spirulina	74.4	11.5	4.7	9.2	33.4-39.9	(Ross *et al.*, 2010:2237)
Chlorella	75	9.9	7.8	7.3	38.1	(Ross *et al.*, 2010:2237)
Duckweed	72.53	8.75	13.31	5.41	33.95	(Xiu *et al.*, 2010:1298)
Brown macro algae	70-74	7.8-7.9	14-17	3.8-4	32-34	(Anastasakis and Ross., 2014: 550)
Soybean straw and sunflower oil*	84	9.55	2.52	0.01	34.38	(Chen *et al.*, 2010:4606)
Soybean straw and sunflower oil*	83.87	13.68	2.25	0.017	43.8	(Chen *et al.*, 2011:1940)

*Co-deoxyliquefaction processes

2.7.2) Catalysts

By definition, a catalyst is a compound added to a mixture in small amounts, to accelerate a reaction without being consumed by the reaction. Catalysts are one of the major determinants of product yield. Catalysts used in liquefaction assisted trans-esterification reactions can be either homogeneous or heterogeneous. Homogeneous catalysts are catalysts that are in the same phase as the reactants and are most commonly used for trans-esterification, because they can facilitate faster reactions compared to heterogeneous catalysts (Lam & Lee *et al.*, 2012:685). Heterogeneous mixtures are substances that are not in the same phase and are less well studied, despite the fact that this type of catalyst can be recycled and re-used/ recovered several times, which is financially beneficial (Lam & Lee *et al.*, 2012:685).

The catalyst can either be an acid, base or enzymes; the latter has not been widely used due to the high cost of enzymes. An acidic catalyst triggers a slower reaction that can tolerate free fatty acids (FFA) with no saponification and requires more alcohol (Leung *et al.*, 2010:1088). A base catalyst triggers a rapid reaction (up to 4000 times faster when compared to an acid catalyst) (Leung *et al.*, 2010:1088). Base catalysts function at lower temperatures and pressures and can be used efficiently in industrial processes (Leung *et al.*, 2010:1088). Typical

base catalysts are hygroscopic (absorb water) and are less tolerant toward FFA's. The presence of FFA's or water in the solution results in a saponification, a reaction in which an alkali metal, like sodium or potassium binds to a fatty acid and forms soap. This is detrimental to the reaction and reduces diesel yield, since the soaps are emulsifying agents that hinders the separation of glycerol (Sharma & Singh, 2009:1648; Leung *et al.*, 2010:1088).

A high moisture level in the feedstock cleaves the triglyceride and the catalyst can then react with the FFA's, reduces the amount of catalyst available for the reaction and increases soap formation. Residual moisture in the feedstock can be removed through drying. High amounts of FFA's in the oil can be reduced with an acid catalysed trans-esterification reaction. The FFA's in the sample should be reduced too below 1wt% before the base trans-esterification to biodiesel is done (Leung *et al.*, 2010:1087). Generally, hydroxides, carbonates and bicarbonates are used as catalysts for hydrothermal liquefaction (Anastasakis & Ross, 2011:4879 ; Zhu *et al.*, 2006:391). No single catalyst has been proven to give the highest biodiesel yield, although potassium hydroxide (KOH) and sulphuric acid (H_2SO_4) have been widely used. Catalyst free production using supercritical methanol has also been successfully utilised (Kouzu *et al.*, 2007:2798).

Chen *et al.* (2011:1940) showed that the major determinant of product distribution during liquefaction assisted trans-esterification was temperature and then the type of catalyst used. The choice of a catalyst is dependent on the amount of free fatty acids and water present in the feedstock. Higher catalyst loadings decrease the gas yield and biochar production and increase the oil yield (Chen *et al.*, 2011:1935; Anastasakis & Ross, 2011:1879). The catalyst loading for each process should be optimized, but using published data as guidelines, a catalyst loading between 1-10% should be ideal and 2% is a good starting point.

2.7.3) Biomass loading

During liquefaction, the biomass loading determines the amount of raw material available for reaction and conversion. Some studies suggested that biomass loading is second only to temperature regarding its influence on product yield and distribution (Anastasakis & Ross,

2011:4880). The biomass loading determines the alcohol to oil molar ratio: decreasing the biomass loading increases the alcohol to oil ratio and the surface of exposure of the lipids (Ghoreishi & Moein, 2013:27). The supercritical point of the reactant/product mixture was reduced when the surface of exposure was increased (Ghoreishi & Moein, 2013:27; Patil *et al.*, 2011:121 ; Anitescu & Bruno, 2012: 138). Optimum yields are achieved when the solvent solute mixture is in supercritical state because the (pseudo) critical point removes the interface layer between phases resulting in optimum mixing and maximum reactants exposure levels (Anitescu & Bruno, 2012: 138). Under supercritical conditions, the methanol breaks down the intermolecular hydrogen bonding, resulting in the reduction of the polarity and dielectric constant of methanol, allowing it to function as a free monomer (Patil *et al.*, 2011:119). This allows methanol to act as a solvent and a hydrogen donor that stabilise free radical production to promote the formation of oil, inhibiting secondary decomposition of oil and repolymerisation (Zhou *et al.*, 2012:2345).

Higher alcohol volumes and lower biomass loading can inhibit polymerisation and thermal degradations by diluting reactants and free radical production (Levine *et al.*, 2013:561). Exceeding the critical temperature can also lead to the breakdown of bio-oil and FAME (Ghoreishi & Moein, 2013:27; Patil *et al.*, 2011:121; Jin *et al.*, 2014:344). On the other hand, a biomass loading that is too high obstructs and clogs the reaction (Ghoreishi & Moein, 2013:29).

When higher biomass loadings of up to 10wt% are used, the molecules rearrange so that there is an increase in H/C content, a reduction in oxygen content and an increase in the energy density of the biomass (Anastasakis & Ross, 2011:4878). Increasing the biomass loading leads to a decrease in the product density (Widayat *et al.*, 2013:69). Lower O/C molar ratios were observed, suggesting that biomass contains an *in -situ* hydrogen donor or hydrogenating agents that can contribute towards the upgrading of the product to improve the oxidative stability of the liquid product (Chen *et al.*, 2011:1939). Furthermore, biomass has a high natural content of plant gums (phosphatides) that increases viscosity and causes gumming or plugging of filters (Aresta *et al.*, 2005: 138).

At lower biomass loadings, biochar with high carbon content, high heating value and low ash content is produced (Anastasakis & Ross, 2011:4878). Lower biomass loadings should also

yield lower levels of solid residue due to the fact that there is more solvent to dissolve monomers (higher alcohol to oil molar ratio) (Behrendt *et al.*, 2008: 672). Higher biomass loadings can be in excess and yield lower char content when compared to lower biomass loadings (Amin *et al.*, 2009:16; Anastasakis & Ross, 2011:2880; Behrendt *et al.*, 2008:672). Different results have been reported, for instance, a 16wt% biomass loading yielding 580 $g.kg^{-1}$ oil and 420 $g.kg^{-1}$ residue, and a 30wt% biomass loading delivering a 550 $g.kg^{-1}$ oil yield and 450 $g.kg^{-1}$ residue (Karagoz *et al.*, 2006:92). Other studies found that a 10wt% biomass loading is ideal for wet microalgae, delivering conversions of between 720 $g.kg^{-1}$ and 900 $g.kg^{-1}$ (Patil *et al.*, 2011:1402). According to Zhou *et al.* (2012:2342), a 9wt% biomass loading is ideal as it delivered 311 $g.kg^{-1}$ bio-oil and 416 $g.kg^{-1}$ biochar. Karagoz *et al.* (2006:92) obtained a production of 86 $g.kg^{-1}$ oil and 420 $g.kg^{-1}$ biochar with a 16wt% biomass loading, and when a 30wt% biomass loading was employed, they achieved low yields of 64 $g.kg^{-1}$ oil and 450 $g.kg^{-1}$ biochar. Widayat, *et al.* (2013:72) produced optimum yields with a 28wt% biomass loading, while Yang *et al.* (2014:941) found that yields increased to 152 $g.kg^{-1}$ when a 100wt% biomass loading was systematically reduced to 30wt% and again decreased to a 25wt% biomass loading.

This simply indicates that the terms 'high' and 'low' biomass loading change according to the different biomass type, temperature, alcohol, catalyst and holding time. Each one of these applications requires a different optimisation.

2.7.4) Mixing/ Stirring

Stirring is necessary for effective mass transfer during liquefaction. Stirring ensures the dispersion of active material throughout the reaction chamber (Galadima & Maruza, 2014:3). Higher rates of stirring enable a more complete reaction more rapidly, because it determines the extent of contact between catalyst and reactants (Galadima & Maruza, 2014:3). A rate of stirring that is too high may result in side reactions where as a slow rate of stirring may cause a very slow reaction (Galadima & Maruza, 2014:3). Better results have been achieved with mechanical than magnetic stirring (Sharma & Singh, 2009:1648). However, under the high pressure and temperature conditions of a supercritical fluid the rate of stirring does not have

an effect on yield distribution, this is probably due to the increased solubility of solvent in critical or near critical temperatures that present as a single phase (Madras *et al.*, 2004:2029).

2.7.5) Particle size and shape

The correct particle size and shape is necessary to achieve high oil or biodiesel yields, and is dependent on a trade-off between heat and mass transfer effects (Madras *et al.*, 2004:2029). The smaller the fragments are, the greater the surface of exposure, increasing contact between the particles and the solvent. This makes mixing more efficient, leading to a lower required alcohol to oil ratio (Wagner & Haas, 2011:1220).

2.7.6) Residence time

The residence (holding) time in a batch system refers to the time the autoclave is kept at constant temperature. Increasing the holding time allows repolymerisation and condensation reactions to take place, but extended residence times tend to favour gas production, resulting in a further break down of the desired product and a reduction in oil yield (Anastasakis & Ross, 2011:4879; Chen *et al.*, 2011:1938). A higher temperature generally reduces the required residence time (Hidalgo *et al.*, 2013:192). Galadima and Maruza (2014:3) reviewed reaction times and found reaction times of between 60- and 180 min to be efficient, on the other hand, Jin *et al.* (2014:345) came to the conclusion that shorter reaction times of between 0- and 120 min are effective. The reaction time must be adequate for the reaction to take place, but not so long that it promotes side reactions. Reaction time has less of an effect on yield when compared to temperature that produces a relative constant yield after a certain temperature has been reached (depending on temperature and type of biomass), with most of the conversion of product occurring during the heating up towards the reaction temperature (Jin *et al.*, 2014:345; Chen *et al.*, 2011:1938). Residence times exceeding 15 min favoured gas production and reduced yields in some studies (Chen *et al.*, 2011:1938). Yang *et al.* (2014:941) found a 20 min residence time to be optimal, longer times reduced bio-oil yields and increased gas yields.

2.7.7) Solvent

The solvent can either be acidic, alkaline or organic. Acid solvents can facilitate the breakdown of the glycosidic bonds of cellulose at 220°C, causing the depolymerisation to glucose and sugar derivatives (Behrendt *et al.*, 2008:669). Alkaline media triggers the conversion of celluloses and hemicelluloses into organic acids by rupturing the glycosidic bonds (Behrendt *et al.*, 2008:669). Hemicelluloses depolymerise to monosaccharides at 120°C and are converted to furfural and acid derivatives (Behrendt *et al.*, 2008:669). The lignin α-OH, α-ether and α-O groups are converted to benzylium ions that are then condensed or sulfonated (Behrendt *et al.*, 2008:669). Lignin hydroxyl groups favour ester and ether bond ruptures to produce stilbene-derivatives (Behrendt *et al.*, 2008:669).

The type of alcohol used for reactions is largely determined by financial means. Organic solvents, such as methanol, trigger the homolytic rupture of C-O and C-C bonds in biomass (Behrendt *et al.*, 2008:669). Methanol, or similar alcohols that are small simple alcohols, are used since they serve well as reactants, as polar catalysts have a poor miscibility in larger alcohols (Wagner & Haas, 2011:1220). Methanol or ethanol are used most often, but methanol is preferred because it is more cost effective, has improved separation from water and is readily available. The alcohol used, should dissolve the catalyst easily to facilitate the reaction with triglycerides. The use of ethanol provides a more environmentally friendly reagent that is less toxic than that of methanol, but it requires long reaction times (Georgogianni *et al.*, 2008:507; Sharma & Singh, 2009:1649). Ethanol is more soluble in oil than methanol and does not deactivate enzyme catalysts (Madras *et al.*, 2004:2030).

2.8) Supercritical extraction

A fluid heated to temperatures and pressures above its critical point, is called a supercritical fluid. Supercritical fluids undergo drastic changes in their physicochemical properties at a point of transfer between the phases of fluids and gasses, this is called the supercritical point. Supercritical fluids have the densities of fluids and the flow properties of gasses, giving these

supercritical fluids the ability to diffuse through a solid matrix in the same manner as a gas, but with the solvent strength of a liquid to extract solutes from the solid matrix. This improved mass transfer afforded by supercritical fluids is useful for improved extractions.

2.8.1) A description of the supercritical extraction process

The solvent strength of a compound is expressed as the solubility parameter. This parameter is the square root of the cohesive energy density, suggesting that the solvation strength is directly related to fluid density. The density of the supercritical fluid above its critical point is very sensitive to small changes in temperature and pressure, allowing for the control of solvent solubility through temperature and pressure. The changes in solubility are not uniform for all compounds and can have a positive or negative effect on solubility.

2.8.2) Supercritical CO_2 extraction

Carbon dioxide (CO_2) is a small linear gas molecule that is non-flammable, non-toxic and environmentally benign. The gas has a moderately low critical pressure of 73 Bar and a low critical temperature of 31°C (304 K). Supercritical carbon dioxide is miscible in a variety of organic solvents and is easily recovered; making it an ideal compound to save energy and to use during the extraction practices of heat labile compounds. Compared to hydrocarbon solvents, carbon dioxide has no dipole moments or weaker Van der Waals forces. Most research on supercritical CO_2 has focused on lower temperature extractions of between 40- and 80°C (Copalan *et al.*, 2003:109; Aresta *et al.*, 2005: 137), and extensive research on supercritical CO_2 indicated that it is effective for extracting non-polar solutes and for the enhanced solubilisation of more polar moieties by the addition of ethanol or methanol (Copalan *et al.*, 2003:105). Copalan *et al.* (2003:108) proved that supercritical CO_2 is an autocatalytic agent for glycerol-lysis under ambient conditions. Supercritical CO_2 has been successfully used in the trans-esterification of triglycerides, with the use of lipases (Novozyme SP 435). Furthermore, it is an agent to facilitate the upgrading of bio-oil by facilitating the full hydrogenation of methyl esters formed from vegetable oils. It is also employed as a

solubilisation agent and a reaction agent for de-oxygenations, decarbonylation, and the decarboxylation of FAME (Copalan *et al.*, 2003:112).

Supercritical CO_2 has been studied for its use in the extraction of oil from seed oils (Stahl *et al.*, 1980:1153). Stahl *et al.,* (1980:1155) found that a high oil content of the seeds and the changing solubility behaviour of lipids at high pressures reduced the yield of extraction and showed that the yield was independent of the gas flow rate. In an unrelated study by Aresta *et al.* (2005:136), they compared liquefaction to supercritical CO_2 extraction. The comparison indicated that thermochemical liquefaction was more efficient in the extraction of lipids and requires temperatures of 350˚C to 395˚C for optimum oil production, but lower temperatures are required to keep long chain fatty acids intact. Liquefaction utilises high temperatures and can tolerate wet biomass. On the other hand, supercritical CO_2 requires only moderate temperatures and cannot tolerate moisture (Aresta *et al.*, 2005:139). The low permeability and composition of some solid materials require prior milling or cracking open of the husks. Madras *et al.* (2004:2033) reported that almost complete conversion was obtained when using supercritical ethanol or methanol, but only a 30% conversion was achieved when supercritical CO_2 was used with an enzyme catalyst. Ethanol had a higher conversion due to the increased solubility parameter, closer to the solubility characteristics of oils while methanol deactivated enzymes (Madras *et al.*, 2004:2029).

To increase the solubility of higher molecular weight compounds in supercritical CO_2, co-solvents may be added. These can be polar or non-polar moieties that interact with the desired product to increase its solubility, for instance the use of methanol to extract cholesterol. Kucuk (2001:363) studied the liquefaction of biomass using supercritical gas extraction with a 10% NaOH catalyst in supercritical methanol, ethanol and acetone to deliver yields of 40%, 45.6% and 47.8%, respectively. Erzengin & Kucuk (1998:1203) investigated the liquefaction of sunflower stalks by using supercritical extraction with an organic solvent such as methanol, ethanol and acetone and found that the yields increased with the use of a catalyst (10% NaOH).

Chapter 3: Experimental

This chapter provides a description of the experimental work conducted. The chemicals and feedstock used are described in section 3.1. The experimental methods used are explained in section 3.2 and the analysis procedures are provided in section 3.3.

3.1) Chemicals and feedstock

A list of all the chemicals used in this study is provided in Table 3.1. All chemicals were used as obtained from the supplier without further purification.

Table 3.1: List of chemicals used

Component	Supplier	Purpose	Purity
Cyclohexane	Sigma Aldrich	FTIR	99.5%
Dichloromethane	Sigma Aldrich	GC Analysis	99%
Dodecane	Sigma Aldrich	GC Analysis	>99%
Methyl Nonaoate	Sigma Aldrich	GC Analysis	97%
Trimethyl Sulfonium Hydroxide Solution (TMSH)	Sigma Aldrich	Oil GC Analysis	0.25M in methanol
Potassium Hydroxide	Ace chemicals	Biodiesel Production	>99.8%
Methanol	Ace chemicals	Biodiesel Production	99.5%
Carbon dioxide	Afrox	Biodiesel production	99%
n-Hexane	Sigma Aldrich	Biodiesel production	99%
Calcium carbonate	Sigma Aldrich	Biodiesel production	> 99.8%
Sulphuric acid	Ace chemicals	Biodiesel production	98%

3.1.2) Biomass selection and preparation

The sunflower is a wide spread agricultural crop in South Africa. The distribution of sunflower across South Africa is mainly found, but is not limited to the Highveld (North West, Free State, Gauteng, Limpopo and Mpumalanga provinces) (South African Department of Agriculture, 2010). Biodiesel research has been done for over four decades in South Africa, specifically on the use of sunflower seeds as biofuel feedstock were thus selected as a

baseline feedstock for this study. Sunflower seeds from a draught resistant strain with lower oil content were acquired from a local farmer in the North West Province, South Africa. The seeds were collected from a harvester in a 50 kg grain pack and transported by car to the university premises.

The seeds were milled with a hammer mill to a seed size of between 0.8 mm- and 15 mm before they were dried overnight at 100˚C. The biomass was then quartered (see Figure 3.1) and sealed in plastic airtight containers of 20 g+-0.2 g each until used.

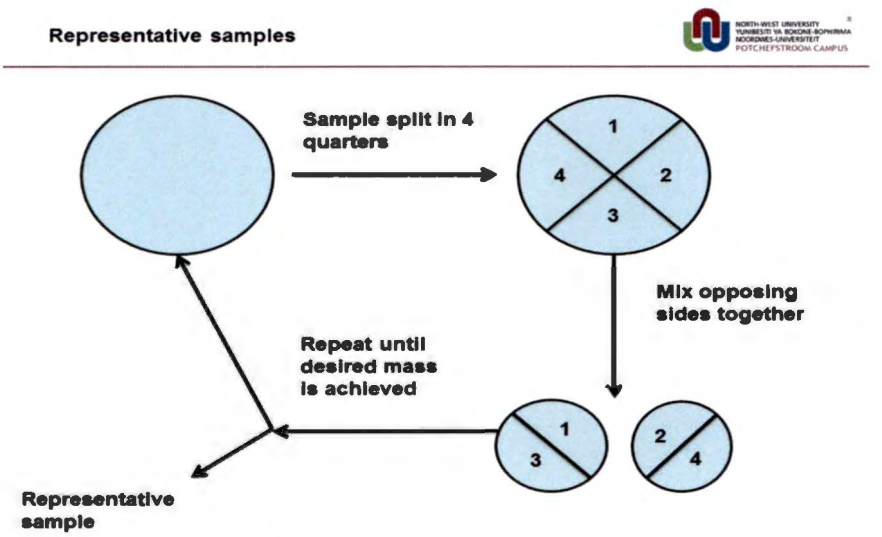

Figure 3.1: Quartering process followed in order to deliver a uniform feedstock

A compositional analysis of dried, milled seeds was done by the Agricultural Research Council, Irene, South Africa and is provided in Table 3.2.

Table 3.2: Compositional analysis of dried sunflower seeds (wt% on dry basis)

Component	Percentages (wt%)
Dry matter	92.21
Moisture	7.79
Ash	3.21
Crude Protein	16.12
Fat (ether extraction)	37.66
Neutral detergent fibre (NDF)	30.27
Acid Detergent Fibre (ADF)	25.85
Acid detergent lignin (ADL)	12.58
Cellulose (ADF-ADL)	13.27
Hemicellulose (NDF-ADF)	4.42
Lignin (ADL)	12.58

3.2) Methods and graphic

Figure 3.2 contains a schematic and graphical representation of the experimental setup used in this study.

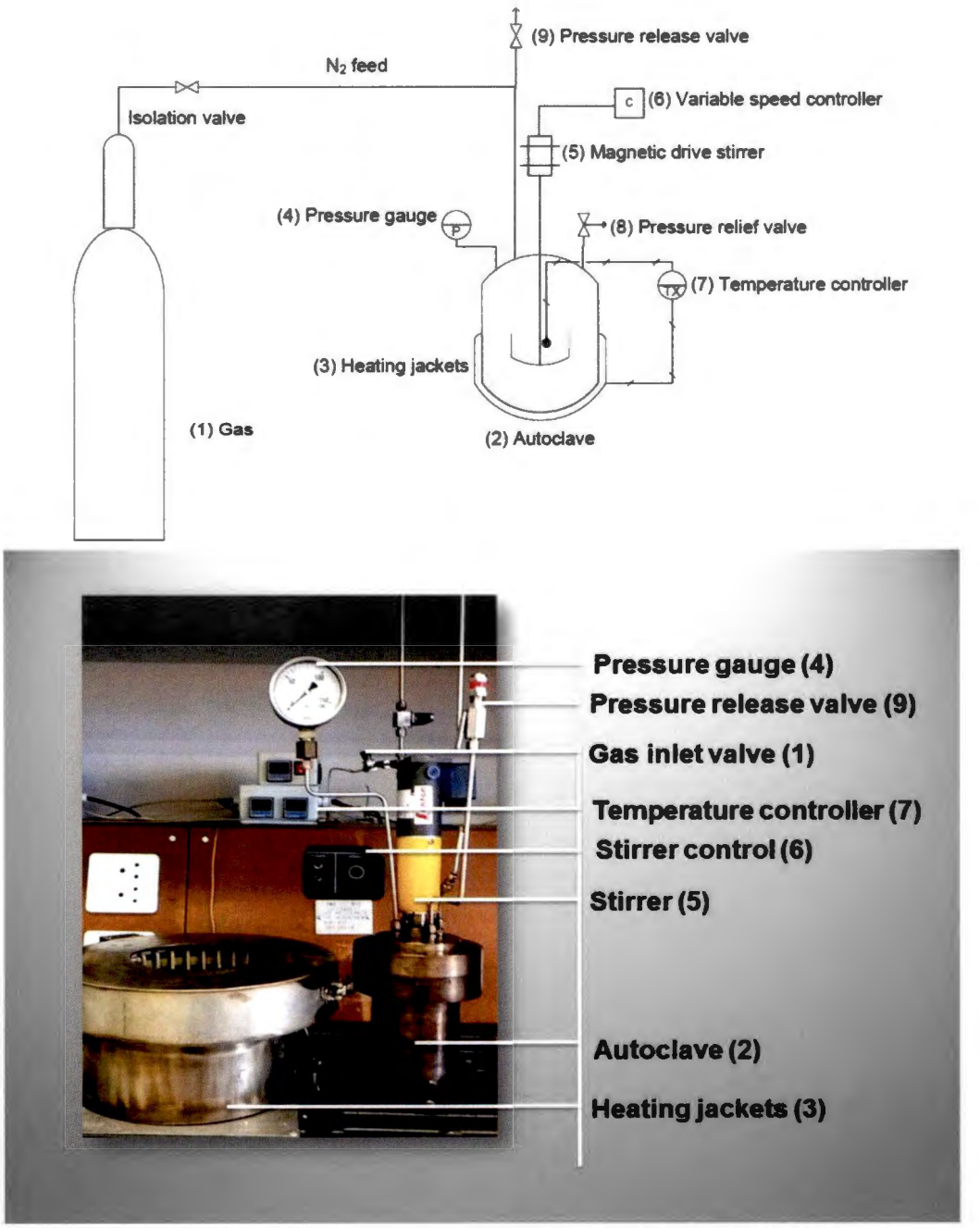

Figure 3.2: Liquefaction reactor used in this study

During this study, the influence of three parameters on the oil and biodiesel yields was investigated, i.e. the temperature (320°C to 360°C with intervals of 10°C), type of catalyst, potassium hydroxide (KOH), sulphuric acid (H_2SO_4) and calcium carbonate ($CaCO_3$) at catalyst loading of 20 $g.kg^{-1}$ biomass and the biomass loading (150 $g.kg^{-1}$, 200 $g.kg^{-1}$, 250 $g.kg^{-1}$ by weight in solvent).

Liquefaction was done according to standard methods (Barnard, 2009:67). A stainless steel 316 autoclave, with dimensions of 0.954 L in volume, 150 mm in length and an internal diameter of 90 mm, was used for all the experiments (see Figure 3.2)

In a typical experiment, an autoclave (6) was loaded with the designated mass of biomass (15wt%, 20wt%, or 25wt%), 100 ml of methanol and 2 g catalyst. The autoclave (6) was then sealed and the screws tightened with an 80 Nm torque wrench. The gas inlets (2) were opened and the reactor was purged for 10 min with CO_2 gas to remove residual air. The pressure was then raised to 0.5 MPa and kept at that pressure for 15 min to check for gas leaks, before the temperature was increased to the desired holding temperature. The heating jackets (7) raised the temperature to the desired reaction temperature and were regulated by a temperature controller (3). Once the reaction temperature was reached, the stirrer (300 rpm) (4 and 5) was switched on and left stirring for the holding time of 15 min. At the end of the experiment, the heating jackets (7) were opened and the system was left to cool under a fan till the temperature was below 30°C. The autoclave is a closed system, thus the pressure and temperature are dependent on the gas law. The autoclave was fitted with a pressure relief valve with a relief pressure of 11 MPa.

After of the reaction was completed, the contents of the autoclave were removed and processed to recover the crude bio-oil and the biochar. The experimental procedure followed is shown in Figure 3.3.

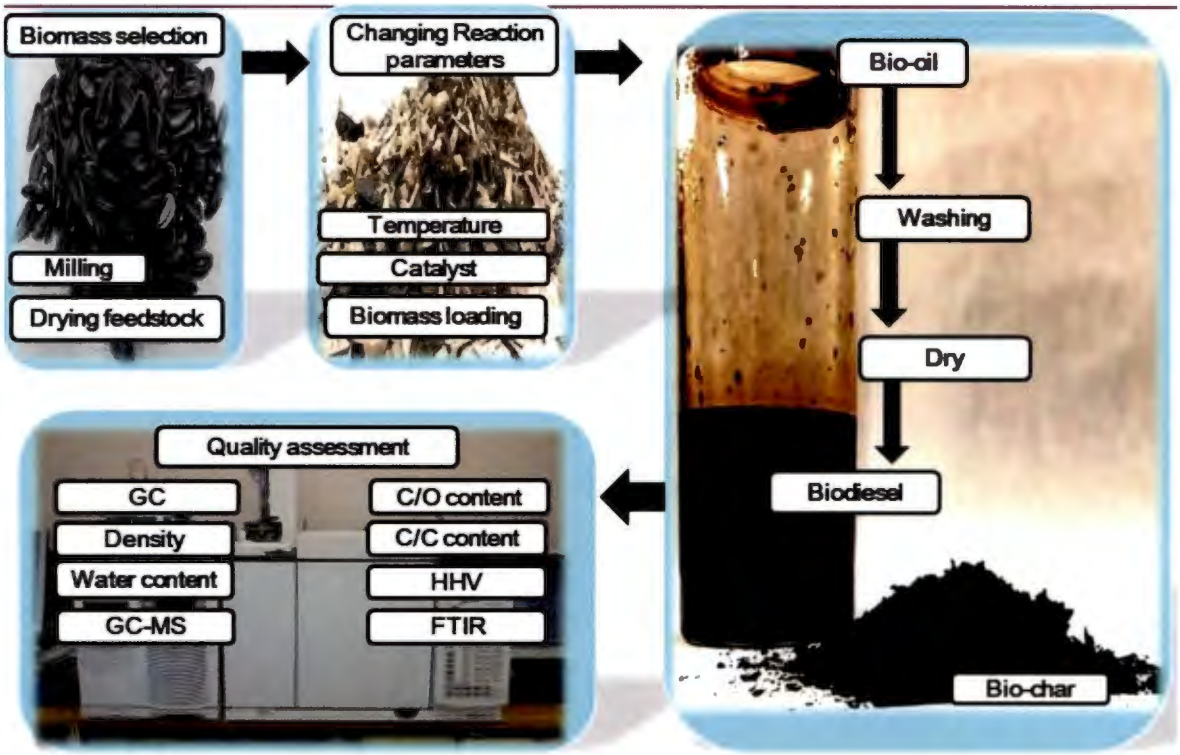

Figure 3.3: Experimental procedure.

After cooling down, the pressure was released and 20 ml of acetone was added to the oil and the mixture was stirred for 15 min to allow all oil substances to dissolve (Qu *et al.*, 2002:599). The end product was removed and filtered under vacuum through Whatman no. 40 filter paper. The residue (biochar) was dried overnight at 105˚C. The bio-oil was water washed in a mixing volume to volume ratio of crude oil and 95˚C water. After settling, the water phase was decanted and discarded. The washing procedure was repeated three times. An improved washing technique used later utilised n-hexane for oil recovery before the water washing cycle commenced. An n-hexane ratio of 1 volume of n-hexane to 2 volumes of crude oil was used for n-hexane washing. The end product was then placed on a hotplate and stirred for an hour at 80˚C to remove all n-hexane before being oven dried overnight at 105˚C.

The yields were calculated as the mass of bio-oil product over the mass of biomass and expressed as grams bio-oil per kilogram of biomass ($g.kg^{-1}$). The fatty acid methyl esters

(FAME) yields were calculated as the weight fraction of bio-oil consisting of fatty acid esters and expressed as gram FAME per kilogram of biomass ($g.kg^{-1}$)

3.3) Analysis

3.3.1) Compositional analysis (FAME and crude oil)

The biodiesel samples were filtered through a 0.45 µm and a 0.2 µm syringe filter before a 100 µL of biodiesel sample was transferred into a sample vial and the mass recorded. Methyl nonanoate which was used as an internal standard was added (20 µL) and the mass of the mixture was recorded. The mass of the internal standard was calculated: $m_{IS} = m_{combined} - m_{biodiesel}$. The sample was then diluted to approximately 1 ml using dichloromethane (DCM) and briefly vortexed. The samples were prepared in a similar way for the gas chromatography (GC) (Agilent 7820A) and gas chromatography coupled mass spectrometry (GC-MS) (Agilent 7890A).

Gas chromatography was used to determine the composition of fatty acid methyl esters (FAME) in the produced oils. The instrument was equipped with an Agilent 5975C auto-injector, HP-88 (100 m) column and a flame ionization detector (FID). The operating conditions used for GC analysis are summarised in Table 3.3.

Table 3.3: Conditions used for gas chromatography

GC protocol	
Carrier	Helium
Carrier flow rate	40 $ml.min^{-1}$
Air flow rate	400 $ml.min^{-1}$
Make up He flow	1.0 $ml.min^{-1}$
Linear viscosity	35 $cm.s^{-1}$
Split ratio	35 $cm.s^{-1}$
Injection volume	1.0 µL
Inlet temperature	250°C
Inlet pressure	381.98kPa
Oven programming	100°C; 5min
FID detector	350°C
Solvent	Dichloromethane

The gas chromatography coupled mass spectrometry (GC-MS) analysis was used to identify the components in the bio-oil that was not FAME. The instrument was equipped with an Agilent 5975C auto-injector, HP-5 (30 m) column and MSD triple axis detector. The operating conditions used for the analysis are provided in Table 3.4.

Table 3.4: Conditions used for GC-MS analysis

GC-MS protocol	
Column	HP-5 MS 30 m
Carrier	Helium
Carrier gas velocity	35 $cm.s^{-1}$ Constant flow
Inlet	Split-split less, operating as split less
Inlet Temperature	250°C
Inlet Pressure	21.5 kPa
Oven programming	35°C; 2 min, Ramp at 5°C until 320°C
Detector	Agilent 5975C MS With triple axis detector
Solvent	Dichloromethane

3.3.2) Structural analysis

Fourier transform infrared spectroscopy (FTIR) was performed using an Eralytics Eraspec fully automated fuel analyser to identify the functional groups present in the crude oil. The samples were scanned and the absorbance plotted against the wavelength that ranged from 600 cm^{-1} to 3000 cm^{-1}. The lowest absorption peaks were used as transmittance peaks to identify functional groups. This device also measured the density with an oscillating U-tube and calculated the cetane value and aromatic content.

Karl Fischer coulometry was used to determine the water content of the diesel or oil using the coulometric method with a cell diaphragm (Metrohm Karl Fischer coulometer). The cells were dried until a constant drift of less than 10 ppm was maintained. A dry syringe was purged 3 times with the sample before 1 g of sample was added to the titration cell and analysed.

The ultimate analysis was performed using a C-440 Elemental analyser with a CE 490 interface (EAT Exeter analytical, Inc.). Acetaldehyde with a composition of 71.09wt% C, 6.71wt% H, 10.36wt% N and 11.84wt% O was used as a standard for calibration.

The Calorific value (LHV) of the 1 g sample was measured using a Bomb calorimeter (IKA C5003) with a KV600 water cooling system, and C5000 control package and a cotton thread flint (C710.4). Benzoic acid (IKA C723) was used as calibration standard.

Chapter 4: Results and discussion

4.1) Introduction

The results obtained from the *in -situ* trans-esterification by liquefaction and supercritical carbon dioxide are discussed in this chapter. The influence of the catalyst type, reaction temperature and biomass loading on the bio-oil, biochar and fatty acid methyl esters (FAME) yields is discussed in sections 4.2, 4.3 and 4.4, respectively. An analysis of the crude bio-oils is provided and discussed in section 4.4. Concluding remarks are given in section 4.5.

The experimental error associated with these experiments was determined by repeating one experiment five times at fixed conditions (H_2SO_4 catalyst with 200 $g.kg^{-1}$ biomass loading at 330°C for 15 min with CO_2 atmosphere and methanol as solvent). The experimental error was calculated as 9.08% for a 95% confidence level and is presented in Figures 4.1 and 4.4 with vertical error bars.

4.2) Effect of the catalyst

4.2.1) Bio-oil and FAME yields

The effect of the catalyst type on the bio-oil, FAME and biochar yields was investigated by using three different catalysts: potassium hydroxide (KOH), sulphuric acid (H_2SO_4) and calcium carbonate ($CaCO_3$) at a catalyst loading of 20 $g.kg^{-1}$ during *in -situ* trans-esterification. The effect of the catalyst type on the bio-oil, FAME and biochar yield is given as a function of temperature in Figure 4.1, 4.2 and 4.3, respectively.

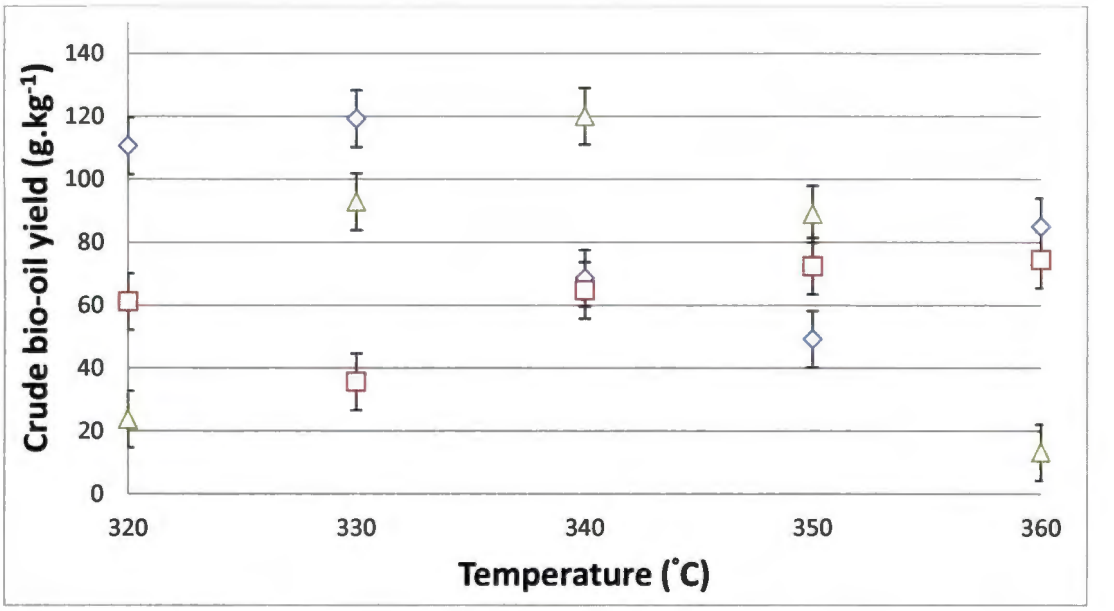

Figure 4.1: Effect of catalysts KOH (◊), H_2SO_4 (□) and the $CaCO_3$ (Δ) on bio-oil yield (See raw data in appendix).

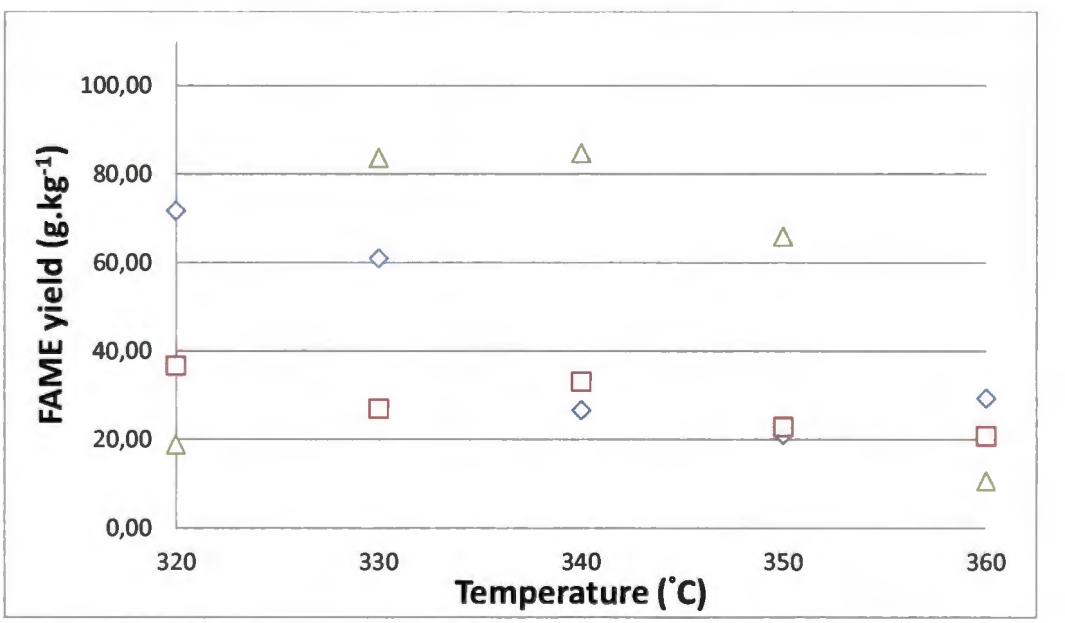

Figure 4.2: Effect of catalysts KOH (◊), H_2SO_4 (□) and $CaCO_3$ (Δ) on FAME yield catalysts (See raw data in appendix).

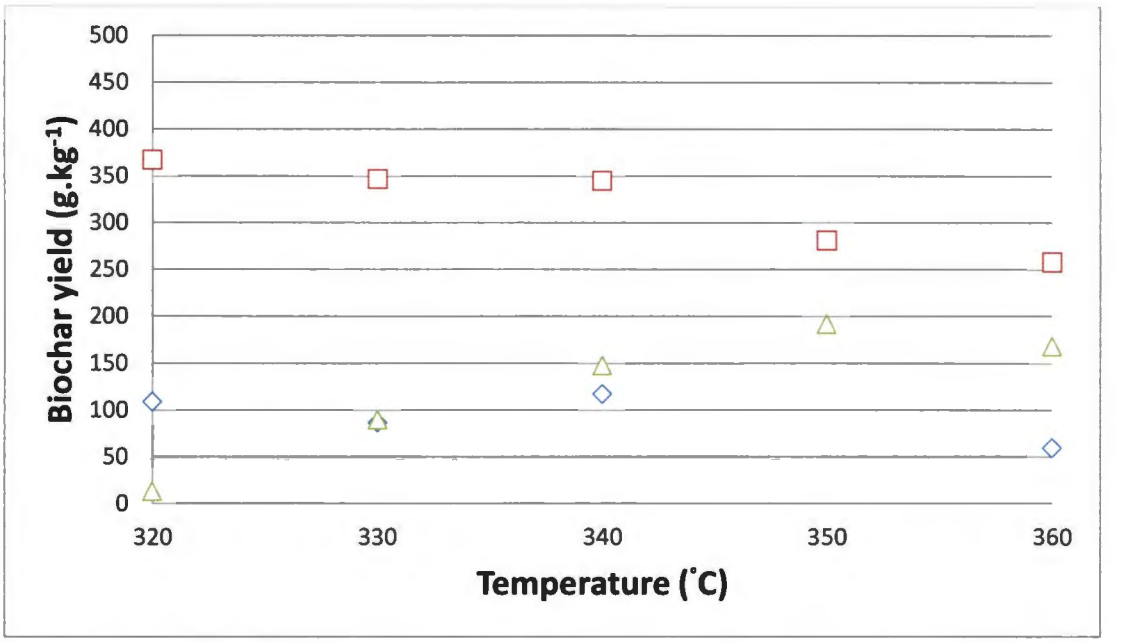

Figure 4.3: Effect of catalysts KOH (◊), H_2SO_4 (□) and the $CaCO_3$ (Δ) on Biochar production (See raw data in appendix).

From Figures 4.1, 4.2 and 4.3 it is evident that bio-oil yields of 110.55 $g.kg^{-1}$ and the highest FAME yields of 71.72 $g.kg^{-1}$ were obtained when using KOH as a catalyst at 320°C. A bio-oil yield of 120 $g.kg^{-1}$, a FAME yield of 84.71 $g.kg^{-1}$ and a biochar yield of 192.2 $g.kg^{-1}$ were obtained in the presence of $CaCO_3$ at 340°C. The primary product formation led to an initial increase in bio-oil yield from 320°C to 330°C, followed by the primary breakdown of bio-oil from 330°C to 350°C and reformation of bio-oil from 350°C to 360°C.

The first increase in bio-oil yield between 320°C and 330°C when KOH is used as a catalyst is attributed to the thermal breakdown of the basic biological components of the cell and the formation of bio-oil. The primary breakdown between 330°C to 350°C is brought about by the cracking of the bio-oil products to smaller compounds through isomerisation, dehydration and fragmentation reactions (Mazaheri *et al.*, 2013:749). The increase in the bio-oil yield between 350°C and 360°C is caused by condensation through cyclisation, hydration and repolymerisation reactions triggered by a high temperature and pressure (Mazaheri *et al.*, 2013:749). Alkaline catalysts are especially effective against cellulose and lignin biomolecules during liquefaction by hydrolysing the intra- and inter-molecular hydrogen bonds, as this causes swelling of the biomass that increases the surface of exposure of the cellulose to

reactants (Mazaheri *et al.*, 2013:750; Tian *et al.*, 2014:942; Centi *et al.*, 2011:24). The catalyst inhibits the formation of stable bonds such as carbon-carbon bonds that lead to the formation of gas products (Mazaheri *et al.*, 2013:750; Tian *et al.*, 2014:942). KOH promotes the water-gas shift reaction ($CO+H_2O = CO_2+H_2$) that leads to the production of H_2 and the consumption of H_2O (Jazrawi *et al.*, 2013:272). The hydrogen produced by the water-gas shift reaction and potassium (alkali metal with one electron to donate) stabilise free radicals to promote the formation of bio-oil. KOH is a base catalyst that increases the pH of the solution, resulting in compound ionisation or dissociation of biomolecules, and leading to an abundance of charged particles. This causes a reduction in the dielectric constant of the solution and increases the solubility of different compounds. The high number of molecules present increases the possibility of dehydration reactions during temperature increases and the production of water molecules leads to the formation of soap and favours the solubility of organics in the aqueous phase (Anastasakis & Ross, 2011:4879). The low and constant char yield obtained when using KOH as catalyst, supports the notion that alkali salts do not favour the formation of biochar and are conducive to the formation of gasses and to a lesser extent bio-oil (Amin *et al.*, 2009:16 ; Zhou *et al.*, 2010:4057-4058).

The reaction mechanism of calcium carbonate proposed by Murakami *et al.* (2015:91) is an ion-exchange reaction with carboxylic acids in a mixture of the calcium-ions during coal gasification.

$$CaCO_3 = Ca^{2+} + CO_3^{2-} \quad (1)$$

$$Ca^{2+} + 2(\text{-COOH}) = Ca(\text{-COOH})_2 + 2H^+ \quad (2)$$

$$2H^+ + CO_3^{2-} = H_2O + CO_2 \quad (3)$$

During $CaCO_3$ catalysed liquefaction, there is a complex set of reactions taking place simultaneously. The Ca-ion is an alkali earth metal with the potential to donate two electrons and can act as a simple Bronsted base catalyst. The Ca-ion is believed to participate via Lewis acid complexation to the carbonyl group of the triglyceride (Suppes *et al.*, 2001:145). The latter, combined with the fact that the carbonate ion is a strong base, results in increased alkoxide -intermediate formation that serves as a catalyst for trans-esterification. The carbonate ion can also break down to form CO and CO_2, creating a stable atmosphere as

shown in equation 3 (Galadima & Maruza, 2014:1; Suppes *et al.*, 2001:145). This explains the higher FAME yield obtained with $CaCO_3$ as a catalyst compared to the yield with KOH as a catalyst at higher temperatures.

During the ion exchange reaction in equation 2, weak organic acids are produced from the interaction between the Ca-ion and the carboxylic acids (originate from triglyceride degradation). These acids can contribute to the catalysis of trans-esterification or, under harsh conditions it may reduce yield via a dehydration reaction producing Ca-containing emulsifying agents (Manabe *et al.* 2001:10102). Murakami *et al.* (2015: 91) showed that most of the carboxylic acid ends up as CO_2, but the carboxylic acid also originates from other sources that have not been determined yet, suggesting multiple potential Ca-ion exchange reactions. The combinations of these reactions accelerate the degradation of the feedstock and favour gasification, which explains the decrease in bio-oil and FAME yields at higher temperatures (Murakami *et al.*, 2015:94).

Both alkaline catalysts produced relatively low biochar yields. According to Zhou *et al.* (2010:4058) alkaline catalysts suppress biochar formation and increase gas formation. This is in agreement with the low char and oil yields obtained in the presence of the alkaline catalysts in this study.

From Figures 4.1, 4.2 and 4.3, it is clear that with sulphuric acid (H_2SO_4) as a catalyst the highest yields in the presence of H_2SO_4 were 36.61 $g.kg^{-1}$ FAME, 61.15 $g.kg^{-1}$ bio-oil and 367.25 $g.kg^{-1}$ biochar at a temperature of 320°C. The breakdown of bio-oil occurred from 320°C to 330°C and the formation of bio-oil occurred from 330°C to 360°C.

The breakdown of biomass occurred due to the acidic catalyst hydrolysing and breaking down carbohydrates to polar water soluble compounds (Yang *et al.*, 2014:940). The increase of bio-oil was caused by reactive intermediates from the mixture produced during the decomposition of biomass through depolymerising, recombining and repolymerising to form bio-oil (Tian *et al.*, 2014: 941)

Sulphur is detrimental to lignin gasification, slowing down and even halting the gasification process and therefore, it is considered as a poison in coal and biomass gasification (Osada *et al.*, 2007:1405). Sulphur binds to aromatics, such as benzene and toluene (both present in

abundance), to form tars. Sulphur also binds to trace elements, especially metals, forming metal sulphide complexes on any surface. These sulphur containing species bind and form inactive sulphur masses (Yung *et al.*, 2009:1884). This was evident in this study through the high biochar yields obtained due to the formation of lignosulfonates, a causative agent of gumming. Sulphuric acid is also a strong diprotic acid. A single molecule will donate two protons to the solution, decreasing the pH and triggering the binding of hydrogen atoms by biomolecules and the reduction of charged particles in solution. This will result in desorption of complexes that will contribute to biochar formation and decreased bio-oil formation.

Sulphuric acid had the highest char production, peaking at 367.25 $g.kg^{-1}$ at the lowest temperature of 320°C that gradually reduced in a linear fashion as the temperature increased. The yield of char should, and did decrease gradually with increase in temperature (Zhou *et al.*, 2010:4057). Yang *et al.* (2014:938) reported a 150 $g.kg^{-1}$ bio-oil yield from the sulphuric acid catalysed liquefaction of *Enteromorpha prolifera*, while Wagner & Haas (2011:1219) used sulphuric acid catalysed *in -situ* trans-esterification to yield 900 $g.Kg^{-1}$ FAME. Widayat *et al.*, (2013:72) produced optimum yields of 536 $g.kg^{-1}$ FAME from rubber seeds using a H_2SO_4 catalyst. This is significantly higher than the best yield obtained with sulphuric acid as a catalyst in this study.

4.3) Effect of temperature

The effect of temperature on the bio-oil and FAME yields was investigated with a single catalyst ($CaCO_3$), by varying the temperatures between 320°C and 360°C. The bio-oil yield was calculated as the mass of bio-oil obtained per kilogram of sunflower seed used. The FAME yield was calculated as the mass of FAMEs per kilogram of sunflower seed. The effect of temperature on the bio-oil and FAME yields using $CaCO_3$ as catalyst is presented in Figure 4.4.

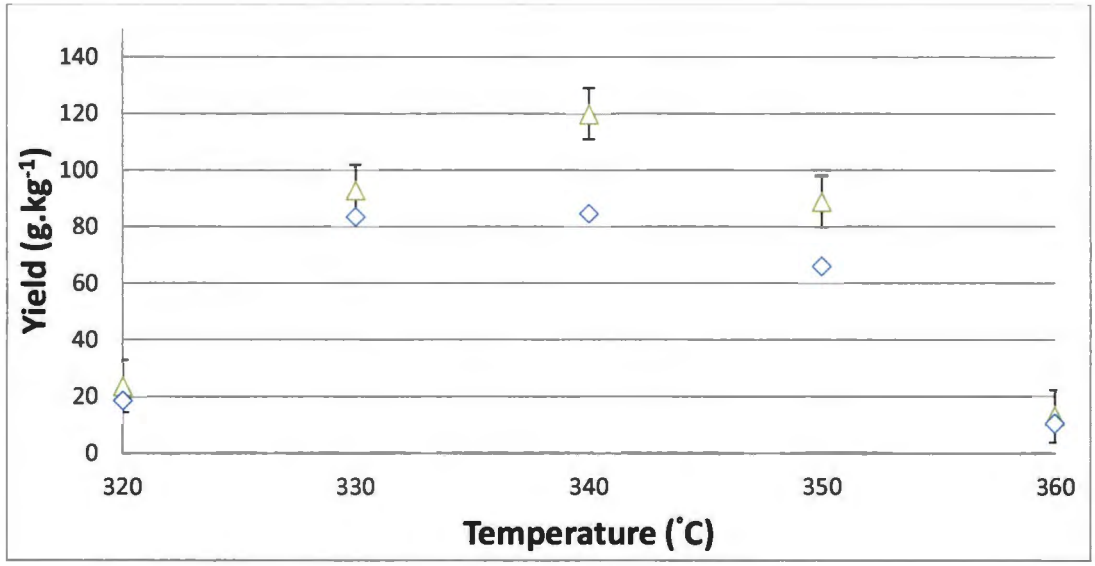

Figure 4.4: Effect of temperature on bio-oil (Δ) and FAME yield (◊) in the presence of a $CaCO_3$ catalyst (See raw data in appendix).

From Figure 4.4 it is clear that the highest bio-oil yield of 120 $g.kg^{-1}$ and FAME yield of 84.71 $g.kg^{-1}$ were obtained at 340˚C. There was an initial increase in bio-oil production as temperature increased until 340˚C, followed by a decrease in yield with an increase in temperature beyond 340˚C when cracking of bio-oil into smaller compounds resulted in yield reduction (Wang *et al.*, 2013:511). Other than cracking, the reduction in yield beyond 340˚C can be attributed to the condensation, cyclisation, and repolymerisation of the oil to form new compounds in biochar and dehydration, decarboxylation and deoxygenating facilitating depolymerisation into gas (Zhou *et al.*, 2010:4057). This tendency of bio-oils is consistent with the results reported by Wang *et al.* (2013:511-512) during liquefaction of *Litsea cubeba*, Qu *et al.* (2002:597) observed the same trend with direct liquefaction of *Cunnighamia lenceolata* and Zhou *et al.* (2010:4057) identified this tendency in marine macro algae, in which bio-oil yield increased until 300˚C before declining at 320°C.

Jin *et al.* (2014:343) found that 428.24 $g.kg^{-1}$ FAME can be delivered at an optimum temperature of 270˚C using a $ZnCl_2$ catalyst, while higher temperatures resulted in a reduction of FAME yields. Levine *et al.* (2013:559) reported ester losses at 275˚C and higher. Losses are attributed to thermal degradation that causes FAMEs to degrade to unsaturated compounds and then repolymerise to form biochar or be cracked to gasses above 300˚C. In experiments conducted by Patil *et al.* (2012: 826), ester production increased and peaked at 255˚C (850 $g.kg^{-1}$) and then reduced as temperatures increased. The lowered ester content at higher

temperatures was attributed to FAMEs becoming thermally unstable through cis- trans isomerisation of the carbon-carbon double bonds. The trans-esters are naturally unstable and break down through the autoxidation of compounds (Patil *et al.*, 2012: 825; Ghoreishi & Moein, 2013:27). Anitescu & Bruno (2012:140) showed that FAMEs start to degrade after approximately 6 min in supercritical fluids. The generation of esters under supercritical conditions changes the composition of the solution. Large changes in ester concentrations reduce the supercritical temperature of the reactant/product mixture and increases solubility. The increased solubility leads to favourable conditions for lower molecular weight compounds that cause the breakdown of bio-oil, including the breakdown of esters.

The breakdown of biomass occurs through the interactions between solvent and biomass or through thermal degradation (Barnard, 2009:44). Thermal degradation occurs through electron excitations that directs the creation of free radicals. The effect of temperature is the single largest influence on free radical production that is determined by the complexity of the biomass composition and the oxygen content of the biomass collectively (from which the free radicals are produced). The rate of free radical production increases with temperature, i.e. increased temperature accelerates the breakdown of biomass (Barnard, 2009:45). Sugar and protein degrade at lower temperatures. Carbohydrates primarily break down to sugars at temperatures around 200˚C when glycosidic linkages are attacked by free radicals, this results in dehydration, decarboxylation, decarbonylation and cleaving of molecules into smaller soluble fragments (Erzengin & Kucuk, 1998:1205). Protein degradation occurs when the C/N peptide bond hydrolyses and breaks the protein down to amino acids. Amino acids start to become unstable at 300˚C (especially positively charged glutamine and arginine). Lignocelluloses are decomposed at higher temperatures before the mixture stochastically condenses to other macromolecules (Kucuk, 2001:367). This thermal breakdown to lower molecular weight water soluble molecules explains why lower yields were obtained at higher temperatures. The decrease in yield in this study was only observed beyond 340˚C, while researchers report that individual components of biomass degrade at much lower temperatures, i.e. carbohydrates at 200˚C, amino acids at 300˚C and lipids at 260˚C to 280˚C.

Consistent with literature (Barnard, 2009:131; Shuping *et al.*, 2010:5408; Yu *et al.*, 2011:242-243; Yang *et al.*, 2004:32; Ross *et al.*, 2010:2242; Jin *et al.*, 2014:345 Patil *et al.*, 2012: 826; Levine *et al.*, 2013:556) low bio-oil and FAME yields were obtained in this study. The low

FAME yields are attributed to the low bio-oil yields. After an improved washing method including n-hexane extraction during water washing, was added, the FAME yield increased to 495 $g.kg^{-1}$ achieved at 330°C with a $CaCO_3$ catalyst. This is comparable to yields obtained and reported on in literature. Although the prior processing and oil extraction steps were eliminated in this direct or *in –situ* application production process, there was a need to include it in the washing stages due to the wide distribution of molecules present in the product mixture.

A summary of the bio-oil and biochar yields obtained at different temperatures, with different catalysts is provided in Table 4.1.

Table 4.1: Biochar and bio-oil yields from different temperatures using different catalysts (See raw data in appendix).

	Temperature (°C)				
KOH	**320**	**330**	**340**	**350**	**360**
Biochar ($g.kg^{-1}$)	**108.95**	**86.95**	**117.65**	**261.45**	**59.5**
Bio-oil ($g.kg^{-1}$)	**110.55**	**119.25**	**68.6**	**49.25**	**85**
H_2SO_4	**320**	**330**	**340**	**350**	**360**
Biochar ($g.kg^{-1}$)	**367.25**	**347.4**	**345.55**	**281.8**	**258.6**
Bio-oil ($g.kg^{-1}$)	**61.15**	**35.65**	**64.75**	**72.45**	**74.45**
$CaCO_3$	**320**	**330**	**340**	**350**	**360**
Biochar ($g.kg^{-1}$)	**13.35**	**90.1**	**147.9**	**192.2**	**168.55**
Bio-oil ($g.kg^{-1}$)	**23.75**	**92.85**	**120**	**88.95**	**13.2**

From Table 4.1 it is clear that there was a net increase in bio-oil production and a net decrease in biochar production with an increase in temperature. Biochar production reduced with increasing temperature as thermal degradation caused depolymerisation of solid residues to bio-oil or biogas (mainly H_2O, CO and CO_2). The repolymerisation of compounds is highly dependent on temperature, and as temperature increases, the formation of smaller volatile compounds is favoured (Zhou *et al.*, 2010:4057).

Sadaka *et al.* (2014:553), Zhou *et al.* (2010:4057) and Anastasakis and Ross (2011:4879) found that biochar production reduced as temperature increased.

These results conform to literature reported that char formation occurs at lower temperatures. As temperature increase, processes favouring bio-oil and gas production increases, reducing the char yield (Zhou *et al.*, 2010:4057; Anastasakis & Ross, 2011:4879; Sadaka *et al.*, 2014:553). The char yields obtained in this study were lower than those found in literature, i.e. 826 $g.kg^{-1}$ (Sadaka *et al.*, 2014:563), by using switchgrass as a feedstock 505 $g.kg^{-1}$ (Patil *et al.*,2011:118) from algae and 250 $g.kg^{-1}$ (Anastasakis & Ross, 2011:4878) also from algae.

4.3.1) Elemental analysis

The effect of temperature and catalyst type on the composition of bio-oil and biochar obtained in this study, is given in Table 4.2

Table 4.2: Elemental analysis of bio-oil and biochars at different temperatures with different catalysts

	Bio-oil							
Sample	Temperature (°C)	Carbon %	Hydrogen %	Nitrogen %	Oxygen %	H/C	O/C	HHV
KOH	320	74.57	10.72	1.48	14.10	1.71	0.14	40.12
	330	78.76	11.40	1.30	9.71	1.72	0.09	42.50
	340	78.76	11.38	1.58	9.75	1.72	0.09	42.46
	350	85.97	12.07	2.75	9.60	1.67	0.08	45.87
	360	79.23	10.97	2.04	9.35	1.65	0.09	42.04
H_2SO_4	320	79.15	11.15	3.34	7.05	1.68	0.07	42.28
	330	80.90	11.10	5.45	8.56	1.63	0.08	42.79
	340	78.43	11.00	5.29	8.10	1.67	0.08	41.82
	350	78.66	11.18	4.48	8.67	1.69	0.08	42.15
	360	76.73	10.95	6.20	9.88	1.70	0.10	41.18
$CaCO_3$	320	59.44	7.33	1.87	32.40	1.47	0.41	30.25
	330	78.78	11.13	1.80	19.55	1.68	0.19	42.12
	340	78.25	10.96	2.27	10.04	1.67	0.10	41.71
	350	74.27	10.37	1.93	14.07	1.66	0.14	39.53
	360	79.72	11.15	2.72	8.11	1.67	0.08	42.46

	Biochar							
Sample	Temperature (°C)	Carbon %	Hydrogen %	Nitrogen %	Oxygen %	H/C	O/C	HHV
KOH	320	48.09	3.83	2.56	45.51	0.95	0.71	25.20
	330	56.20	4.69	3.11	36.00	0.99	0.48	26.25
	340	68.08	6.03	3.59	22.30	1.06	0.25	30.29
	350	67.48	5.38	4.08	23.05	0.95	0.26	30.66
	360	49.66	3.94	2.25	44.15	0.95	0.67	30.46
H_2SO_4	320	64.71	7.19	3.04	25.06	1.32	0.29	28.73
	330	58.66	4.63	3.94	32.77	0.94	0.42	27.98
	340	80.51	7.31	4.50	7.67	1.08	0.07	31.35
	350	59.87	5.71	3.70	30.72	1.14	0.39	31.39
	360	71.97	6.91	3.55	17.57	1.14	0.18	31.82
$CaCO_3$	320	40.43	3.48	0.75	55.43	1.03	1.03	20.89
	330	62.42	5.41	3.27	28.90	1.03	0.35	29.47
	340	61.12	5.45	2.76	30.66	1.06	0.38	28.12
	350	61.13	6.37	2.29	30.20	1.24	0.37	29.37
	360	61.65	6.18	2.67	29.51	1.19	0.36	29.96
S.A. Coal		70.40	3.95	1.61	8.05	0.67	0.09	
Crude oil		82.00	7.10	4.90	5.40	1.03	0.05	

Biomass		43.7	5.6	45.6	1.8	1.53	0.03	29.61

It is evident from Table 4.2 that the carbon content of the bio-oil increased to 75-80wt% and the hydrogen content of the bio-oil increased to 10-12wt% when compared to the original biomass. The nitrogen content of the bio-oil increased when elevating the temperatures, but remained tenfold lower than the nitrogen content of the sunflower seeds. The oxygen content decreased to 9wt% with increasing temperature. An average H/C ratio of 1.70+-0.05 and an O/C ratio of 0.09+- 0.02 was observed for all the bio-oil produced. The higher heating value (HHV) increased with increasing temperature and a relatively high heating value was obtained for most of the bio-oils.

It is also clear from Table 4.2 that the carbon content of the biochar increased to 40-60wt% and the hydrogen content remained unaffected at approximately 5wt% in agreement with the results obtained by Jin *et al.*(2014:346), when compared to the composition of the sunflower seeds. The average nitrogen content of the biochar decreased at elevated temperatures and the oxygen content increased with increasing temperatures and was 20wt% on average. The high oxygen content of the biochar is due to catalysts precipitating in the char and lowering the quality. The biochars showed an H/C ratio of 1.10+-0.15 and an O/C ratio of 0.3 to 0.8 on average. Both the HHV and the carbon content increased with increasing temperatures.

The reduction in oxygen content in the produced bio-oil is indicative of the decarboxylation, deoxygenation and dehydration reactions promoted by an increase in temperature (Xiu *et al.*, 2010:1298; Jin, 2014:374).

The increase in nitrogen content indicates that higher temperature promoted the degradation of protein to NO_x-gasses (Jazrawi *et al.*, 2013:273; Jin *et al.*, 2014:347).

When biomass is subjected to thermal processing, an increase in the H/C ratio and a decrease in the O/C ratio are desirable when compared to the original material (Anastasakis & Ross, 2011:4880). The O/C ratios increased in both the char and bio-oil products when compared to the ratios in sunflower seed. This is due to the introduction of oxygen to the system through

the catalyst. The H/C ratio of bio-oil improved and that of biochar decreased when compared to the sunflower seeds.

The oxygen and nitrogen content of the bio-oil remain high and requires further processing for deamination and de-oxygenation. The temperature improved the H/C and O/C ratios and the HHV of the bio-oil when compared to the sunflower seeds, but there is little variation between the elemental compositions in the temperature intervals (320°C to 360°C). This corresponds with the results that were reported by Zhou *et al.* (2012:2345) and Jazrawi *et al.* (2006:276). The more severe the reaction conditions, the higher the carbon, hydrogen and nitrogen content, the lower the oxygen content.

The heating value of a substance is a measure of the energy per unit of volume or mass of a substance when burned (Sanli et al., 2014:850). A higher heating value is indicative of more energy stored in a fuel and is measured by an oxygenated bomb calorimeter as the heat released during the complete combustion of 1 g of fuel at 101,3 kPa at 298 K (Sanli et al., 2014:851; Demirbas, 2007:1744 and 2009:633).

The obtained HHV for all the oils and chars were high when compared to the values obtained in literature (Demirbas, 2007:1744 and 2009:635; Yang *et al.*, 2004:32; Sanli et al., 2014:852-853; Anastasakis & Ross, 2011:4880; Chen *et al.*, 2011:1940). The oil with the highest heating value (45.87 $g.kg^{-1}$) was prepared with KOH as catalyst at a reaction temperature of 350°C. This HHV is higher than the value obtained for upgraded bio-oil (34.38 $MJ.kg^{-1}$) in literature (Chen *et al.*, 2010:4606). The higher carbon and hydrogen contents and lower oxygen content correlated with an increase in HHV. Both the bio-oils and biochars had produced a high oxygen and nitrogen content when compared to values obtained in literature (Zhou *et al.*, 2012:2345).

For bio-oil and biochar, the loss of structural water acts as a hydrogen and oxygen sink as lignocellulose degrades (Sadaka *et al.*, 2014:553). The loss of structural water and volatiles and the lower O/C and higher H/C ratios provide the bio-oil and biochar with a composition closer to that of fossil fuels (Sadaka *et al.*, 2014:559).

Potassium hydroxide promotes deoxygenation via decarboxylation (Tian *et al.*, 2014:943). This combined with the water-gas shift reaction ($CO+H_2O = CO_2+H_2$) lead to the production of H_2 and the consumption of H_2O (Jazrawi *et al.*, 2013:272). Additionally, KOH introduced O- and

H- groups into the system from the catalysts during liquefaction. This serves as an explanation for the increase in hydrogen content and the lower oxygen content in KOH catalysed reactions.

For $CaCO_3$ catalysed reactions, a very high oxygen content, that was the result of the introduction of O- groups during liquefaction by the catalyst (donates 3 oxygen molecules per mole), was apparent. The catalyst promoted denitrogenation but had little effect on the carbon and hydrogen content, with minimal hydrogenation and deoxidation as is evident from the low HHV's (Jin, *et al.* 2014:348).

The carbon and hydrogen content of the oils and chars prepared in the presence of sulphuric acid as catalyst remained constant, despite the promotion of hydrogenation by the catalyst and the donation of two hydrogen atoms to the solution. The nitrogen content in the presence of sulphuric acid was the highest of all catalysts. Amino acids react with sugars in what is known as the Maillard reaction, to form N-containing cyclic compounds (Tian *et al.*, 2014:944). Most of these compounds are washed out during the organic phase when using other catalysts but in the case of sulphuric acid catalysed reactions, sulphur as discussed earlier, binds to aromatics (which now contain nitrogen), leading to an increased nitrogen content in the bio-oil and biochar (Yung *et al.*, 2009:1884). The use of sulphuric acid as catalyst did not reduce the HHV of oil significantly as is evident from other studies (Yang *et al.*, 2014:942).

Biochar tends to vary a lot more than bio-oil in terms of composition and HHV (Anastasakis & Ross, 2014:550). Catalysts tend to lower char quality by precipitating as solid mass, increasing carbon content and decreasing oxygen content (Anastasakis & Ross, 2011:4881; Chen *et al.*, 2011:1935). The chars from the sunflower seeds in this study compare well to the biochars from switchgrass (59.6% C, 4.7% H, 31.3% O, 1% N) (Sadaka *et al.*, 2014:558). These are considered low quality when compared to the chars obtained from soybean straw (72.37% C, 3.42% H, 3.13% N and 21.08% O) and for sunflower seeds (72.13% C, 3.25% H, 2.84% N and 2.78% O), although these were subjected to additional processing (Chen *et al.*, 2010: 4607). An oxygen content of 5.5 -8.6wt% is considered low (Anastasakis & Ross, 2011:4879).

The effect of a catalyst is dependent on the type of catalyst, as indicated by the abovementioned results, in which none of the three selected catalysts delivered the same trends regarding biochar production. Catalysts precipitate in char, increasing the mineral

content and lowering the quality. Catalysts tend to lower char quality, decrease the gas yield and increase the oil yield (Chen *et al.*, 2011:1935; Anastasakis & Ross, 2011:4881).

4.4) Effect of biomass loading

4.4.1) Bio-oil, FAME and biochar yields

The effect of biomass loading was investigated by using three different biomass loadings of 150 $g.kg^{-1}$ solvent (15wt%), 200 $g.kg^{-1}$ solvent (20wt%) and 250 $g.kg^{-1}$ solvent (25wt%) with a 20 $g.kg^{-1}$ $CaCO_3$ catalyst loading at a constant temperature of 330 °C. The effect of biomass loading on the biochar, bio-oil and FAME yields is provided in Figure 4.5.

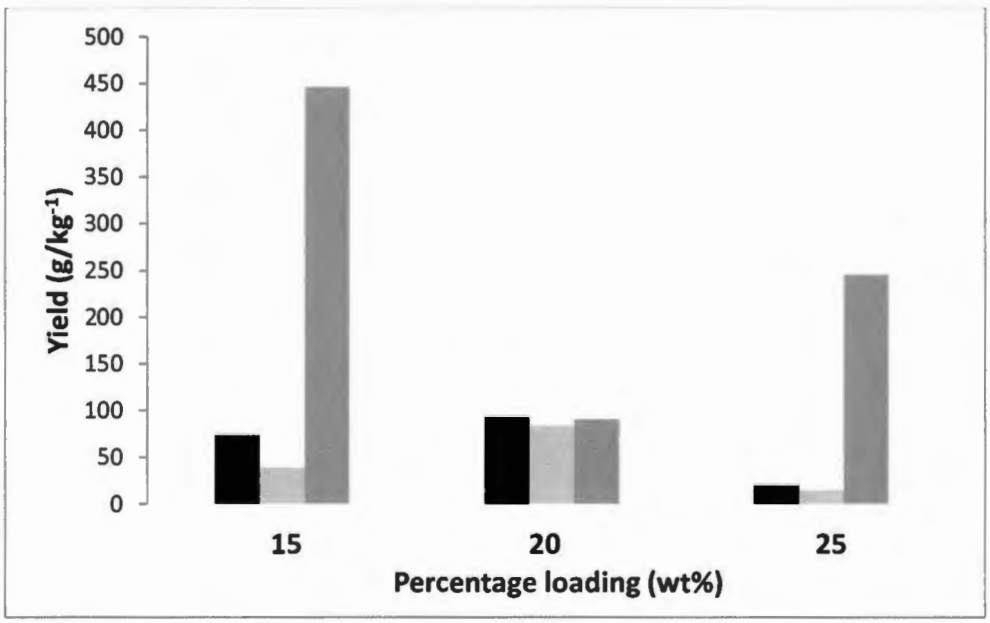

Figure 4.5: The influence of biomass loading on bio-oil ($g.kg^{-1}$), FAME ($g.kg^{-1}$) and biochar yield ($g.kg^{-1}$) (See raw data in appendix).

The best results were achieved at a biomass loading of 200 $g.kg^{-1}$ for bio-oil (92.85 $g.kg^{-1}$) and FAME (83.63 $g.kg^{-1}$) yield and 150 $g.kg^{-1}$ for biochar production (446.1 $g.kg^{-1}$).

Lower biomass loadings should yield lower levels of solid since more solvent is available for solvolysis (higher alcohol to oil molar ratio) (Behrendt *et al.*, 2008:672). However, several studies, including this one have identified that higher biomass loadings yield lower char

content when compared to lower biomass loadings (Amin *et al.*, 2009:25; Anastasakis & Ross, 2011:4878; Behrendt *et al.*, 2008:672).

The trends in the results of this study where similar to that found in other studies, in that both under and over loading proved to be detrimental to yields. The biomass loading determined the alcohol to oil molar ratio, decreasing the biomass loading increased the alcohol to oil ratio and the surface of exposure of the lipids (Ghoreishi & Moein, 2013:27). The supercritical point of the reactant/product mixture was reduced when the surface of exposure was increased (Ghoreishi & Moein, 2013:27; Patil *et al.*, 2011:121; Anitescu & Bruno, 2012:138). Optimum yields are achieved when the solvent solute mixture is in supercritical state because the (pseudo) critical point removes the interface layer between the phases of the optimum mixing and maximum exposure levels (Anitescu & Bruno, 2012:138). The methanol breaks down intermolecular hydrogen bonding under supercritical conditions resulting in the reduction of the polarity and dielectric constant of methanol, allowing it to function as a free monomer (Patil *et al.*, 2011: 191). This allows methanol to act as a solvent and a hydrogen donor that stabilises radical production to promote the formation of oil, inhibiting secondary decomposition of oil and repolymerisation (Zhou *et al.*, 2012:2345). Higher alcohol volumes can also inhibit polymerisation and thermal degradations by diluting reactants and free radical production (Levine *et al.*, 2013:261). Exceeding the critical temperature can also lead to the breakdown of bio-oil and FAME (Ghoreishi & Moein, 2013:27; Patil *et al.*, 2011:121; Jin *et al.*, 2014:344). The opposite occurs when the biomass loading is too high and the reaction becomes obstructed at high doses (Ghoreishi & Moein, 2013:29).

This explains the low bio-oil yields at low biomass loadings of 150 $g.kg^{-1}$ in which the sample was diluted to such an extent that biomass only partially degraded, leading to high biochar yields. At high biomass loadings the reaction chamber became obstructed and unstabilized free radical formation led to excess gas production. This was confirmed by the elemental analyses that showed a low carbon and high oxygen content.

4.3.2) Compositional analysis

The influence of the three biomass loadings on carbon, hydrogen, nitrogen and oxygen content is given in Table 4.3.

Table 4.3: Elemental composition at different biomass loadings in the presence of 20 $g.kg^{-1}$ $CaCO_3$ as catalyst and a reaction temperature of 330°C (See raw data in appendix).

	Bio-oil							
	Biomass loading ($g.kg^{-1}$)	Cwt%	Hwt%	Nwt%	Owt%	H/C Molar ratio	O/C Molar ratio	HHV
Biomass loading	150	74.75	10.14	2.46	13.91	1.62	0.14	39.37
	200	78.78	11.13	1.80	19.55	1.68	0.19	42.12
	250	52.77	3.77	5.76	37.70	0.85	0.08	22.96
	Biochar							
	150	55.89	5.63	2.31	36.17	1.20	0.49	28.52
	200	62.42	5.41	3.27	28.90	1.03	0.35	29.42
	250	62.42	6.42	2.79	26.57	1.23	0.32	29.67
South African coal	-	70.40	3.95	1.61	8.05	0.67	0.09	39.06
Crude oil	-	82.00	7.10	4.90	5.40	1.03	0.05	37.42
Sunflower seeds	-	43.70	5.60	45.60	1.80	1.53	0.03	29.61

It is clear from Table 4.3 that the biomass loading appeared to have had a major influence on the elemental compositions of both the bio-oil and biochar. All three biomass loadings, when compared to the raw biomass, had an increased carbon, hydrogen, and oxygen content and HHV, as well as a reduced nitrogen content of the bio-oil and biochar. These results agree with results in the literature indicating that C, H, N, O and HHV are influenced by biomass loading when compared to raw biomass (Zhou *et al.*, 2012:2350; Yang *et al.*, 2014:942; Anastasakis & Ross, 2011:4880 ; Qu *et al.*, 2002:597). These results contradict findings by Jazrawi *et al.* (2006:273) indicating that biomass loading should not have an adverse effect on bio-oil quality.

The increase in carbon and hydrogen content is due to the promotion of denitrogenation, hydrogenation and deoxygenation, as evident from increasing HHVs (Jin *et al.*, 2014:347). A loss of carbon at a biomass loading of 250 $g.kg^{-1}$ is attributed to gasification in the form of CO and CO_2 (Valdez et al., 2011:3239; Chen *et al.*, 2011:1939).

Lower biomass loadings favoured the hydrogenation of bio-oil as indicated by the high hydrogen content (Jin *et al.,* 2014:348). The oxygen content is reduced due to the reduction in carbonyl content by the formation of CO and CO_2 gas products, as well as H_2O (Chen *et al.*, 2011:1939). Nitrogen concentrations reduced, but remained high (7-9wt%). This indicative of the contribution of protein to the oil. When compared to the biomass, the reduction of nitrogen, is due to denitrification to NO_x, NH_4 and other water soluble compounds (Jin *et al.*, 2014:347; Zhou *et al.*, 2012:2346). The reduction in the C/H ratio in chars at a biomass loading of 200 $g.kg^{-1}$ is partly due to the repolymerisation to oils and formation of benzenes and phenols as confirmed by GC-MS (discussed in section 4.4) (Chen *et al.*, 2011:1940; Zhou *et al.*, 2012:2350).

These results contradict findings by Zhou *et al.* (2012:2344) and Patil *et al.* (2011:121) indicating that char production is independent of the biomass loading. Biomass loading has a much greater effect on yield, than catalyst loading. It is second to temperature regarding the influence it has on the product (Anastasakis & Ross, 2011:4879).

4.4) Product characterisation

This section covers all the reaction parameters studied and the effect of these parameters on the energy content and composition of the product from *in-situ* trans-esterification liquefaction reactions.

4.4.1) Compositional and structural analysis

This compositional and structural analysis was done by GC-MS and FTIR. The GC-MS results are presented in Table 4.4 and then the FTIR and GC-MS results are discussed. Fourier Transform Infrared Spectroscopy (FTIR) is a quantitative method used to identify functional groups and compounds corresponding to their absorption/transmittance (Magida, 2013).

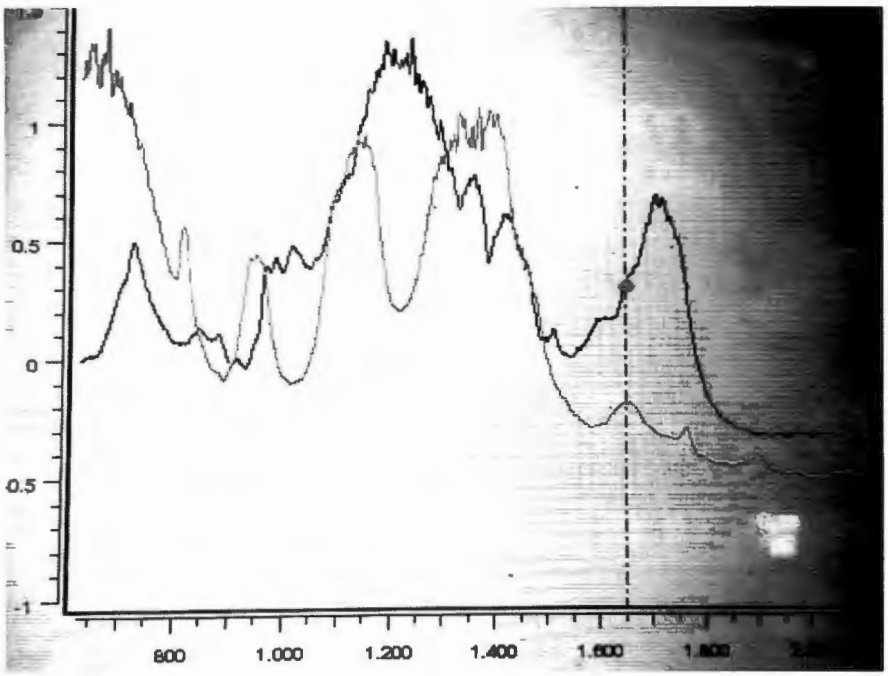

Figure 4.6: FTIR absorbance spectrum of a sample prepared at 330°C using $CaCO_3$ as catalyst.

Table 4.4: Summary of FTIR peaks (Ozcimen & Karaosmanoglu, 2004:784; Zhou *et al.*, 2012:2345-2346; Silverstein, 1981)

Wavelength (cm^{-1})	Active Group
1600-1800	Carbonyl Groups (C=O Vibrations)
1705-1725	Acyclic Ketones
1705-1850	Cyclic Ketones
1735-1750[1]	Esters (C=O Vibrations)
1377, 1456	Esters (C-H Bending)
1735-1750	Carboxylic Acids (C=O Vibrations)
1377	Aromatics (C-O Stretching)
650-900	Aromatics (C-H Bending)
700-850, 1450-1600	Aromatics (C=C Stretching)
1000-1100	Alcohols (C-O Stretch)
1400-1600	Amines (C-N Stretch)
1515-1560	Nitro- Compounds (N-O Stretch

The GC-MS analysis was performed to identify the specific molecules in the bio-oil samples. More than 110 peaks were identified per run and Table 4.4 is a compound list of tentative identities.

Table 4.5: Molecules identified by GC-MS

Toluene	**9,12-octadecadienoic acid (Z,Z)-,methyl ester**
Hexanal	**10,13-Octadecadienoic, methyl ester**
Pentanoic acid, 2,4-dimethyl-,methyl ester	**6,9-Octadecadienoic, methyl ester**
Octanoic acid,2-methyl-,methyl ester	**5-nonadecen-1-ol**
Nonanoic acid methyl ester	**9,15-Octadecadienoic, methyl ester**
Nonanoic acid	**9,11-Octadecadienoic, methyl ester**
Benzenepropanoic acid methyl ester	**5-dodecyne**
Benzenepropanoic acid	**9-octadecenoic acid (Z) methyl ester**
Pentadecanoic acid,14 methyl, methyl ester	**10-octadecenoic acid methyl ester**
Hexadecanoic acid, 14-methyl, methyl ester	**7-octadecenoic acid methyl ester**
Hexadecanoic acid	**Hexadecanoic acid,16-methyl, methyl ester**
8,11-octadecadienoic acid, methyl ester	**Hexadecanoic acid, 15-mthyl, methyl ester**
9,12-octadecadienoic acid,(Z,Z)-, methyl ester	**Heptadecanoic acid,16-methyl, methyl ester**
8-octadecenoic acid, methyl ester E	**Cyclopentanetridecanoic acid, methyl ester**
9-octadeccenoic acid, methyl ester Z	**Decanoic acid methyl ester**
6-octadeccenoic acid, methyl ester Z	**Phenol, 4-ethyl-2methoxy-**
9,12-octaddecadien-1-ol,(Z,Z)-9-eicosyne	**Phenol, 2-methoxy-4-propyl**
Octadecanoic acid	**10-undenoic acid methyl ester**
7,10-octadecadienoic acid, methylester	**Methyl tetra decanoate**

From the GC-MS analysis, three major types of compounds where identified, i.e. phenols, methyl esters and carboxylic acids. Higher temperatures and alkali catalysts produced a wider range of molecules. The methyl ester production of C5-C20 remained consistent with all catalysts, and as temperatures increased shorter chain production was favoured (Amin *et al.*, 2009:15). The methyl ester hydrocarbon chains consisted of even and uneven numbers, straight and branched as well as saturated, mono- and polyunsaturated hydrocarbon chains. The ketones and phenols present were derived from the decomposition of lignocellulose. Increasing temperature further increased the ketone and phenol content, indicating the breakdown of lignocellulose (Anastasakis & Ross, 2011:4879). Lignin is composed of phenyl propane, a rich source of phenolic compounds (Karagoz *et al.*, 2004:362). Other Indole, pyrole and phenol derivatives may also originate from cyclic amino acids such as phenylalanine and tryptophan (Ross *et al*, 2010:2238). The presence of pyran derivatives is thought to be due to

the presence of methanol (Karagoz *et al.*, 2004:237). Reaction conditions may have contributed to the abundance of certain types of smaller molecular weight compounds and aromatics, e.g. Increases in temperature increased the ketone fraction through cyclopentenone derivatives (Amin *et al.*, 2009:15; Anastasakis & Ross, 2011:4880).

The most common compounds were aromatics such as ethyl/methyl-benzenes (toluene and styrene); aliphatic compounds such as pentadecene, cycloalkanes and oxygenated hydrocarbons (hexanal), C5 -C20 methyl esters and several different amides (Anastasakis & Ross, 2011:4880; Wang *et al.*, 2013:514). The amide content, also identified with FTIR, is indicative that part of the oil production stems from non-lipid components such as protein (various reactions with ammonia after protein deamination) and lignocellulose (Wang *et al.*, 2013:514; Anastasakis & Ross, 2011: 4879). Fatty acid esters were produced from fats and waxes, as was identified with FTIR and GC-MS (Karagoz *et al.*, 2004:237). Biomass derived oil is extremely complex in this study it contained 30% cyclic compounds, indicating that phenols are the major destinations for many constituents (Karagoz *et al.*, 2004:241). An increase in phenol content is promoted by the formation of smaller molecules after primary breakdown, then depolymerisation into unsaturated compounds and cyclic, aromatic and polycyclic compounds (Anastasakis & Ross, 2011:4879). This explains the high cyclic contents identified by both GC-MS and FTIR.

GC-MS and FTIR analysis detected and converged in an abundance of aromatic compounds, stemming from unsaturated, branched, uneven hydrocarbon chains. The bio-oil can therefore be classified as paraffin-like, but a high level of methyl esters means that the bio-oil should be classified as low quality diesel oil (Yang *et al.*, 2004:32).

4.5) Diesel properties

Some properties of the diesel produced in the presence of the calcium carbonate catalyst are provided in Table 4.6.

Table 4.6: Characterisation of prepared biodiesel

Biodiesel property	End product	ASTM standards	Literature
FAME yield ($g.kg^{-1}$) (min)	900.71	95.6	20-990
Bio-oil yield ($g.kg^{-1}$)	550	-	50-900
Water content (%) (min)	0.15	0.005	0.001-5
Cetane number (min)	88.7	51	49
Density (kg/m^3)	0.9199	860-900	880-993
Biodiesel HHV ($MJ.Kg^{-1}$)	39.599	-	20-43
Char HHV	27.803	-	10-25

During the washing stages large amounts of oil were lost due to a lack of or very slow separation. The water washing was chosen in order to keep production costs at a minimum, but due to the vast array of molecules produced, emulsification effectively nullified separation and reduced the yield. Hexane extraction was introduced into the washing step of the highest FAME yielding parameter. This proved to be very advantageous, since 9 successive runs with this improved washing yielded an average of 555 $g.kg^{-1}$ of oil produced per run. This unequivocally proves that organic solvents improve yields that would not have been obtained in the absence of the solvent (Valdez *et al.*, 2011:3243). These results compares well to results in the literature that obtained bio-oil yields that varied between 100 $g.kg^{-1}$ and 700 $g.kg^{-1}$ and were composed of fatty acids, fatty acid methyl esters, ketones, various aromatics, phenols, aldehydes, etc. (Yang *et al.*, 2004:32; Shuping *et al.*, 2010:5409).

The viability of a novel biodiesel production process has been asserted. Table 4.7 directly compares the raw biomass to the biodiesel end product and shows that the energy content has been concentrated.

Table 4.7: Raw material vs. best results (Yield, FAME, C/O, C/H, HHV)

	Mass Yield	FAME Yield	Cwt%	Hwt%	Owt%	Nwt%	O/C	H/C	HHV ($MJ.kg^{-1}$)
Sunflower seed	-	-	43.7	5.6	45.6	1.8	0.03	1.53	29.61
Bio-oil	92.85 $g.kg^{-1}$	900.71 $g.kg^{-1}$	78.78	11.13	19.55	1.80	0.19	1.68	41.97

Various different methyl esters (straight, branched and cyclic), phenols, carboxylic acids, amines and nitro-containing compounds were identified by GC-MS and FTIR. The products of *in -situ* trans-esterification by liquefaction and supercritical carbon dioxide can compete with fossil fuels regarding the HHV. However, low quality biodiesel was produced that requires secondary processing to reduce the cyclic compounds and oxygen content. These findings contribute to the current growing bed of knowledge to overcome the blatant lack of innovative uses for the existing technology.

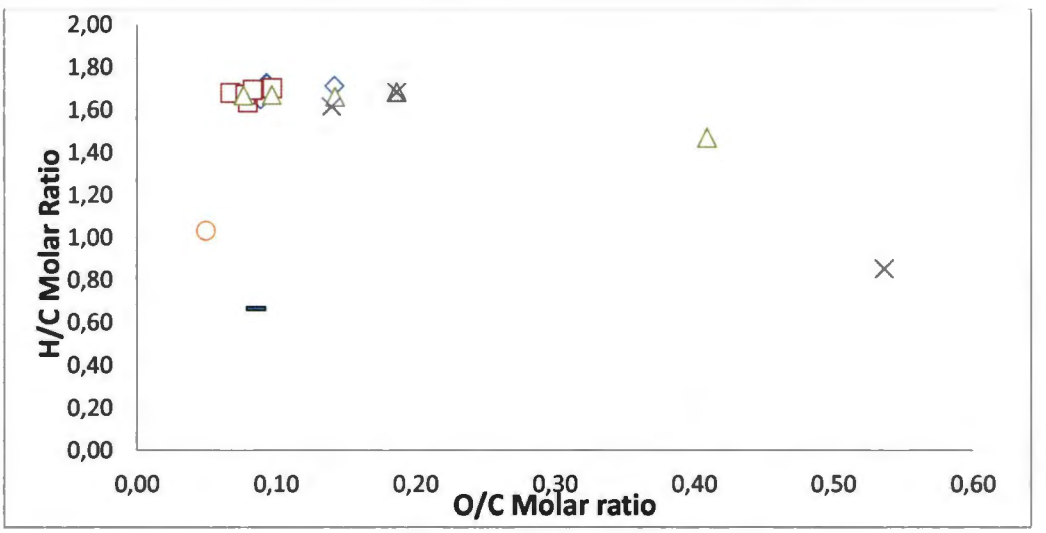

Figure 4.7: Van Krevelen diagram of bio-oil. Catalysts, KOH (◊), H_2SO_4 (□) and $CaCO_3$. (Δ), as well as biomass loading (x), Crude oil (o), South African Coal (-) are represented.

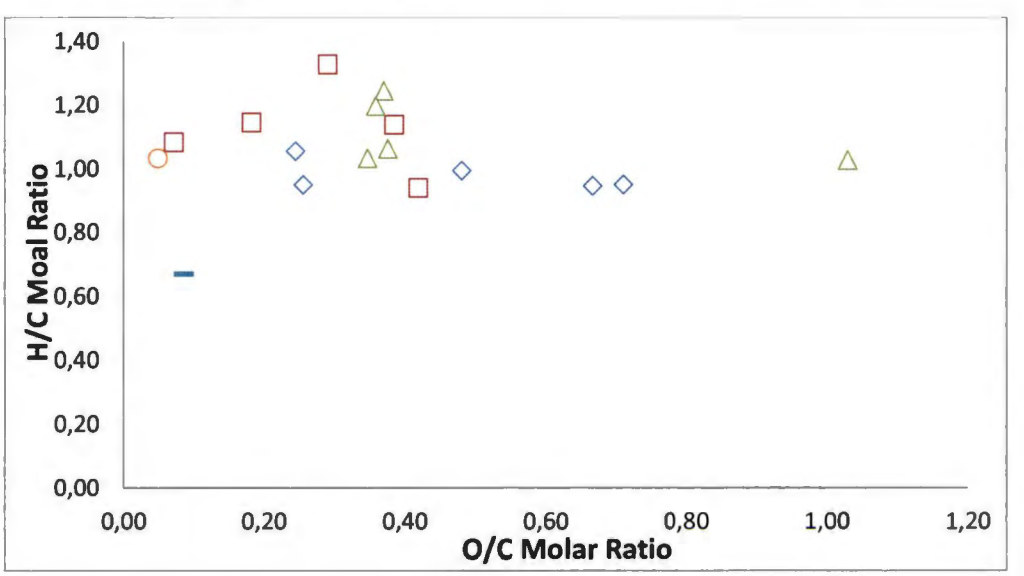

Figure 4.8: Van Krevelen diagram of biochar. Catalysts, KOH (◊), H_2SO_4 (□) and $CaCO_3$. (Δ), as well as biomass loading (x), Crude oil (o), South African Coal (-) are represented.

A Van Krevelen diagram (Figures 4.7 and 4.8) is a plot of the C/H molar ratios against the C/O content. This is used to show the change in oil quality relative to specific fossil fuels. The HHV tend to increase at points closer to the upper left hand corner of the diagram i.e. higher carbon-hydrogen ratios and lower oxygen ratios improve the HHV. These plots indicate that bio-oil results clustered around a threshold point of oxygen. The bio-oil can be compared to crude oil (Figure 4.7). The biochar results (Figure 4.8) re-affirmed the notion that the different catalysts delivered scattered results for biochar.

5) Conclusion

The goal of this study was to combine the high temperatures and pressures of liquefaction, with the cost effective thinking of *in -situ* trans-esterification and the improved solubility of supercritical extraction in an attempt to optimise *in -situ* biodiesel production. This involved an investigation into the consequences created by the reaction parameters of temperature, catalyst and biomass loading and their effects on mass yield, FAME yield, energy content, elemental and structural composition.

5.1) Influence of catalyst

The influence of the catalyst was third to temperature and biomass loading in influencing the yield of bio-oil, FAME and biochar. The three catalysts delivered products that were different from one another in terms of yield and elemental composition. The alkaline catalysts favoured gas production and suppressed biochar production. The acid catalysts favoured biochar production and had the greatest positive influence on elemental composition. The catalysts had a major effect on Bio-oil, FAME and biochar production and the elemental composition. The carbon, hydrogen, nitrogen and oxygen contents were all influenced differently by the different catalysts, with the oxygen content being particularly sensitive. The HHV indicated that catalysts play an indirect role in energy content, by influencing the oxygen content. From the results the researcher deduced that calcium carbonate was the superior catalyst. These results emphasise that the highest yields are not necessarily the best yields.

5.2) Influence of temperature

The effect of temperature is the major contributing factor affecting bio-oil, FAME and biochar yields. The results obtained at the different temperatures compared well to results obtained in literature in which the optimum temperature varies widely between 300˚C and 400˚C. An increase in temperature led to an increase in oil yield and conversion (to biodiesel), but only up to a certain point, after which thermal degradation increased and gas production was

favoured. There was an apparent threshold limit to the effect of temperature on yield and elemental composition due to thermal degradation (seen across different scientific fields, such as biochemistry, where enzyme function improves up to the point where the temperature causes denaturation of the same enzyme). Temperature is the first parameter that should be optimised.

5.3) Influence of biomass loading

Biomass loading had an adverse effect on yield which, by proportion exceeded the effects of catalysts, but not those of temperature. Three different biomass loadings where tested and the results illustrated that both under- and overdosing of the reaction mixture is detrimental to a reaction. Biomass loading had a major effect on the product distribution and should be optimised after temperature.

5.5) Recommendations

- The researcher recommends that a large scale parallel experiment is done to analyse different catalysts and catalyst dosages.
- It is recommended that an investigation is performed to determine the composition of the water phase after washing.
- The researcher recommends that an investigation into different solvents.
- It is recommended that an investigation is launched to determine the effect of different reaction atmospheres.
- IT is recommended that the gas yields are analysed to complete the mass balance.

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Appendix:

In this section, the experimental data that was used in this study is given. Table A lists the mass yields of both the bio-oil and biochar for all parameters tested. Table B provides a summary of the elemental analysis of bio-oil and biochar at different temperatures with different catalysts, which can also found on page 53, as well as how it was calculated. The calorific values (LHV), as measured with a bomb calorimeter and calculated HHV is provided in Table C. All the tables include the data-processing of the raw values.

Table A: Raw Yields of Bio-Oil and Biochar

Variables		Bio-oil				Biochar	
Catalyst	**Temperature (°C)**	**Yield by weight as measures (g.20g^{-1} biomass)**	**Biocrude yield (g.kg^{-1}) (i)**	**Fame yield (%) (ii)**	**FAME (g.kg^{-1}) (iii)**	**Yield by weight as measures (g.20g^{-1} biomass)**	**Biochar yield (g.kg^{-1}) (iv)**
KOH	**320**	2.211	110.55	64.876	71.72	2.179	108.95
	330	2.385	119.25	51.106	60.94	1.739	86.950
	340	1.372	68.600	38.672	26.53	2.353	117.65
	350	0.985	49.250	42.686	21.02	5.229	261.45
	360	1.700	85.000	34.467	29.30	1.19	59.500
H_2SO_4	**320**	1.223	61.150	59.871	36.61	7.345	367.25
	330	0.713	35.650	75.331	26.86	6.948	347.40
	340	1.295	64.750	51.005	33.03	6.911	345.55
	350	1.449	72.450	31.413	22.76	5.636	281.80
	360	1.489	74.450	27.833	20.72	5.172	258.60
$CaCO_3$	**320**	0.475	23.750	79.296	18.83	0.267	13.350
	330	1.857	92.850	90.071	83.63	1.802	90.100
	340	2.400	120.00	70.593	84.71	2.958	147.90
	350	1.779	88.950	74.139	65.95	3.844	192.20
	360	0.264	13.200	80.261	10.59	3.371	168.55
Biomass loading (wt%)	**15%**	**330**	1.103	73.537	52.016	38.25	8.922	446.10
	20%	**330**	1.857	92.850	90.071	83.63	1.802	90.100
	25%	**330**	1.178	20.000	70.593	14.12	4.915	245.75

i) Biocrude yield = (yield by weight * 50)
= (20g*50)
= yield in g.kg^{-1}

ii) % FAME as indicated by GC

iii) FAME (g.kg^{-1}) = % Fame *10
=Yield in g.kg^{-1}

iv) Bio-char yield = (yield by weight * 50)
= (20g*50)
= yield in g.kg^{-1}

Table B: Elemental analysis of bio-oil and biochars at different temperatures with different catalysts (also found on page 53).

	Bio-oil							
Sample	Temperature (°C)	Carbon%	Hydrogen %	Nitrogen %	Oxygen%	H/C	O/C	HHV (i)
KOH	320	74.57	10.72	1.48	14.10	1.71	0.14	40.12
	330	78.76	11.40	1.30	9.71	1.72	0.09	42.50
	340	78.76	11.38	1.58	9.75	1.72	0.09	42.46
	350	85.97	12.07	2.75	9.60	1.67	0.08	45.87
	360	79.23	10.97	2.04	9.35	1.65	0.09	42.04
H_2SO_4	320	79.15	11.15	3.34	7.05	1.68	0.07	42.28
	330	80.90	11.10	5.45	8.56	1.63	0.08	42.79
	340	78.43	11.00	5.29	8.10	1.67	0.08	41.82
	350	78.66	11.18	4.48	8.67	1.69	0.08	42.15
	360	76.73	10.95	6.20	9.88	1.70	0.10	41.18
$CaCO_3$	320	59.44	7.33	1.87	32.40	1.47	0.41	30.25
	330	78.78	11.13	1.80	19.55	1.68	0.19	42.12
	340	78.25	10.96	2.27	10.04	1.67	0.10	41.71
	350	74.27	10.37	1.93	14.07	1.66	0.14	39.53
	360	79.72	11.15	2.72	8.11	1.67	0.08	42.46

	Biochar							
Sample	Temperature (°C)	Carbon%	Hydrogen %	Nitrogen %	Oxygen%	H/C	O/C	HHV(ii)
KOH	320	48.09	3.83	2.56	45.51	0.95	0.71	25.20
	330	56.20	4.69	3.11	36.00	0.99	0.48	26.25
	340	68.08	6.03	3.59	22.30	1.06	0.25	30.29
	350	67.48	5.38	4.08	23.05	0.95	0.26	30.66
	360	49.66	3.94	2.25	44.15	0.95	0.67	30.46
H_2SO_4	320	64.71	7.19	3.04	25.06	1.32	0.29	28.73
	330	58.66	4.63	3.94	32.77	0.94	0.42	27.98
	340	80.51	7.31	4.50	7.67	1.08	0.07	31.35
	350	59.87	5.71	3.70	30.72	1.14	0.39	31.39
	360	71.97	6.91	3.55	17.57	1.14	0.18	31.82
$CaCO_3$	320	40.43	3.48	0.75	55.43	1.03	1.03	20.89
	330	62.42	5.41	3.27	28.90	1.03	0.35	29.47
	340	61.12	5.45	2.76	30.66	1.06	0.38	28.12
	350	61.13	6.37	2.29	30.20	1.24	0.37	29.37
	360	61.65	6.18	2.67	29.51	1.19	0.36	29.96
S.A. Coal		70.40	3.95	1.61	8.05	0.67	0.09	
Crude oil		82.00	7.10	4.90	5.40	1.03	0.05	
Biomass		43.7	5.6	45.6	1.8	1.53	0.03	29.61

(i) HHV= (0.3361C+1.419H-(0.1532-0.0007O) (Zhou *et al.*,2012:2344).

(ii) Measurements as determined from the LHV obtained from the bomb calorimeter HHV=(LHV+6%).

Table C: Calorific values as tested with the bomb calorimeter and calculated HHV.

	LHV			**HHV = (LHV+6%)**		
Temperature (°C)	**KOH** ($MJ.kg^{-1}$)	**H_2SO_4** ($MJ.kg^{-1}$)	**$CaCO_3$** ($MJ.kg^{-1}$)	**KOH** ($MJ.kg^{-1}$)	**H_2SO_4** ($MJ.kg^{-1}$)	**$CaCO_3$** ($MJ.kg^{-1}$)
320	23.772	27.104	19.706	25.198	28.730	20.888
330	24.766	26.397	27.803	26.252	27.981	29.471
340	28.577	29.577	26.529	30.292	31.352	28.120
350	28.922	29.612	27.707	30.657	31.389	29.369
360	28.736	30.023	28.26	30.460	31.825	29.956
Average	26.954	28.542	26.001	28.572	30.255	27.561
15%	26.907			28.521		
25%	27.99			29.669		
Biomass	27.937			29.613		
Biodiesel	39.599			41.975		