

Preparation of bio-oil from *Scenedesmus acutus* using thermochemical liquefaction in a 1 L reactor

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Abstract

Biomass from microalgae is a potential feedstock for biofuels production. It poses no threat to food security as it does not compete with agricultural crops for arable land. *Scenedesmus acutus* was used as feedstock to produce bio-oil in a large liquefaction reactor. The influence of reaction temperature (280–360°C), reaction atmosphere (N₂ or CO₂) and solvent on bio-oil yield, C-16 fatty acid yield and oil properties were investigated. Oils were characterised using gas chromatography, Fourier transform infrared (FTIR) spectroscopy and ultimate analysis. Higher bio-oil yields were obtained in a CO₂ atmosphere (250 g.kg⁻¹ dry microalgae) than in a N₂ atmosphere (210 g.kg⁻¹ dry microalgae) whilst higher C16 fatty acid concentrations (600 g.kg⁻¹ bio-oil) were recorded in N₂ atmosphere compared to oil prepared in a CO₂ atmosphere (500 g.kg⁻¹ bio-oil). The oil yield increased to a maximum at 320°C, after which there were no significant changes. Highest bio-oil yields (425 g.kg⁻¹ dry microalgae) were obtained in ethanol as solvent. FTIR spectroscopy and ultimate analysis showed that proteins present in the feedstock were degraded by breakage of peptide linkages, and nitrogen present in the oils is peptide fragments from protein degradation. The carbon content of all produced oils was high, but the hydrogen content was low, leading to low hydrogen/carbon ratios. Energy consumption and energy efficiency calculations showed that liquefaction in both reaction atmospheres results in a net energy gain, and a CO₂ atmosphere is best for high energy efficiency.

Keywords: microalgae, atmosphere, reaction temperature, yields, C-16 fatty acid

Highlights

- Higher bio-oil yields obtained in a CO₂ atmosphere than N₂.
- N₂ atmosphere resulted in a higher C-16 content of the oil than in a CO₂ atmosphere.
- Significantly higher bio-oil yields were obtained in ethanol as solvent.
- There was a lower energy consumption rate value for oils prepared in a CO₂ atmosphere.

1. Introduction

There is a growing interest in the development of renewable and sustainable energy sources that could serve as an alternative to fossil-derived fuels. Microalgae biomass is a promising photosynthetic source of renewable energy and has several advantages as a biofuel feedstock, such as the ability to capture CO₂, high oil productivity (exceeding some of the best oil crops) and having fast growth rates. Moreover, microalgae biomass is tolerant of marginal land and thus does not compete for arable land with agricultural crops (Vlaskin *et al.* 2018; Saber *et al.* 2016; Lam and Kee, 2012). A thermochemical conversion process, such as hydrothermal liquefaction, can be used for bio-oil production from algal biomass. Bio-oil from microalgae can be upgraded into useful liquid fuels and used for extraction of valuable chemicals (Galadima and Muraza, 2018; Saber *et al.* 2016). Hydrothermal liquefaction (HTL) is a decomposition reaction carried out in a water media at a lower temperature range (200–400°C) and pressures between 5 and 20 MPa, keeping the solvent still in a liquid state, often with alkaline catalysts present. Under these conditions, bio-oil is produced, and the oil can be used for heating or improved to liquid transport fuel (Chen *et al.*, 2009). The key principle of the process is to produce oil products of increased hydrogen-to-carbon (H/C) ratios and decreased oxygen-to-carbon (O/C) ratios relative to those present in the original feedstock, and therefore high calorific values (Xu and Lancaster, 2008). Other liquefaction products are biochar and syngas; however, bio-oil has been considered here as the product of interest.

Microalgae liquefaction in batch reactors has been studied by various researchers (Vlaskin *et al.*, 2018; Wang *et al.*, 2018; Saber *et al.*, 2016; Xu and Savage, 2014) to produce bio-oil using various microalgae species, under various reaction condition and variables (i.e., reaction temperature, catalyst type, holding time and reactor volumes). In general, these studies focused primarily on the liquefaction of microalgae in a water medium and inert (N₂) atmosphere. Vlaskin *et al.* (2018) successfully obtained high bio-oil yields (44 wt.%) from the liquefaction of different samples of *Arthrospira platensis* (freshly harvest, dried and frozen samples) at 300°C, at 60 minutes holding time in small autoclaves as reactors. Wang *et al.* (2018) investigated the catalytic hydrothermal liquefaction of microalgae *Nannochloropsis* and observed maximum bio-oil yields (48,23 wt.%) using Ni/TiO₂ as catalyst at 300°C, at 30 minutes holding time. Saber *et al.* (2016) performed hydrothermal liquefaction of *Nannochloropsis sp.* carried out in a 500 ml batch reactor, at a range of temperatures (210°C; 230°C; 250°C) for 60 minutes, with the addition of a nano catalyst (Ni/SiO₂), zeolite and sodium carbonate

(Na₂CO₃), and achieved highest bio-oil yields (30 wt. %) at 250°C using nano-Ni/SiO₂ as catalyst. Xu and Savage (2014) performed the liquefaction of *Nannochloropsis sp.* (in slurry form) in mini-batch reactors at 350°C (20 minutes holding time) and obtained high bio-oil yields (44 wt.%). Other researchers (Jena *et al.*, 2011; Ross *et al.* 2010; Shuping *et al.*, 2010) have also demonstrated that reasonable oil yields with good fuel properties can be obtained from liquefaction of other algal species (*Spirulina plantensis*, *Chlorella vulgaris* and *Dunaliella tertiolecta*, respectively) using alkali catalyst (i.e., Na₂CO₃) at 350°C, 60 minutes holding time.

The dielectric constant of water changes when it is heated under pressure, which changes its solubility and reactive properties (Kruse and Dinjus, 2007; Jena *et al.*, 2011). The higher reactivity of water under sub-critical conditions can be used during hydrothermal liquefaction (HTL) to convert plant materials into solid, liquid and gaseous fractions through a combination of hydrolysis, decarboxylation, aromatisation and dehydration reactions (Marx and Piyo, 2014; Toor *et al.*, 2011). The use of HTL is advantageous in that it takes place in the water phase, and hence requires no drying of feedstock, and the higher content of lipids, proteins, fibre and carbohydrates results in higher yields of biofuels (Jin *et al.*, 2013; *et al.*, 2008; Biller and Ross, 2011).

Bio-oil is an oxygen-rich, dark, viscous, crude oil-like liquid obtained as one of the products of HTL. It has been shown to contain phenols (Akhtar and Min, 2011), water soluble organic acids (Kruse *et al.*, 2013), n-alkanes and polyaromatic hydrocarbons (Brown *et al.*, 2010). Bio-oil yields are directly related to the lipid content in the feedstock (Jin *et al.*, 2013). The glycerol backbone of lipids present in microalgae can be converted to several hydrocarbon components that will be distributed in the water and organic fractions of the oil as well as the gas phase (Barreiro *et al.*, 2013). The lipids form fatty acids as part of the oils fraction, and, in the presence of methanol or ethanol as solvent during thermochemical liquefaction, the fatty acids can be converted to methyl or ethyl esters (Huang *et al.*, 2011; Yuan *et al.*, 2011). Proteins in the feedstock are hydrolysed to amino acids, which undergo decarboxylation to ammonia and organic acids, which in turn can further be converted through polymerisation to form a wide variety of nitrogen-containing hydrocarbons (Toor *et al.*, 2011). Carbohydrates degrade mostly to water soluble fractions, but some of the smaller fractions can repolymerise to form heavier hydrocarbons that will contribute to the bio-oil fraction (Barreiro *et al.*, 2013, Biller and Ross, 2011). The lignin fraction of the biomass contributes to the biochar fraction from thermochemical liquefaction (Marx and Piyo, 2014).

The characteristics of the crude bio-oil depend on the characteristics of the biomass feedstock (Shuping *et al.*, 2010) as well as the processing conditions (temperature, pressure, reaction time, solvent, reaction atmosphere) (Barreiro *et al.*, 2013). Many studies have reported the influence of temperature (200–380°C) on the bio-oil yield and composition (Jena *et al.*, 2011; Huang *et al.*, 2011; Duan *et al.*, 2013; Jin *et al.*, 2013). Liquefaction begins at temperatures above 280°C (depending on the feedstock), and below this temperature only water-soluble components, extractives, and volatile organics are obtained (Kruse *et al.*, 2013). After the onset of liquefaction, the bio-oil yield increases up to a maximum temperature (usually around 360–380°C), after which a decrease in yield is observed as the temperature exceeds the supercritical temperature of water. The unique solvent properties of supercritical water benefit the formation of lighter organic fractions through cracking, condensation, and dehydration of longer oil molecules (Duan *et al.*, 2013; Barreiro *et al.*, 2013), causing a decrease in oil yield.

Most studies on the influence of reaction holding time on the bio-oil yield and composition have observed an initial increase in bio-oil yield with an increase in holding time from 5 to 30 minutes followed by a decrease in oil yield at longer holding times (Zou *et al.*, 2009; Shuping *et al.*, 2010; Akhtar and Amin; 2011; Li *et al.*, 2012). During liquefaction, longer holding times are necessary at low temperatures and shorter holding times at high temperatures to achieve the same bio-oil yield (Barreiro *et al.*, 2013). The composition of the bio-oil is dependent on the holding time (Jin *et al.*, 2013). The decrease in bio-oil yield at longer holding times have been attributed to repolymerisation and cracking reactions to form gas and biochar (Zou *et al.*, 2010; Jin *et al.*, 2013), decomposition of bio-oil (Li *et al.*, 2012; Akhtar and Amin, 2011), and loss of light fractions from the bio-oil during the bio-oil recovery steps (Zhou *et al.*, 2010). Alkali metal earth catalysts (NaOH, KOH, Na₂CO₃) have been reported to suppress biochar formation during liquefaction, but this does not increase the bio-oil yield significantly (Huang *et al.*, 2011; Shuping *et al.*, 2010). Biller and

Ross (2011) showed that Na₂CO₃ selectively increased the oil yield from carbohydrates, but not from proteins. The addition of an alkali catalyst increases the light water-soluble fractions and gas fractions, which could lead to decreased bio-oil yields (Li *et al.*, 2012; Anastasakis and Ross, 2011).

Li *et al.* (2012) suggested that the ash content of the feedstock influences the effect the addition of an alkali catalyst will have on the bio-oil yield from microalgae. Li *et al.* (2012) found that an increase in biomass to water ratio leads to an increase in overall product yield, but a decrease in bio-oil yield during the liquefaction of marine microalgae. Akhtar and Amin (2011) suggested that the hydrothermal liquefaction process behaves like pyrolysis at high biomass to water ratios. Jena *et al.* (2011) reported an increase in bio-oil yield with an increase in biomass to water ratio from 10% to 20% during the liquefaction of microalgae.

Many different micro and macro algae species have been investigated for bio-oil production. In this study the influence of temperature, reaction atmosphere and solvent on bio-oil yields and characteristics during the thermochemical liquefaction of *Scenedesmus acutus* in a one-litre reactor was investigated. To the author's knowledge, this is the first study to evaluate the liquefaction of *Scenedesmus acutus* as feedstock for bio-oil production and in a large reactor.

2. Materials and methods

2.1 Materials

Scenedesmus acutus used in this study was obtained from the Institute of Chemical Technology at the Nelson Mandela University (33°59'54.88" S 25°40'19.90" E) in dry powder form. The microalgae powder was kept in airtight containers at room temperature for the duration of the study. Nitrogen (N₂) (ultra high purity) and carbon dioxide (CO₂) (technical grade) were obtained from Afrox gas company. Solvents used in this study were all of 99% purity and were obtained from ACE chemical company. Solvents were used as received without prior purification. Some properties of solvents used in this study are listed in Table 1.

Table 1: Some properties of solvents used in this study.

Solvent	Density (kg.m ⁻³) ^a	Boiling point (K) ^b	Critical temperature (K)	Critical pressure (MPa)	Dielectric constant	Hydrogen atoms
Water (distilled)	1000	373	647	22	78.54	2
Methanol	791	338	512	6.4	32.6	4
Ethanol	789	352	514	8.1	24.6	6
Iso-propanol	785	277	508	4.8	18.3	8
Acetone	791	329	508	4.8	20.7	6

Notes: a. At standard temperature and pressure (STP). b. At sea level.

2.2 Experimental setup and procedure

Liquefaction experiments were carried out in a standard SS316 stainless steel high-pressure autoclave (Sawayama *et al.*, 1995; Dote *et al.*, 1996). The autoclave has a working volume of 950 mL, an inside diameter of 90 mm, and a height of 150 mm, and is equipped with a high-pressure stirrer and heated with an electrical heating jacket. In each experiment, a desired amount of *Scenedesmus acutus*, distilled water (or alcoholic solvent) and potassium hydroxide (KOH) were fed into the autoclave. The dosage of KOH was equal to 5 wt.% of microalgae. In all experiments, the autoclave was agitated using a magnetic stirrer drive at 720 rpm speed, which was set by a variable speed controller to ensure homogeneous reactions. After the completion of the experiment, the heating jackets were removed, and the autoclave could cool to room temperature. Following liquefaction, chloroform was used to dissolve all organic compounds in the crude extract in the autoclave whilst stirring. The mixture was vacuum filtered using Whatman no. 3 filter paper to remove solid residues. A separating funnel was used to separate the aqueous and organic phases. The organic phase containing the oil extract was decanted into a pre-weighed round-bottom flask and the flask was used in a vacuum distillation set-up to evaporate the chloroform from the oil extract at 70°C. The round-bottom flask containing the purified oil sample was then weighed to determine the oil weight by mass difference. The influence of temperature was investigated by varying the reaction temperature from 280°C to 360°C with 20°C intervals. Experiments were conducted in either a N₂ or a CO₂ atmosphere.

2.3 Calculation of energy and mass balance parameters

The bio-oil yield was calculated as the ratio between the mass of dry bio-oil obtained and the mass of dry microalgae powder used in the experiment. An energy efficiency parameter (ER) and an energy consumption rate (ECR), as defined by Jena *et al.*, (2011), were calculated according to Equations 1 and 2 (Minowa *et al.*, 1995).

$$ER = \frac{\text{Energy content of bio-oil}}{\text{Energy content of algae}} - \frac{\text{Mass of dry bio-oil}}{\text{Mass of dry feedstock}} \times 100 \quad (1)$$

$$ECR = \frac{x_{\text{water}} C_{p,\text{water}}(T_{\text{HTL}} - T_{\infty}) + (1 - x_{\text{water}}) C_{p,\text{algae}}(T_{\text{HTL}} - T_{\infty})}{rYH_0} \quad (2)$$

where ER is the liquefaction energy efficiency, ECR is the energy consumption, Y is the bio-oil yield, and

H₀ is the higher heating value of *Scenedesmus acutus*. The value of r was taken to be 0.6 for calculation purposes (Jena *et al.*, 2011). X_{water} is the fraction of water in the liquefaction reactor, T_{HTL} is the reaction temperature and C_{p, water} and C_{p, microalgae} are the heat capacities of water and microalgae, respectively. The heat capacity of microalgae was taken to be equal to that of water (4.18 kJ.kg⁻¹.k⁻¹) due to the low biomass loadings used in this study.

The higher heating values (HHV) were calculated according to Beckman's equation (Equation 3) (Channiwala *et al.*, 2002).

$$HHV \text{ (MJ/kg)} = 0.352 C + 0.944 H + 0.105 (S - O) \quad (3)$$

2.4 Analytical methods

An Agilent 7890 GC equipped with an Agilent 7683B auto-injector, a HP-5 column (100 m × 320 μm × 0.25 μm) and a flame ionisation detector were used to determine the composition of bio-oil obtained during liquefaction. The bio-oil was methylated into the methyl esters by a trimethyl sulfonium hydroxide solution. The amount of fatty acid from the bio-oil was determined through standard calibration curves and the amount of fatty acid yield (wt. %) was calculated as the ratio of the mass of measured fatty acid to the initial mass of *Scenedesmus acutus* used. The conditions under which the oils were analysed on the gas chromatography are summarised in Table 2.

Table 2: Operating conditions for gas chromatography analyses

Inlet temperature	250°C
Injection volume	1.0 μL
Oven programming	175°C for 10 min; 210°C for 5 min; 230°C for 5 min
Detector flame ionisation detector	350°C
H ₃ flow rate	40 ml/min
Air flow rate	400 ml/min
Make up helium	1.0 ml/min

FTIR analyses were used to determine the main organic constituents of the bio-oil samples, based on the absorption peaks of the functional groups present in the bio-oil. The bio-oil was applied as a droplet to a potassium bromide pellet and 30 scans were obtained and processed on a Bruker spectrometer. The carbon (C), hydrogen (H), nitrogen (N), sulphur (S) and oxygen (O) present in the extracted bio-oil were determined by a FLASH organic 2000 elemental analyser.

3. Results and discussion

3.1 Bio-oil yield and composition

The influence of reaction temperature on the total bio-oil yield at different reaction atmospheres is shown in Figure 1. The graph shows that the bio-oil yield stabilised rapidly in a CO₂ atmosphere with only an insignificant increase in oil yield with an increase in temperature, whereas in a N₂ atmosphere, a steady increase in oil yield was observed and peaked at 320°C. A similar trend was observed by Wang *et al.* (2018), who noted an increase in bio-oil yield that reached a peak at 300°C and no significant increase was observed beyond that temperature. In contrast, Jena *et al.* (2011) observed an increasing trend in oil yields with an increase in reaction temperature (from 200°C to 350°C). At subcritical temperatures, the unique ionic properties of water allow for the hydrolysis of a wide range of compounds (Shuping *et al.*, 2010). As the temperature is increased, the free radical fractions formed during hydrolysis start to repolymerise to form biochar and/or gas.

The role of the reaction atmosphere during liquefaction is to stabilise the free radical fragments and inhibit polymerisation reactions from taking place, thereby increasing the oil yield. This was observed in this study, with higher bio-oil yields obtained with CO₂ than with N₂ as atmosphere. Lower bio-oil yields in a N₂ atmosphere could be attributed to the low reactivity of the N₂ comparatively (Akhtar and Amin, 2011). Liquefaction in a CO₂ environment is thus conducive to the formation of bio-oil, as it promotes the deoxygenation and hydrodeoxygenation pathways for the degradation of the biomass. The ultimate analysis will also show the lower oxygen content of bio-oil prepared in a CO₂ atmosphere compared to the oxygen content of the

original biomass and bio-oils prepared in a N₂ atmosphere.

Palmitic fatty acid (C-16:0) was the main fatty acid detected in the prepared bio-oils. The influence of reaction temperature and atmosphere on the Palmitic (C-16:0) lipid content of the prepared bio-oils is shown in Figure 2.

As shown in Figure 2, C-16 fatty acid content increases as temperature increases, and a N₂ atmosphere resulted in a higher C-16 content of the oil than did a CO₂ atmosphere. The high concentrations of C-16 present in the bio-oils supports the notion that cellulose and hemicellulose in the plant material degrade to form part of the bio-oils. As the temperature is increased, the ionic nature of the solvent is increased, leading to increased hydrolysis. C-16 fatty acids are formed during the hydrolysis stage, prior to depletion of hydrogen that supports the ongoing formation of free radical fragment. CO₂ acts as a stabiliser for the free radicals and inhibits the formation of heavier fragments that could condensate to the biochar fraction. Consequently, higher yield of C-16 fatty acid concentrations is obtained in a N₂ atmosphere, as the hydrolysis products are less stable than in a CO₂ atmosphere.

The role of the solvent in thermochemical liquefaction is to dissolve the biomass and act as a hydrogen donor for the hydrolysis reactions (Baloch *et al.* 2018; Singh *et al.* 2015). The bio-oil obtained from different feedstocks using different solvents during liquefaction is determined to a large extent by the properties of the solvent as well as the process parameters. The influence of different solvents on the bio-oil yield during thermochemical liquefaction of *Scenedesmus acutus* was investigated using different solvents in a CO₂ atmosphere. The results are shown in Figure 3.

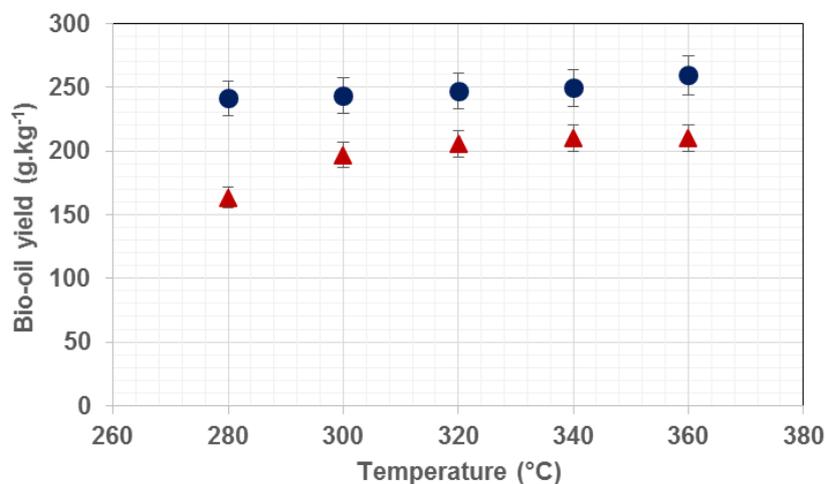


Figure 1: Effect of reaction temperature on the total bio-oil yield at a biomass loading of 60 g.kg⁻¹ in water and a KOH loading of 50 g.kg⁻¹ (▲ – N₂ atmosphere, ● – CO₂ atmosphere).

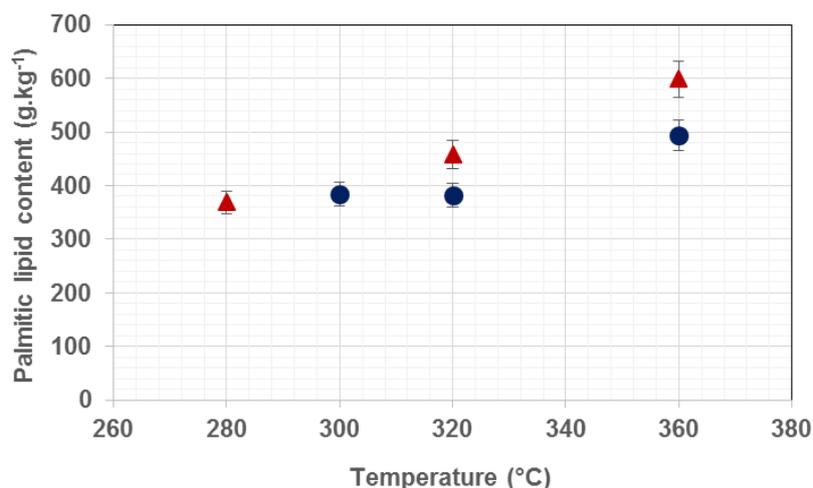


Figure 2: Effect of reaction temperature on the C-16 fatty acid content at a biomass loading of 60 g.kg⁻¹ in water and a KOH loading of 50 g.kg⁻¹ (▲ - N₂ atmosphere, ● - CO₂ atmosphere).

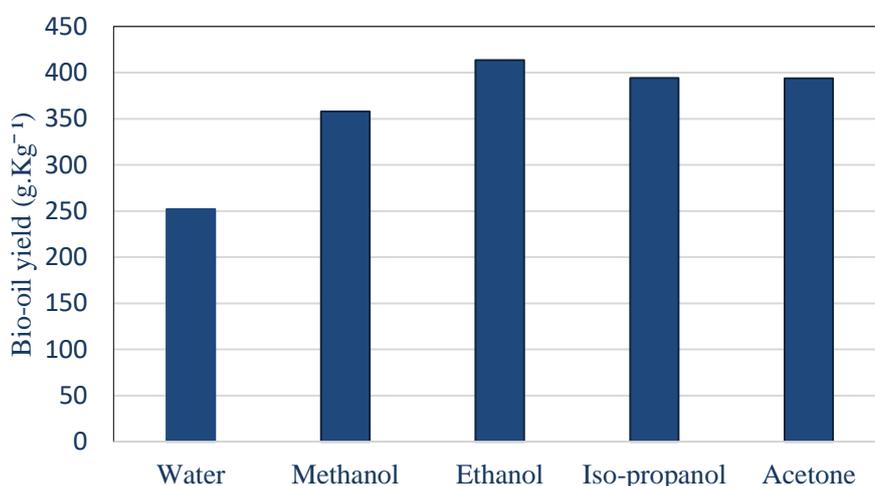


Figure 3: Effect of solvent type on the bio-oil yield at a constant biomass loading of 60 g.kg⁻¹ at 360°C.

As shown in Figure 3, significantly higher bio-oil yields were obtained in ethanol as solvent than in any of the other solvents investigated. Water as solvent resulted in the lowest bio-oil yield. These results are consistent with findings by Baloch *et al.* (2018). Solvents act as hydrogen donors and, as such, the presence of active hydrogen from the solvent plays a role in further stabilisation of liquefaction intermediates, thereby inhibiting them from forming compounds that are more difficult to decompose, leading to high bio-oil yields (Singh *et al.*, 2018). Comparing the properties of the various solvents, all solvents except water were in a supercritical state during liquefaction, and this explains the much lower yield obtained in water as solvent. A comparison of the different listed properties of the solvents (see Table 1) showed that there is no clear relationship between the bio-oil yields and most of

the properties, although the di-electric constant and the number of hydrogen atoms in the molecule has a weak linear relationship with the bio-oil yields. Hydrogen from the solvent and the polarity of the solvent both play a role in the initiation of solvolysis and hydrolysis and it thus makes sense that there should be some relationship, albeit weak, between these properties and the amount of oil formed. The polar nature of the solvent during liquefaction can both sustain fragmentation as well as stabilise fractions to produce more oil.

3.2 Compositional analysis of oils

3.2.1 FTIR analysis

The main functional groups and the related classes of compounds present in the *Scenedesmus acutus* feedstock and the bio-oil are represented in Table 3.

Table 3: Identification of prominent peaks on FTIR spectra of bio-oils prepared at 360°C in either nitrogen or carbon dioxide atmosphere.

Wave no.	Characteristic vibration	Reference
3150–3500	Weak C-H stretching associated with carbohydrates in oils (only present in oils)	Parshetti <i>et al.</i> , 2013; Jin <i>et al.</i> , 2013
3325, 3450, 3500	Primary amines and C-N stretching associated with peptides in microalgae (only present in feedstock)	Huang <i>et al.</i> , 2011; Jena <i>et al.</i> , 2011
2850, 2925	C-H and O-H stretching associated with methyl and methylene alkane groups (present in both oil and feedstock)	Jin <i>et al.</i> , 2013; Duan and Savage, 2011
2300–2400	Aliphatic –C-H and C=O stretching associated with fatty acids (only present in oils)	Yuan <i>et al.</i> , 2011
1700, 1725, 1650, 1600	–C=O and –C-O stretching associated with fatty acids (present in both oils and feedstock)	Duan and Savage, 2011; Shuping <i>et al.</i> , 2010
1550, 1300	Polypeptides (only present in feedstock)	Huang <i>et al.</i> , 2014
1515	–C-H stretching associated with cellulose and polar compounds (only present in oils)	Jena <i>et al.</i> , 2011
1400	–C-H ether vibrations associated with oils (only present in oils)	Das <i>et al.</i> , 2009
1377	–C-N associated with fractions from peptide linkages (only present in oils)	Yuan <i>et al.</i> , 2011
1270	–C-O ether stretching associated with oils and fats (only present in oils)	Huang <i>et al.</i> , 2014
1050	Si-O-C organic silicone associated with cell walls (only present in feedstock)	Giordano <i>et al.</i> , 2001
940–950	–C-H stretching of alkyl substituted compounds (only present in oils)	Duan <i>et al.</i> , 2013
550	Aliphatic iodol compounds (only present in feedstock)	Yang <i>et al.</i> , 2014; Gómez-Jacinto <i>et al.</i> , 2010
400–500	–S-S stretching of polysulfides (only present in feedstock)	Barreiro <i>et al.</i> , 2014

The C-N and primary amine peaks at 3325, 3450, 3500, 1550 and 1300 cm^{-1} associated with proteins and polypeptides in the feedstock is not present in the bio-oil spectra, and a peak at 1377 cm^{-1} that is associated with peptide fraction is only present in the bio-oils. This indicates that the proteins present in the microalgae feedstock were completely broken down through the breakage of peptide linkages, leading to the –C-N fractions in the oils (Yuan *et al.*, 2011). This is confirmed by ultimate analysis (in Table 4) that showed high concentrations of nitrogen in the feedstock, but less in the bio-oils. Weak aliphatic methyl and methylene peaks at 2850 and 2925 cm^{-1} in the feedstock are present as prominent peaks in the oils. This is linked to –CH₃ groups attached to fatty acid chains. Single peaks between 1600 and 1700 cm^{-1} in the feedstock become broader, stronger peaks in the bio-oil. These peaks are associated with the –C=O and –C-O vibrations of fatty acids. There are some oils present in the microalgae feedstock as C-16 fatty acids, but as the organic material is liquefied

and some fatty acid methyl esters are forming, the peaks become more prominent. The fatty acids present in the bio-oils are also evidenced by the peaks at 2300–2400, 1400, 1270 and 940–950 cm^{-1} . The presence of organic iodine in microalgae have been reported (Yang *et al.*, 2014; Gómez-Jacinto *et al.*, 2010). In this study, the presence of an iodide compound in the microalgae was observed at a wave number of 550 cm^{-1} . The oils were not analysed for iodine presence in this study, so it is not clear if the iodine was distributed to the oils or the chars. Polysulfide stretching was observed at wave numbers of 400–500 cm^{-1} in the feedstock, but not in the oils. This could indicate the organic sulphur present in the microalgae was removed as SO_x during liquefaction (Barreiro *et al.*, 2014). Silicone normally associated with cell walls was observed at a wave number of 1050 cm^{-1} in the feedstock but was not present in the bio-oil spectra. This is an indication that cell walls were completely broken down during the liquefaction process. Comparison of the spectra of the feedstock and the oils thus confirmed that

plant cell walls were broken down completely and organic matter was liquefied to fatty acids (primarily C-16) and gas components containing SO_x and NO_x.

3.2.2 Ultimate analysis

The ultimate analysis of the raw microalgae and the bio-oil product was conducted to evaluate the extent of deoxygenation in the liquefaction of *Scenedesmus acutus* and the HHV of the bio-oil products. The elemental composition of the raw microalgae and of the bio-oil are presented in Table 4. The elemental composition of the bio-oil shown were obtained at 360°C, at a 60 g.kg⁻¹ biomass loading in water under N₂ and CO₂ atmospheres. The HHV, energy efficiency (ER) and energy consumption (ECR) parameters for each of the oils are presented in Table 5.

In Table 4, it can be observed that the oxygen and sulphur contents were lower in the bio-oil than in the raw microalgae; this is an indication that deoxygenation and desulphurisation of the bio-oils took place and agrees with the FTIR compositional analysis that showed the absence of sulphur peaks in the oils. The lower nitrogen content in the oils compared to the feedstock agrees with the FTIR analysis. The carbon content of the oils was significantly improved compared to the microalgae feedstock, but the H/C ratio was not significantly increased. From Table 5, the HHV of both oils are significantly higher than that of the feedstock. The energy efficiency calculations showed that more energy of the original feedstock was retained in the oils prepared in a CO₂ atmosphere than in those prepared in a N₂ atmosphere. The energy consumption ratio for oils prepared in both atmospheres was below 1, indicating net energy being produced by the liquefaction process. The lower ECR value for oils prepared in a CO₂ atmosphere showed that CO₂ is better for bio-oil production than N₂, even though the HHV of the oils produced in N₂ were slightly higher.

4. Conclusions

Scenedesmus acutus was successfully degraded in a one-litre liquefaction reactor. Both carbohydrates and fats were converted to a C-16 fatty acid rich oil. It was shown that not much is gained in terms of bio-oil yield by increasing the liquefaction temperature beyond 320°C, indicating that the liquefaction process is essentially finished at 320°C. Higher bio-oil yields were obtained in a CO₂ atmosphere than in a N₂ atmosphere, but higher C-16 fatty acid yields were obtained in a N₂ atmosphere. Higher bio-oil yields were obtained when using ethanol as solvent compared to methanol, iso-propanol, or acetone. Deoxygenation and hydrodeoxygenation pathways were confirmed with Fourier transform infrared spectroscopy and ultimate analysis and it was shown that all oils produced had a high carbon content. The low hydrogen content in the original microalgae feedstock and the oils resulted in low hydrogen/ carbon ratios. However, high yield of C-16 fatty acid could make the upgrading of oils to improve the hydrogen/carbon ratio an economical prospect. Overall, it has been demonstrated that thermochemical liquefaction results in positive energy balance and that a CO₂ atmosphere is ideal for high oil yields and high energy efficiency.

Author contributions

H. Baloyi: Primary research, data analysis and analytical discussions, and preparing final article.

S. Marx: Project supervision, technical editing and review of the draft paper prepared from an original mini dissertation.

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Table 4: Elemental composition of the raw microalgae and bio-oil

	wt.% C	wt.%H	wt.% N	wt.% S	wt.%O	H/C	O/C
Raw microalgae	43.92	6.95	6.44	0.53	20.85	0.16	0.47
N ₂ atmosphere	67.70	8.33	5.26	0.00	7.91	0.12	0.11
CO ₂ atmosphere	62.30	7.04	3.10	0.03	3.71	0.11	0.06

Table 5: Higher heating values (HHV), energy efficiency (ER) and energy consumption (ECR) for feedstock and bio-oil prepared at 360°C.

	HHV (MJ.kg ⁻¹)	ER	ECR
Raw microalgae	19.89	-	-
N ₂ atmosphere	30.86	33	0.56
CO ₂ atmosphere	28.19	35	0.47

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