ORIGINAL PAPER

TITLE: Co-liquefaction of hypersaline *Tetraselmis* sp. microalga and cow manure for biocrude production and as means for solid waste treatment

Abstract

This paper investigated hydrothermal coliquefaction of the microalga *Tetraselmis* sp. and cow manure at different mix ratios and the characterisation of produced biocrude. The carbon and nitrogen balances across the reactor and energy recovery were also elucidated. The study was conducted using a 1L batch reactor at 300^{o}C and 350^{o}C at constant reaction time of 10min using ~16w/v% solids loading. The results showed that irrespective of reaction temperature, there were substantial influence on yield and properties of biocrude. Importantly, there were up to 60% reductions in nitrogen content of biocrude, which could be due to synergistic effect from interactions of feedstock molecules during liquefaction. These findings suggest blending of *Tetraselmis* sp. and cow manure could improve biocrude quality, while simultaneously treating waste.

1. Introduction

Hydrothermal liquefaction (HTL) of microalgae biomass to biocrude is a promising process for biofuel production (Huang *et al.*, 2019). Importantly, HTL avoids use of energy- intensive in drying of feedstock, as required in other thermochemical methods such as pyrolysis (Eboibi et al., 2014). However, due to current challenge in production cost of microalga, its treatment in a single-stream could negatively impact future commercialization of the process (Brilman *et al.*, 2017; Giaconi *et al.*, 2017). In addition to improving yield and quality of biocrude, presence of heteroatoms such as nitrogenous and oxygenated compounds in biocrude have remained an issue. Thereby inducing replacing conventional fossil crude in nearest future. Therefore, more scientific research on suitable feedstock selection as co-feeds alongside algae liquefaction and means of reducing heteroatom's is necessary.

Due to potential synergistic impact on yield and quality of biocrude, low logistics costs for feedstock collection and transportation, it is believed that co-liquefaction of different organic biomass in a single-stream is advantageous compared to liquefaction of individual feedstock (Yang *et al.*, 2019). Co-liquefaction of algae and solid organic waste, in this instance animal manure has been suggested to enhance economic sustainability of HTL-algae-biofuel (Giaconi *et al*., 2017; Lam *et al*., 2019).Importantly, co-liquefaction has the potential to enhance yield and properties of biocrude through adjusting the biochemical composition of feedstock mixtures (Yang *et al*., 2017). This would not only improve the yield and quality of biocrude (Xu *et al*., 2019;Yuan *et al*., 2019) but also addresses issues relating to handling and disposal of manure (Eboibi *et al*., 2015). As manure management has been a concern to agro industries (Usapein and Chavalparit, 2017).

Approximately 921m metric tons of wet manure was produced from 77.6m animal units of cattle in the United States (USDA, 2007). Although, manure is applicable on fields as traditional/or suitable methods for manure management, increases in urbanization and strict environmental policies seems to make this option limited (Saba *et al*., 2018). As this option may lead to increase in greenhouse gas and particulate emissions (Baldé *et al*., 2016). Manure contains essential nutrient (such as phosphorus and nitrogen), its improper management could lead to run offs, affecting water quality (such as eutrophication),public health and surrounding ecosystems (Cantrell *et al*., 2017; Sharpley, 1981).Therefore, co-liquefaction of microalga and animal manure has broad application potentials; provide resource recovery, reduction in carbon footprint, and in waste treatment to mitigate environmental pollution (Huang *et al*., 2019; Wu *et al*., 2017).

A review of the scientific literature showed limited study on co-liquefaction of microalgae and cow manure, though there has been a reported study on individual liquefaction of cow manure and algae. Few studies have evaluated the co-liquefaction of microalgae with other organic biomass as co-feedstock. These include co-liquefaction of microalgae and; microalgae(Jin *et al*., 2013); agricultural waste (Chen *et al*., 2019; Wang *et al*., 2019); sewage sludge (Xu *et al*., 2019); polypropylene (Wu *et al*., 2017) and the use of mixed algae strains (Dandamudi *et al*., 2019; Hietala *et al*., 2019) and model compounds (Feng *et al*., 2019; Zhang *et al*., 2016). The results of these studies are contradictory, generally; the effects are either synergistic when combined feeds yields are higher than individual feed (Gai *et al*., 2015; Xu *et al*., 2019) or antagonistic (Brilman *et al*., 2017; Chen *et al*., 2019) when reverse is the case. Also, the elemental carbon and nitrogen content distributions are inconsistent (Yang *et al*., 2019). This development therefore suggests more scientific research investigation.

In addition, previous studies used pulverised algae and combined feeds for their experimental studies. Such practices are may be acceptable at laboratory scale unlike in commercial scale. The properties and structure of the feeds may be altered prior to liquefaction, which may affect output and quality. The use of freshly harvested microalgae seems to present real-life scenario unlike pulverised algae. Therefore the main aim of present study is to fill the knowledge gap.

2. Materials and method

2.1 Materials

Freshly harvested hypersaline *Tetraselmis* sp. alga (TA), and cow manure (M) was used in the present study. TM was grown and cultivated in outdoor open raceway ponds. Its cultivation and harvesting has been reported elsewhere (Isdepsky1and Borowitzka1, 2019; Fon-Sing *et al*., 2014). Cow manure (CM) was obtained from a local farm at Thandallam, Chennai, India.

2.2. Methodology

HTL experiment were conducted using a 1L batch high-pressure reactor made of Inconel at reaction temperature of 300°C and 350°C at constant reaction time of 15min, using biomass feedstock containing ~20%w/w solids. Typically, for individual run, 500g of either TA or CM was loaded in the reactor. For co-liquefaction studies, TA and CM were mixed in ratio 04:01, 03:02, 01:01, 02:03 and 01:04. The production and separation procedures were carried out in accordance with previous reports (Eboibi *et al*., 2014; Wang *et al*., 2019). For repeatability, each experimental run was carried out in triplicate, and the average result reported.

2.3. Analysis

The elemental and biochemical composition of TA and CM, including that of previous studies are shown in Table 1. A Variol III Elemental Analyser System GmbH was used to determine the weight percentages ofcarbon (C), hydrogen (H), nitrogen (N) and sulfur (S) following ASTM D-5291 method. The oxygen content was estimated by subtraction from the combined mass of CHN and S.

After product separation, the primary product biocrude yield was determined in wt% on an ash free dry wt. basis. The solid residue and dissolved aqueous solids were estimated in weight percent by relating their mass yield to that of feed loaded in the reactor (Theegala and Midgett, 2012). The gas phase yield was estimated by difference by subtracting the combined mass yield of biocrude, solid residue and dissolved aqueous solids from unity.

Based on the data from the elemental analysis, the higher heating value (HHV) was estimated using Eq. (1), proposed by Chinnawala and Parikh, (2002), while the molar atomic ratios of H/C, O/C N/C were estimated in accordance to previous reports (Alba *et al*., 2012; Eboibi *et al*., 2019).

$$HHV\ \left(\frac{MJ}{kg}\right) = 0.3491C + 1.1783H + 0.1005S - 0.1034O - 0.0151N - 0.0211A \qquad (1)$$

where C represents carbon, H hydrogen, S sulfur, O oxygen, N nitrogen and A ash, on dry basis.

The amount of energy recovered (ER. %) was estimated using Eq. (5).

$$ER\ = \frac{HHV\ \left(\frac{MJ}{kg}\right) of\ biocrude * weight\ of\ product\ \ (g)}{HHV\ \left(\frac{MJ}{kg}\right)\ of\ feed * weight\ of feed\ (g)} \times 100\% \qquad (5)$$

It should be noted that the external work applied for heating the reactor was not considered in Eq. (2), but by relating the HHVs and mass of biocrude to that of initial biomass load fed to the reactor (Biller and Ross, 2011). The C and N recovery (%) was calculated by using the elemental mass balances across the reactor (Eboibi *et al*., 2014).

Table 1: Biochemical and elemental composition of feedstocks

Feedstock	Biochemical composition						Elemental composition							Reference
	Carbohydrate	Proteins	Lipids	Cellulose	Hemicellulose	lignin	C	H	N	S	O	H/C	HHV	
Tetraselmis sp.[a]	22	58	14	-	-	-	42	6.8	8	3	40.2	1.94	18.3	Present study
Cow manure[b]	58	11	15	-	-	-	35.6	6.5	2.1	0.6	55.2	2.19	14.3	
Spirulina plantensis	23.9	70.3	5.8	-	-	-	46.9	6.9	10.7	NR	35.5	1.77	18.7	Feng *et al*., (2019)
α-Cellulose	-	-	-	99.6	-	-	44.4	6.1	0	NR	40.3	1.67	16.0	
Nannochloropsis sp.	12.4	36.4	19.0	-	-	-	49.2	7.2	6.2	NR	36.3	NR	20.5	Zhang *et al*., (2016)
Spirulina plantensis	10.8	48.5	5.8	-	-	-	45.6	6.6	10.	NR	36.7	NR	18.4	
Cyanidioschyson merolae	37.8	47.8	4.3	-	-	-	48.1	5.1	9.9	1.2	35.5	NR	18.1	Dandamudi *et al*., (2017)
Galdiera sulphuraria	42.2	45.1	3.2	-	-	-	42.4	3.9	9.4	1.3	42.9	NR	16.4	
Spirulina sp.	29.3	26.8	11.0	-	-	-	34.5	5.1	3.4	NR	24.2	NR	NR	Jin *et al*., (2013)
Entermorpha prolifera	18.2	23.8	5.0	-	-	-	28.0	4.5	3.8	NR	10.7	NR	NR	
Chlorella pyrenoidosa	20.6	57.6	10.6	-	-	-	47.2	8.3	8.9	0.3	NR	NR	22.6	Chen *et al*., (2019)
Sweet potato residue	20.7	1.7	NR	25.9	12.7	34.5	47.2	7.2	0.2	NR	40.5	NR	20.4	

[a] Eboibi *et al*., (2014)

[b] Eboibi *et al*., (2015)

Results and discussion

3.1. Mass yields

The yields obtained from HTL of individual and combined feed is presented in Figure 1. The treatment at 300^{o}C (Fig. 1a), with 10min reaction time using 20% solid loading led to 32 to 42, 15 to 24, 12 to 15 and 21-34wt% biocrude, solid residue, dissolved aqueous solids and gas phase yields, respectively. An increase in reaction temperature from 300^{o}C to 350^{o}C (Fig. 1b), led to substantial increase in biocrude yield when compared to that obtained at 300^{o}C. For individual treatment, biocrude from TA increased from 42wt% to 58wt%, and from 32wt% to 42wt% for CM. In contrast, the solid residue reduced from 24wt% to 16wt% and 14wt% to 10wt% for TA and CM, respectively. This variation in yields with respect to reaction temperature could be attributed to promotion of decomposition of reactants including polymerisation of intermediates (Wu *et al.*, 2017).Similar trend was found for the combined feeds; as there were general reduction in solid residue from 24wt% to 13wt%, and 15wt% to 11wt% for dissolved aqueous solids. However the gas phase yields increased numerically at all conditions. The relative increase in biocrude yields from co-liquefaction could be due algae containing alkali salts. Alkali salt is known to enhance biomass macromolecules degradation into biocrude. Hence, blending of TA with biomass materials with or without alkali metals would improve biocrude yields.

The low biocrude yield and relatively high solid residue yields obtained at 300^{o}C compared to at 350^{o}C could be due to insufficient conversion of the feedstock. It is possible some bonds were unbroken due to perhaps inadequate reaction temperature and or reaction time. In addition, the biochemical composition (protein, carbohydrate and lipids) of the feedstocks could have influenced the yields. Microalgae containing higher lipids potentially produce higher biocrude and lower solid residue yields unlike algae containing higher carbohydrates. Consequently, coliquefaction of algae with high lipids and with other biomass of higher carbohydrate would lead to lower biocrude yield (when compared to HTL of individual algae) and higher yield (when compared with HTL of the other biomass with higher carbohydrate component).Carbohydrate has been reported to contribute little to the overall bio-crude yield, and at the sometime neutralizes the negative effect of protein and enhance the performance of HTL by the Maillard reaction at an optimal ratio (Zhang *et al.*, 2016).

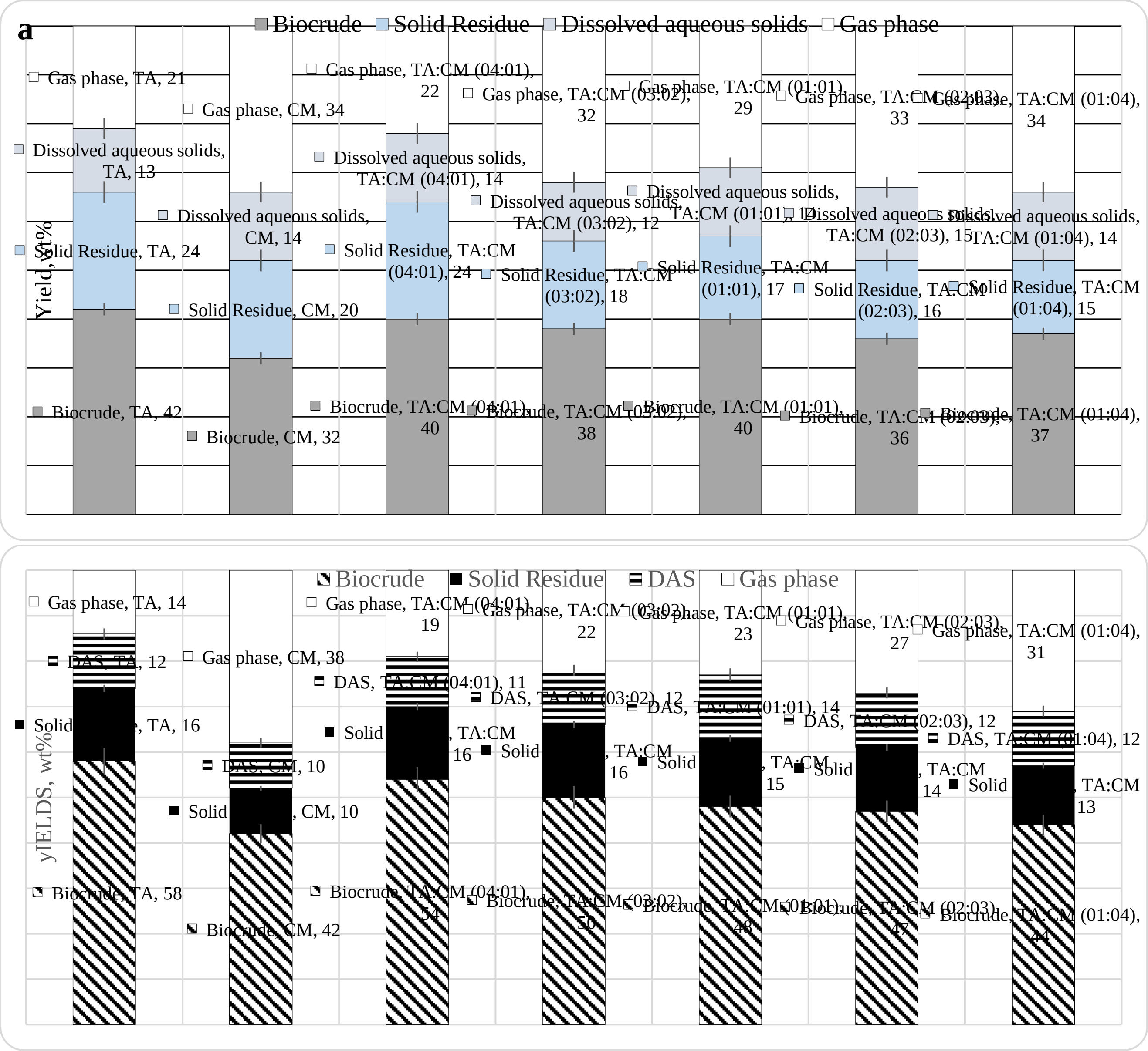

Fig. 1: Mass yields from co-liquefaction of *Tetraselmis* sp. alga (TA) and cow manure (CM) at 10min constant reaction time. A: 300^{o}C, B: 350^{o}C.

This therefore suggests that for optimal biocrude yield, the composition of feedstocks needs to be varied, in order to achieve a suitable feedstock mix ratio. This study has shown that blending different feedstock have substantial positive effects on distribution and potentially enhances the biocrude yield. This is advantageous of using all kinds of available biomass wastes (Yuan *et al*., 2019) for biofuel production. Although, there are limited data on the synergistic effect (SE) on co-liquefaction of algae and cow manure, a SE of -4.2 to ~7 was found and which is within the range of previous related studies (shown in Table 2). Yang *et al*., (2019) reported that co-liquefaction of microalgae with other biomass types would most likely lead to SE of about 2.2 to 8.7wt% on biocrude yield.

Table 2: Summary of some previous research investigation on co-liquefaction of algae and other biomass feedstocks

Feedstock	Optimum reaction condition	IY biocrude yield, wt.%	Optimal mixed ratio	Mixed feedstock biocrude yield, wt.%			Biocrude properties								Reference
							Carbon		Nitrogen		ER,%		HHV(MJ/kg)		
	T (ºC), T(min), SL(wt.%)			Expt.	Cal	SE	IY	Col	IY	CoL	IY	CoL	IY	CoL	
Spirulina plantensis	300, 30	35.6	2:1	40.3	24	+16	NR	NR	NR	NR	50	82	33	30	Feng *et al*., (2019)
α-Cellulose		14.0					NR		NR		28		28		
Nannochloropsis sp.	280, 60	37.9	1:1	28.96	33.4	-4.4	72.7	73.2	4.6	6.7	66.4	54.4	35.9	36.5	Zhang *et al*., (2016)
Spirulina plantensis		29.2					70.9		7.2		54.2		34.1		
Cyanidioschyson merolae	300ºC, 30, 20	18.9	80:20	25.5	NR	NR	78.0	74.4	3.7	7.6	NR	NR	33.8	35.2	Dandamudi *et al*., (2017
Galdiera sulphuraria		14.0					76.6		6.2		NR		36.4		
Mixed-culture algal strain	300, 60, 25	27	1:3 25:75	35.7	NR	NR	59.4	35.7	2.5	2.5	52	37	25.8	17.8	Chen *et al*., (2014)
Swine manure		40					76.6		3.5		83		38.8		
Spirulina sp.	340, 40,	24.7	1:1	21.6	24.8	+3.2	74.2	74.3	4.1	5.3 ^	56	54	35.5	35.3	Jin *et al*., (2013)
Entermorpha pro.		14.6					74.1		4.5		35.2		34.9		
Chlorella pyre.	300, 60	40.5	4:1	40.4	38.1	-2.3	73.5	75.2	7.6	7.1	NR	67	35.3	35.4	Chen *et al*., (2019)
Sweet potato waste		37					70.8		0.3		NR		29.6		
Chlorella sp.	340, 30, ~10	~22	1:1	26.8	22.1	+4.7	72.0	70.8	6.5	6.5	41.6	57.8	34.1	33.4	Xu *et al*., (2019)
Sewage sludge		~24					71.8		6.0		52.7		34.2		
Dunaliella tertiolecta	340, 40, 10*	28	2:8	27.42	30.7	+3.3	NR	NR	NR	NR	NR	NR	NR	NR	Wu *et al*., (2017)
Polypropylene		1.82					NR		NR		NR		NR		
Desmodesmus sp.	350, 10, 10	53	1:1	29	36	-7	NR	NR	6	4.2	71	~43	33	32	Brilman *et al*., (2017)
Pine wood		24					NR		1		40		30		

As shown in Table 2, Feng *et al.*, (2019) investigated co-liquefaction of a low-lipid microalgae *Spirulina* sp. and α-Cellulose at reaction temperature of 300°C at 30min reaction time. At an optimal 2:1 mix ratio, SE of ~16wt% on biocrude yield was obtained. Operating at a reaction temperature of 340°C, 30min reaction time and using 10w/v% solids content, Xu *et al.*, (2019) reported SE of 4.7wt% at 1:1 mix ratio. Similarly, Jin *et al.*, (2013) reported SE of 3.2wt% on biocrude yield from the co-liquefaction of *Spirulina* sp. and *Entermorpha prolifera* at 340°C, 40min at ratio 1:1. In contrast to these SE, some investigations (Brilman *et al.*, 2017 and Zhang *et al.*, 2016) have shown negative effect on biocrude yields following co-liquefaction (Table 2). The reported AE were majorly due to the biochemical composition of the feedstock. For example Chen *et al.*, (2019) and Dandamudi *et al.*, (2017) used had feedstock with high carbohydrate and/or low lipids for their studies (Table 1), hence the low biocrude yield (shown in Table 2). Nevertheless, most reported studies have shown improved quality of the biocrude obtained from co-liquefaction when compared to individual feedstock, which will be discuss in next session. Importantly, more scientific research investigation are required to understand the underlying mechanism for the observed co-liquefaction effects.

3.2. Elemental composition of biocrude

The elemental carbon, hydrogen, nitrogen, sulfur and oxygen content for biocrudes obtained at different mix ratio and reaction temperature is shown in Fig. 3. As illustrated in Fig. 3, the reaction temperature and co-liquefaction of *Tetraselmis* algae (TA) with cow manure (CM) had substantial impact on the elemental content of the biocrudes. Generally, operating at 350°C (high temperature (HT)) led to an increase fractionation of elemental carbon and hydrogen in resultant biocrudes when compared to that at 300°C (low reaction temperature (LT)).Co-liquefaction of TA and CM was found to have substantial effects on the elemental distribution in biocrudes, irrespective of reaction temperature.

Furthermore, higher mass of TA in the feedstock mix led to numerical increase in carbon and hydrogen content in biocrude when compared to individual CM. Leading to enhanced energy density of biocrude, thus improving biocrude quality. For example, irrespective of reaction temperature, biocrude obtained from mix ratio 04:01 (TA:CM) led to 72w/w% of carbon compared to 68w/w% from 01:04 (TA:CM) mix ratio. However, for individual liquefaction of TA, co-liquefaction led to relatively low carbon and hydrogen content and high oxygen content in resultant biocrude. Suggesting that co-liquefaction promotes deoxygenation and decarboxylation reactions. Chen *et al.*, (2019) reported substantial reductions in oxygen and improved carbon recovery in produced biocrude.

More CM in the feedstock mix was found to substantially reduced N- content in biocrude when compared to individual liquefaction of TA. As shown in Fig. 3, an increase in CM mass in the blend CM and TA, the lower the nitrogen content of biocrude. This led to about 50% reduction in N-content in resultant biocrude. Suggesting combining biomass feedstock of lower nitrogen with algae would reduce the recovery of nitrogen, thus reduction in NO_x emissions upon combustion. Wang *et al*., (2019)reported similar trend, as more dosage of sweet potato residue (SPR) biomass in blend of *Chlorella pyrenoidosa*and SPR led to reduction of N-content from 6.77w/w% to 3.20w/w%. They conducted their co-liquefaction studies at 300^{o}C and 60min reaction temperature and reaction time, respectively.

Similarly, Chen *et al*., (2019) investigated the co-liquefaction of *Chlorella pyrenoidosa* and potato biomass at 300^{o}C and 60min and reported substantial reduction in N-content of biocrude from7.6w/w% to 4.4%.Algae biomass are generally known to contain high nitrogen, which is one of the important challenges of HTL-algae biofuel (Eboibi et al., 2015, Tang *et al*., 2020). Therefore, this approach could help to address this issue. Furthermore, based on the data presented in Figure 3, the optimal mix ratio of TA:CM (01:01) was found more suitable for more recovery of element in biocrude, except oxygen which was found for 04:01. However, since one of the objectives was also to improve biocrude yield, via co-liquefaction, a TA CM ratio of 03:02 was found more suitable for higher biocrude yield at 350^{o}C.

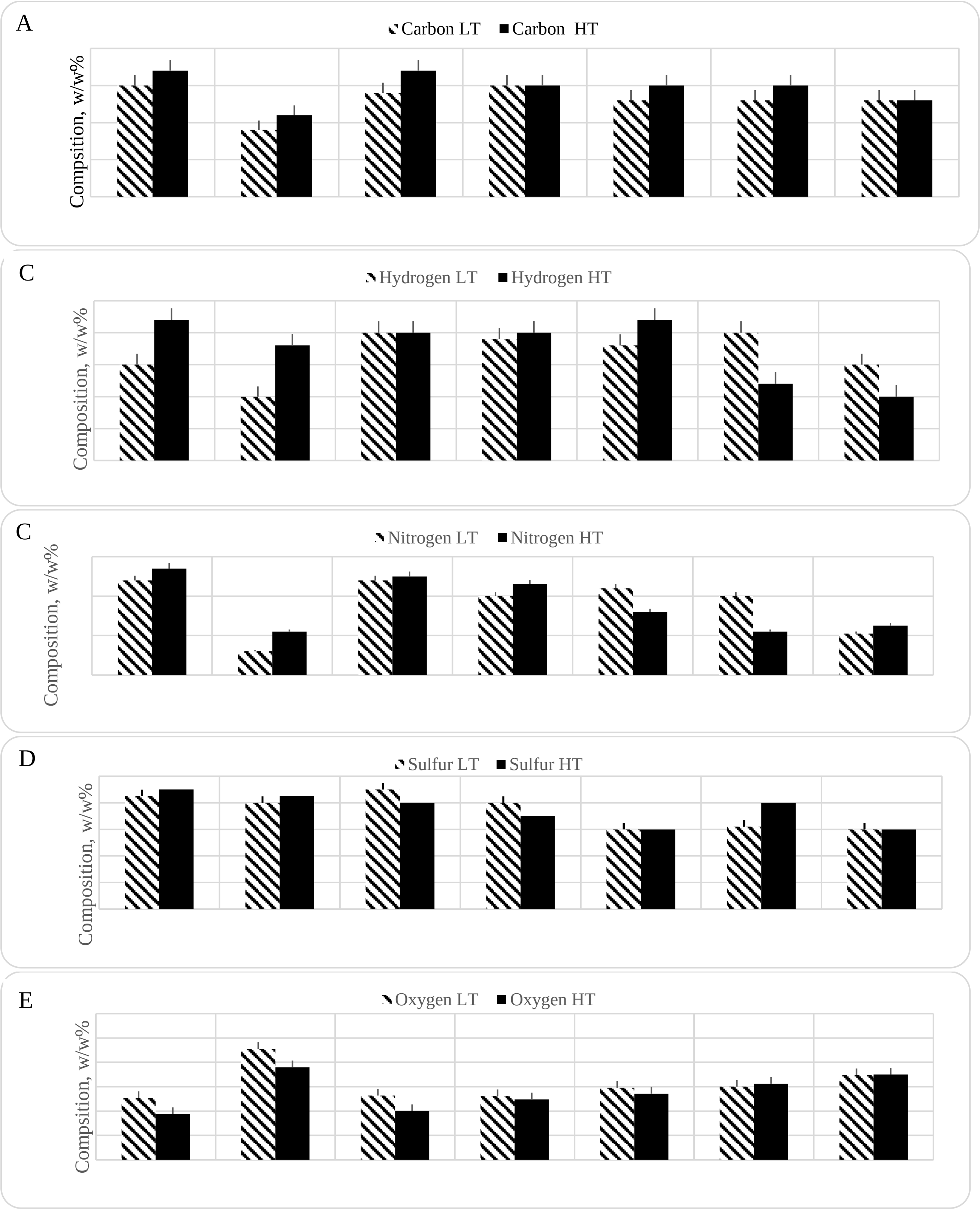

Fig. 2: Elemental composition of biocrude obtained at different feedstock mixing ratio

A: Carbon content B: Hydrogen content. C: Nitrogen content. D: Sulfur content. E: Oxygen content. LT: low reaction temperature (300^{o}C). HT: high reaction temperature (350^{o}C)

3.3. Effects of co-liquefaction on molar atomic ratios

The H/C, O/C and N/C molar atomic ratios of biocrudes obtained from individual TA and CM and at different mixing ratios are presented in Fig. 4 (the Van Krevelen diagram). As illustrated in Fig. 4A, the biocrude derived from *Tetraselmis* sp. (TA) had a better H/C and O/C ratio when compared to those obtained from cow manure and the mix feeds, however, substantially lower than that of petrocrude, with H/C close to 2 and O/C close to unity. Also, the biocrudes obtained from different mix ratios in have improved O/C atomic ratios and in most cases relatively enhanced H/C ratios. The improved O/C and H/C atomic ratios could be majorly due to dehydration and deoxygenation reactions. This finding suggests that co-liquefaction of microalgae and other organic biomass such as cow manure could improve the quality of biocrude.

The H/C with N/C atomic ratio of biocrudes (Fig. 4B) further shows the advantages of co-liquefaction. Due to CM having lower nitrogen content (Table 1), biocrude obtained from CM has a better H/C and N/C ratio when compared to that of other biocrudes from TA and mix feeds. Interestingly, biocrude H/C and N/C atomic ratios of mixed feedstocks were found to be lower when compared with that of individual TA. Apparently, this finding has shown the importance of blending feedstock of lower N-content with algae, as it potentially reduces the nitrogenous compounds in produced biocrude. Such practices are seen to improve biocrude quality. It could be concluded that addition of feedstock with no/or low nitrogen content, in this case CM with microalgae enhances Mannich reaction, hence the reduction in N-content of produced biocrudes.

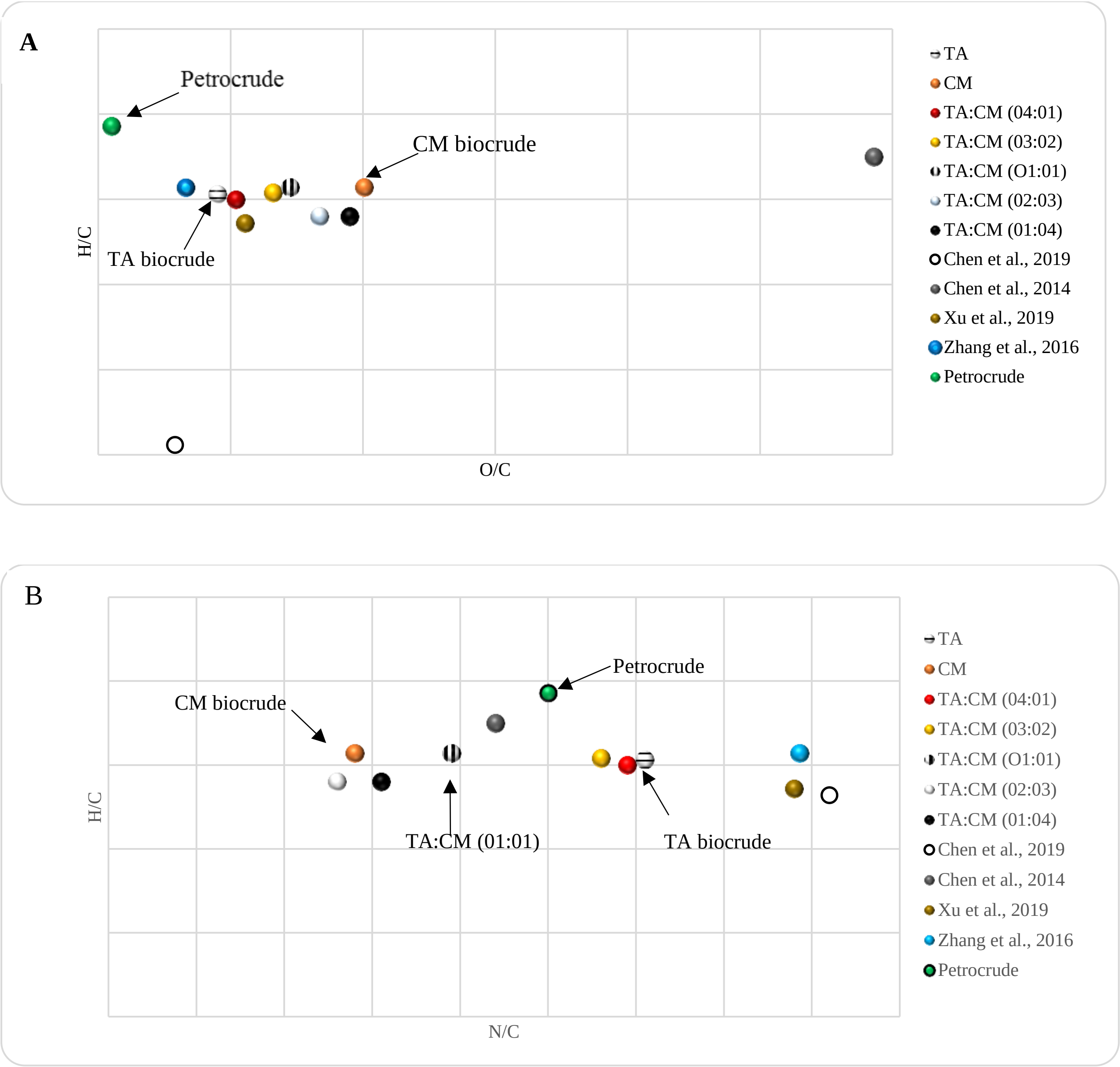

Fig.3: Van Krevelen diagram showing atomic ratios after co-liquefaction at 350^{o}C.

A: Hydrogen-to-carbon (H/C) against oxygen-to-carbon ratio (O/C). B: Hydrogen-to-carbon ratio against nitrogen-to-carbon (N/C) ratio.

3.4. Carbon and nitrogen recovery

One of the factors to determine quality of biocrude is the amount of carbon and nitrogen recovered in the biocrude. As more amount of carbon recovery (CR) and lower nitrogen recovery (NR) in the biocrude, the better the quality. Higher amount of CR the denser is the energy and performance. Also, lower amount of N-content in biocrude the better the quality. However, biocrude denitrification has remains an important challenge in its quality (Chen *etal*. 2019). Consequently, scientific research effort towards nitrogen reduction in bio crude would of interest. The amount of CR and NR in biocrudes obtained from mixed and individual feedstock is illustrated in Fig. 5.

Based on the data presented in Fig 5A, more carbon were recovered in biocrude at higher reaction temperature (HT (350^{o}C)) when compared to that at lower reaction temperature (LT (300^{o}C)).CR were found to correspond with the yield in biocrude, as an increase biocrude, the more CR in biocrude. Similarly, NR was found to follow similar trend for CR with respect to reaction temperatures. This finding seems to be in agreement with previous studies on decomposition of nitrogen with respect to reaction temperature. Decomposition of nitrogen in biocrude following HTL has been shown to increase with an increase in reaction temperature (Eboibi *et al*., 2014)

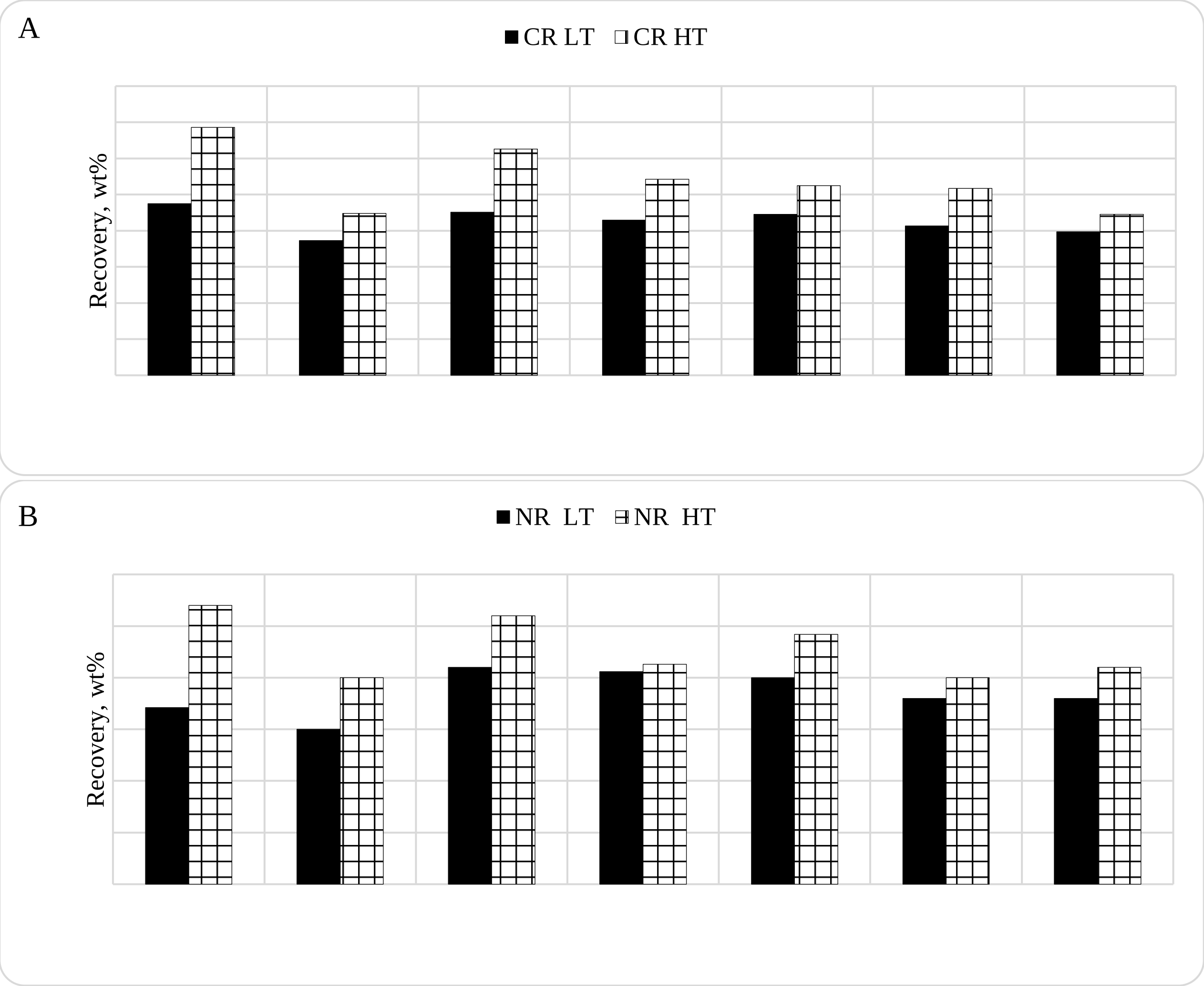

Fig. 4: Carbon and nitrogen recovery from co-liquefaction of TA, CM and at different ratios

CR: carbon recovery. NR: nitrogen recovery. LT: low temperature (300^{o}C). HT: high temperature (350^{o}C). TA: *Tertraselmis* sp. algae. CM: Cow manure

As shown in Fig.5, biocrude obtained from mixed feedstock had relative lower amount of CR and NR in biocrude, especially when compared to TA biocrude. However, when compared with CM biocrude, CR and NR from mixed feedstock were higher. Suggesting quality and yield of biocrude derived co-liquefaction is dependent on combined feedstock. In this present study, NR reduces thus improving the quality of biocrude as the proportion of CM increased in the mixed feedstock. However, CR in biocrude relatively decreases, reducing the energy density of the biocrude. Nevertheless, using TA and CM as mixed feedstock for HTL-biocrude seems more advantageous, in terms of lower NR, potentially reduction in pollution and a viable option for waste management.3.5. Energy density: Energy recovery and HHV

Another important factor in terms of biocrude quality and HTL reaction efficiency is the amount of energy recovery (ER) and HHV (Biller and Ross, 2011, Wang et al., 2019).The ER and HHV of biocrudes are presented in Fig. 6. As illustrated in Fig. 6 both reaction temperature and co-liquefaction had substantial effects on the energy recovery and HHVs of the biocrudes. For individual feedstock, about 60% and 45% ER was achieved for TA and CM at 300^{o}C reaction temperature (LT). ER increase to 76% for TA, and to 50% for CM, with an increased in reaction temperature (HT) to at 350^{o}C. This increase was found to correspond with biocrude yields and consistent with previous reports as shown in Table 2. For example, Prestigiacomo *et al.*, (2019) reported ER of 44.5% to 57.8% for *Chorella vulgaris*, and 44.7% to 57.2% for sewage sludge when operating at 325^{o}C and 30min reaction time.

Importantly, co-liquefaction of CM with TA generally led to improved ER. It was found that more mass of CM in the mixed feedstock led to decrease in ER, the maximum ER was obtained at 04:01 ratio, the minimum at 01: 04. In addition, ER from co-liquefaction of TA and CM at all mass ratio (except for 02:03 and 01:04), and at HT and LT, were found higher than ER mean value of individual liquefaction of TA and CM. Had it been there was no synergistic effect, the ER obtained from co-liquefaction of TA and CM at mass ratio 04:01 (72%), 03:02 (68%) and 01:01(65%) could have been equal to the mean value (62%) of ER from individual liquefaction of TA and TM. At optimal mix ratio of 1:1 for *Spirulina* sp and *Entermorpha pro.* Jin *et al*., (2013) reported mean value of 45.7% ER, however, 54% ER was achieved for mixed feedstocks.

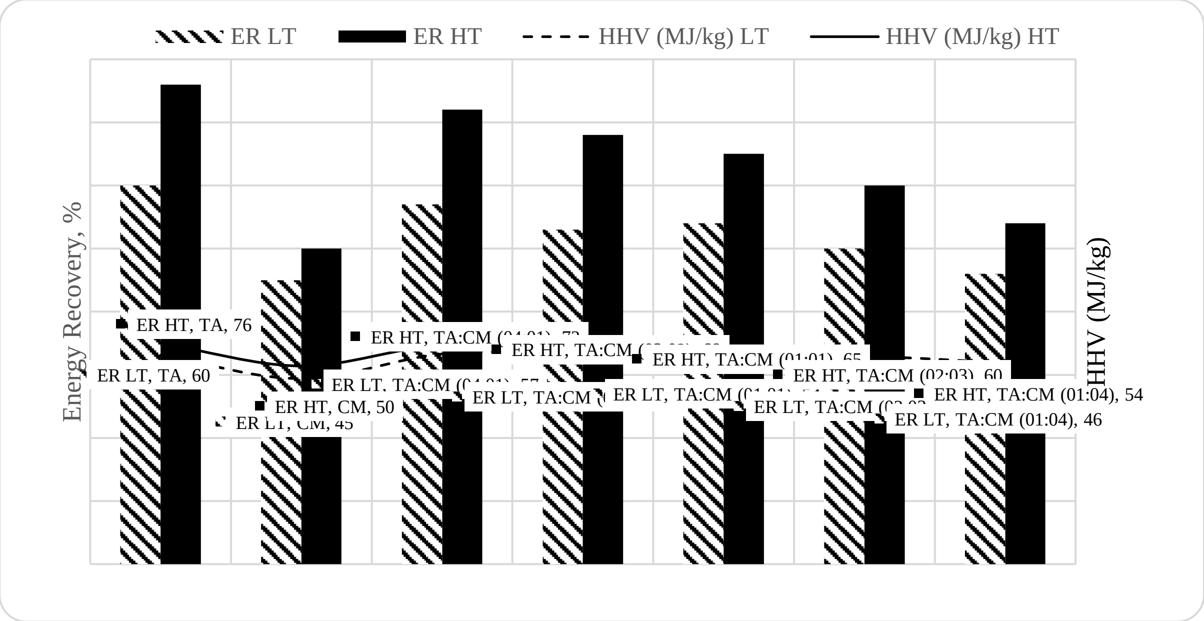

Figure 5: Energy recovery and higher heating value

Moreover, biocrudes HHV were found to be between 29MJ/kg to 35MJ/kg. The HHVs suggest that higher carbon led to improve energy density, while higher oxygen content led to decrease in resultant biocrudes. Co-liquefaction of TA and CM seems improve the HHV of biocrudes (31-34MJ/kg) when compared to HHV of biocrude obtained from individual CM (29MJ/kg).The biocrudes HHV were found to be similar to the average HHV of 33.9MJ/kg reported by (Xu *et al*., 2019) and within range of 32-34.7MJ/kg reported for individual HTL of *Spirulina* sp. swine manure and digested sludge (Leng *et al*., 2018). This findings show that synergistic effect occurs during co-liquefaction, improving the quality of biocrude.

Conclusion

This study investigated hydrothermal co-liquefaction of *Tetraselmis* sp. and cow manure for bio-crude production at different feedstock mix ratio. The study showed that co-liquefaction substantially has impact on yield and quality, importantly reducing the nitrogen content of resultant biocrude. Using cow manure as co-feedstock for HTL could be a viable option towards reducing environment pollution while simultaneously producing biofuel.

Reference

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