Life Cycle Design of an Algal Biorefinery Featuring Hydrothermal Liquefaction: Effect of Reaction Conditions and an Alternative Pathway Including Microbial Regrowth

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ABSTRACT: Algae are an appealing source for bioenergy due to their high yields relative to terrestrial energy crops. The high cost of production, however, has prohibited commercialization. Hydrothermal liquefaction is a technology that converts more of the algae into oil than alternative technologies, thereby reducing the amount of expensive pond infrastructure and energy required for cultivation. We incorporate recent experimental results into an analysis that models the economic and life cycle performance of an algal biorefinery across a range of reaction conditions. Two strategies are explored: one pathway with gasification of the aqueous waste products for onsite energy recovery and another pathway featuring cultivation of Escherichia coli on the aqueous products and recycling of the biomass back through the reactor for boosted oil yields. We found that the maximum net energy ratio of 1.9 and minimum global warming potential of 1.0 kg CO₂e L⁻¹ oil occurred with gasification, along with the minimum reaction temperature explored, 250 °C, and reaction times close to 1 h. The optimal economic and occupied land results occurred at a maximum temperature of 400 °C and with the shortest reaction time explored of 5 min. The cost of algal oil at these conditions was $1.64 L⁻¹ oil (or $263 bbl⁻¹). For the regrowth pathway, the land footprint could be further reduced by 10%, and the optimal cost could be reduced to $1.59 L⁻¹ oil. Forgoing gasification had a significantly detrimental effect on the other two metrics. Given the importance of economics to an algal biorefinery’s operations, this could be a viable option.

KEYWORDS: Algae, Hydrothermal liquefaction, Life cycle assessment, Biofuel, Escherichia coli

INTRODUCTION

Algal biomass is a promising source of bioenergy that could potentially produce billions of liters of biofuel annually in the United States.1,2 An appealing characteristic of algae is that they could theoretically occupy significantly less land than terrestrial biofuel crops due to their high growth rate, and the use of engineered ponds enables algae to be cultivated on marginal land that could not otherwise be used for agriculture.3−7 Despite substantial investment from the government and within the private sector, a scalable and economically viable means for producing algal biofuel has not emerged. The primary economical hurdle is the high cost to build and operate the ponds,8−11 and a major technical hurdle is a means to convert the algal biomass into a usable transport fuel.12,13 This article focuses on methods to advance a technology, hydrothermal liquefaction (HTL), that could address both of these obstacles by providing a means to convert wet algal biomass to biocrude oil while reducing the amount of pond infrastructure required to produce a given quantity of oil.

A life cycle assessment (LCA) by Lardon et al. illustrated that the solvent-based oil extraction technologies used for other energy crops such as soybean cannot be relied upon for algae; the amount of energy required to dry the biomass to levels typical of terrestrial crops would exceed the energy content in the algal oil.14 Several studies have demonstrated the viability of “wet” solvent extraction strategies to separate the lipid fraction of the algal biomass as an oil that can be upgraded via transesterification to a biodiesel product.14,15 This approach still has not been implemented at commercial scale, and efficient recovery of the solvent (typically hexane) has proven to be a challenge.12 Another limitation of solvent extraction is that the yield of oil per unit of biomass is limited to the fraction of the biomass that is lipid. Currently, the most economical strategy for cultivating phototrophic algae is an open paddle-wheel...
mixed pond, and the species grown in these ponds typically have just 10–25% lipid content.

By contrast, HTL converts a portion of the carbohydrate and protein fractions of the algal biomass into oil in addition to the lipid fraction. This process works by converting a wet (15–20% solids) algal slurry under high temperature and pressure (e.g., 350 °C, 16.5 MPa) into a variety of products including a biocrude oil. Unlike gasification, the HTL process occurs in a closed vessel to preserve the solution in the aqueous phase by high pressure. HTL mimics the way fossil crude oil was formed in nature, by geologic heat and compression of plant tissue, but does so in minutes rather than in millions of years. In addition to the oil that is formed, HTL produces solids, gas, and soluble products remaining in the water, or the “aqueous phase”.

Use of the nonproduct portions of the algal biomass (e.g., lipid extracted algae (LEA) remaining after oil extraction or the aqueous phase products from HTL) is significant and has the potential to improve the economics and life cycle impacts of the biorefinery. Frank et al. performed an LCA that demonstrated the benefits of solvent-based lipid extraction compared to HTL due to the relatively high amount of nonproduct portion of the algae available for energy recovery via gasification. To date, the work by Frank et al. has been the only LCA performed on an algal biofuel product featuring HTL, and the authors acknowledged the limitations of their analysis due to limited experimental results.

Recent research exploring in detail the range of products from HTL across a range of conditions has enabled a more thorough analysis rooted in experimental results. The objective of this study is to incorporate the reaction network model developed by Valdez et al. into an LCA that evaluates the life cycle impacts of an algal biocrude oil from HTL across a range of reaction conditions. The study will examine the effect of HTL reaction conditions ranging from 250 to 400 °C and 5–90 min. Given the importance of reducing costs to enable commercialization of algal biofuels, this analysis will evaluate an economic metric in addition to the life cycle metrics of energy return on investment (EROI, or the energy in the algal oil divided by the total life cycle nonrenewable energy inputs), global warming potential (GWP), and occupied land.

In addition to being the first study to analyze the sensitivity of LCA performance to the HTL reaction conditions, this study will compare the viability of two pathways: one that assumes that catalytic hydrothermal gasification (CHG) is used to recover thermal and electrical energy from the aqueous phase products and a second strategy featuring intermediate growth of microbial biomass to boost oil yields, proposed by Nelson et al. While several studies have theorized schemes for onsite energy recovery from the nonproduct portion of the algal biomass either by anaerobic hydrothermal gasification or hydrothermal gasification, none has explored the possibility of utilizing the aqueous phase products from HTL to grow a secondary source of biomass for HTL processing. Nelson et al. demonstrated that Escherichia coli (E. coli) can be grown on the aqueous phase products from HTL, and Valdez et al. demonstrated recovery of crude bio-oil from E. coli biomass via HTL with yields similar to those of algae (i.e., 29% yield for E. coli compared to 38% for Nannochloropsis sp. for the same conditions). Therefore, the proposed regrowth pathway could incorporate cultivation of microbial biomass such as E. coli to recycle carbon and thereby yield more oil per unit of initial algal biomass. Growth of microbial biomass on the aqueous phase could also serve as a preprocessing step enabling nutrient recycle back to the algae pond, as other HTL research has shown that direct recycle of aqueous phase to algae growth operations can be problematic due to suspected toxicity and/or nutrient availability.

The Energy Independence and Security Act of 2007 (EISA) has targeted the production of 36 billion ethanol-equivalent gallons of biofuel annually by 2022. Of those targets, 21 billion gallons are to be non-corn starch-derived “advanced biofuels”. To qualify as an advanced biofuel, the final product must have 50% less life cycle greenhouse gas emissions relative to conventional fossil gasoline. This analysis will therefore examine the life cycle performance of the biocrude oil produced by HTL in the context of these policy objectives.

**MATERIAL AND METHODS**

A challenge of conducting biofuel LCAs is that results and data from multiple sources must be aggregated into a single model with the premise that the results derived from the independent scenarios remain valid in the integrated system. In some key areas (i.e., energy recovery schemes and nutrient recycle), these assumptions must remain in question until they have been examined experimentally. This study features an interdisciplinary collaboration between experimentalists and environmental systems analysts in an attempt to derive results that are realistic. Not all aspects of the model have been verified in the laboratory, as processes such as algae cultivation and harvesting remain the specialty of others and have technical challenges that are independent of oil extraction and conversion.

**Experimental Data Sources Used.** This research builds upon work performed by Valdez et al. characterizing the products from HTL and a recent study featuring a model that predicts the biocrude yield of *Nannochloropsis* sp. across the two-dimensional design space of HTL performed in 4.1 mL batches between 250 and 400 °C and from 5 to 90 min. The model also calculates the yield of other HTL product fractions, such as gas, solids, and aqueous phase products. Accurate information about the aqueous phase is particularly important to this analysis as its constituents will be used either for energy recovery via gasification for the CHG pathway or growth of secondary *E. coli* biomass for the regrowth pathway. Details about these two pathways will be explained later in the report. Data from Valdez et al. allows for determination of the amount of carbon remaining in the aqueous phase and specifies the fraction that is organic versus inorganic.

In addition to the data set characterizing HTL, this analysis also incorporates results from Nelson et al. for modeling the secondary growth of microbial biomass. Nelson characterized E. coli growth on the aqueous phase products from an HTL reactor, specifically the fraction of organic carbon removed as a function of the level of dilution. A separate study demonstrates the feasibility of converting the *E. coli* to biocrude via HTL. The microbial biomass exhibited biocrude yield of approximately 29% compared to a yield of 38% from *Nannochloropsis* sp. from HTL at the same single set of reaction conditions. There is no model that can predict a detailed product distribution from HTL of *E. coli* over the full design space of 250–400 °C and 5–90 min, so this study will assume the oil yield and product distribution are the same for *E. coli* as the algal biomass. The elemental composition of the *E. coli* and algae will be used to determine the mass balance of elemental flows through the biorefinery.

**Data Sources for Life Cycle Modeling.** The upstream processes of algae cultivation and dewatering were based on the operational assumptions outlined by Frank et al. in the Greenhouse Gases, Regulated Emissions, and Energy Use in Transportation (GREET) model published by Argonne National Laboratory and a subsequent report including collaboration from the National Renewable Energy Laboratory and the Pacific Northwest National Laboratory. This harmonized study by Davis et al. assimilates data from multiple research institutions. Although it does incorporate some assumptions that have not been validated empirically, it provides a foundation that has been cross-examined by the collaborators. To align with the
The CHG pathway assumes energy can be recovered from the organic carbon within the aqueous phase solution by catalytic hydrothermal gasification. In this process, the nutrients are, in principle, able to be nearly entirely recycled while the organic carbon is converted to biogas.\(^\text{19}\) Combustion of the biogas in a combined heat and power (CHP) generator allows for onsite production of thermal and electrical energy, thereby reducing fossil energy imports. Modeling of CHG is based on the study by Frank et al., which assumes 33% electrical efficiency and 76% total efficiency for the CHP generator.\(^\text{19}\) This energy recovery scheme will be used for the CHG pathway, but an alternative regrowth pathway featuring secondary biomass cultivation for boosted oil yields is also explored, as will be discussed later in this study.

The biocrude obtained from HTL is assumed to be upgraded by hydrotreating. The crude oil is heated and processed with hydrogen for the removal of O and N as H\(_2\)O and NH\(_3\), respectively.\(^\text{19,31}\) The hydrogen demand is modeled on the formula outlined by Li and Savage.\(^\text{32}\) The product from this stage represents the functional unit for this analysis, a liter of upgraded algal oil.

**Modeling Framework.** Figure 1 illustrates the structure of the core model and the differences between the CHG pathway and the regrowth pathway. Mass balances around each of the process units (represented by rectangles in Figure 1) serve to track nutrient flows and calculate energy and water demand. The core model was assembled in Microsoft Excel but featured intermediate data sets that were derived in MATLAB.

Determining how the algal biomass was altered after undergoing the HTL process was crucial to the model. To determine the yields of the product fractions from the HTL reactor, a predictive model derived by Valdez et al. was developed by solving a system of first-order differential equations to fit replicate sets of experimental data.\(^\text{34}\) Valdez et al. used a constrained nonlinear solver in MATLAB to minimize the value of the squared difference between the experimental and predicted values. The resulting function was implemented to predict the distribution of the original algal biomass into the HTL products of light biocrude, heavy biocrude, solids, gas, and aqueous phase constituents. Details about this distribution can be found in the aforementioned harmonized study, the algal biomass is assumed to be grown in an open pond with circulation provided by a paddle wheel mixer, and flue gas is assumed to be the CO\(_2\) source. Although operating assumptions and parameters were modeled based on GREET and Davis et al., an independent Excel-based analysis was conducted to enable expansion of the model into the full HTL design space analyzed in this study and exploration of the microbial regrowth pathway. The life cycle impact conversion factors can be found in the Supporting Information.

The thermal energy input requirements for HTL are based on the assumption that the fluid is heated from 25 °C to the final temperature (which ranges from 250 to 400 °C), and the final enthalpy is estimated using that of a saturated water vapor. The calculation also assumes that the enthalpy of the algal biomass is half that of the water and that 85% of the heat is recovered by a heat exchanger during the cooling and expansion step. The model assumes that a semicontinuous separation tank after the HTL reactor allows the solid, aqueous, and organic phases to naturally phase separate without the need of a solvent.

Algae yield is modeled as 13.2 g m\(^{-2}\) day\(^{-1}\), which is also in alignment with the report by Davis et al. when seasonal variation in productivity is considered. The pond is harvested at a concentration of 0.5 g L\(^{-1}\) and then concentrated to 10 g L\(^{-1}\) by flocculation and settling. Dissolved air flotation and centrifugation accomplish secondary dewatering to ultimately achieve a final concentration of 150 g L\(^{-1}\). This concentration deviates from that assumed by Davis et al., 200 g L\(^{-1}\), but it aligns with the experimental conditions used by Valdez et al. The authors found that the yields of the HTL product fractions were not significantly affected by the concentration of the incoming algae slurry over this small range.\(^\text{20}\) A consequence of this lower concentration is that a greater volume of fluid must be processed, which increases the energy input requirements. Another deviation of this model from that of Davis et al. is the use of *Nannochloropsis* sp. which is the biomass source used by Valdez et al. in the generation of their predictive model. The biomass is therefore assumed to contain 14% lipid, 59% protein, and 20% carbohydrates\(^\text{20}\) to be consistent with the experimental work.

Figure 1. Sankey diagram illustrating the mass flows of carbon, nitrogen, and phosphorus for (A) the CHG pathway, which assumes energy recovery from the aqueous phase products of HTL via catalytic hydrothermal gasification, and (B) the regrowth pathway featuring cultivation of a secondary biomass, *E. coli*, on the nutrients available in the aqueous phase products that can be processed via HTL to boost the net oil yield. Both figures show mass flows for 350 °C and 60 min reaction times. The mass flows shown for water recycling reflect the fraction of biomass that is recirculated with the water due to imperfect separation.
Solid products, though always in low yields, are more abundant at low temperatures and short reaction times. These are treated as unconverted biomass and therefore assumed to have the same composition as the original biomass fed into the HTL reactor. Gas yields are also low, and the gas produced during HTL is assumed to be entirely CO₂, which is consistent with other studies for gasification of biomass, particularly at lower temperatures. At higher temperatures, especially above the critical temperature of 374 °C for water, hydrocarbons such as methane, ethylene, and ethane are present. At the highest temperature considered, 400 °C, the gas phase is still primarily CO₂. The elemental composition of the biocrude constituents is well characterized, meaning the elemental composition of the aqueous phase products can be calculated by mass balance. The Dulong formula was used to estimate the higher heating value (HHV) of the biocrude oil based on its elemental composition.

Only a portion of the carbon in the aqueous phase is available for gasification or secondary biomass growth, and this fraction depends on the reaction conditions. For example, more of the carbon is inorganic at the more severe reaction conditions, with a maximum inorganic fraction of 47% observed at the highest temperature and longest duration. The study by Valdez et al. reported the fraction of carbon utilized by the cell culture. The maximum organic carbon utilization (45%) occurred with E. coli growing in 10 vol % aqueous phase medium. The effect of this value on results is explored in a sensitivity analysis included in the Supporting Information. Growth in concentrations above 40 vol % aqueous phase was found to be too toxic for E. coli, a higher tolerance threshold than has been exhibited by algae.

It is assumed that cultivating E. coli in a fed-batch culture results in a harvested concentration of 40—60 g L⁻¹, and the microbial biomass can be combined with algal biomass in the centrifugation step used for dewatering the algae for concentration to 150 g L⁻¹ prior to conversion in the HTL reactor. Fed-batch cultures of E. coli have been demonstrated to grow to concentrations denser than those projected in this analysis, with measurements in excess of 100 g L⁻¹ reported.

The C:N:P ratio of the aqueous phase solution compared to that of the E. coli indicates that carbon will be the limiting nutrient, which was confirmed by Nelson et al. in their analysis. Of the fraction of organic carbon utilized by the bacteria, a portion is respired as CO₂ during cell maintenance rather than being synthesized into biomass. Modeling for this study was based on the biomass yield to substrate utilization ratio for carbon-limited E. coli reported by Chen and Strevett.

This analysis assumes that there will be no energy recovery from CHG in the regrowth pathway. Recall that the E. coli consume at most 45% of the organic carbon, meaning the majority is still theoretically available for energy recovery via a technology such as CHG or anaerobic digestion. However, unlike the standard pathway, the aqueous phase products were diluted and cooled for cultivation of the microbial biomass. The amount of energy required to reheat and concentrate the solution to the level necessary for CHG was determined to be in excess of the potential energy recovered.

The elemental mass flows through each of the processes were calculated by mass balance. Recall that the aqueous phase composition was calculated by difference given the biocrude and biomass compositions and assumptions about the solid and gas products from HTL. An illustration of the mass flows for the elements C, N, and P is shown for the CHG pathway in Figure 1a and for the E. coli regrowth pathway in Figure 1b. The mass flows shown in Figure 1 represent baseline HTL conditions of 350 °C and 60 min.

HTL reactions at 350 °C for 60 min are commonly used in the literature, so this study will compare the life cycle impacts at these baseline conditions to those across the full design space (250—400 °C, 5—90 min). Uncertainty was estimated by implementing a Monte Carlo simulation using the RiskSim Excel plugin tool with triangular...
probability distributions and 10,000 trials for each simulation. Details about the parameters and the range of values incorporated into the simulation can be found in the Supporting Information.

Cost assumptions were adapted from the techno-economic model implemented in the harmonized study published by Davis et al.38 Each line item in the harmonized model was normalized by the operational parameters of their baseline facility and then classified into three groups: costs that scale in proportion to the (1) pond size ($ ha−1 yr−1), (2) algal biomass throughput ($ kg−1 biomass), and (3) total oil produced ($ L-oil−1). Costs for the HTL and CHG reactors39 were added to the line items of the harmonized model, while the cell rupture, solvent extraction, and anaerobic digestion costs were removed. Capital costs were annualized using straight line depreciation, assuming a 35% tax rate and no residual value at the end of the equipment lifetime. Depreciation schedules were determined using Davis et al.38 based on equipment lifetime: 7 years for general equipment, 20 years for power-related equipment, and 30 years for plant-related costs. While other models have used the modified accelerated cost recovery system (MACRS) to depreciate equipment, the straight-line depreciation method was chosen to spread costs evenly over each year. A table showing the financial modeling details can be found in the Supporting Information.

■ RESULTS AND DISCUSSION

Catalytic Hydrothermal Gasification Pathway Results. The EROI for the CHG pathway is depicted in Figure 2a for the full range of HTL reaction conditions. These results illustrate the significance of the HTL reaction conditions on the life cycle energy balance, with EROI results varying from as low as 1.2 (at 250 °C, 5 min) to as high as 1.9 (at 250 °C, 50 min, marked with a black square). The GWP results (included in the Supporting Information) track closely to the EROI, with the minimum value of 1.0 kg CO2e L-oil−1 occurring at 250 °C and 42 min. The highest oil yield from HTL is 42%, occurring at 400 °C and 5 min, but at the higher temperatures a greater portion of the biomass is also converted to CO2. Furthermore, an increased oil yield results in less of the initial algal biomass ending up in the aqueous phase solution where it could be converted into useful energy via CHG. Avoiding electrical energy inputs in particular is beneficial in terms of the GWP and EROI impact metrics, as electricity has a relatively high life cycle energy burden. The thermal and electrical energy produced by CHG is consumed on site, thereby reducing the amount of fossil energy that is required to be consumed. There is no surplus electricity sent back to the grid.

A biorefinery featuring HTL reaction conditions with relatively high temperatures and long reaction times results in an aqueous phase containing a higher fraction of carbon in the inorganic form rather than organic. Inorganic carbon is not available for energy recovery via CHG which is why the EROI drops significantly as temperatures and reaction times are increased.

Figure 2b illustrates the effect of the HTL reaction conditions on the economics of the biorefinery. The lowest costs occur at reaction conditions of 400 °C and 5 min (marked with a black square), where the oil would cost $1.65 L-oil−1. The fact that the optimum occurs at this point indicates that reducing the amount of infrastructure required plays a more important role in the economics than reducing the amount of energy inputs required, which occurs at conditions where more CHG energy can be produced such as the optimum indicated in Figure 2a. This cost represents a 9% reduction compared to the cost of $1.81 L-oil−1 at standard conditions (350 °C, 60 min). Conditions with low temperatures and short reaction times, conversely, have significantly higher costs because a relatively large portion of the initial biomass remains as solids, and therefore, the oil yields are low. The land occupation metric tracks closely with the economic results because both are driven primarily by the size of the pond required for cultivation. Compared to standard conditions, optimizing for the highest oil yield reduces the land footprint by 11%.

Figure 3 compares the four metrics over three sets of reaction conditions: standard conditions (350 °C, 60 min), optimal conditions for the EROI metric (250 °C, ~60 min), and optimal conditions for economics (400 °C, 5 min). In this figure, the EROI metric has been inverted to be displayed instead as EROI−1, so that for each of the four metrics a lower number is preferable. A design trade-off is present then because the second set of conditions has the lowest energy input requirement but 7% greater cost compared to the third set of conditions. To minimize the cost of producing algal oil using HTL, conditions that maximize oil yield should be sought. To maximize the EROI or minimize the GWP, conversely, conditions that produce high proportions of aqueous phase solution that is rich in organic carbon are ideal. Maximizing the oil yield does offer energy savings by reducing the amount of algal biomass required to produce the functional unit and therefore the energy inputs associated with cultivation. At conditions with relatively high yields of high quality aqueous phase, the amount of energy that can be recovered via CHG more than offsets the increased energy inputs for cultivation. A figure in the Supporting Information illustrates this phenomenon.

E. coli Regrowth Pathway Results. The previous results illustrate the trade-off between high oil yields (which minimizes the cost and land footprint) and high energy recovery via CHG of the aqueous phase (which minimizes the EROI and GWP).
Switching to a pathway featuring *E. coli* regrowth on the aqueous phase rather than energy recovery essentially amplifies this trade-off. That is, using the aqueous phase to produce additional microbial biomass (which is then converted to oil along with the algal biomass via HTL) boosts the total oil yield per unit of algal biomass. Doing so has a drawback because it eliminates the possibility of energy recovery via CHG.

The greatest boost to the oil yield, 21%, occurs at conditions of 250 °C and reaction times near 60 min where the aqueous phase product fraction is large and the fraction of inorganic carbon present in the aqueous phase is low. Adding *E. coli* regrowth in these conditions would increase the total oil yield from 39% to 46% of the initial algal biomass. The greatest total oil yield for the regrowth pathway occurs at 400 °C and 5 min. At these conditions, the amount of boost is less (just 11%), but the baseline is higher so the initial oil yield of 42% is elevated to 47% of the initial algal biomass.

Given that onsite energy recovery is not considered for the regrowth pathway, the conditions for maximum net oil yield also correspond to the best performance for the four metrics considered in this analysis. The GWP results track closely with the EROI results, and the land occupation results track closely with the economic results, as was the case with the standard pathway. By utilizing microbial regrowth, the land footprint could be reduced by 10% relative to the optimal scenario for the CHG pathway or 21% relative to standard conditions for the CHG pathway. Figure 4a and b show contour plots for the same result metrics that were reported in Figure 2 but for the regrowth pathway rather than the CHG pathway. The optimal sets of conditions for the EROI and cost metrics are represented by a black circle for clarity.

Figure 5 compares two sets of reaction conditions for each of the two pathways. The first set of reaction conditions is the standard scenario (350 °C and 60 min), and the second set is the optimal for the regrowth pathway (400 °C and 5 min). These results show that the lowest-cost scenario for the standard pathway can be further reduced from $1.65 L^{-1}$ to $1.61 L^{-1}$ by incorporating *E. coli* regrowth. Doing so reduces the EROI by 44% and increases the GWP 88%, again due to the elimination of onsite energy production via CHG.

The greenhouse gas reduction threshold to qualify as an advanced biofuel for the EISA production targets require a 50% reduction in GHGs relative to conventional fossil fuels. Adjusting for the energy intensity of the upgraded algal oil, the equivalent GWP target is shown in Figure 5 by the dashed line. The results show that the increase caused by switching from CHG to instead facilitate *E. coli* regrowth could jeopardize the fuel’s qualification as an advanced biofuel.

Table 1 summarizes the optimal conditions for each of the metrics considered in this analysis.
This study analyzed only the use of wild type *E. coli* for boosting oil yields via a regrowth pathway. In the corresponding model, over half of the organic carbon present in the aqueous phase is unused, which provides a fertile venue for further research on improving biomass growth with the aqueous phase. Microbial species other than *E. coli* could correspond the same utility. Examples that have been examined in aqueous phase. Microbial species other than *E. coli* were to be converted into microbial biomass were to be grown on the aqueous phase without dilution, it is possible that the carbon concentration would be increased from the baseline (45%) to 75% or 100%, for that could be possible is 1.0, providing only as much energy in the material should be used prudently. For example, if the fraction of organic carbon that could be converted into microbial biomass were to be increased from the baseline (45%) to 75% or 100%, for example, the cost of algal oil could be further reduced from $1.61 to $1.55 or $1.50 L-oil$^{-1}$, respectively.

**Significance of Aqueous Phase Recovery.** Recovery of aqueous phase products from HTL is instrumental to the overall life cycle performance and economics of the biorefinery, so the material should be used prudently. For example, if nothing were done with the aqueous phase, the greatest EROI that would be possible is 1.0, providing only as much energy in the algal oil as was invested to produce it. By using CHG, conversely, the EROI could be increased to 1.9. If CHG of the aqueous phase solution is feasible, it would be an attractive option to realize the energy return on the investment and increase the carbon footprint significantly. This analysis did not consider energy recovery from the spent aqueous phase, that is, the solution remaining after *E. coli* growth that still contains valuable nutrients and organic carbon. If a microorganism were able to be grown on the aqueous phase without dilution, it is possible that the carbon concentration would be sufficient to recover energy as was modeled for the CHG pathway. Alternatively, it is possible that recycling the spent aqueous phase directly to the pond could induce mixotrophic algae growth providing another means for boosted yields, if the species were capable of such metabolisms.

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### Table 1. Summary of Optimal Conditions for Each of the Metrics Considered in This Analysis and for Both Pathways Examined

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<th>CHG pathway</th>
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<td>opt. conditions</td>
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<td>400 °C, 5 m</td>
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</tbody>
</table>

**The most likely values are indicated in bold, and the 25–75% range of Monte Carlo simulation results are shown in brackets.**

### References


(19) Frank, E. D.; Elgowainy, A.; Han, J.; Wang, Z. Life cycle comparison of hydrothermal liquefaction and lipid extraction pathways to renewable diesel from algae. *Mitigation Adapt. Strategies Global Change* 2013, 18, 137–158.


