Industrial Symbiosis: Corn Ethanol Fermentation, Hydrothermal Carbonization, and Anaerobic Digestion

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Dedication

This thesis is dedicated to my family. I wouldn't be the person I am today without your love and support.

Abstract

The production of dry-grind corn ethanol results in the generation of intermediate products, thin and whole stillage, which require energy-intensive downstream processing for conversion into commercial co-products. Alternative treatment methods, specifically hydrothermal carbonization of thin and whole stillage coupled with anaerobic digestion were investigated to determine if they provide an opportunity to recover some of this value. By substantially eliminating evaporation of water, reductions in downstream energy consumption from 65-73% were achieved, while hydrochar, fatty acids, treated process water, and biogas co-products were generated, providing new opportunities for the industry. Processing whole stillage in this manner produced the four co-products, eliminated centrifugation and evaporation, and substantially reduced drying. With thin stillage, all co-products were again produced, as well as a high quality animal feed. Anaerobic digestion of the undiluted aqueous product stream from thin stillage hydrothermal carbonization reduced chemical oxygen demand (COD) in this product stream by more than 90% and converted 83% the initial COD to methane. Internal use of this biogas could entirely fuel the HTC process and reduce natural gas overall usage.

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Part I. Literature Review

1.1 Hydrothermal Carbonization (HTC)

1.1.1 Introduction and Origins

Hydrothermal carbonization (HTC) is a thermochemical process that utilizes temperature and pressure to increase the carbon content of an organic material in the presence of water. HTC is similar to naturally occurring geological processes that result in the formation of coal, and was first investigated in the early 1900's. Modern HTC reactions occur at moderate temperatures of 180-250°C (Mumme et al., 2011) and autogenous pressures in a closed vessel. Reaction times vary substantially depending on the substrate and heating mechanism, but generally are on the order of a few hours. The process generates a carbonized (increased C:O ratio) coal-like material known as hydrochar along with an aqueous fraction containing soluble products that can easily be separated by filtration. Carbonization reactions at aforementioned conditions primarily occur through a dehydration mechanism, generating little gaseous products; nevertheless, decarboxylation resulting in CO_2 production can contribute to carbonization as well.

Other thermochemical conversion technologies such as hydrothermal liquefaction, pyrolysis, and gasification employ higher temperatures, and as a result reaction products are substantially changed. Hydrothermal liquefaction, which produces a petroleum replacement bio-oil or bio-crude, utilizes a temperatures between 280-370°C with pressures of 10-25 MPa (Toor et al., 2011). Conversely, pyrolysis occurs above the supercritical point of water between 400-500°C in the absence of oxygen (Libra et al., 2011). Pyrolysis produces a mixture of biochar, bio-oil, and gases; specific distribution of

these products depends on reaction conditions and substrate characteristics (Balat et al., 2009). Gasification employs the highest temperatures ranging from 600-1000°C in the presence of an oxidizing agent (Kumar et al., 2009). Gaseous products from gasification known as syngas primarily consists of CO, H_2, CO_2, and CH_4 (Wang et al., 2008). Syngas can be used directly for heat and power or upgraded catalytically to chemicals or liquid fuels. In general, pyrolysis and gasification are conducted on dry substrates.

1.1.2. Hydrothermal Carbonization of Insoluble Biomass

Early hydrothermal carbonization research was conducted with lignocellulosic materials to develop a better understanding of the natural coalification process. Bergius and Spect began heating peat and cellulose in water under pressure in the early 1900s. Bergius noted that many others had tried to heat cellulose or wood in order to make a product similar to coal; never, however, had reactions been conducted in liquid water under pressure. In his Nobel lecture of 1932, Bergius explained that at temperatures between 290-350°C an exothermic reaction of two parts cellulose would yield two parts CO_2, five parts water, and a powdery substance that was essentially $C_{10}H_8O$ (Bergius, 1932).

A 1960 research article summarized and built upon many of the early HTC findings (Schuhmacher et al., 1960). In the article it was reported that cellulose (44% carbon) was converted to a coal-like material that was 81% carbon in a 39% yield. This high level of carbonization was only achieved at severe conditions, i.e., 340°C for 3 hours, and produced a 20% yield of CO_2. At less extreme conditions, i.e., 225°C for 3 hours, a 63% yield of 51% carbon material was produced yielding 1% CO_2. Similar

carbonization of lignin and wood was achieved and the authors concluded that both cellulose and lignin could participate in coal formation. In addition, reactions with glucose generated a material very similar in composition to that created from cellulose, which supported the conclusion that cellulose was hydrolyzed to intermediate products.

As hydrothermal carbonization research progressed, the focus shifted from understanding coalification to understanding and quantifying soluble intermediate products. Bobleter was a part of numerous studies that sought to answer these questions, and made use of a sophisticated continuous flow system (Bobleter et al., 1976). The optimum temperature and flow rate for cellulose degradation to glucose was shown to be 265°C at 12 cm^3/min, yielding 52% glucose. At flow rates below 11 cm^3/min, glucose yields decreased whereas concentrations of 5-hydroxymethylfurfural and furfural increased (Bonn et al., 1983). Lignin and hemicellulose were degraded at 180°C. In order to overcome this temperature discrepancy a two-stage HTC process was investigated with aspen wood. In the first hydrolysis stage (180°C), 43% of the starting aspen wood was degraded to soluble products. The second stage (265°C) converted 52% of the remaining material to soluble products (Bonn et al., 1983). At the time, promising applications of this research included a method to pretreat lignocellulosic material for ethanol production (Hörmeyer et al.,1988). In contrast, recent efforts have revisited production of insoluble hydrochar from lignocellulosic materials for use as a carbon-neutral coal replacement in addition to numerous other applications (Titirici et al., 2007; Titirici & Antonietti, 2010; Funke et al., 2010).

1.1.3 Hydrothermal Carbonization of Soluble Biomass

Considerable effort has been directed towards HTC research on soluble saccharides such as sucrose or glucose, both to investigate novel syntheses of carbon nanostructures and to determine HTC mechanisms. Hydrochar yields from glucose and sucrose are highly dependent on initial concentrations and reaction conditions, but in general, yields have been shown to be between 20-40% (w/w) (Sevilla & Fuertes, 2009). Saccharide hydrochars are made up of monodispersed microspheres. More specifically, glucose hydrochars contained 1-2 μm diameter microspheres (Mi et al., 2008), whereas sucrose hydrochars contained 1-5 μm diameter microspheres (Wang et al., 2001). The size of the microspheres within each respective range was dependent on HTC reaction conditions. Research areas that have expanded upon these observations include 1) attachment and encapsulation of metal on the carbon network (Huang et al., 2007), 2) carbon networks structured using sacrificial metal (Deng et al., 2006), and 3) co-deposition of carbon and metal followed by oxidative removal of carbon generating hollow metal spheres (Yang et al., 2007). These specialized materials all produced via HTC of saccharides, have a multitude of commercial application as catalysts or electrodes.

The vast amount of HTC research on simple saccharides in water has provided enough information to piece together a generally accepted mechanism for the carbonization and hydrochar building processes. The first step that occurs with insoluble polysaccharides is hydrolysis of glycosidic linkages, resulting in soluble oligo, di, and monosaccharides. Autoionization of water at hydrothermal conditions catalyzes initial

hydrolysis, and further hydrolysis is catalyzed by hydronium ions from organic acids (formed by fragmentation on saccharides) (Garrote et al., 1999). Monosaccharides that are not fragmented can be intramolecularly dehydrated, primarily forming furans, i.e, furfural and 5-hydroxymethylfurfural. Dehydration products and monomers then participate in condensation and polymerization reactions, which occur through intermolecular dehydration or aldol condensation, forming soluble polymers (Sevilla & Fuertes, 2009). The appearance of oxygen groups such as C=O can be attributed to intramolecular dehydration of vicinal hydroxyl groups forming enols that tautomerize into ketones and aldehydes. Intermolecular dehydration contributes to aromatization of soluble polymers, developing aromatic clusters. It has been suggested that the formation of hydrochar microspheres follows a nucleation growth mechanism consistent with the LaMer model (Sun & Li, 2004). Therefore, when aromatic clusters reach a saturation point, a burst nucleation occurs and the resulting particle grows outward by diffusion of reactive moieties. Species in the solution are linked to the growing microsphere through reactive oxygen functional groups present on both surfaces, i.e., hydroxyl, carbonyl, and carboxylic. This hypothesis is supported by the presence of stable ether and pyrone groups in the core of the resulting hydrochar microsphere (Sevilla & Fuertes, 2009). At the conclusion of HTC reactions two principle products remain: insoluble hydrochar microspheres, and a soluble fraction consisting of small organics such as organic acids, furans, and aldehydes.

1.1.4 Hydrothermal Carbonization of bioenergy feedstocks and liquid waste streams

The presence of water is vital for HTC reactions and this makes it uniquely suited for processing high moisture-content biomass or waste streams that also contain high levels of organic carbon. With increasing concerns about global warming, emphasis has been placed on moving away from fossil fuels to renewable energy sources. Energy or chemicals derived from renewable biomass have been projected to play a role in this transition (Perlack et al., 2005) , but the high water content in biomass presents a problem. Evaporation of water requires 1100 BTU/lb, and this can result in a negative energy balance for renewable biomass resources that require extensive drying. HTC processing eliminates all or much of the drying requirement by allowing reactions to occur with the material as received followed by the separation of hydrochar by filtration. A number of nonconventional low cellulosic materials have been investigated, including microalgae (Heilmann et al., 2010; Heilmann et al., 2011b), distillers grains (Heilmann et al., 2011a), manures and wastewater sludge (Libra et al., 2011), and anaerobically digested maize silage (Mumme et al., 2011). As previously mentioned HTC research on lignocellulosic materials is ongoing, but because of the relatively high temperatures needed to hydrolyze cellulose, other low-cellulose biomass feedstocks that carbonize at less severe reaction conditions may provide a more advantageous energy balance.

Complex substrates make it difficult to determine mechanistic details about hydrochar generation, but individual macromolecule components, i.e., carbohydrates, triglycerides, and proteins, have provided some insight. Carbohydrates are considered to be the primary reactant for HTC and are central to hydrochar formation. Proteins are

hydrolyzed into constituent peptide fragments and amino acid monomers. These protein fragments may undergo Maillard reactions with carbohydrates that contain a reactive aldehyde group, giving rise to a variety of small heterocyclic compounds that can be integrated into hydrochars through the mechanism described in section 1.1.3. Additionally, proteins add insoluble mass that is subsequently collected with hydrochar through denaturation and insolublization (Heilmann et al., 2011a). In contrast to both carbohydrates and proteins, triglycerides do not contribute to hydrochar formation. Instead, triglycerides are hydrolyzed, and the resulting fatty acids are sorbed to the insoluble hydrochar product. Although it remains unclear how fatty acids are sorbed to hydrochar, they have been shown to be readily extractable with an organic solvent (Heilmann et al., 2011b). It should be noted however, that even seemingly analogous substrates can have different HTC outcomes with respect to hydrochar composition and yield.

In order for HTC to be an industrially relevant bioprocessing or waste treatment operation, both the resulting hydrochar and aqueous product must be useful. Hydrochars, due to carbonization, have heating values on par with bituminous coal (Heilmann et al., 2010), and have a default application as renewable solid fuel. Other higher value applications have been suggested for hydrochars, including use as a soil amendment, concrete or asphalt additive, activated carbon analog, and CO_2 sorbent (Libra et al., 2011). Structures of hydrochars derived from complex biomass or waste streams may not be as predictable as those from simple saccharides, yet these complex substrates,

particularly those containing proteins, could generate useful hydrochar products as a result of nitrogen functionalities incorporated into the resulting hydrochars.

Aqueous products generated along with hydrochars have historically received less attention, but in the context of bioprocessing or waste treatment, equal emphasis must be placed on these products so one waste stream is not traded for another. Carbon contained in hydrochars is generally between 40-60% of that in the starting biomass, leaving most of the remaining carbon in the form of soluble products in the liquid fraction minus minor amounts of CO_2 produced. Additionally, it has been shown that the majority of nitrogen and phosphorus in the system remains in the aqueous product (Heilmann et al., 2011a; Heilmann et al., 2011b). Direct uses of these liquid products have included water and nutrient recycle for algae growth and liquid fertilizer for terrestrial crop production. Another option is to anaerobically digest the liquid fraction, thereby treating the stream and generating biogas that could be used to run the HTC process.

1.2 Anaerobic digestion

1.2.1 Introduction

Anaerobic digestion is a process in which a community of microorganisms cooperatively breakdown organic material in the absence of oxygen, forming biogas primarily composed of methane and carbon dioxide. Industrially, anaerobic digestion is used as a method to reduce organic carbon levels in a range of wastewaters (e.g., sewage, manure, and brewery) while generating biogas fuel (i.e., methane).

Conversion of soluble and insoluble organic molecules into biogas requires multiple organisms and metabolic steps. First, macromolecules are hydrolyzed into

simple soluble products by extracellular enzymes. For complex substrates, this can be a rate-limiting step (Li & Noike, 1992). Next, the products from the first step are fermented into short-chain organic acids, alcohols, CO_2, and hydrogen. Specifically, production of short chain fatty acids is known as acidogenesis. Intermediate volatile fatty acids and alcohols are then converted by hydrogen- producing acetogenic bacteria into acetate, CO_2, and hydrogen in a step called acetogenesis. Thermodynamically, acetogenesis is unfavorable if the partial pressure of hydrogen in the system is above 1×10^{-3} atm (Zinder, 1990). Additionally, a small amount of acetate is generated from CO_2 and hydrogen by homoacetogens (Mackie & Bryant, 1981). Methanogenesis is the final step in the process, in which methanogenic *Archaea* convert acetate and CO_2 and hydrogen into methane and CO_2. Methane production is used as an indicator of overall system performance because methanogenesis is considered to be the major rate limiting step in digestion. Acetotrophic methanogens account for nearly three quarters of the total methane production The remaining methane is produced by hydrogenotrophic methanogens using CO_2 as the electron acceptor and hydrogen or formate as the electron donor (Khanal, 2009).

1.2.2 Environmental factors and operating conditions

Environmental factors play a critical role in the success or failure of anaerobic systems. Anaerobic systems, and specifically methanogens, are very sensitive to pH deviations from the optimum range of 6.8 – 7.4. Even a moderate change in pH can result in the failure of a reactor. A decrease in pH can occur from excessive CO_2 production or volatile fatty acid accumulation. Low pH further contributes to the accumulation of

volatile fatty acids by lowering methanogenic activity, increasing the partial pressure of hydrogen and inhibiting acetogenesis (Khanal, 2009). Similarly, methanogenic activity is inhibited above pH 8 due to an increase in the free ammonia concentration (Chen et al., 2008). Alkalinity mitigates pH fluctuations by buffering anaerobic systems. Most healthy reactors maintain an alkalinity capacity between 1000 – 5000 mg/L as $CaCO_3$ (Tchobanoglous et al., 2003), and a volatile fatty acid to alkalinity ratio of 0.1 – 0.25 (Khanal, 2009).

Temperature is another environmental factor that influences the overall performance of anaerobic systems. Optimum temperature ranges for methanogens are of 5 – 15°C, 35 – 40°C, and ~55°C for psychrophilic, mesophilic, and thermophilic methanogens respectively. Temperatures outside these optimal ranges are survivable but result in decreased methanogenic activity (Lettinga et al., 2001). Currently most commercial reactors are operated under mesophilic conditions. Mesophilic reactors are more stable and have shorter startup times than thermophilic reactors (Khanal 2009). Nevertheless, methane production rates are 25 – 50% higher in thermophilic reactors than those in mesophilic reactors. Thermophilic treatment also results in the destruction of pathogens (Smith et al., 2005) and antibiotic resistance genes (Ghosh, Ramsden, LaPara, 2009). Nonetheless, additional heating requirements associated with maintaining thermophilic conditions can negate some of the energy benefits gained from increased methane production.

Anaerobic digestion, similar to other biological processes, requires macro and micronutrients and is subject to inhibition or toxicity. The minimum ratio of nitrogen and

phosphorus related to chemical oxygen demand (COD) (COD:N:P) has been shown to be 350:7:1 for high organic loading rates of 0.8-1.2 kg COD/kg VSS day and 1000:7:1 for organic loading rates of < 0.5 kg COD/kg VSS day (Henze, Harremos, 1983). Other important macronutrients include sulfur, potassium, calcium, magnesium, and iron. Small amounts of micronutrients (i.e., Cr, Co, Cu, Mn, Mo, Ni, Se, V, and Zn) and growth factors (i.e., vitamins, amino acids, purines, and pyrimidines) are also required for microbial growth (Angelidaki & Sanders 2004). Ammonia is beneficial at low levels, but has been shown to be inhibitory above free (unionized) ammonia concentrations of 700 mg/L (I Angelidaki, Ahring, 1994). Likewise, heavy metals, short chain fatty acids (more inhibitory when unionized), and long chain fatty acids can become inhibitory or toxic if certain threshold concentrations are exceeded. Additional sources of toxicity include refractory or recalcitrant organic compounds and sulfide (again, more inhibitory when unionized). It should be noted, however, that the levels of certain compounds that are inhibitory to a given community depends largely on the degree of acclimation to that compound. Multiple studies have shown that with proper acclimation, anaerobic cultures can tolerate levels of various compounds (i.e., ammonia, sulfide, and recalcitrant organic compounds) previously thought to be toxic (Khanal 2009).

1.2.3 Anaerobic digestion of Thin Stillage

Anaerobic digestion of thin stillage, a byproduct of the corn ethanol industry containing approximately 93% water, has attracted interest as a method to treat and recycle process water while generating renewable biogas. Thin stillage has a high organic content ranging between 50 – 100 g/L COD, a low pH of 4-5, and zero alkalinity. It does,

however, contain nearly all of the required nutrients for sustained anaerobic treatment. In the early 1980's Seely and Spindler (1981) and Stover et al. (1983) studied the anaerobic digestion of thin stillage. These studies demonstrated the potential of thin stillage as a feedstock for digestion. One noteworthy finding of Stover et al. (1983) was that at higher thin stillage concentrations with less alkalinity added and influent pH values of (3-5), effluent alkalinity remained normal (4880 mg/L $CaCO_3$). This phenomenon was later explained by Khanal (2009) as in situ alkalinity generation. When protein present in thin stillage is hydrolyzed, it gives rise to an organic acid and ammonia; the ammonia then reacts with CO_2 to form ammonium bicarbonate, which acts as buffer.

Changes in the corn ethanol process that have improved the biodegradability of thin stillage and advances in anaerobic technology have renewed interest in the digestion of thin stillage. A 2008 study by Schaefer and Sung achieved steady-state thermophilic digestion of undiluted thin stillage at hydraulic retention times (HRT) of 30, 20, and 15 days using continuously stirred tank reactors (CSTR). The study indicated that by using biogas generated in digestion a typical 100 million gallon/year ethanol plant could reduce natural gas consumption by 43 – 59%, saving around $10 million/year in energy costs (Schaefer, Sung, 2008). Another 2008 study reported stable thermophilic digestion at a 10 day HRT using anaerobic sequencing batch reactors (ASBR). The average total chemical oxygen demand (TCOD) reduction was 92.2% and a 51% reduction in natural gas requirements was predicted for a 100 million gallon/year ethanol plant. Cobalt supplementation was found to be necessary, however, for prolonged digestion (Agler et al. 2008). In 2012, Andalib et al. advanced the state of the art by successfully digesting

undiluted thin stillage with an HRT of 3.5 days and an organic loading rate of 29 kg COD/m^3 day while reducing TCOD by as much as 88%. At steady state, this equated to the production of 40 L CH_4/ L thin stillage. In order to achieve these results a high rate anaerobic fluidized bed reactor (AFBR) was utilized under mesophilic conditions. Most studies mentioned the potential benefits to the ethanol industry of recycling digester effluent as process water for fermentation, but little research has been done to support this hypothesis. One report analyzed thin stillage digester effluent with HPLC, observing that compounds known to inhibit yeast fermentation were either not present or at low enough levels not to inhibit fermentation, but no actual fermentation trials were performed (Alkan-Ozkaynak & Karthikeyan, 2011). Regardless of the potential energy or water consumption benefits, anaerobic digestion of thin stillage has not been broadly adopted.

1.2.4 Thermal Hydrolysis

Thermal hydrolysis pretreatment, similar to hydrothermal carbonization, has been shown to improve anaerobic digestion. Thermal hydrolysis pretreatment improves digestion by increasing the rate of hydrolysis, which can be the rate-limiting step in the anaerobic digestion of complex organic matter, and by increasing the total amount of organic matter solubilized (Li, Noike, 1992). Both thermal hydrolysis and HTC employ temperature and autogenous pressure in the presence of water. Optimum temperatures for thermal hydrolysis are between 160 – 180°C (Wilson Novak, 2009), which just borders HTC conditions. Above pretreatment temperatures of 180°C, biodegradability has been shown to drop due to the production of recalcitrant Maillard compounds (Bougrier,

Delgenès, Carrère, 2008). The extent of this drop in biodegradation is not well understood, and merits further investigation, especially for substrates outside of wastewater sludge.

1.3 Summary and Research Needs

Hydrothermal Carbonization is a technology uniquely suited for treating high moisture content biomass or waste. Little attention, however, has been paid to the aqueous products created. Coupling hydrothermal carbonization with anaerobic digestion is a novel research area that offers an advantageous way to generate beneficial products but it is untested in practice. With respect to HTC, designed experiments varying reaction conditions must be conducted on specific substrates of interest to accurately determine product yields, elemental distribution within those products, and effects on downstream processing. Thermal hydrolysis pretreatment research has provided some insight as to what might be expected upon the HTC of other wastes but the effect of production and separation of hydrochar on digestion is largely unknown.

Part II. Research Paper

Introduction

Corn ethanol production in the U.S. has risen steadily, reaching 13.9 billion gallons in 2011 and rapidly approaching the amount mandated by the Energy Independence and Security Act (15 billion gallons by 2015). Use of corn in the production of ethanol has been controversial for a number of reasons, including energy and water consumption. Substantial improvements, however, have been achieved with the dry-grind ethanol process, which represents more than 80% of total US production

capacity (http://www.afdc.energy.gov/fuels/ethanol_production.html). Currently, net energy ratios, or energy output/energy input, of 1.61-1.64 have been reported (Liska et al., 2009), and water use has decreased to 3-4 gallons of water per gallon of ethanol produced (Aden, 2007). Nevertheless, further reductions in energy and water use should be possible through alternative downstream processing.

When fermentation has finished and ethanol has been removed by distillation, the residue material is known as whole stillage. Whole stillage (WS) is centrifuged to generate thin stillage (TS) as the centrate and wet distillers grains as the centrifugate. Thin stillage has a high moisture content (>90%) and only a portion of it, 10-50% (Liu & Rosentrater, 2012), can be added back into fermentation as backset due to the buildup and subsequent toxicity of low molecular weight organic compounds, inorganic salts, and solids (Jacques K et al., 1999; Alkan-Ozkaynak & Karthikeyan, 2011). The remainder of thin stillage can be evaporated to obtain condensed distillers solubles, which is generally re-combined with wet distillers grains, dried to a moisture content of <10%, and sold as dried distillers grains with solubles (DDGS), a shelf-stable animal feed and an important co-product for the industry. Energy intensive evaporation and drying of these intermediate products has been estimated to account for 40-45% of the thermal energy and 30-40% of the electrical energy required in the dry grind ethanol production process (Meredith, 2003).

Hydrothermal carbonization (HTC) is a thermochemical process that is especially well-suited for high moisture content biomass materials. Biomass is heated in water in a confined system at subcritical temperatures of generally < 250 ^{o}C. Two products are

created that are separated by filtration: a hydrochar and an aqueous filtrate containing dissolved products. The vast majority of water is removed from the system by filtration and not by high energy evaporation; therefore, net energy output is generally positive. Our recent efforts (Heilmann et al., 2011a) examining HTC of DDGS generated hydrochars with fuel value and other uses, as well as nutrient-rich aqueous filtrate streams. Carbohydrates and proteins were shown to be involved in the chemical formation of the hydrochar, while lipids were hydrolyzed to fatty acids that sorbed to the hydrochar. These fatty acids could be readily extracted from the hydrochar for subsequent use (Heilmann et al., 2011b). Despite these previous results, a lack of predictably exists in HTC reactions due to the complexity of substrates and chemistry. This makes extrapolation to other substrates, even seemingly analogous ones, difficult.

Anaerobic digestion could improve the overall energetics of ethanol manufacturing by displacing a portion of natural gas with biogas and reducing water consumption by allowing recycle back into fermentation. Efficient digestion of undiluted thin stillage has been achieved by others (Schaefer & Sung, 2008; Agler et al., 2008; Andalib et al., 2012), with a hydraulic retention time (HRT) as brief as 3.5 days (Andalib et al., 2012). Furthermore, substitution of biogas could provide a 43-59% reduction in the natural gas requirement for an ethanol plant. It has also been suggested that the effluent from digested thin stillage could be employed as backset without disrupting ethanol production (Alkan-Ozkaynak & Karthikeyan, 2011). Because thin stillage contains diverse nutrients and minerals, only trace cobalt supplementation is likely to be required

for long term digestion (Agler et al., 2008). Despite potential benefits, anaerobic digestion of stillage intermediates has failed to gain traction in the industry.

Integration of HTC and anaerobic digestion could allow for the generation of new products (hydrochar and fatty acids), while anaerobic digestion of the hydrochar filtrate stream could provide biogas and decreased water use. Even so, as determined by investigations of the thermal hydrolysis of wastewater sludge (Bougrier et al., 2008), treatment with temperatures above 190 °C decreased the subsequent digestibility due to formation of Maillard compounds. The extent of this decrease in biodegradability is largely unknown with thermal hydrolysis temperatures above 210 °C, especially for substrates other than wastewater sludge. Additionally, when wastewater sludge undergoes thermal hydrolysis, the entire stream is subjected to digestion, and thus it remains unclear what the effect of generating and removing the solid hydrochar product will have on digestion.

The overall objective of this research was to determine if HTC and anaerobic digestion could be effectively integrated into the dry-grind corn ethanol process to provide hydrochar, fatty acids, biogas, and treated process water. Two separate scenarios are proposed (Figure 1b and Figure 1c) compared to the conventional dry-grind process (Figure 1a). The use of whole stillage for an HTC feed stream has the advantage of completely eliminating the need for centrifugation and evaporation; no animal feed product would remain, however. Compared to conventional processes, the use of thin stillage as an HTC feed stream eliminates evaporation and minimizes drying, while continuing to produce an animal feed product (DDG). Because the current market for

animal feed is strong, more emphasis was placed on the utilization of thin stillage. For this study, the specific objectives included: 1) establish alternative downstream processing methods for stillage intermediates that are less energy intensive, 2) determine if water consumption can be reduced by recycling more process water following HTC or anaerobic digestion, and 3) generate additional co-product options.

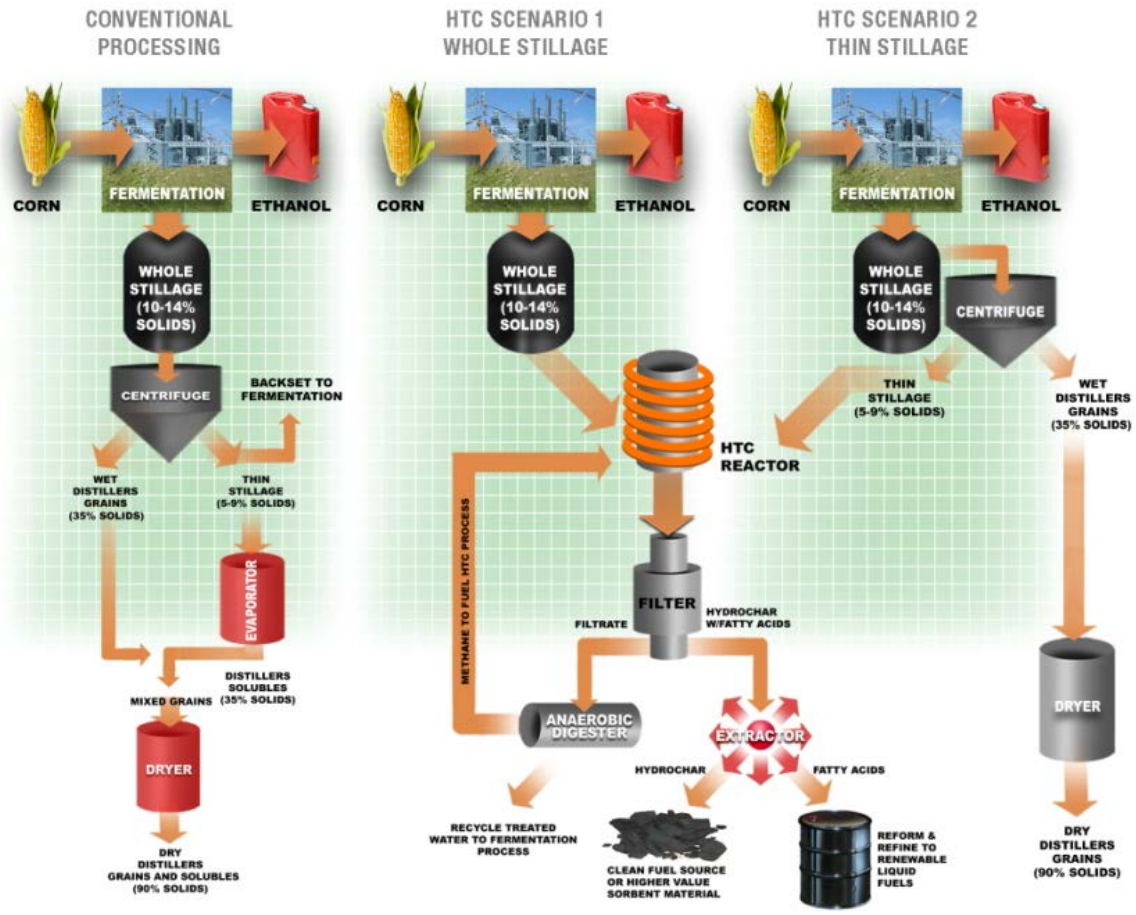

Figure 1. (A) Simplified schematic of a conventional dry-grind corn ethanol plant. (B) Whole stillage scenario wherein centrifugation, evaporation, and a majority of drying is eliminated. (C) Thin Stillage scenario wherein evaporation is eliminated and dry distillers grain can continue to be utilized as an animal feed.

Materials and Methods

Hydrothermal Carbonization Procedure

Procedures for HTC of both whole stillage and thin stillage were the same, with the exception of cooling temperature. Samples of authentic whole stillage and thin stillage were obtained from Al-Corn Clean Fuels (Claremont, MN). All reactions were conducted in a 450 mL stirred stainless steel reactor available from Parr Instruments, Inc. (Moline, IL). Heating was applied using an inductive heating system available from LC Miller, Co. (Monterey Park, CA). Thin or whole stillage were poured into the reactor, stirred at 88 rpm, and heated to 220 ^{o}C for 75 minutes. The pressure in the reactor after the reaction period was 2.34 MPa, and the apparatus was cooled using a fan. When the unit had cooled to at least 35 ^{o}C for thin stillage or 45°C for whole stillage, the reactor was disassembled, the contents were filtered, and the hydrochar was washed thoroughly with distilled water. The sterile filtrate was stored at 4°C, and moist hydrochar was freeze-dried to obtain dry hydrochar. Extraction of fatty acids from the hydrochar was accomplished using methyl t-butyl ether (MTBE). The extracted hydrochar was dried overnight at room temperature at 40 ^{o}C followed by additional drying at < 1 Torr. MTBE was removed from the extract using a rotary evaporator.

Design of Experiments

Reaction conditions examined with thin stillage were temperatures from 200 – 240 ^{o}C, times from 0.5 – 2.0 h, and percent solids values from 5.3 – 9.1 (Table I). Whole stillage experiments were conducted with temperatures from 200 – 230 ^{o}C, times from 0.5 – 2.0 h, and percent solids values from 10.0 – 14.0 (Table II). The three percent solids levels employed with thin stillage were obtained by atmospheric distillation of water from the as-received thin stillage (7.2% solids) until a percent solids level of 9.1% was

achieved. The lowest concentration (5.3% solids) was obtained by dilution of the as-received thin stillage with distilled water. In the course of conducting experiments with whole stillage and despite refrigerated storage, it was noted that percent solids values changed with time. Because of this, an additional sample of whole stillage was freeze-dried, homogenized to a powder, and stored at -20 ^{o}C prior to use. This dry material was reconstituted to 10.0, 12.0, and 14.0% solids by the addition of distilled water and homogenization with a Waring blender.

Hydrochar and Fatty Acid Analysis

Elemental analyses, heats of combustion, and ash contents were determined by Huffman Laboratories, Inc. (Golden, CO). Surface areas of hydrochars were determined by the Chemical Engineering Department, University of Minnesota. HPLC analyses of filtrates were conducted using an Agilent 1200 HPLC equipped with an Aminex-87H column (300 mm x 7.8 mm) at 50 ^{o}C using an isocratic program of 5 mM sulfuric acid and a flow rate of 0.5 mL/min, with 1% sodium propionate as an internal standard. Total fat, fatty acid profiles, and percent triglyceride values were determined by Medallion Labs Inc. (Minneapolis, MN).

Data Analysis

Results from designed experiments were analyzed using JMP Pro 9 software, and the full statistical analysis is available in the appendix A.1.

Substrate and Inoculum Characterization

Prior to digestion experiments, the total solids for the inoculum (see below) was determined gravimetrically at 103° C, while total solids for thin stillage and HTC filtrates

were determined via freeze-drying due to their low initial pH (Angelidaki et al., 2009). Volatile solids levels were determined gravimetrically at 550 °C. Total chemical oxygen demand (TCOD) values were determined using Hach COD Test Kits (ultra-high range, Hach Co., Loveland, CO) according to the procedure of Jirka and Carter (1975). Samples were not blended prior to COD testing as a result of foaming. pH was measured before and after batch experiments using a Thermo Orion 330 pH meter. Alkalinity of substrates was determined by the Soil Sciences Analytical Laboratory at the University of Minnesota.

Anaerobic Digestion Experiments

Biochemical Methane Potential (BMP) assays were used as a test of biodegradability of the HTC filtrate streams and the initial thin stillage, according to the procedure of Owen et al. (1979). No additional nutrients or buffers were added, however. Three dilutions, 35%, 65%, and 100%, of untreated thin stillage and HTC filtrate (also called substrate below) were examined in triplicate. Triplicate controls consisted of inoculum only or 100% HTC filtrate without an inoculum added. An inoculum-to-substrate ratio of 7:3 was employed in all experiments. Inoculum was removed from source reactors (description in A.2) anaerobically and allowed to degas for two days prior to use. Serum bottles (160 mL) were filled with 21 mL of inoculum and fed 9 mL of substrate. Millipore water was utilized in lieu of inoculum or the substrate in the two controls. Immediately after inoculation, bottles were purged with N_2 for two minutes, sealed with butyl rubber stoppers, and allowed to equilibrate at the incubation temperature (37 °C) for one hour before being degassed to initiate the experiment. The

BMP assays were performed at 37 °C while mechanically shaken at 90 rpm. Biogas production and methane production were measured once a day for the first 8 days and another 7 times in the following 21 days (total experimental period of 30 days). Biogas production was measured by the volumetric displacement of the barrel of a wetted glass syringe. Biogas was subsequently exhausted from the assays to re-establish atmospheric pressure. The methane content of the biogas was measured via GC (see A.3) immediately after establishing atmospheric pressure in the bottles. After 30 days the bottles were opened and pH, total solids, volatile solids, and TCOD were measured. Details of the BMP analysis are given in A.4.

Results

HTC Experiments

Hydrothermal carbonization and subsequent filtration of thin and whole stillage resulted in the production of solid carbonized hydrochars, sorbed fatty acids, and a filtrate stream (Tables I and II). The carbon content of thin stillage and whole stillage increased from 44.3% and 46% to center point averages of 63.9% and 65.15%, respectively. Unextracted hydrochars had high heats of combustion and lost ca. 13-15% on extraction, similar to what has been observed in the literature with DDGS as the HTC substrate (Heilmann et al., 2011a). Ash contents of hydrochars were reduced compared to starting stillages, and the ash of thin stillage hydrochar was about six times that of the whole stillage hydrochar. As with HTC of microalgae (Heilmann et. al., 2010) and DDGS (Heilmann et al., 2011a), phosphorus and nitrogen were primarily located in the filtrate streams.

Table I. Reaction conditions, elemental analysis, and yields of thin stillage and resulting extracted hydrochars

Process Conditions			Elemental Analysis[a]						Hydrochar	Fatty Acid	HHV	
Temp (°C)	Time (h)	% Solids	%C	%H	%N	%O	%S	%P	% Yield[b]	% Yield[c]	BTU/lb[d]	% Ash[e]
TS	—	—	44.35	7.85	2.79	37.06	0.29	1.87	—	—	8339	12.9
200	0.5	5.3	60.45	6.83	9.26	22.79	—	—	4.15 (5.7)	46.91	—	—
200	0.5	9.1	61.48	6.62	8.84	22.52	—	—	6.38 (8.9)	45.40	—	—
200	2.0	5.3	66.11	6.68	7.03	19.33	—	—	6.04 (9.0)	66.09	—	—
200	2.0	9.1	65.86	6.54	6.99	19.24	—	—	7.63 (11.3)	42.97	—	—
220	1.25	7.2	65.28	6.49	6.40	—	—	—	6.08 (9.0)	59.09	—	—
220	1.25	7.2	62.50	6.22	6.27	17.43	0.39	1.21	6.09 (8.6)	47.19	12647(14477)	5.23
220	1.25	7.2	63.98	6.34	6.31	—	—	—	6.08 (8.8)	51.30	—	—
240	0.5	5.3	59.68	5.78	6.19	16.16	—	—	4.38 (5.9)	43.78	—	—
240	0.5	9.1	59.60	5.80	6.25	15.32	—	—	6.68 (9.0)	51.15	—	—
240	2.0	5.3	48.66	4.98	4.10	13.07	—	—	4.14 (4.5)	32.96	—	—
240	2.0	9.1	51.45	5.22	4.55	12.12	—	—	7.10 (8.2)	63.57	—	—

[a] % based on dry weights of starting thin stillage and extracted hydrochars

[b] Yield based on dry weights. Values in parentheses are % of total carbon recovered in extracted hydrochar

[c] Yield based on thin stillage dry solids with 17.52 % fatty acids and an extract concentration of 84% fatty acids

[d] HHV of unextracted hydrochar in parentheses

[e] Thin stillage value from liturature (Kim, et al., 2008)

Table II. Reaction conditions, elemental analysis, and yields of whole stillage and resulting extracted hydrochars

Process Conditions			Elemental Analysis[a]					Hydrochar % yield[b]	Fatty Acid % Yield[c]	HHV BTU/lb[d]	% Ash[e]
Temp (°C)	Time (h)	% Solids	%C	%H	%N	%S	%P				
WS	—	—	45.99	7.58	4.03	0.34	1.11	—	—	8738	5.0
200	0.5	10	59.45	6.80	7.19	—	—	18.45 (23.8)	75.97	—	—
200	0.5	14	59.21	6.69	6.87	—	—	20.78 (26.7)	81.58	—	—
200	2	10	63.10	6.52	5.50	—	—	18.07 (24.8)	93.38	—	—
200	2	14	63.15	6.55	5.69	—	—	19.65 (26.9)	92.61	—	—
215	1.25	12	64.85	6.52	5.72	—	—	17.55 (24.7)	91.23	—	—
215	1.25	12	65.54	6.62	5.90	—	—	17.65 (25.2)	90.75	—	—
215	1.25	12	65.01	6.63	5.52	0.47	0.18	17.72 (25.0)	90.16	11819(13889)	0.83
215	1.25	12	65.28	6.73	5.48	—	—	17.79 (25.2)	89.27	—	—
230	0.5	10	64.51	6.51	5.96	—	—	16.00 (22.4)	82.35	—	—
230	0.5	14	64.88	6.54	6.07	—	—	17.33 (24.4)	88.71	—	—
230	2	10	66.66	6.64	5.60	—	—	15.49 (22.4)	97.60	—	—
230	2	14	65.48	6.53	5.61	—	—	16.68 (23.7)	101.79	—	—

[a] % based on dry weights of starting whole stillage and extracted hydrochars

[b] Yield based on dry weights. Values in parentheses are % of total carbon recovered in extracted hydrochar

[c] Yield based on whole stillage dry solids with 12.84 % fatty acids and an extract concentration of 84% fatty acids

[d] HHV of unextracted hydrochar in parentheses

[e] Whole stillage value from liturature (Kim, et al., 2008)

Triglycerides present in thin and whole stillage were hydrolyzed and sorbed onto hydrochars as fatty acids (yields in Tables I and II). Total gravimetric fat values, percent triglyceride, and fatty acid contents measured by GC of digested freeze-dried thin stillage were 19.41%, 18.32%, and 17.52% and for whole stillage 14.81%, 13.42%, and 12.84%. Fatty acid components for both stillages were essentially the same, with palmitic (16:0), stearic (18:0), oleic (18:1), linoleic (18:2) and linolenic (18:3) acids comprising 14.5%, 1.9 %, 25.3%, 55.0%, and 1.4% respectively, for a total of 98.1%. The weight-average molecular weight used for mass computations of fatty acids present in extracts was 276 g/mole. HTC treatment had little effect on fatty acids in the system, as the fatty acid profile of the extract from the center point whole stillage hydrochar was essentially unchanged from the profile of starting whole stillage and contained the following fatty acids: 16:0, 13.9%; 18:0, 2.0%; 18:1, 25.6%; 18:2, 51.5%; and 18:3, 1.2% (97.0% of the fatty acids present).

Batch Anaerobic Digestion

Table III. Initial characteristics of inoculum and substrates

	Total Solids % w/w	Volatile Solids % w/w	pH	Alkalinity mg $CaCO_3$/L
Thin Stillage	5.72 ± 0.003	4.99 ± 0.02	4.71	0
HTC Filtrate	4.02 ± 0.02	3.36 ± 0.01	4.82	0
Inoculum	3.47 ± 0.02	2.30 ± 0.02	8.16	9803.3

Biochemical methane potential (BMP) assays established that undiluted HTC filtrate generated from thin stillage at 220°C could readily be digested. Substrate and inoculum characterization determined that HTC processing reduced total solids and TCOD of thin stillage by ca. 30% and 45%, respectively, while maintaining a volatile

solid to total solid ratio of 0.84 compared to 0.87 for thin stillage (Table III). Specific methane production, corrected at STP, from all the BMP assays is shown in Figure 2. Total inoculum methane production was low (18.5 mL at STP), which was subtracted from the values presented in Figure 2. Unaltered methane production data are given in A.5. Initially, none of the treatments appeared to go through an acclimation or lag phase prior to methane production. From Day 7 to Day 12, however, there was a notable lag in methane production in the 100% HTC treatment, with two of the three reactors producing methane at reduced rates and with increased standard deviations. All of the dilutions of either the thin stillage or HTC filtrate treatments (35%, 65%, and 100%) produced essentially the same quantity of total methane per gram of initial COD, indicating that the undiluted samples were not toxic.

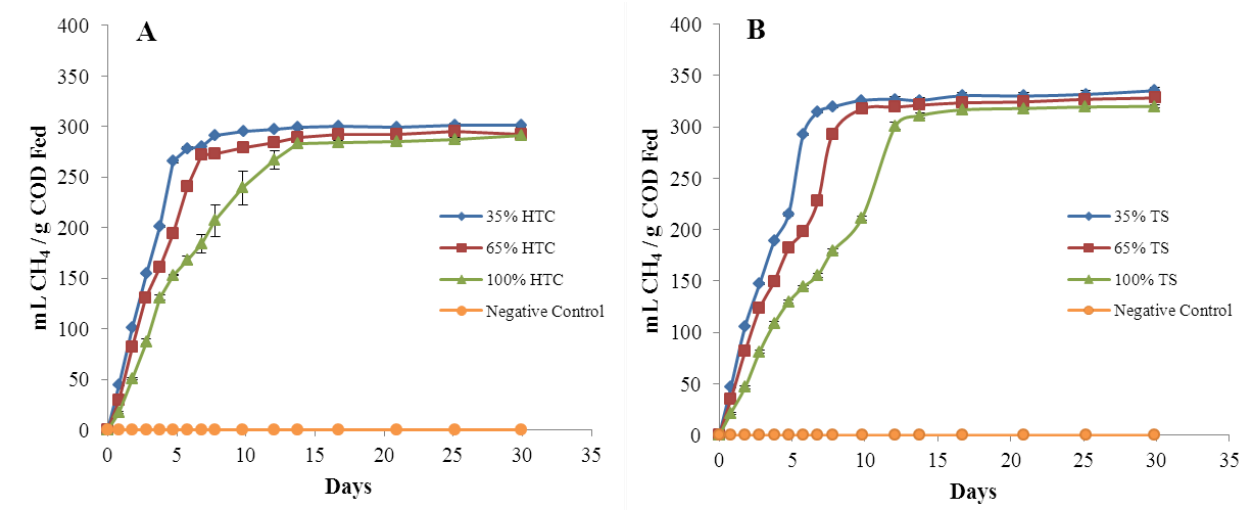

Figure 2. Specific methane production at STP of HTC filtrate generated at 220°C from TS (A) and untreated TS (B) from BMP assays. Background methane production from inoculum has been subtracted and error bars represent standard deviation.

Furthermore, thin stillage and the HTC filtrates, again at all dilutions, proved to be highly biodegradable, as evidenced by greater than 90% reductions in total COD over the incubation period (Table IV).

Table IV. Total Chemical Oxygen Demand

Treatment	Initial mg/L	Final mg/L[a]	% Removal
35% HTC*	17547 ± 970	1600 ± 927	91 ± 5
65% HTC*	32587 ± 970	2500 ± 894	92 ± 3
100% HTC	50133 ± 970	4367 ± 702	91 ± 1
35% TS*	31181 ± 484	1300 ± 707	96 ± 2
65% TS*	57908 ± 484	1933 ± 976	97 ± 2
100% TS	89089 ± 484	4633 ± 1978	95 ± 2
Negative control*	15040 ± 970	13167 ± 115	12 ± 6
Inoculum control*	24795 ± 342	20100 ± 436	19 ± 2

* Initial COD values calculated using dilution factor

[a] Values calculated by subtracting inoculum control

In addition, 91% and 83% of the initial COD was converted into methane for the 100% thin stillage and 100% HTC filtrate samples, respectively (Figure 3). Multiple comparisons using a studentized range ($\alpha = 0.05$) demonstrated that there were statistically significant differences between the HTC filtrates and the corresponding untreated thin stillage samples with respect to the percent of initial COD converted to methane (Figure 3).

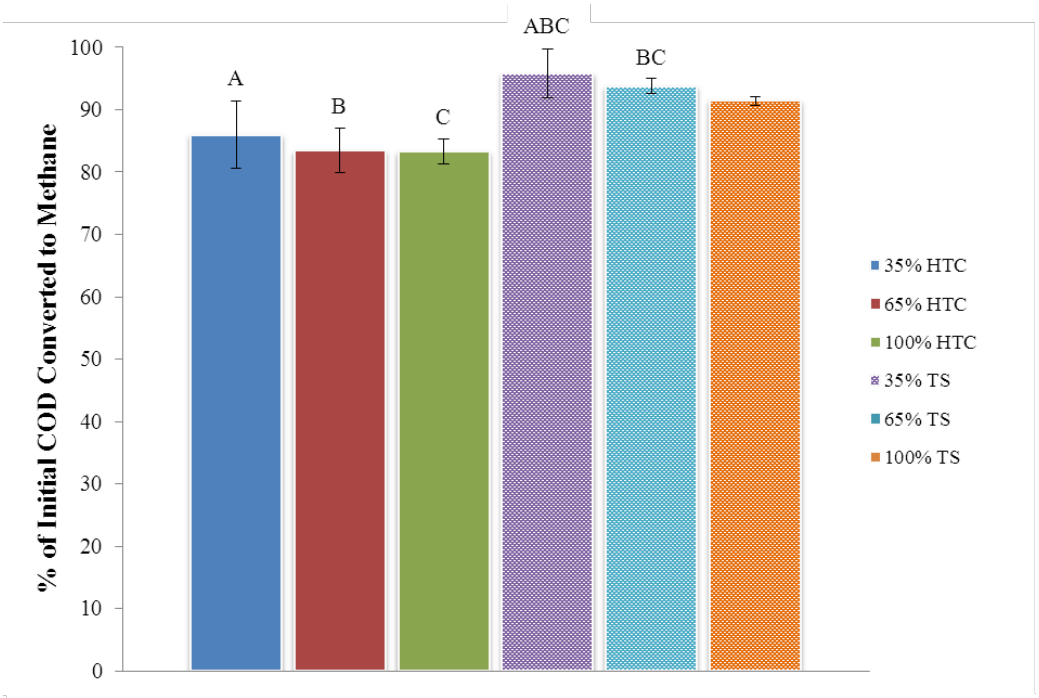

Figure 3. Percent of initial COD converted to methane. Methane to COD conversion was calculated using measured total methane production excluding background methane from inoculum and the theoretical value 0.35 L CH_4 = 1 g COD. Corresponding letters represent significant differences by studentized range (α = 0.05). Error bars represent standard deviation.

Discussion

Energy Analysis

Energy analysis was conducted by comparing the requirements for HTC versus those of conventional downstream processing to generate a shelf stable product, defined as 90% solids. Conventional energy input values were obtained from a literature report (Rausch & Ronald, 2006), whereas HTC energy input values were computed from an enthalpy balance. A detailed set of assumptions and computations are available in A.6 and A.7. The results showed that HTC processing would reduce energy consumption by 73% for thin stillage and 65% for whole stillage compared to conventional downstream processing operations. For the thin stillage scenario, the preliminary evaluation of methane production rates indicated that the anaerobic digestion of HTC filtrate could

provide all of the energy needed to fuel the HTC process and displace additional natural gas requirements.

Interestingly, initial rate constants of methane production were lowered by HTC processing at 220°C. Over the initial linear phase (5 days) of methane production, HTC filtrate had an initial rate constant of 38.00 mL CH_4 $gVS^{-1}d^{-1}$ while thin stillage with a corresponding initial COD value had an initial rate of 44.66 mL CH_4 $gVS^{-1}d^{-1}$ at STP (Thin stillage initial rate constant was calculated by using a polynomial fit model S.4). Consequently, HTC treatment generated organic compounds that were more difficult to degrade relative to initial compounds present in thin stillage. Thin stillage was readily digestible without pretreatment because it had undergone enzymatic hydrolysis prior to fermentation and contained acetic acid which provided acetate, the primary carbon and energy source for methanogenesis. Other pretreatment methods such as sonication offered little or no benefit to thin stillage digestion (Schaefer & Sung, 2008). In an effort to improve the rate of methane production, thermophilic digestion could be investigated since metabolic rates of methanogens can double (Khanal, 2009), and in this instance there would be no need to heat the influent HTC stream.

HTC Process Water Backset

Recycle of process water could occur with the filtrate directly after HTC or after anaerobic digestion of the filtrate. The choice of direct recycle versus digestion followed by recycle depends on a number of factors. Lactic acid, acetic acid and glycerol will be present in HTC filtrate streams and may negatively impact both the growth of yeast and the fermentation process. Indeed, the growth of yeast in fermentation can be inhibited

when lactic acid, acetic acid, and glycerol levels are present in excess of 0.80%, 0.05%, and 1.0%, respectively (Jacques K et al., 1999). In our experiments, lactic acid concentrations were well below the 0.80% limit, ranging from 0.16-0.23% with whole stillage and 0.18-0.32% with thin stillage. Acetic acid levels, however, exceeded 0.05% in some cases, with the whole stillage samples ranging from 0.07-1.0% and the thin stillage samples ranging from 0.02-0.07%. As anticipated, glycerol levels were high as a result of extensive hydrolysis of triacylglycerides, ranging from 0.84-1.03% with whole stillage samples and 0.96-1.50% with thin stillage samples. From this analysis there would seem to be little benefit in recycling HTC filtrates directly. After anaerobic digestion, however, it is likely that the filtrate will contain few if any of these compounds (Alkan-Ozkaynak & Karthikeyan, 2011) and may be effectively recycled. The possibility does exist that Maillard compounds or other inhibitory by-products may be formed in the HTC process as well; this could also cause toxicity in direct recycle applications and possibly in the recycle of digested filtrates as well. This requires additional research.

Hydrochar and HTC Mechanism

The mechanism of carbonization with thin stillage was explored via the construction of a Van Krevelen diagram (Van Krevelen, 1950) in which changes in H/C and O/C ratios are plotted for starting biomass and coalified products. With this technique, a slope of 2 indicates a dehydration process, while slopes < 2 provide evidence for decarboxylation. When the H/C versus O/C ratios were plotted for starting thin stillage and thin stillage derived hydrochars obtained at the various reaction conditions (data in Table I), all of the hydrochars were located within a relatively tight cluster and

the slope of the line ($R^2 = 0.98$) between the starting thin stillage and the center of the cluster was 2.09, indicating that dehydration was the principal carbonization process occurring under the reaction conditions. Interestingly, decarboxylation was the principal mechanistic pathway identified in a related study of HTC of anaerobically digested maize silage (Mumme et.al, 2011). A slope of 0.90 supported the authors' conclusion that substantial decarboxylation had occurred with that substrate during carbonization.

Hydrochar having a significantly decreased O/C ratio relative to starting biomass can be utilized as a carbon-neutral fuel having a coal-like high heating value. Low extracted yields and relatively high nitrogen contents (Tables I and II), however, suggested that other applications may be more suitable. As with other hydrochars (Chen et al., 2011), the BET surface area of the thin stillage center point hydrochar was quite low (2.2 m^2/g). In addition, virtually all of the hydrochars generated in this research possessed high nitrogen contents, especially one thin stillage hydrochar with N = 9.3% (Table I), derived from proteins, peptide fragments, and amino acids that participated in hydrochar formation by denaturation and precipitation (Heilmann et al., 2011a) and Maillard reactions (Zerong, 2010). In addition to carboxylic acid groups that are commonly present on hydrochar surfaces (Chen et al., 2011), a significant quantity of this nitrogen content may be present as basic amine groups on the surface of hydrochars, The presence of both acidic and basic functional groups could provide for mixed bed ion exchange behaviors and unexpected utility for these materials. Future research is needed to explore this and other options for hydrochar use.

Other Products

HTC processing did not appreciably alter the composition of fatty acids present in either whole or thin stillage; notably, even the polyunsaturated acid, linoleic acid, underwent only a 1% change in concentration. Similar low conversions of fatty acids under HTC conditions have been reported (Wilson & Novak, 2009). Fatty acid yields, however, were markedly different with the two stillages. Thin stillage gave lower yields of fatty acids compared to whole stillage, which was largely attributed to the relatively small quantities of hydrochar formed from the thin stillage. Because hydrochar sorbs the fatty acids, hydrochar yields of only 4.2 – 7.1% for the thin stillage compared to 15.5 – 20.8% for the whole stillage were believed to be partially responsible. This was apparently not the only explanation, however, as reproducibility of fatty acid yields in the replicate thin stillage center point experiments was poor, i.e., 47-59%, despite having the same quantity of sorbing hydrochars produced in all three experiments. This suggested that the reaction workup procedure may have been a factor, and variations in the cool-down procedure, e.g., time and final temperature achieved, may have played roles in the fatty acid yield. It was determined that a final product mixture temperature of 45 ^{o}C provided an optimum fatty acid yield of 68% for thin stillage center point conditions. This modification was applied in the workup of the whole stillage materials in Table II, resulting in equivalent fatty acid yields in the four replicate center point experiments. A possible explanation of this phenomenon was that a ripening process may have occurred upon hydrochar cooling, involving both the sorption and desorption of fatty acids from the hydrochars. At temperatures below 45 ^{o}C, possible formation of relatively stable

micelles of fatty acids in the aqueous suspending medium may have competed with the collection of fatty acids on the hydrochars, causing a decrease in fatty acid yields.

Corn oil from thin stillage or condensed distillers solubles has become a valuable co-product in the dry grind ethanol industry. One of the more prevalent technologies to remove the oil from condensed distillers solubles is centrifugation, with typical oil yields of ca. 30% (private communication from Randall Doyal of Al-Corn Clean Fuels). By contrast, fatty acids extracted from thin stillage and whole stillage hydrochars were obtained in 68% and 90% yields, respectively. These fatty acids can be converted directly into biodiesel or other liquid transportation fuels, providing yet another benefit of the HTC process.

The practicality of either HTC scenario depends to a considerable extent on the animal feed market. With HTC of thin stillage, an animal feed, DDG, is produced that is equivalent to or may actually exceed DDGS in feed quality as a result of its higher protein and lower lipid content (Dahlen et al., 2011).

Linear Regression analysis

The orthogonal design of these experiments facilitated a linear regression analysis that identified the significance of time, temperature, and concentration (percent solids) input variables on specific outputs. Equations with an R^2 of at least 0.95 are shown in Table V (full statistical analysis A.1).

Table V. Linear regression equations having R^2 values > 0.95 that were obtained from designed stillage experiments of Tables I and II.

Thin Stillage Equations [a]:
Extracted Hydrochar % Yield: $Y = 5.89 – 0.24\ X_1 + 0.42\ X_2 + 1.14\ X_3 – 0.37\ X_4$
Extracted Hydrochar % Nitrogen: $Y = 6.56 – 1.38X_1 – 0.98X_2$
Whole Stillage Equations [b]:
Extracted Hydrochar % Yield: $Y = 17.76 – 1.43X_1 – 0.33X_2 + 0.80X_3 – 0.17X_5$
Fatty Acid % Yield: $Y = 89.61 + 3.36X_1 + 7.10X_2 + 1.92X_3 – 1.07X_6$

X_1 = temperature, X_2 = time, X_3 = solids, X_4 = temperature*time, X_5 = temperature*solids, X_6 = time*solids
[a,b] = All variables have dimensionless units

Thin stillage hydrochar yields decreased with temperature and temperature*time interaction, but increased with time and concentration. Similarly, whole stillage hydrochar yield decreased with temperature and temperature*concentration interaction, and increased with concentration. Whole stillage hydrochar yield, however, decreased with time. With both whole stillage and thin stillage, increased yields were obtained at higher solids input. Although this value could be controlled to some extent for whole stillage by distillation conditions and for thin stillage by centrifuge conditions, it is impractical for manufacturers to attempt to control solids input substantially. The equation for percent nitrogen in the thin stillage hydrochar indicated that percent solids had no significant effect, while temperature and time had large negative effects. Whole stillage fatty acid yields were maximized by increasing temperature and time. Overall, lower temperatures and retention times are favored for continuous processing because they reduce the size and the pressure rating of the equipment required.

Conclusion

Hydrothermal carbonization and subsequent anaerobic digestion of stillage intermediates can be successfully combined to generate biogas, treated process water, hydrochar, and fatty acids. The first objective of this study was to improve the overall energy balance of dry-grind corn ethanol production by modifying downstream processing of stillage intermediates. Eliminating the need for evaporation and some of the drying considerably reduced operational energy use; furthermore, on-site production of biogas would reduce natural gas requirements. Reducing water consumption was the second objective, and HPLC analysis indicated that HTC filtrates would require treatment in order to increase backset. Anaerobic digestion of the filtrate consumed greater than 90% of the TCOD and converted more than 80% of that initial COD to methane. A final objective was to develop options with respect to new or improved co-products. Thin stillage fatty acid yields of 68% are a marked improved compared to the current industry average of 30%. Using thin stillage in such an HTC process would also provide an animal feed product higher in protein and lower in fat than DDGS. Hydrochar was shown to have a good fuel value, but due to surface functionality may be better suited for higher-value applications such as a mixed bed ion exchange material. In closing, although a more detailed techno-economic analysis is necessary to determine the economic viability of the proposed model, this process appears promising for increasing the sustainability of the dry-grind corn ethanol industry.

Part III. Recommendations

3.1 Hydrothermal Carbonization

3.1.1 Optimal Reaction Conditions and Products

HTC reaction conditions dictate composition and thus function of the resulting hydrochar and aqueous product stream. Linear regression equations generated from the full factorial experiments are useful tools that can optimize reaction conditions, within the design parameters, to maximize beneficial characteristics and yields. Currently, there exists a level of uncertainty about the desired characteristics of hydrochar because the final application of the material remains unidentified. Until the most suitable application is determined, it seems logical to conduct reactions at or near the lowest temperatures evaluated for both thin and whole stillage as this will increase hydrochar yield and minimize recalcitrant compounds in the aqueous fraction. Reaction time is a variable that is less clearly defined, but it should be kept in mind that shorter reaction times benefit continuous or larger scale batch processing. Additionally, percent solids had a large effect on certain outputs; however, this factor cannot be easily manipulated and was included in designed experiments to represent normal fluctuations that appear in the ethanol industry. When HTC product applications are clear, an economic assessment will be needed in conjunction with linear regression equations to optimize product distributions based on market values.

3.1.2 Future Research

Future research on HTC of stillage intermediates requires a focus on the utility of hydrochar, and the mechanism of triglyceride hydrolysis and sorption. Hydrochars generated from stillage intermediates must have high value due to relatively low yields; otherwise it becomes difficult to justify coupling HTC with anaerobic digestion instead of

anaerobic digestion independently. Nevertheless, there are a number of promising applications for stillage hydrochars such as a CO_2 sorbent (Zhang et al., 2012) or as an electrode material for supercapacitors (Wei et al., 2011). Both of these applications require activation of the hydrochar to generate pore structure, and it seems increasingly likely that this will be a necessary step for high value applications. One property of stillage hydrochars that is beneficial in theory is the high nitrogen content. In the current study only gross nitrogen content was reported. To fully appreciate the material's usefulness surface functional groups, including oxygen containing groups, need to be quantified. Another HTC research area that merits further investigation is the mechanism by which fatty acids are sorbed to hydrochar. Understanding this mechanism may help improve yields and further explain the cooling temperature phenomena observed at 45°C.

3.2 Anaerobic Digestion

3.2.1 Future Research

Anaerobic digestion of HTC filtrates is still a new area of research with many possibilities to explore. Although thermophilic digestion has the advantage of higher metabolic rates it is hindered by additional heating requirements. This is not the case with HTC filtrates. Even with heat exchanges, liquid products would leave the HTC system above 55°C, making thermophilic digestion a viable option. Biochemical Methane Potential (BMP) assays should be conducted with stillage filtrates at thermophilic conditions to determine methane production rates and any toxicity or inhibition effects. Once BMP assays are completed and compared, a decision can be made about the optimal temperature for larger scale digestion. With commercial application in mind,

moving to a high rate continuously fed system would be the next step in development. Since a short hydraulic retention time will be important to the ethanol industry, a high rate reaction design such as anaerobic fluidized bed reactor that can separate hydraulic retention time from solids retention time would be an appropriate choice. During the start-up phase microbes can be acclimated gradually, increasing the likelihood that recalcitrant compounds will be degraded. Another reason it is important to move to a continuously fed system is that representative digestate is essential for small scale fermentation trials. Fermentation trials will ascertain the amount of digestate that can be backset without effecting ethanol production. Many reports have suggested that digestates can be recycled to fermentation, but to my knowledge no experimental data has been published.

3.3 Summary of Research Needs

If HTC of whole or thin stillage is ever to be an industrially relevant process resulting hydrochars must have high value applications and subsequent anaerobic digestion of aqueous filtrates should produce recyclable or dischargeable process water instead of creating a new liquid waste stream. Thermally and or chemically activating hydrochar is a new and promising area of research for high value applications. In regards to digestion, high rate continuously fed reactor experiments are necessary to determine methane production rates and produce digestate for fermentation studies. Conducting larger scale anaerobic digestion research is the key to developing a sound energy and water balance for the overall process. In conclusion, the coupling of HTC and anaerobic digestion to treat downstream stillage intermediates has been proven as technically

feasible, but more investigation is required to decide if it is practical and economically beneficial.

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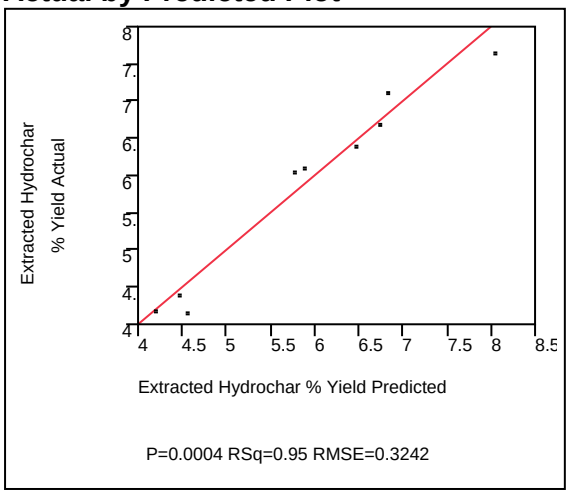
Appendix

A.1

Thin Stillage Linear Equations:

Response Extracted Hydrochar % Yield
Actual by Predicted Plot



Summary of Fit

RSquare	0.954504
RSquare Adj	0.924173
Root Mean Square Error	0.324192
Mean of Response	5.886364
Observations (or Sum Wgts)	11

Analysis of Variance

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	4	13.230050	3.30751	31.4699
Error	6	0.630605	0.10510	**Prob > F**
C. Total	10	13.860655		0.0004*

Lack Of Fit

Source	DF	Sum of Squares	Mean Square	F Ratio
Lack Of Fit	4	0.63053788	0.157634	4729.034
Pure Error	2	0.00006667	0.000033	**Prob > F**
Total Error	6	0.63060455		0.0002*
				Max RSq
				1.0000

Parameter Estimates

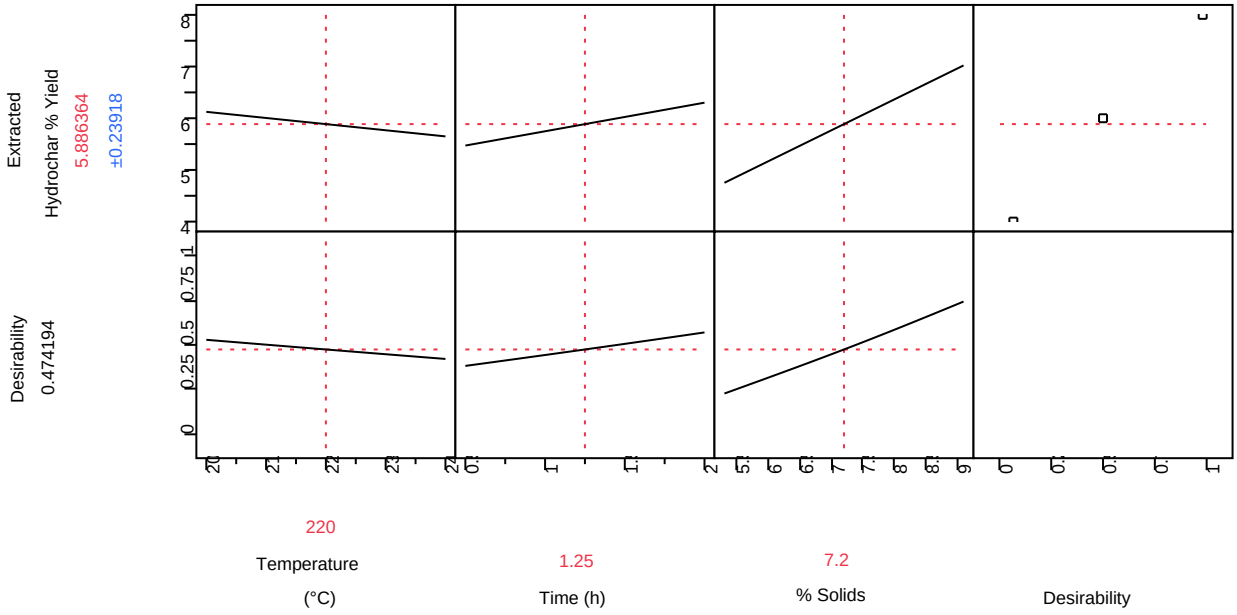
Term	Estimate	Std Error	t Ratio	Prob>|t|
Intercept	5.8863636	0.097748	60.22	<.0001*
Temperature (°C)(200,240)	-0.2375	0.114619	-2.07	0.0836
Time (h)(0.5,2)	0.415	0.114619	3.62	0.0111*

Term	Estimate	Std Error	t Ratio	Prob>|t|
% Solids(5.3,9.1)	1.135	0.114619	9.90	<.0001*
Temperature (°C)*Time (h)	-0.37	0.114619	-3.23	0.0180*

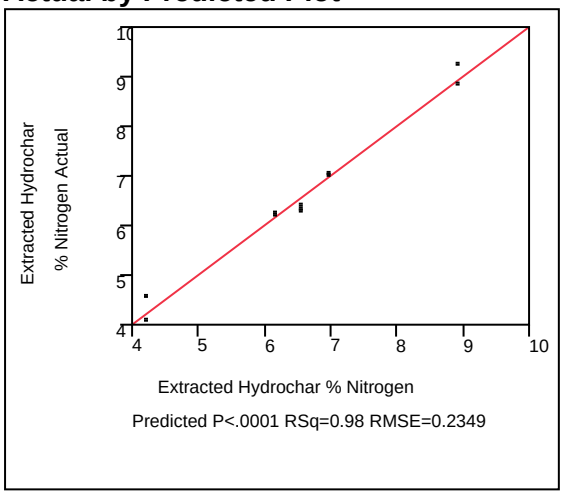
Sorted Parameter Estimates

Term	Estimate	Std Error	t Ratio	t Ratio	Prob>|t|
% Solids(5.3,9.1)	1.135	0.114619	9.90		<.0001*
Time (h)(0.5,2)	0.415	0.114619	3.62		0.0111*
Temperature (°C)*Time (h)	-0.37	0.114619	-3.23		0.0180*
Temperature (°C)(200,240)	-0.2375	0.114619	-2.07		0.0836

Prediction Profiler



Response Extracted Hydrochar % Nitrogen
Actual by Predicted Plot



Summary of Fit

RSquare	0.981134
RSquare Adj	0.976418
Root Mean Square Error	0.234865
Mean of Response	6.562727
Observations (or Sum Wgts)	11

Analysis of Variance

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	2	22.949725	11.4749	208.0225
Error	8	0.441293	0.0552	**Prob > F**
C. Total	10	23.391018		<.0001*

Lack Of Fit

Source	DF	Sum of Squares	Mean Square	F Ratio
Lack Of Fit	2	0.24037652	0.120188	3.5892
Pure Error	6	0.20091667	0.033486	**Prob > F**
Total Error	8	0.44129318		0.0944
				Max RSq
				0.9914

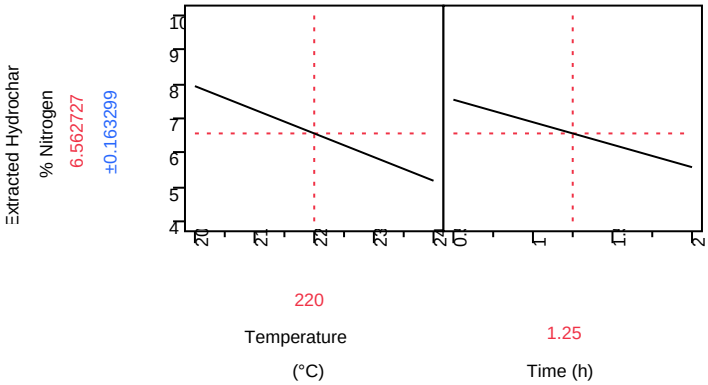
Parameter Estimates

Term	Estimate	Std Error	t Ratio	Prob>|t|
Intercept	6.5627273	0.070815	92.67	<.0001*
Temperature (°C)(200,240)	-1.37875	0.083037	-16.60	<.0001*
Time (h)(0.5,2)	-0.98375	0.083037	-11.85	<.0001*

Sorted Parameter Estimates

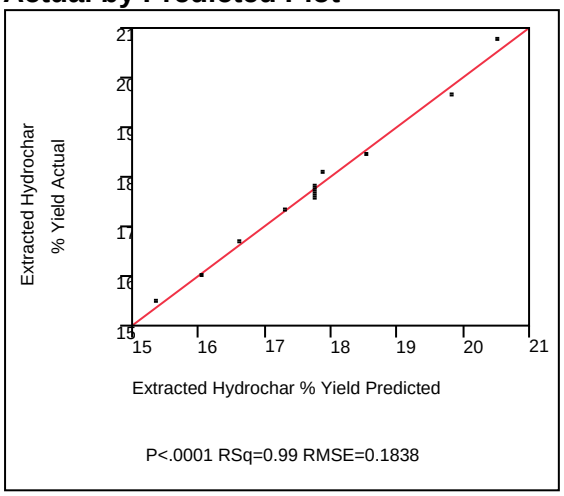
Term	Estimate	Std Error	t Ratio	t Ratio	Prob>|t|
Temperature (°C)(200,240)	-1.37875	0.083037	-16.60		<.0001*
Time (h)(0.5,2)	-0.98375	0.083037	-11.85		<.0001*

Prediction Profiler



Whole Stillage Linear Equations:

Response Extracted Hydrochar % Yield
Actual by Predicted Plot



Summary of Fit

RSquare	0.989683
RSquare Adj	0.983788
Root Mean Square Error	0.183815
Mean of Response	17.76333
Observations (or Sum Wgts)	12

Analysis of Variance

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	4	22.688550	5.67214	167.8738
Error	7	0.236517	0.03379	**Prob > F**
C. Total	11	22.925067		<.0001*

Lack Of Fit

Source	DF	Sum of Squares	Mean Square	F Ratio
Lack Of Fit	4	0.20504167	0.051260	4.8858

Source	DF	Sum of Squares	Mean Square	F Ratio
Pure Error	3	0.03147500	0.010492	Prob > F
Total Error	7	0.23651667		0.1117
				Max RSq
				0.9986

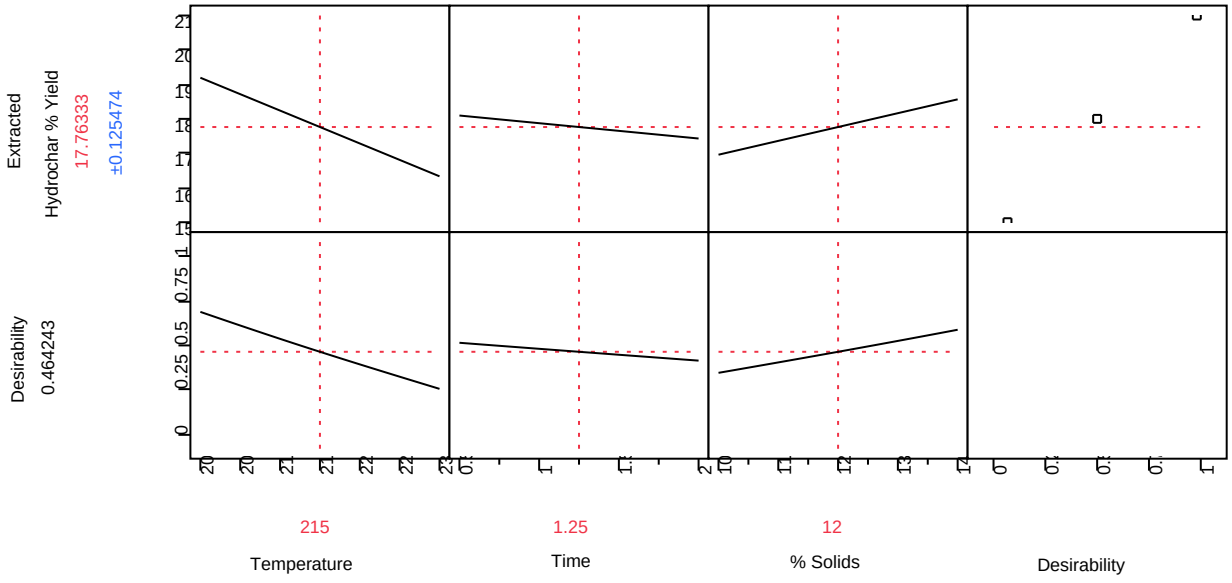
Parameter Estimates

Term	Estimate	Std Error	t Ratio	Prob>|t|
Intercept	17.763333	0.053063	334.76	<.0001*
Temperature(200,230)	-1.43125	0.064989	-22.02	<.0001*
Time(0.5,2)	-0.33375	0.064989	-5.14	0.0013*
% Solids(10,14)	0.80375	0.064989	12.37	<.0001*
Temperature*% Solids	-0.17375	0.064989	-2.67	0.0318*

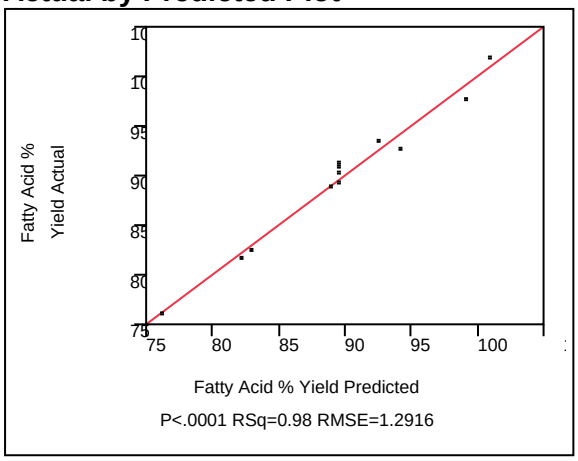
Sorted Parameter Estimates

Term	Estimate	Std Error	t Ratio	t Ratio	Prob>|t|
Temperature(200,230)	-1.43125	0.064989	-22.02		<.0001*
% Solids(10,14)	0.80375	0.064989	12.37		<.0001*
Time(0.5,2)	-0.33375	0.064989	-5.14		0.0013*
Temperature*% Solids	-0.17375	0.064989	-2.67		0.0318*

Prediction Profiler



Response Fatty Acid % Yield
Actual by Predicted Plot



Summary of Fit

RSquare	0.978525
RSquare Adj	0.966253
Root Mean Square Error	1.291628
Mean of Response	89.61667
Observations (or Sum Wgts)	12

Analysis of Variance

Source	DF	Sum of Squares	Mean Square	F Ratio
Model	4	532.11695	133.029	79.7393
Error	7	11.67812	1.668	**Prob > F**
C. Total	11	543.79507		<.0001*

Lack Of Fit

Source	DF	Sum of Squares	Mean Square	F Ratio
Lack Of Fit	4	9.541242	2.38531	3.3488
Pure Error	3	2.136875	0.71229	**Prob > F**
Total Error	7	11.678117		0.1742
				Max RSq
				0.9961

Parameter Estimates

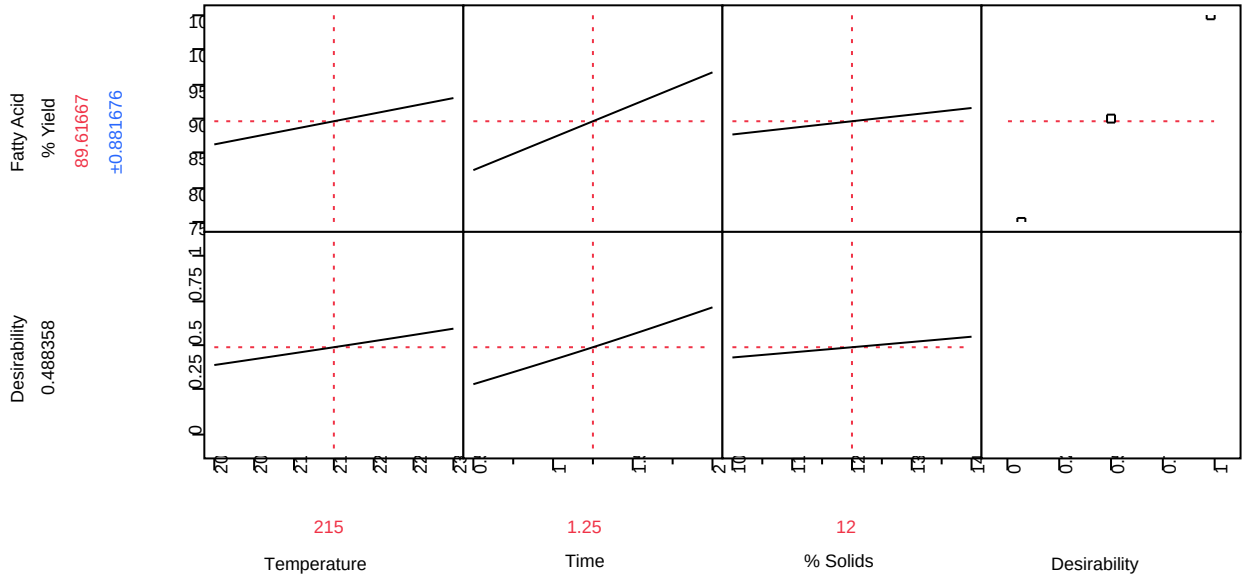
Term	Estimate	Std Error	t Ratio	Prob>|t|
Intercept	89.616667	0.372861	240.35	<.0001*
Temperature(200,230)	3.36375	0.456659	7.37	0.0002*
Time(0.5,2)	7.09625	0.456659	15.54	<.0001*
% Solids(10,14)	1.92375	0.456659	4.21	0.0040*
Time*% Solids	-1.06875	0.456659	-2.34	0.0518

Sorted Parameter Estimates

Term	Estimate	Std Error	t Ratio	t Ratio	Prob>|t|
Time(0.5,2)	7.09625	0.456659	15.54		<.0001*
Temperature(200,230)	3.36375	0.456659	7.37		0.0002*

Term	Estimate	Std Error	t Ratio	t Ratio	Prob>|t|
% Solids(10,14)	1.92375	0.456659	4.21		0.0040*
Time*% Solids	-1.06875	0.456659	-2.34		0.0518

Prediction Profiler



A.2

Source Reactor Operation and Configuration for Anaerobic Digestion

Two, three liter source reactors were initiated to provide inoculum for batch experiments. Start-up consisted of mixing two liters of digester sludge from a mesophilic cow manure digester with one liter of fresh cow manure and purging with N_2. Reactors were sealed with rubber stoppers and gastight valves for feeding and wasting. Magnetically stirred source reactors were operated semi-continuously at mesophilic conditions (37°C) with an HRT/SRT (solids retention time) of 45 days. Cow manure feed was screened (1mm mesh) in order to prevent obstruction of influent and effluent flows.

A.3

GC Method

Gas chromatography was conducted using a 6890 Series Hewlett Packard GC with a molecular sieve 13x packed column (Model: HP 19096C-008). Operating conditions included: thermal conductivity detector temperature of 250 °C, helium carrier gas flow rate of 20.0 mL/min, and an oven temperature of 75 °C. Methane standards were used to produce standard curves for determining % methane of biogas.

A.4

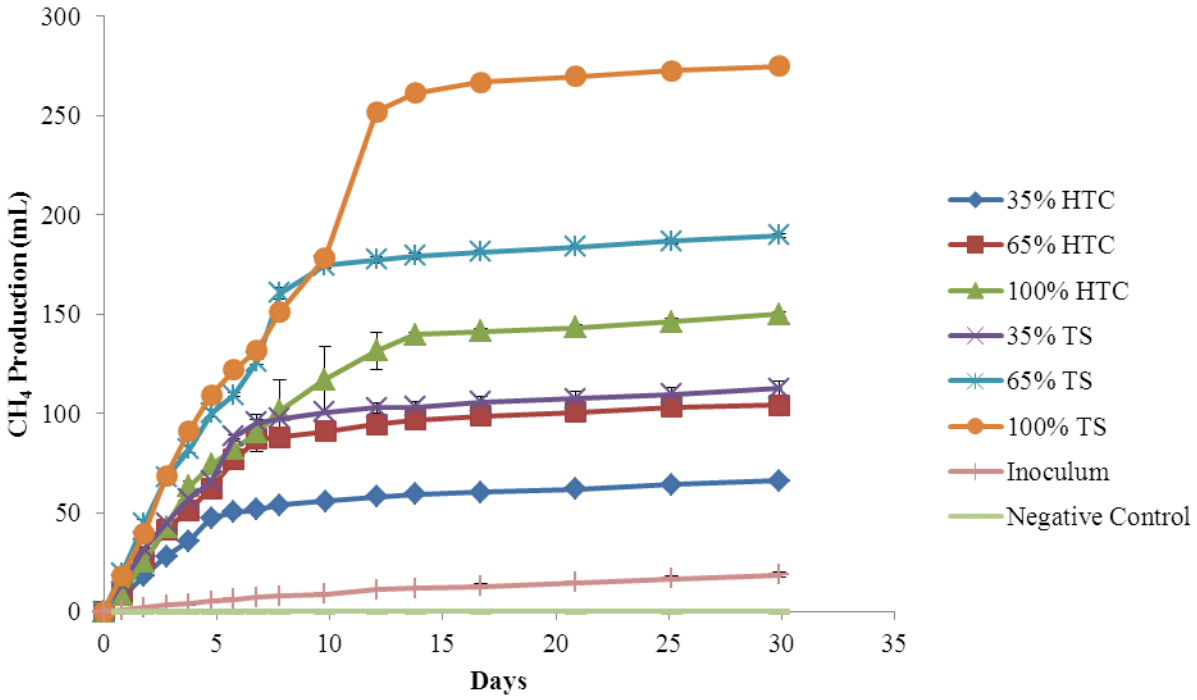
Biochemical Methane Production (BMP) Analysis

Methane production was computed after each sampling by multiplying the (volume of the bottle + volume of biogas removed) by the measured % methane and subtracting residual methane from the previous sampling point. Total methane production was calculated by converting these values to STP (0°C and 1 ATM) and adding them together. Final TCOD values used to calculate the measured COD reduction were determined by subtracting the TCOD of the inoculum control from TCOD values of all treatments in which inoculum was added. Percentages of initial COD degraded to methane were derived through multiplying the measured value of total methane at STP by the theoretical computation $\frac{0.35\ L\ CH4}{1\ g\ COD}$ (Khanal 2008). Initial rate constants (mL CH_4/g VS $_{Inoculum}$) were calculated from the initial linear portion of methane production (5 days) by linear regression analysis. In order to compare initial rate constants for TS and HTC filtrates on an equal COD basis, TS initial rates were plotted over COD and fitted with a second degree polynomial function. The polynomial function was solved for an initial COD value of

50133 mg/L corresponding to the 100% HTC value and allowed for direct comparison between the two.

A.5

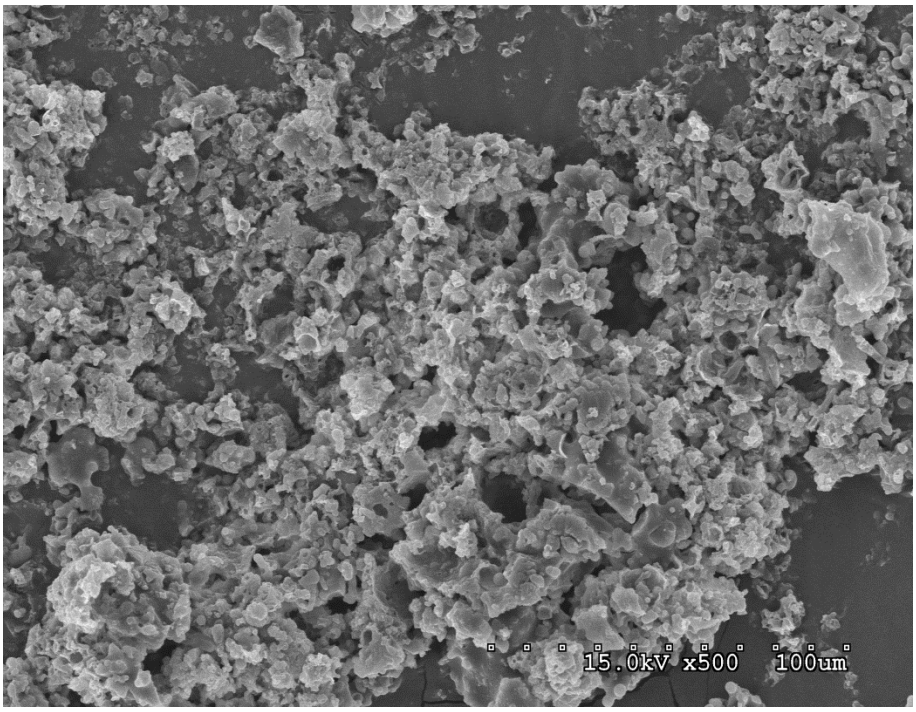
Total Methane Production



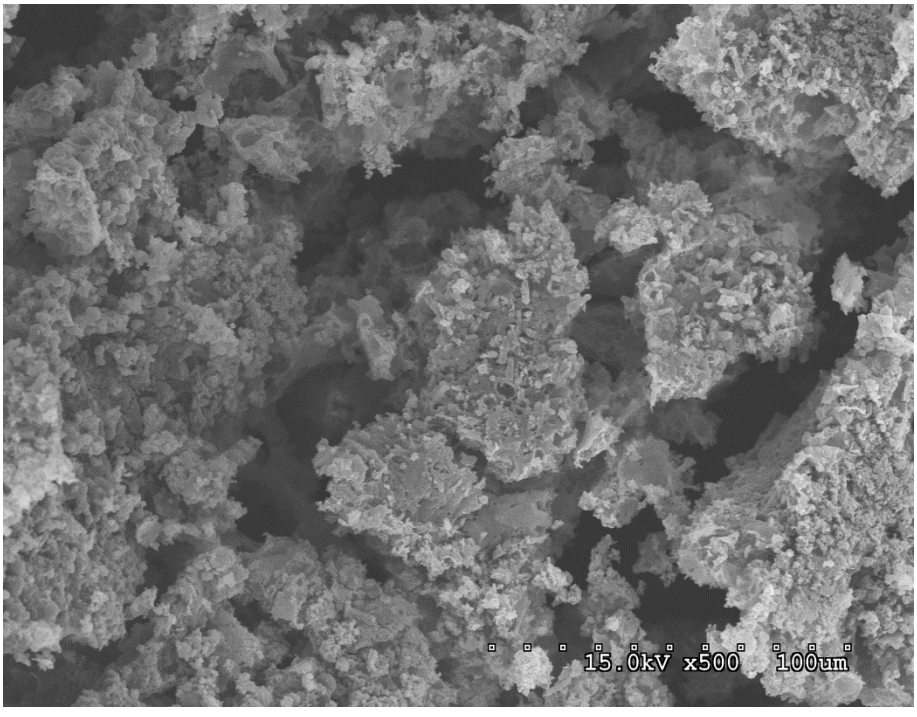
A.6

SEM Images

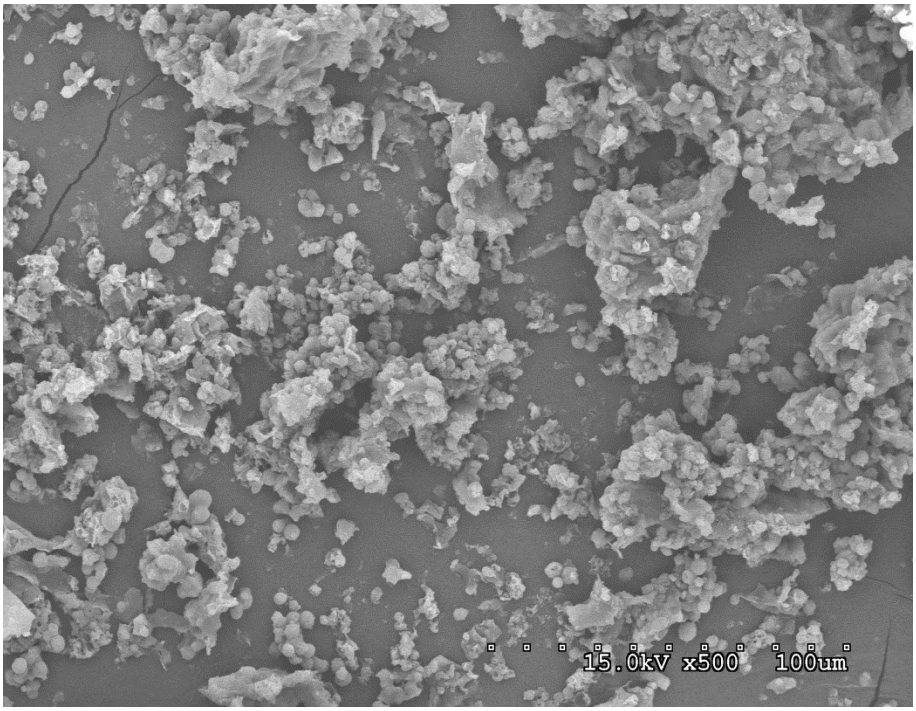
Unextracted Thin Stillage



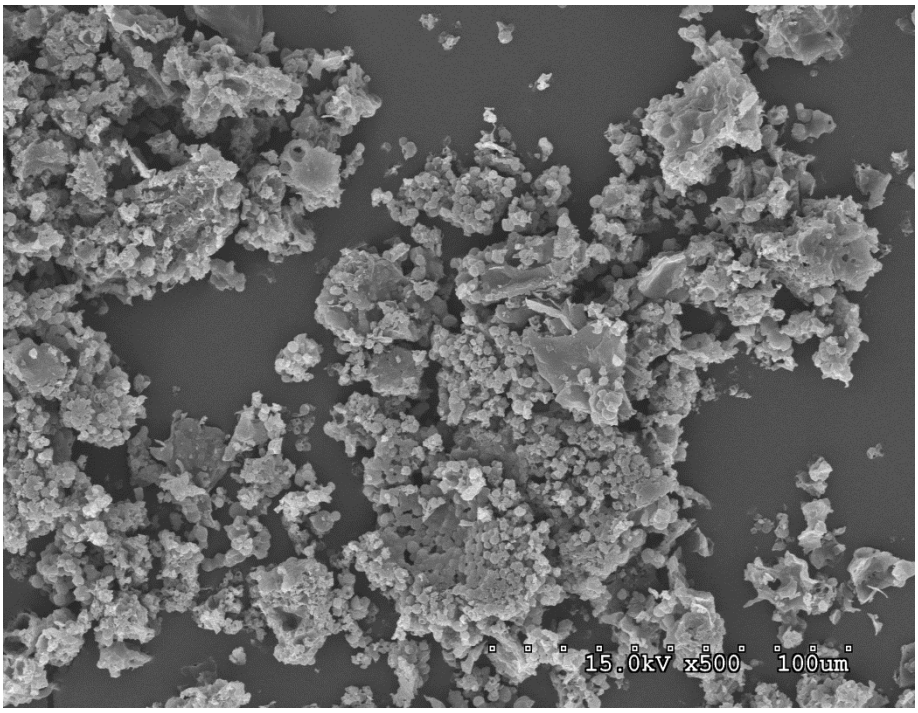
Extracted Thin Stillage



Unextracted Whole Stillage



Extracted Whole Stillage



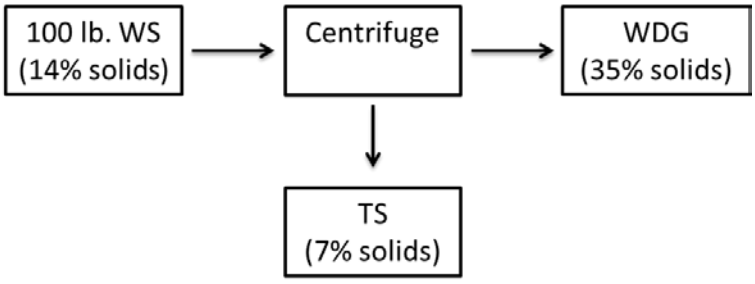
A.7

Comparing the energy consumed by conventional methods and HTC to produce a shelf stable product (shelf stable product is defined as 90% solids)

Conventional Whole Stillage Processing

Assumptions:

- Thin Stillage (TS): 7% solids
- Whole Stillage (WS): 14% soilds
- Wet Distillers Grains (WDG): 35% solids
- Condensed Distiller Solubles: 35% solids
- Dried Distillers Grains and Solubles (DDGS): 90% solids
- All Wet Distillers Grains are used to produce DDGS
- 30% of the Thin Stillage is backset to the front end of the fermentation process
- Centrifuge: 3 BTU/lb_{H2O} [1]
- Evaporator (triple effect): 559 BTU/lb_{H2O} [1]
- Rotary Dryer: 1500 BTU/lb_{H2O}



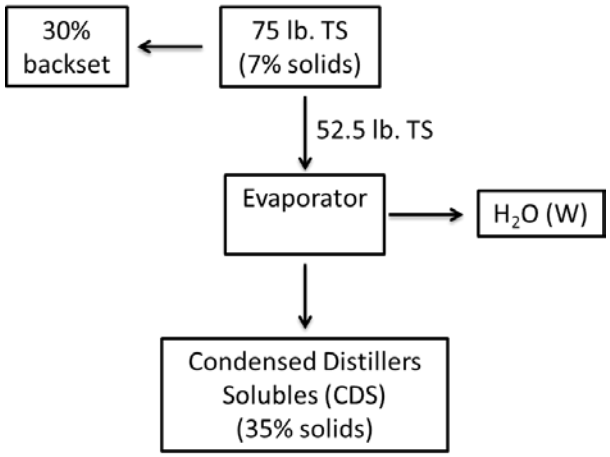
Material balance: WS = TS + WDG
Solids balance: 0.14WS = 0.07TS + 0.35WDG
Water balance: 0.86WS = 0.93TS + 0.65WDG

TS = WS – WDG
.14WS = .07(WS – WDG) + .35WDG
.14(100 lb.) = .07(100 lb.) - .07WDG + .35WDG
14 lb = 7 lb - .28WDG

7 lb = .28WDG
WDG = 7 lb/.28 = 25 lb.
TS = 100 lb. – 25 lb. = 75 lb.

Energy consumed by centrifuge

86 lb_{H2O} in starting WS $\times \frac{3\ BTU}{lbH2O}$ = 258 BTU

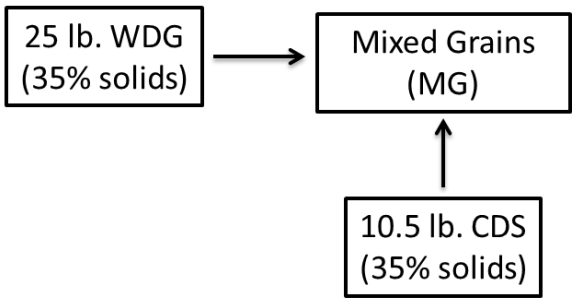


Material balance: TS = CDS + W
Solids balance: 0.07TS = 0.35DS
Water balance: 0.93TS = 0.65DS + W

DS = .07TS/.35
DS = .07(52.5 lb.)/.35 = 10.5 lb.
W = .93(52.5 lb.) - .65(10.5 lb.) = 42 lb.

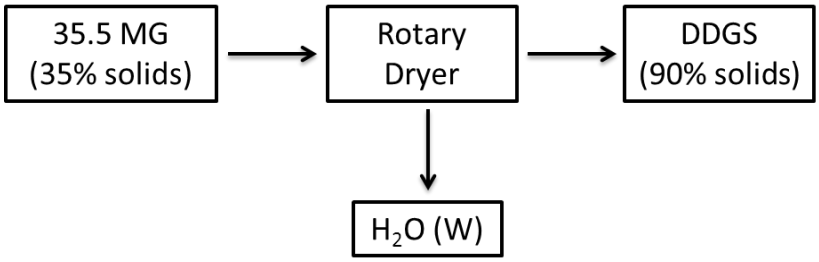
Energy Consumed by Triple Effect Evaporator

42 lb_{H2O} $\times \frac{559\ BTU}{lbH2O}$ = 23478 BTU



Material balance: WDG + DS = MG
Solids balance: 0.35WDG + 0.35DS = 0.35MG

MG = WDG + DS
MG = 25 lb. + 10.5 lb. = 35.5 lb.



Material balance: MG = W + DDGS
Solids balance: 0.35MG = 0.90DDGS
Water balance: 0.65MG = 0.10DDGS + W

DDGS = .35MG/.90
DDGS = .35(35.5 lb.)/.90 = 13.81 lb.
W = .65MG - .10DDGS
W = .65(35.5 lb.) - .10(13.81 lb.) = 21.69 lb.

Energy Consumed by Rotary Dryer

$21.69\ lb_{H2O} \times \frac{1500\ BTU}{lbH2o} = 32535\ BTU$

Total Energy

32535 BTU + 23478 BTU + 258 BTU = 56271 BTU

$\frac{56271\ BTU}{100\ lb.of\ WS} = 562.71\ BTU/lb\ WS$

HTC Processing of Whole Stillage

Assumptions:

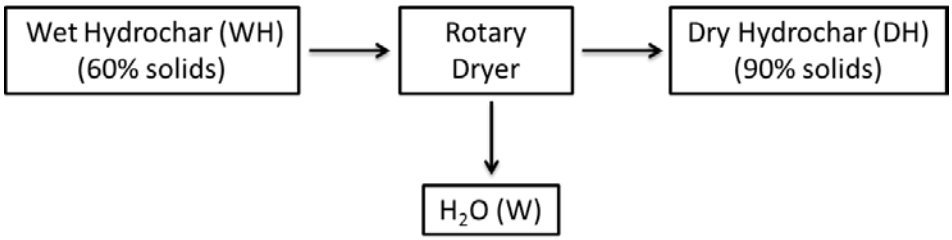
- Whole Stillage (WS) - 14% solids
- 50% heat recovery through use of heat exchangers
- 32% mass yield of unextracted hydrochar

- Hydrochar - 60% solids
- Rotary Dryer – 1500 BTU/lb_{H2O}

100 lb. WS $\times \frac{0.4535\ kg}{1\ lb.}$ = 45.36 kg WS

$q = mC_p\Delta T$
q = 45.36 kg (4.3395 kJ/kg °C) (200°C - 25°C) = 34446.95 kJ
34446.95 kJ × .5 (heat recovery) = 17223.48 kJ
17223.48 kJ $\times \frac{1\ BTU}{1.055\ kj}$ = 16325.57 BTU

100 lb. WS × .14 solids = 14 lb. solids
14 lb. × .32 char yield = 4.48 lb. hydrochar



Material balance: WH = W + DH
Solids balance: 0.6WH = 0.9DH
Water balance: 0.4WH =W + 0.1DH

WH = .9DH/.6
WH = .9(4.48 lb)/.6 = 6.72 lb.
W = .4WH - .1DH
W= .4(6.72 lb.) - .1(4.48 lb.)= 2.24 lb. H_2O

2.24 lb_{H2O} $\times \frac{1500\ BTU}{lbH2O}$ = 3360 BTU

Total Energy

16325.57 BTU + 3360 BTU = 19685.57 BTU

$\frac{19685.57\ BTU}{100\ lb.of\ WS}$ = 196.86 BTU/lb. WS

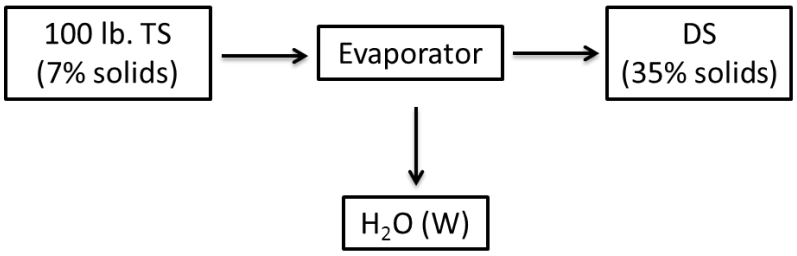
Comparing Convention to HTC

$\frac{196.86\ BTU/lb.WS}{562.71\ BTU/lb\ WS}$ = 0.350 × 100 = 35.0%

Conventional Processing of Thin Stillage

Assumptions:

- Thin Stillage (TS) - 7% solids
- Distiller Solubles (DS) - 35% solids
- Dried Distillers Solubles (DDS) - 90% solids
- All Thin Stillage is used to produce Dried Distillers Solubles
- Evaporator (triple effect) – 559 BTU/lb_{H2O} [1]
- Rotary Dryer – 1500 BTU/lb_{H2O}

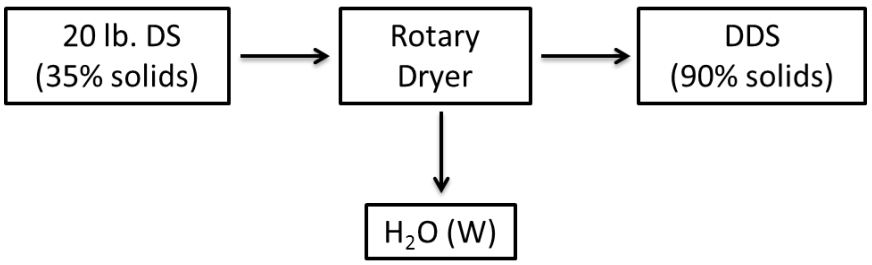


Material balance: TS = W + DS
Solids balance: 0.07TS = 0.35DS
Water balance: 0.93TS = W + 0.65DS

DS = .07TS/.35
DS = .07(100 lb.)/.35 = 20 lb.
W = .93(100 lb.) - .65(20 lb.)= 80 lb.

Energy Consumed by Triple Effect Evaporator

80 lb_{H2O} × $\frac{559\ BTU}{lbH2O}$ = 44720 BTU



Material balance: DS = W + DDS
Solids balance: 0.35DS = 0.9DDS
Water balance: 0.65DS = W + 0.1DDS

DDS = .35DS/.9
DDS = .35(20 lb.)/.9 = 7.78 lb.
W = .65(20 lb.) - .1(7.78 lb.) = 12.22 lb.

Energy Consumed by Rotary Dryer

$12.22\ lb_{H2O} \times \frac{1500\ BTU}{lbH2o} = 18330\ BTU$

Total Energy Consumed

18330 BTU + 44720 BTU = 63050 BTU

$\frac{63050\ BTU}{100\ lb.of\ TS} = 630.5\ BTU/lb\ TS$

HTC Processing of Thin Stillage

Assumptions:

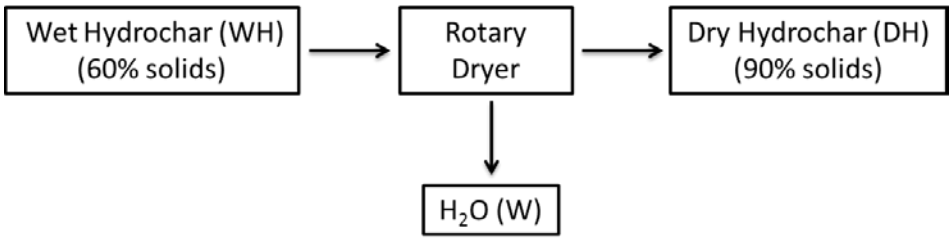
- Thin Stillage (TS) - 7% solids
- 50% heat recovery through use of heat exchangers
- 17% mass yield of dry unextracted hydrochar
- Hydrochar - 60% solids
- Rotary Dryer – 1500 BTU/lb_{H2O}

$100\ lb.\ TS \times \frac{0.4535\ kg}{1\ lb.} = 45.36\ kg\ TS$

$q = mC_p\Delta T$

q = 45.36 kg (4.3395 kJ/kg °C) (200°C - 25°C) = 34446.95 kJ
34446.95 kJ × .5 (heat recovery) = 17223.48 kJ
17223.48 kJ × $\frac{1\ BTU}{1.055\ kj}$ = 16325.57 BTU

100 lb. TS × .7 solids = 7 lb. solids
7 lb. × .17 char yield = 1.19 lb. hydrochar



Material balance: WH = W + DH
Solids balance: 0.6WH = 0.9DH
Water balance: 0.4WH =W + 0.1DH

WH = .9DH/.6
WH = .9(1.19 lb.)/.6 = 1.79 lb.
W = .4WH - .1DH
W= .4(1.79 lb.) - .1(1.19 lb.)= 0.597 lb. H_2O

0.597 lb_{H2O} × $\frac{1500\ BTU}{lbH2O}$ = 895.5 BTU

Total Energy

16325.57 BTU + 895.5 BTU = 17221.07 BTU

$\frac{17221.07\ BTU}{100\ lb.of\ TS}$ = 172.21 BTU/lb. TS

Comparing Conventional to HTC

$\frac{172.21\ BTU/lb.TS}{630.5\ BTU/lb\ TS}$ = 0.268 × 100 = 27.3%

A.8

Anaerobic Digestion of HTC Filtrate

Thin Stillage Flow [a]	190000 L/h
Thin Stillage TCOD	89.089 g/L
HTC Filtrate Flow	~190000 L/h
HTC Filtrate TCOD	50.133 g/L
Methane Yield [b]	0.26715 L/g TCOD
Methane Production Rate	2279873 L/h
Methane Lower Heating Value at STP [c]	35.846 kJ/L
Energy Production Rate	$9.12 * 10^7$ kJ/h
	$8.65 * 10^7$ BTU/h
HTC Processing Energy Requirement	172.21 BTU/lb TS
Density of TS	~1 kg/L
HTC Energy Consumption Rate	$7.21 * 10^7$ BTU/h
Net Energy Production Rate	$1.43 * 10^7$ BTU/h

[a] Value from Agler et al., 2008

[b] Methane yield from the 12th day of 100% HTC BMP assay

[c] Value from Khanal 2008

Appendix References

Agler MT, Garcia ML, Lee ES, Schlicher M, Angenent LT. 2008. Thermophilic anaerobic digestion to increase the net energy balance of corn grain ethanol. Environmental science & technology 42(17):6723–9.

Khanal SK. 2009. Anaerobic Biotechnology for Bioenergy Production. Iowa: Wiley-Blackwell.

Rausch KD, Ronald BL. 2006. The future of coproducts from corn processing. Applied Biochemistry and Biotechnology 128:47-86.