Effect of reaction time and algae loading on water-soluble and insoluble biocrude fractions from hydrothermal liquefaction of algae

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ABSTRACT: Hydrothermal liquefaction (HTL) of algae produces a water-insoluble organic phase (biocrude) and an aqueous phase that also contains algae-derived organic compounds (water-soluble biocrude). We determined the influences of the HTL reaction time and algae loading (wt% biomass in algae paste) on the yields and compositions of these two biocrudes. To the best of our knowledge this is the first detailed study of the water-soluble biocrude and how its yield and composition vary with processing conditions. Increasing the HTL reaction time from 10 to 60 min led to lower yields of water-soluble biocrude (from 6.4 to 3.6 wt%) and higher yields of water-insoluble biocrude (from 38 to 41 wt%). The total biocrude yield, however, was insensitive to the processing time. Longer HTL processing times also led to fewer and lighter GC-elutable compounds in the water-soluble biocrude. Increasing the algae loading from 8.6 to 18.7 wt% led to higher yields of water-insoluble biocrude (from

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29 to 44 wt%) and lower yields of water-soluble biocrude (from 6.6 to 2.6 wt%). The increase in the water-insoluble portion was much greater than the decrease in the water-soluble portion, so the total biocrude yield increased with loading. Increasing the wt% of algae in the slurry placed in the reactors also produced biocrudes with greater heating value. HTL of algae paste with higher biomass content had the twin benefits of providing more total biocrude and a higher quality biocrude. Reaction time and algae loading can be used to manipulate the proportions of water-soluble biocrude and biocrude that spontaneously separates from the aqueous phase after HTL.

Keywords: algae, hydrothermal liquefaction, biocrude, biorefinery

1. Introduction

Hydrothermal liquefaction (HTL) converts wet organic material (e.g., biomass) into energy-dense biocrude via chemical reactions in and with hot compressed liquid water [1,2]. HTL is more appealing for processing wet feedstocks (e.g., algae) than are thermochemical conversion methods, such as gasification or pyrolysis, that require the biomass to be dried [3]. HTL of wet algal biomass produces an organic biocrude fraction, an aqueous fraction containing numerous polar organic compounds [4], a gas fraction, and a solid residue. The latter two product fractions generally appear in yields of just a few percent.

Two different approaches can be used to recover the biocrude from HTL. One approach is to allow the biocrude and aqueous phases to separate spontaneously due to their immiscibility and density difference [e.g.,5,6]. The second approach is to add an organic solvent, such as dichloromethane (DCM), to dissolve the biocrude, extract additional organic material from the aqueous phase, and facilitate biocrude recovery as a larger organic phase that separates from the co-existing aqueous and solid phases

[e.g.,7]. The former approach has the advantage of being a simpler and “greener” process as no solvent extraction step is needed and no organic solvent is required. The latter approach has the advantage of producing a modestly higher biocrude yield, since water-soluble material is extracted and recovered in the biocrude fraction. These water-soluble extractables tend to be richer in heteroatoms than the water-insoluble components of the biocrude [8], so this solvent extraction approach tends to produce a biocrude of lower quality (less heating value, more heteroatoms) than the former approach.

More generally, whether or not the water-soluble materials are extracted as biocrude affects aspects of the biorefinery apart from considerations of biocrude quantity and quality. If the DCM-soluble components are removed from the aqueous phase, then that aqueous phase will have a lower concentration of organic compounds. This lower loading could be beneficial if the aqueous phase is recycled directly to algae growth ponds. The reduced organic content would provide a less hospitable environment for growth of undesired invasive heterotrophic organisms in the algae pond. The lower concentration could be a disadvantage, however, if the biorefinery employed a process such as catalytic hydrothermal gasification to convert the dissolved aqueous-phase organic compounds into fuel gases (e.g., CH_4) to provide heat and power for the biorefinery [9]. Removing the DCM-soluble material from the aqueous phase would also hinder recovery of nitrogen and its recycling for algae growth, as more of the N atoms would be removed from the aqueous phase and partitioned into the biocrude fraction.

The discussion above shows that the decision about whether to use an organic solvent for biocrude extraction or whether to rely solely on spontaneous phase separation of the HTL reactor effluent affects several aspects of biorefinery operation, economic viability, and environmental sustainability. To make this decision, one desires to know how solvent extraction affects the quantity and composition of the

biocrude and the composition of the aqueous phase coproduct. To the best of our knowledge, our earlier preliminary work on this topic remains the only account in the archival scientific literature [8]. That account compared the biocrudes and aqueous phases from HTL at a single reaction condition and recovered by the two different approaches. The present study expands upon that work by determining the influence of the HTL conditions on the characteristics of the biocrude that spontaneously separates from the aqueous phase post reaction and the biocrude that can be extracted from the aqueous phase using DCM. This information will be useful in deciding how best to design and operate an algal biorefinery that employs HTL.

2. Experimental section

2.1. Experimental procedures

We used the preservative-free *Nannochloropsis* sp. slurry, reactors, reagents, etc., that have been fully described in our previous report [8]. HTL experiments were carried out at 350 °C under two sets of conditions. One set explored HTL at four different batch holding times (10, 30, 45, and 60 min) and a fixed algae loading of 14.1 wt%. The other set explored HTL at four different algae loadings (8.6, 11.4, 16.5, and 18.7 wt%) at a fixed batch holding time of 20 min.

We loaded the reactor with known amounts of algae slurry and deionized water, sealed the reactor, and then placed it in a Techne Fluidized Sand Bath (Model SBL-2) maintained at 350 °C. When the desired reaction time was reached, we removed the reactor from the sand bath and quenched it in an

81 ambient-temperature water bath. We allowed the reactor to equilibrate at room temperature for one hour
82 before collecting products.

83 We followed the procedure denoted as method 2 (M2) in our previous article [8] to collect, separate,
84 and quantify the biocrude. Briefly, we opened the reactor and directly collected the aqueous phase from
85 the cool, equilibrated reactor by a pipet. We then added DCM to the reactor to dissolve and collect the
86 water-insoluble biocrude (B) in the reactor as well as that adhering to the pipet. Note that the DCM did
87 not come into contact with the aqueous phase co-product. DCM was added in these experiments for
88 convenience and to ensure complete recovery of the water-insoluble biocrude. Its addition would likely
89 be optional in a large-scale biorefinery where the biocrude-water separation could be done continuously
90 and at higher temperatures. Evaporating the DCM by flowing N₂ in a drying apparatus (VertexTM
91 EVAPORATOR) led to isolation of the water-insoluble biocrude.

92 Centrifugation separated the aqueous phase that had been collected from any accompanying solids,
93 which were subsequently dried. We added DCM to the solids-free aqueous phase and mixed it
94 thoroughly to extract the DCM-soluble, water-soluble material. These DCM-soluble and water-soluble
95 compounds constitute the aqueous-phase contribution to the biocrude (B_A). The total mass of the
96 biocrude (B_T) is the sum of the masses of the water-insoluble (B) and water-soluble (B_A) components.
97 We further separated each biocrude into light (hexane-soluble) and heavy (hexane-insoluble) fractions
98 as described previously [10].

99 2.2. Analysis methods

100 Biocrude samples were shipped to Atlantic Microlab, Inc., for elemental analysis. We identified

components of the biocrudes by redissolving them in DCM and analyzing by GC-MS. Nuclear magnetic resonance (NMR) and Fourier transform infrared spectroscopic analysis (FTIR) of biocrudes were also conducted to characterize the biocrude. Detailed information about these instruments and procedures was reported previously [8].

The higher heating value (HHV) of biocrude was estimated by using the Dulong formula,

$$\text{HHV (MJ/kg)} = 0.338C + 1.428(H - O/8) + 0.095S \quad (1)$$

where C, H, O, and S are the wt% of each element in the biocrude.

Additionally, the biocrude yield, the fraction of total biocrude from the aqueous phase, the fraction of light biocrude, and the energy recovery in the biocrude were calculated as follows:

$$\text{Biocrude yield} = \frac{\text{Mass of biocrude}}{\text{Mass of algae in reactor}} \times 100\% \quad (2)$$

$$\text{Fraction of total biocrude from aqueous phase} = \frac{B_A}{B + B_A} \times 100\% \quad (3)$$

$$\text{Fraction of light biocrude} = \left(1 - \frac{\text{Mass of measured } B^H}{\text{Mass of } B}\right) \times 100\% \quad (4)$$

$$\text{Energy recovery of biocrude} = \frac{\text{HHV of biocrude} \times \text{Mass of the biocrude}}{\text{HHV of initial dry algae} \times \text{Mass of the algae feedstock}} \times 100\% \quad (5)$$

Three independent runs were carried out at the same conditions to determine the uncertainties in the experimental data. Results reported herein are average values and their uncertainties are the sample standard deviations.

3. Results and discussions

The section provides results about the biocrude yields and compositions. To the extent possible, the discussion focuses on examining the effects of time and algae loading on the water-soluble biocrude fraction and on characterizing the light and heavy fractions of the water-soluble biocrude, as this report

is the first to consider these topics. The mass balances in the experiments ranged from 90–95 wt%.

3.1. Biocrude yields

Table 1 shows the influence of reaction time on biocrude yields. The yield of water-soluble biocrude (B_A) declined from 6.4 to 3.6 wt% as time increased, but the yield of water-insoluble biocrude (B) rose from 38.2 to 40.5 wt% as time increased from 10–60 min. Accordingly, the total biocrude yield (B_T) was time-invariant at about 44 wt%. Table 1 also shows that the fraction of total biocrude from the aqueous phase decreased from 14.4 to 8.1 wt% as time increased from 10–60 min. Accordingly, one expects the total biocrude collected at short times to be richer in O and N (more water-soluble compounds) than the biocrude collected at longer times. The absence of much variation in the total biocrude yield with time at 350 °C has been observed previously [10]. That the relative contribution of the aqueous phase biocrude to the total amount of biocrude changes with HTL reaction time has not been previously documented.

Table 1 Biocrude yields (wt%) from HTL at 350 °C and different times (14.1 wt% algae loading).

Time (min)	10	30	45	60
Yield of water-insoluble biocrude (B)	38.2±1.4	39.2±1.3	40.1±1.3	40.5±1.5
Yield of water-soluble biocrude (B_A)	6.4±0.5	4.5±0.6	4.6±0.5	3.6±0.4
Total biocrude yield	44.6±1.9	43.7±2.0	44.7±1.8	44.1±1.9
Fraction of total biocrude from aqueous phase (wt%)	14.4±1.6	9.3±0.8	10.2±0.8	8.1±0.7

Table 2 shows that the yield of water-soluble biocrude (B_A) first increased and then decreased from 6.6 to 3.0 wt%, as algae loading increased from 8.6 to 18.7 wt%. In contrast, the yields of

water-insoluble and total biocrude increased from 29.2 to 44.0 wt% and from 34.8 to 47.0 wt%, respectively, as the algae loading increased. The fraction of the total biocrude that was in the aqueous phase declined from 16.1 to 6.4 wt% as the algae loading increased from 8.6 to 18.7 wt%. These results for the total biocrude are in accord with previous investigations with *Nannochloropsis* showing that increasing algae loading from 5 to 35 wt % (for HTL at 350 °C, 60 min) increased the total biocrude yield from 36 to 46 wt % [10].

Table 2 Biocrude yields (wt%) from HTL at 350 °C and different algae loadings (20 min).

Algae loading (wt%)	8.6	11.4	16.5	18.7
Yield of water-insoluble biocrude (B)	29.2±2.1	38.1±1.8	41.1±1.5	44.0±2.4
Yield of water-soluble biocrude (B _A)	5.6±0.5	6.6±0.6	3.7±0.5	3.0±0.4
Total biocrude yield	34.8±2.6	44.7±2.4	44.7±2.0	47.0±2.9
Fraction of total biocrude from aqueous phase (wt%)	16.1±1.4	14.7±1.2	8.2±0.8	6.4±0.4

Overall, both increased time and increased algae loading decreased the yield of water-soluble biocrude. This decrease likely occurred because some of the molecules initially formed from algae HTL contain polar groups and hence enjoy solubility in water. As the reaction progressed, however, such compounds could undergo deamination, decarboxylation, dehydration, and oligomerization. One expects the oligomerization pathways, which involve bimolecular reactions, to become more favorable as the algae loading increases. The net effect is that the molecules became larger and less polar, which rendered them less soluble in water.

The results show that reaction time and algae loading can be used to control the proportions of water-soluble biocrude and biocrude that spontaneously separates from the aqueous phase after HTL. Such control could be useful, for example, to ensure that the aqueous phase retains sufficient organic content for a catalytic hydrothermal gasification unit operation to generate enough fuel gas to meet the heat and power demands of the biorefinery.

3.2. Fraction of light biocrude

Fig. 1 illustrates how the portion of the biocrudes that falls into the light (hexane-soluble) fraction changed with time and algae loading. This metric is a proxy for biocrude quality, as the hexane-soluble compounds would be less polar and of moderate boiling point and thus more desirable for fuel use than would the heavier, more polar, hexane-insoluble compounds. The fractions of the water-insoluble and water-soluble biocrudes that were hexane-soluble increased from 62 to 77 wt% and from 67 to 78 wt%, respectively, as time increased from 10 to 60 min. Recall that the fraction of the total biocrude that was water-soluble decreased from 14 to 8% over this same time range. It seems that the same reactions that rendered less of the biocrude to be water-soluble also rendered more of it hexane-soluble. That is, the reactions that made the molecules less polar both reduced the solubility in water and increased the solubility in hexane.

As algae loading increased from 8.6 to 18.7 wt%, the fraction of light (hexane-soluble) material in the water-insoluble biocrude (B) first increased and then decreased, but that in the water-soluble biocrude (B_A) decreased monotonically from 77 to 51 wt%. Recall that the yield of water-soluble biocrude was lower at the higher algae loadings. This trend, along with that noted here for the light biocrude, indicates

that both the quality and quantity of the water-soluble biocrude declined at the higher algae loadings. It would appear that the chemical reactions that produce compounds that are both hexane-soluble and water-soluble are inhibited at high biomass loadings relative to competing reactions.

Fig. 1. Fraction of light biocrude in B and B_A from HTL at 350 °C: a) different times, 14.1 wt% algae loading, and b) different algae loadings, 20 min.

3.3. Elemental compositions and higher heating values

The yields of light, heavy, and total biocrude are important metrics for assessing HTL, as they relate the amount of product obtained. The compositions of these biocrudes are also very significant, however, as this metric relates to the quality of the product obtained. Biocrudes with more H and C and less O, N, and S are more desirable products.

Table 3 lists the elemental compositions and estimated higher heating values (HHV) of the water-soluble and water-insoluble biocrudes and their heavy (hexane-insoluble) fractions formed at different HTL reaction times. The O content was calculated by difference from the experimental C, H, N, and S values and reserving 1 wt% for other elements not assessed.

Table 3 Elemental composition and HHV of water-insoluble (B) and water-soluble (B_A) biocrudes and their heavy (hexane-insoluble) components (B^H, B_A^H) produced at different HTL reaction times (350 °C, 14.1 wt% algae loading).

Item	Time (min)	C (wt%)	H (wt%)	N (wt%)	S (wt%)	O (wt%)	N/C —	O/C —	HHV (MJ/kg)
Dry algae	—	52.6	7.35	8.58	0.57	27.0	0.152	0.477	24.8
B	10	77.5	10.0	4.85	0.56	7.08	0.063	0.091	39.3
	30	77.5	10.0	4.60	0.48	7.36	0.059	0.095	39.3
	45	78.3	10.0	4.54	0.48	6.64	0.058	0.085	39.6
	60	78.1	10.0	4.48	0.50	6.96	0.057	0.089	39.5
B _A	10	63.4	8.76	10.2	0.20	17.5	0.161	0.276	30.8
	30	62.6	8.72	10.7	0.10	17.8	0.171	0.285	30.4
	45	64.4	9.07	10.0	0.20	16.3	0.155	0.253	31.8
	60	63.1	8.88	10.4	0.16	17.5	0.164	0.277	30.9
B ^H	10	72.6	8.49	6.11	0.63	12.2	0.084	0.168	34.5
	30	74.2	8.60	6.16	0.64	10.4	0.083	0.141	35.5
	45	74.5	8.53	5.86	0.58	10.5	0.079	0.141	35.6
	60	75.8	8.66	5.91	0.52	9.09	0.078	0.120	36.4
B _A ^H	10	63.0	8.04	11.1	0.72	17.1	0.176	0.271	29.8
	30	63.5	7.72	9.52	1.18	18.1	0.150	0.284	29.4
	45	63.7	7.81	9.98	1.19	17.3	0.157	0.271	29.7

Our previous article [8] discussed the differences between the water-soluble and water-insoluble biocrudes, so here we focus on the effect of HTL reaction time on the different biocrudes. To the best of our knowledge, Table 3 presents the first data for the elemental composition of the heavy fraction of the water-soluble biocrude from HTL.

The HTL processing time seemed to have little effect on the composition of the biocrude (B) that spontaneously separated post-liquefaction or on the water-soluble biocrude component (B_A). There are modest differences, but we believe these can be attributed to run-to-run variation in the experimental results. The composition of the heavy fractions of these two biocrudes did show some trends with reaction time, however. The heavy fraction of the water-insoluble biocrude (B^H) showed a modestly increasing carbon content and heating value and a decreasing oxygen content as time increased. The heavy fraction of the water-soluble biocrude showed the highest nitrogen content at the shortest time.

Table 4 displays the elemental compositions and HHVs of the biocrudes and their heavy fractions formed at different algae loadings. The carbon content of both water-soluble and water-insoluble biocrudes was generally higher when produced at higher algae loadings. The oxygen contents tended to be lower, so the heating value of the biocrudes tended to be greater at the higher algae loadings. Thus, performing HTL at the higher algae loadings has the twin benefits of producing higher yields of biocrude and producing biocrude with a higher energy density. We note that Peterson et al. [1], on the basis of process engineering considerations, suggested a target algae loading of 15–20 wt % for HTL.

Table 4 Elemental composition and HHV of water-insoluble (B) and water-soluble (B_A) biocrudes and their heavy (hexane-insoluble) components (B^H , B_A^H) produced at different algae loadings (350 °C, 20 min).

Item	Algae loading (wt%)	C (wt%)	H (wt%)	N (wt%)	S (wt%)	O (wt%)	N/C —	O/C —	HHV (MJ/kg)
B	8.6	76.8	10.0	4.44	0.62	8.11	0.058	0.106	38.9
	11.4	76.4	10.1	4.64	0.59	8.33	0.061	0.109	38.7
	16.5	77.4	10.3	4.64	0.45	7.22	0.060	0.093	39.6
	18.7	77.7	10.2	4.81	0.39	6.90	0.062	0.089	39.6
B _A	8.6	63.5	8.38	10.9	0.28	16.9	0.172	0.266	30.4
	11.4	63.7	8.28	11.2	0.31	16.5	0.176	0.260	30.4
	16.5	68.2	8.69	9.94	0.32	12.9	0.146	0.189	33.2
	18.7	65.2	8.74	10.8	0.34	15.0	0.165	0.230	31.9
B ^H	8.6	74.5	8.77	6.11	0.73	9.92	0.082	0.133	36.0
	11.4	74.7	8.92	6.20	0.72	9.42	0.083	0.126	36.4
	16.5	76.7	9.13	6.07	0.46	7.63	0.079	0.099	37.6
	18.7	76.9	9.04	5.91	0.37	7.76	0.077	0.101	37.6
B _A ^H	8.6	63.1	8.05	10.8	0.79	17.3	0.170	0.274	29.8
	11.4	61.0	8.25	10.3	0.83	19.6	0.169	0.321	29.0
	16.5	63.8	8.26	10.6	0.79	16.5	0.167	0.259	30.5
	18.7	63.2	8.19	10.3	0.91	17.4	0.163	0.275	30.0

207 The influence of algae loading on the heavy fraction of the water-insoluble biocrude (B^H) was
208 essentially the same as its influence on the whole biocrude. The carbon content and heating value were

higher at the higher loadings and the oxygen and sulfur contents were lower at the higher loadings. The algae loading seemingly had no discernible effect on the compositions of the heavy fraction of the water-soluble biocrude.

Table 5 shows the masses, energy densities, and energy recoveries for both biocrudes at two HTL conditions. The biocrudes that spontaneously separates from the aqueous phase post-HTL (B) contained about 60% of the chemical energy in the original algal biomass. The “heavy” fractions of those biocrudes (B^H) contained about 40% of the chemical energy in those biocrudes. The biocrudes extracted from the aqueous phase (B_A) contained 5 – 8% of the chemical energy in the biomass. By performing the DCM extraction of the aqueous phase, this material can be recovered as biocrude and potentially refined into liquid fuels. This extraction step, however, removes material from the aqueous phase that could be converted to fuel gases for heat and power in the biorefinery. The total energy recoveries under these two HTL conditions were 65.8% and 67.5%, respectively. These values are consistent with a previously reported 66% energy recovery for biocrude obtained by contacting DCM directly with the entire post-HTL reaction mixture produced at similar conditions [10].

Table 5 Energy recoveries in the water-insoluble (B) and water-soluble (B_A) biocrudes and heavy fraction (B^H) of

water-insoluble biocrude obtained from HTL at 350 °C.

Condition	Biocrude	HHV of biocrude (MJ/kg)	Mass of biocrude (g)	Dry algae mass (g)	HHV of dry algae (MJ/kg)	Energy recovery of biocrude (%)
30 min,	B	39.3	0.1549	0.4019	24.84	60.9

14.1	wt%	B _A	30.4	0.0160		4.89
loading		B ^H	35.5	0.0600		21.4
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20 min, 11.4		B	38.7	0.1055		59.5
		B _A	30.4	0.0181	0.2765	24.84
wt%		B ^H	36.4	0.0454		24.1
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225 3.4. Composition of Biocrude from Aqueous Phase

226 The biocrude that was extracted from the aqueous phase, B_A, was analyzed by GC-MS. Fig. 2 shows
227 the total ion chromatograms for B_A obtained at different HTL reaction times. The water-soluble
228 biocrude components included compounds tentatively identified as pyrazines, piperidenones,
229 cyclopentanone, amides, pyrrolidinones and pyrrolidinediones, pyrrolidines, phenols, and amines.
230 There are numerous heterocyclic compounds containing N and/or O atoms. Oxygenated compounds can
231 be produced by the degradation of lipids and carbohydrates, whereas protein decomposition could yield
232 numerous nitrogen heterocycles [4]. The presence and prevalence of compounds containing N and O are
233 consistent with the elemental analysis results, which showed that the water-soluble biocrude
234 components are richer in N and O than is the water-insoluble biocrude. A high N content in the biocrude
235 negatively affects its combustion and various other properties [2]. Thus, this higher heteroatom content
236 above implies that the biocrude would be of lower quality.

237 a)

b)

c)

Fig. 2. Total ion chromatograms for water-soluble biocrude obtained from HTL at 350 °C and 14.1 wt% algae loading and different reaction times: a) 10 min, b) 30 min, and c) 60 min.

As the HTL reaction time increased, the relative amounts of the different compounds in the water-soluble biocrude varied. Compounds with retention times longer than 36 min are common in the biocrude formed after HTL for 10 min, but largely absent in the biocrude recovered from HTL at longer times. Likewise, the compounds with retention times before 20 min vanish from the chromatogram for the biocrude formed after HTL for 60 min.

3.5. Functional group compositions

We conducted ^1H - and ^{13}C -NMR analyses of biocrudes to identify the types of structures and functional groups present. Fig. 3 depicts the ^1H NMR spectra of the biocrudes obtained from HTL at 30 min and 350 °C and their light and heavy fractions. These spectra are representative of the features in the biocrudes produced at other reaction conditions investigated. All of the spectra show resonances at around 0.8, 1.2, 2.1, and 7.2 ppm, though the relative height of each peak varied from sample to sample. The peaks at 0.8 and 1.2 are characteristic of terminal methyl groups and methylene groups in alkyl chains, respectively [11]. The peak at about 2.1 ppm is consistent with the resonance expected from protons on carbon atoms α to a carbonyl group. The peaks near 7.2 ppm likely arose from aromatic protons or conjugated dienes [12,13]. The appearance of peaks at these resonances reveals the presence of alkyl moieties, carbonyl functionalities, and aromatic or unsaturated molecules in all of the biocrudes, though in differing amounts. Our previous article discussed the differences between the NMR spectra for the biocrudes that separated spontaneously from the aqueous phase and that was extracted from the aqueous phase, so we focus on a comparison of the light and heavy fractions here. To the best of our knowledge, this is the first report of spectroscopic characterization of light and heavy fractions of

biocrude.

Comparing the spectra for the light and heavy biocrudes in Fig 3c and 3d, one observes peaks around 1.5 and 1.9 ppm in the light biocrude that are not present in the spectrum for the heavy portion of the same biocrude. Resonances at these locations are indicative of protons on tertiary aliphatic carbon atoms and on allylic carbons. Apparently, there is a greater abundance of such structures in the light fraction of the biocrude than the heavy fraction.

Comparing the spectra for the light and heavy components of the biocrude extracted from the aqueous phase (panels e and f) shows that the light fraction has peaks at 2.7 and 2.9 ppm that are absent in the heavy fraction. Resonances from protons that are α to a heteroatom or unsaturated moiety (e.g., benzylic or allylic protons) appear in this region.

c) d)

e) f)

Fig. 3. ^1H -NMR spectra of biocrudes obtained from HTL at 350 °C, 30 min, and 14.1 wt% algae loading: a) water-insoluble biocrude (B), b) water-soluble biocrude (B_A), c) light fraction of water-insoluble biocrude B^L , d) heavy fraction of water-insoluble biocrude B^H , e) light fraction of water-soluble biocrude B_A^L , and f) heavy fraction of water-soluble biocrude B_A^H .

Fig. 4 displays ^{13}C -NMR spectra of the biocrudes and their light and heavy subfractions from HTL at one set of reaction conditions. These spectra are representative of those obtained at other conditions, as the reaction time and algae loading did not influence the appearance of the spectra. Both the water-insoluble and the water-soluble biocrude samples display several peaks in the 0–55 ppm region,

where aliphatic carbon atoms appear [14]. This result is consistent with the high content of aliphatic carbons indicated by the ^1H -NMR spectra (0.5–3.0 ppm). The weak peaks at around 130 ppm are consistent with the presence of alkenyl and aromatic carbon in these biocrudes. Furthermore, all biocrudes in Fig. 3 showed a resonance around 207 ppm, which is in the region (180–215 ppm) representing carbonyl carbons in ketones/aldehydes [15].

The spectra for the light and heavy fractions of the water-insoluble biocrude have several peaks in common, but the light fraction has additional peaks in the region from 0 – 30 ppm, suggesting that it contained a larger variety of compounds with methyl and methylene groups. The spectrum for the water-soluble biocrude showed resonances within the region of 165–180 ppm, where carbon atoms in carboxylic acid groups or esters appear [12,15]. The peaks at about 115 ppm and 141–146 ppm are in the region (95–165 ppm) where aromatic and heteroaromatic carbons appear [15].

c) d)

Fig. 4. ^{13}C -NMR spectra of biocrudes obtained from HTL at 350 °C, 30 min, and 14.1 wt% algae loading. a) water-insoluble biocrude (B), b) water-soluble biocrude (B_A), c) light fraction of water-insoluble biocrude B^L , and d) heavy fraction of water-insoluble biocrude B^H .

4. Conclusions

The work explored the influences of HTL reaction time and algae loading (wt% in slurry loaded into the reactor) on the properties of water-soluble and water-insoluble biocrude and the light and heavy fractions thereof. With increasing time, the yield of biocrude recovered from the aqueous phase co-product declined from 6.4 wt% to 3.6 wt% but the yield of biocrude that spontaneously separated from the aqueous phase after cooling the HTL products increased from 38.2 wt% to 40.5 wt%. Algae loading had a much stronger influence. As the wt% of algae in the slurry feedstock increased, the yield of water-soluble biocrude decreased to 2.55 wt%, but the yield of water-insoluble biocrude increased by nearly 50%. Longer reaction times and higher algae loadings favor reactions that produce molecules that are insoluble in ambient liquid water. Reactions such as decarboxylation, deamination, dehydration, oligomerization, and condensation could be responsible for this shift in the product distribution. The

heating value of the biocrudes showed little variation with HTL processing time, but the heating values of the biocrudes produced at the higher algae loadings always exceeded those of the biocrudes produced at the lower loadings. This trend held for both the water-soluble and water-insoluble biocrude components. The results presented herein demonstrate that the HTL reaction time and the wt% algae in the feedstock affect the relative amounts of water-soluble and water-insoluble biocrude compounds and the heating value of the biocrudes produced.

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