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Enhancing the methane production from untreated rice straw using an anaerobic co-digestion approach with piggery wastewater and pulp and paper mill sludge to optimize energy conversion in farm-scale biogas plants

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Enhancing The Methane Production From
Untreated Rice Straw Using An Anaerobic Co-
Digestion Approach With Piggery Wastewater
And Pulp And Paper Mill Sludge To Optimize
Energy Conversion In Farm-Scale Biogas Plants

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Environmental Technologies for Contaminated Solids, Soils, and Sediments (ETeCoS3)

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Dottore di Ricerca in Tecnologie Ambientali



Degree of Doctor in Environmental Technology

Thèse – Tesi di Dottorato – PhD thesis

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Defended on the 12th of December, 2013

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Erasmus Joint doctorate programme in Environmental Technology for Contaminated Solids, Soils and Sediments (ETeCoS³)

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Abstract

English

The research describes an optimized waste-to-energy technology that utilizes agricultural residues for renewable energy, while reducing global methane emissions and maintaining food security. Laboratory-, pilot- and farm-scale anaerobic batch digesters were evaluated to enhance methane production from the anaerobic digestion of untreated rice straw in dry conditions using a novel co-digestion approach.

An existing farm-scale biogas plant loaded with rice straw and piggery wastewater produced 295 MWh in a 422-day digestion cycle. The long acclimation period (approximately 200 days) and low biogas yield (181 LCH₄/kgVS) could be enhanced by adding anaerobic sludge from the pulp and paper mill treatment process. In a laboratory setting, the addition of the sludge resulted in a specific methane yield of 335 LCH₄/kgVS within 92 days. Hydrolysis of the straw was accelerated, and stable conditions were observed in terms of pH, alkalinity and nutrients. Similar improvements were demonstrated in pilot-scale digesters (1 m³) – a specific methane yield of 231 LCH₄/kgVS was achieved in a 93-day digestion cycle with the sludge compared to 189 days without the sludge. Insufficient mixing within the pilot-scale system caused lower overall methane yields than those obtained in the laboratory-scale digesters.

If sufficient mixing and mesophilic conditions are maintained within the farm-scale system, the co-digestion of rice straw with pig wastewater and paper mill sludge (wet weight ratio of 1:1.25:0.5) has the potential to reduce the retention time to three months (versus 422 days) and increase methane yields to over 300 LCH₄/kgVS.

Italian

Il presente lavoro di ricerca riguarda un sistema di trattamento biologico ottimizzato volto alla trasformazione di residui colturali in energia rinnovabile, garantendo la sicurezza alimentare e contribuendo alla riduzione delle emissioni globali di metano. Sono stati utilizzati digestori batch in scala di laboratorio, scala pilota e scala dimostrativa operanti in condizioni dry al fine di incrementare la produzione di metano a partire da paglia di riso non pre-trattata.

L'impianto in scala dimostrativa alimentato con paglia di riso e reflui suinicoli ha consentito, in un ciclo di digestione della durata di 422 giorni, una produzione di 295 MWh,

mediante la valorizzazione energetica del biogas. Test in scala di laboratorio hanno evidenziato che attraverso l'aggiunta di fango anaerobico proveniente dal trattamento di reflui di cartiera si potrebbero ridurre i tempi di acclimatazione (relativamente elevati, approssimativamente 200 giorni) ed incrementare la produzione specifica di metano (181 LCH₄/kgVS). In particolare l'aggiunta del fango ha determinato una produzione specifica di metano di 335 LCH₄/kgVS in 92 giorni. L'idrolisi della paglia si è conclusa molto più rapidamente e sono state osservate condizioni stabili di pH, alcalinità e contenuto di nutrienti. Miglioramenti analoghi sono stati osservati in digestori in scala pilota (1 m³). In un ciclo di digestione di 93 giorni, mediante l'aggiunta di fango, è stata misurata una produzione specifica di metano pari a 231 LCH₄/kgVS, contro i 189 giorni necessari in assenza di fango. La differenza tra la produzione massima di metano osservata nei digestori in scala laboratorio e pilota è imputabile al ridotto grado di miscelazione nei digestori in scala pilota.

In definitiva, assicurando condizioni mesofile ed un sufficiente grado di miscelazione, la co-digestione della paglia di riso con il refluo suinicolo e il fango di cartiera (rapporto in peso umido pari a 1:1.25:0.5) potrebbe completarsi in un tempo di ritenzione pari a tre mesi (contro 422 giorni), con un incremento della produzione di metano fino a valori superiori a 300 LCH₄/kgVS.

French

Ce travail de thèse présente l'optimisation d'une technologie de valorisation énergétique qui utilise des résidus agricoles pour la production d'énergies renouvelables, tout en réduisant les émissions mondiales de méthane et en garantissant la sécurité alimentaire. Des digesteurs anaérobies à l'échelle laboratoire, pilote et industrielle ont été évalués afin d'améliorer la production de méthane à partir de la digestion anaérobie de la paille de riz non traitée dans des conditions sèches en utilisant une approche nouvelle de co-digestion.

Une installation de production biogaz à l'échelle d'une ferme chargée de paille de riz et d'eaux usées produites par une porcherie génère 295 MWh dans un cycle de digestion 422 jours. La période d'acclimatation relativement longue (environ 200 jours) et le faible rendement en biogaz (181 LCH₄/kg MVS) pourraient être améliorées en ajoutant des boues anaérobies issues d'un procédé de traitement d'effluents de l'industrie papetière. Au laboratoire, l'ajout de la boue conduit à un rendement de méthane spécifique de 335 LCH₄/kgMVS dans les 92 jours.

L'hydrolyse de la paille a été accélérée, et des conditions stables ont été observées en termes de pH, d'alcalinité et de nutriments. Des améliorations similaires ont été démontrés dans des digesteurs à l'échelle pilote (1 m³) - un rendement de méthane spécifique de 231 LCH₄/kgMVS a été obtenu dans un cycle de digestion à 93 jours avec de la boue comparativement à 189 jours sans la boue. Un mélange insuffisant dans le système à l'échelle pilote a causé des rendements de production de méthane inférieurs à ceux obtenus dans les digesteurs l'échelle du laboratoire.

Si les conditions mésophiles et de mélange suffisantes sont maintenues dans le système à l'échelle industrielle, la co-digestion de la paille de riz avec des eaux usées produites par une porcherie et des boues issues d'un procédé de traitement d'effluent de l'industrie papetière (rapport poids humide de 1:1.25:0.5) a le potentiel de réduire le temps de rétention à trois mois (contre 422 jours) et d'augmenter les rendements de production de méthane à plus de 300 LCH₄/kgMVS.

Dutch

Het onderzoek beschrijft een geoptimaliseerde afval-tot-energie technologie welke landbouwresidu gebruikt als bron van duurzame energie, terwijl de wereldwijde uitstoot van methaan wordt verminderend en voedselzekerheid niet in het gedrang komt. Anaërobe batch vergisters op laboratorium-, proef- en boerderijschaal werden geëvalueerd met als doel de methaanproductie van de anaërobe vergisting van onbehandelde rijststro onder droge omstandigheden te verbeteren met behulp van een nieuwe co-vergistingsbenadering.

Een bestaande biogasinstallatie op boerderijschaal, gevoed met rijststro en varkensstal afvalwater produceerde 295 MWh in een 422-daagse vergistingscyclus. De lange acclimatisatieperiode (ongeveer 200 dagen) en lage biogasopbrengst (181 LCH₄/kgVS) konden verbeterd worden door het toevoegen van anaëroob korrelslib van een pulp-en papier fabriek waterzuiveringsproces. Onder laboratoriumomstandigheden leidde de toevoeging van het korrelslib tot een specifieke methaanopbrengst van 335 LCH₄/kgVS binnen 92 dagen. Hydrolyse van het stro werd versneld bij een stabiele pH, alkaliteit en nutriënten concentratie. Vergelijkbare verbeteringen werden aangetoond in vergisters op pilotschaal (1 m³): een specifieke methaanopbrengst van 231 LCH₄/kgVS werd bereikt in een vergistingscyclus van 93 dagen met de toevoeging van het korrelslib, in vergelijking met een cyclus van 189 dagen zonder het

anaërobe slib. Onvoldoende menging in het pilotschaal systeem leidde tot een lagere totale methaanopbrengst dan die verkregen in de laboratoriumschaalvergisters.

Indien voldoende gemengd wordt, en mesofiele omstandigheden worden gehandhaafd binnen het systeem op boerderijschaal, dan heeft de covergisting van rijststro met varkensstal afvalwater en papierfabriek korrelslib (nat gewichtverhouding van 1:1.25:0.5) het potentieel om de retentietijd terug te brengen tot drie maanden (versus 422 dagen) en om de methaanopbrengst tot meer dan 300 LCH₄/kgVS te verhogen.

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CHAPTER 1

Introduction

1.0 Research Context

The major global challenges that exist today involve climate change, the energy crisis, and food security for a growing population. Environmentally sustainable solutions must contribute positively toward solving each of these challenges, while maintaining a cost-effective approach that is practical for large-scale applications. The research presented in the following chapters proposes an optimized waste-to-energy technology that converts agricultural residues into a renewable energy source, while reducing global methane emissions and maintaining valuable land resources for food production.

1.1 Renewable Energy

The move toward renewable energy is motivated by concerns over global warming and increasing costs of fossil fuels. In 2012, 138 countries had established policy targets [1] such as the EU Renewable Energy Directive which aims for a 20% renewable energy share in energy consumption by 2020 [2]. These legislative policies often offer subsidies and economic incentives to promote research, growth and development. A total of 244 billion USD was spent in 2012 on new renewable projects across the globe [1]. Renewable energy is produced from a variety of replenishable sources including solar, wind, biomass, hydropower, and geothermal. On a global scale, electricity produced from hydropower (80.5%) was the major contributor toward the renewable energy share in 2011, followed by wind (10.3), biomass (6.2%), geothermal (1.6%) and solar (1.4%) [3]. Although solar and wind power had significant momentum over the last decade with mean annual growth rates of 46% and 28%, respectively [3], these are considered variable renewables because they fluctuate depending on the environmental conditions. Biomass, however, is a more stable source of energy that can help countries like Denmark reach their long-term energy strategy to become fossil free by 2050 [4].

1.2 Second Generation Biomass

Second generation biomass consists of agricultural residues that remain in the field after the food crop is harvested such as leaves, stems, straw, or husks. Woody crops such as grasses, poplar, willow, and wood chips are also considered to be second generation biomass. Utilization of second generation biomass promotes the use of valuable land resources for food production while still capturing energy from the waste

residues. This alleviates the concern over food scarcity, especially considering the global population will reach 9 billion people by the year 2050, and that 70 to 100% more food will be necessary [5]. Another major concern is the impact of agriculture on global climate change. Agricultural activities such as fermentation of residues in the soil, biomass burning, manure management, etc. constitute approximately 47% of global anthropogenic methane emissions [6]. An effective mitigation strategy is to remove the residues from the field and capture the methane in order to reduce the emission of greenhouse gases, and simultaneously use the methane as an energy source. The advantages of energy recovery from second generation biomass are highlighted in Figure 1-1.

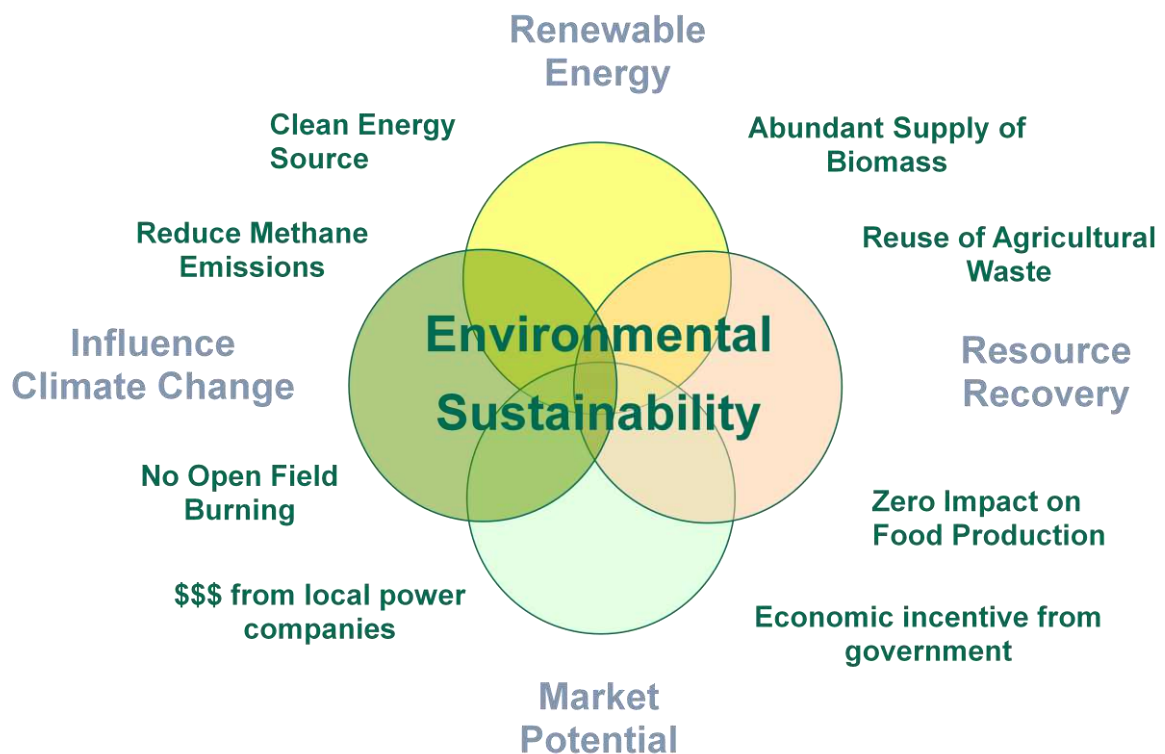


Figure 1-1. Advantages Associated with the Energy Recovery from Second Generation Biomass

1.3 Anaerobic Digestion

One way that biomass can be converted into energy is through anaerobic digestion. Anaerobic digestion is a natural process in which a variety of microorganisms degrade organic matter into several intermediate products that are converted into a renewable energy source known as methane (CH_4). The stages of

anaerobic digestion and general classifications of microorganisms involved are shown in Figure 1-2. Depending on the total solids (TS) concentration of the waste material,

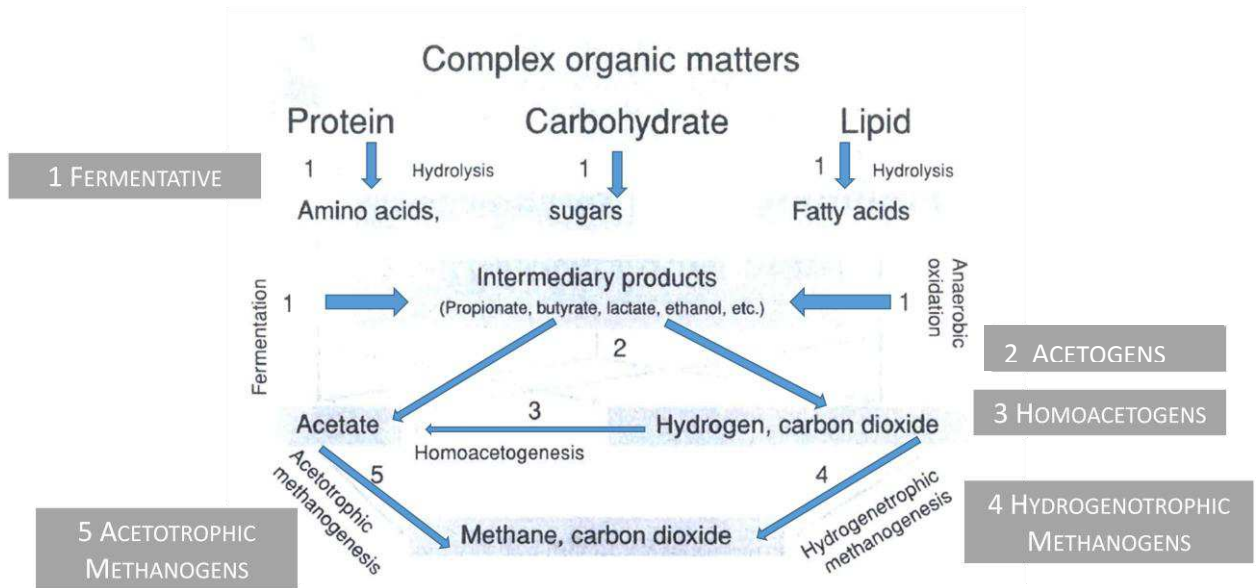


Figure 1-2. Microorganisms Involved in Anaerobic Digestion (adapted from Khanal, 2008 [12])

anaerobic digestion can be applied in wet (< 15% TS), semi-dry (15-20% TS) or dry (>20% TS) conditions. The anaerobic digestion of lignocellulosic biomass like rice straw occurs faster in wet conditions, but the overall methane yield and digestability of the straw is essentially the same in both wet and dry systems [7, 8]. The advantages of dry systems opposed to wet systems include water savings, elimination of wastewater disposal, and reuse of the solid residues as fertilizer. Anaerobic digestion systems can be designed as either batch reactors in which all the substrate/inocula mixture is added at the beginning, or continuously-fed reactors in which the substrate/inocula mixture is added incrementally over time. Batch reactors are much simpler and less expensive (40%), but they have larger volume requirements and need a larger area footprint to place the reactors [9].

Biogas generated from the anaerobic digestion process consists primarily of CH_4 (50 to 65%) and carbon dioxide (35 to 40%), with a balance of nitrogen and trace amounts of hydrogen sulphide and water vapor. CH_4 can be used directly as fuel for cooking and heating, converted into electricity by a combustion engine, or compressed and used as an alternative fuel for motor vehicles. In 2011, 57% of the biogas produced in Europe (*i.e.* 10.1 million tons of oil equivalent) was from biomass sources including decentralized agricultural plants, household wastes and green waste methanation plants

or centralized co-digestion facilities [10]. The production of biogas through anaerobic digestion is considered to be one of the cleanest approaches to recovering energy from biomass [2, 11].

1.4 Research Objectives

The overall goal of the research was to enhance methane production from the anaerobic digestion of untreated rice in dry conditions using a novel co-digestion approach. Laboratory-, pilot- and farm-scale batch digesters were evaluated and specific research objectives were as follows: 1) monitor an existing farm-scale system for rice straw digestion; 2) implement pilot-scale digesters with varying temperature conditions and co-digestion approaches for optimization of the farm-scale plant; and 3) study a novel co-digestion strategy that utilizes both pig wastewater and sludge from the pulp and paper mill industry to enhance methane production. The steps taken to accomplish these objectives are described in the following chapters and depicted as a graphical abstract in Figure 1-3.

A review from existing literature that highlights the motivation and operational strategies for implementing the anaerobic digestion of rice straw is discussed in Chapter 2. Pilot-scale (1 m³) experiments with rice straw co-digested with pig wastewater were conducted to define minimal and optimal conditions required for farm-scale operations and results are reported in Chapter 3. A farm-scale operation that produced electrical energy from rice straw co-digested with pig wastewater was studied and data from this plant is included in Chapter 4. An attempt to enhance methane production from rice straw using anaerobic sludge from the pulp and paper mill treatment process along with pig wastewater was made and the lab results are included in Chapter 5. Chapter 6 highlights a third pilot-scale experiment which incorporates the paper mill sludge as a comparison to the previous pilot-scale digesters to determine if this approach is feasible for the farm-scale plant. In Chapter 7, the overall findings are summarized as well as a discussion on alternative uses for biogas, health concerns with the digestate, and recommendations for future research.

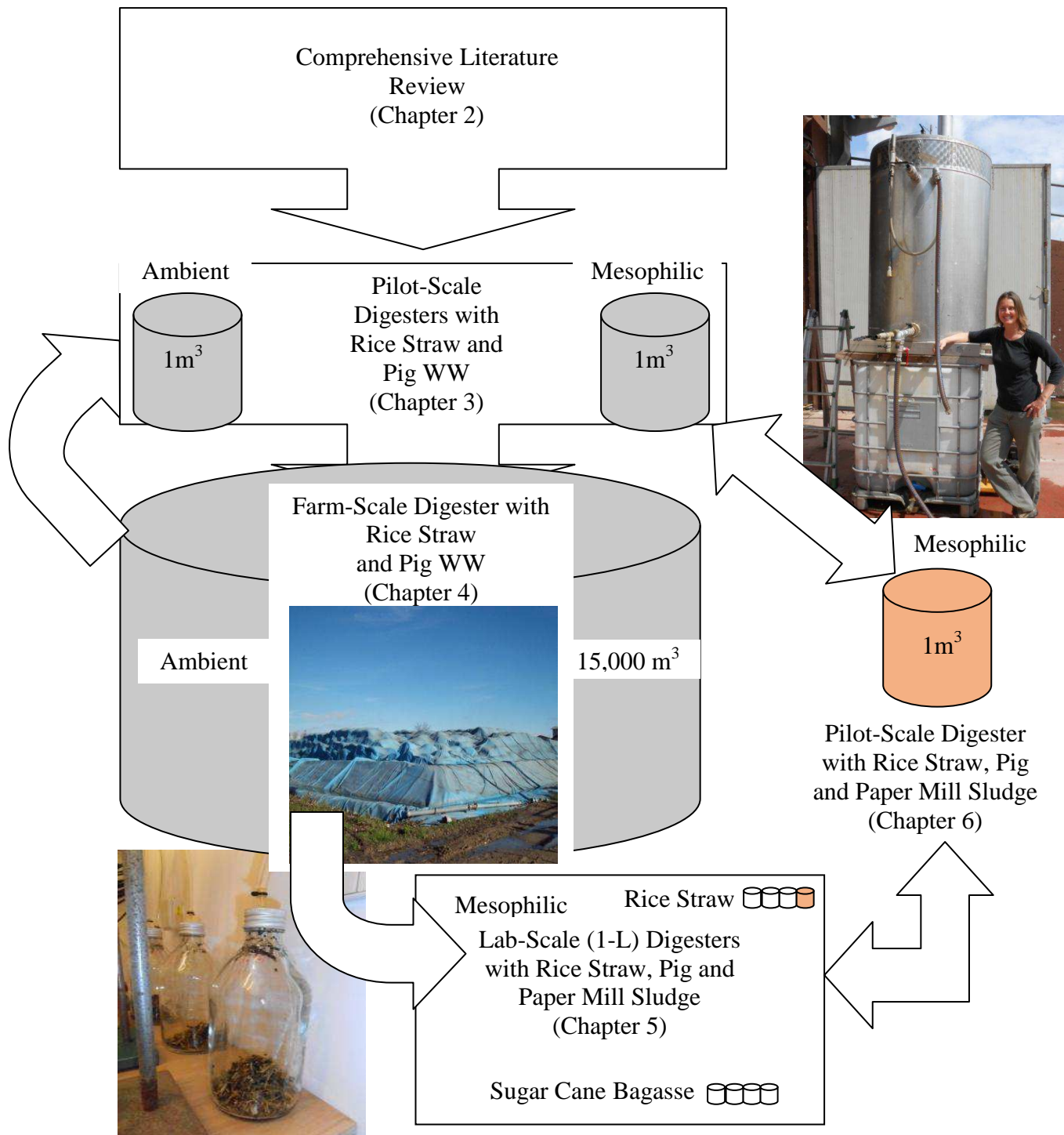


Figure 1-1. Graphical Abstract of Research Activities

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CHAPTER 2

The Anaerobic Digestion of Rice Straw: A Review

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Introduction

Rice is the most important staple food providing nutrition and calorie intake for over half of the world's human population [1], and rice straw is one of the most abundant and renewable energy sources in the world [2]. For every ton of rice harvested, approximately 1.35 tons of rice straw remain in the field with energy potential [3]. Rice is the world's third largest crop behind maize and wheat [1], and the waste product also ranks as the world's third largest agricultural residue [4]. Based on the most recent data available by the Food and Agriculture Organization of the United Nations, a total of 718 million tons of rice were produced in 2012 [5], which equates to approximately 969 million tons of rice straw available worldwide. In 2004, a global annual production of 731 million tons of rice straw was reported [6], but there has clearly been an increasing trend of global rice production in the last decade [7]. Rice straw is a very common agricultural waste and the biogas production potential is appealing to both developed and developing countries. In China, nearly 740 million tons of rice straw were generated in 2006 and approximately 47% of the residues were used for cooking and heating on the household scale [8].

Rice straw is a fibrous, lignocellulosic biomass that remains in the field once the grain is harvested. The straw can be collected and baled once it contains a moisture content below 25%, which can be as soon as 3 to 4 days following harvest depending on climatic conditions [3]. Upon baling, the straw appears as flat fibres with approximate dimensions of 0.5 cm in width and 20 to 60 cm in length. Figure 2-1 contains a photograph of rice straw taken from a cylindrical bale within four months of harvest. Table 2-1 is a summary table showing the typical composition of rice straw [3].

Waste biomass, such as rice straw, can be converted to fuel through biological or thermochemical processes [9]. Biological processes utilize bacteria to convert the biomass into fuel either through anaerobic digestion of organic matter generating methane or through saccharification and fermentation of sugars [1, 10] producing ethanol. Utilizing rice straw for ethanol production has been investigated [6, 10-12], and the global production potential was estimated to be 205 giga-liters (GL), which could replace approximately 147 GL of gasoline [6]. Aerobic composting of rice straw has been evaluated with various other substrates for its use as a fertilizer [13, 14]. Thermochemical processes such as pyrolysis [15, 16], combustion [17, 18], and gasification have also been evaluated as treatment methods of rice straw [1]. However,

Table 2-1. Rice Straw Composition [3]

Feedstock Component	Dry Wt. %
Glucan	38.9
Mannan	0.0
Galactan	0.5
Xylan	20.4
Arabinan	3.4
Lignin	13.5
Extratives	5.3
Ash	18.0
Total	100



Figure 2-1. Photograph of Rice Straw

biological processes require much less energy input when compared with thermochemical processes and can accommodate either wet or dry feedstocks economically on both small and large scales [19]. Financial incentives and legislation are growing in support of this technology, and it is ideal way to recover costs from excess agricultural waste in a farm-scale system. Thus,

the focus of this review article is solely on the potential energy production from rice straw through the anaerobic digestion process.

The decomposition of rice straw (*Oryza sativa*) by means of anaerobic digestion is not a new concept. Rice straw has been studied for the past century by researchers such as Richards and Amoores in the 1920's, Acharya in the 1930's, Kalra, Sun and Hills in the 1980's, and numerous authors in the last 20 years [2, 8, 20-32]. The recent interest in rice straw digestion stems from a global focus on efficiently using renewable energy sources and reducing greenhouse gas emissions contributing to climate change. Common practices such as open-field burning of rice fields or tilling the straw back into the fields contribute significantly to the release of methane into the atmosphere [26, 33, 34].

Historically, rice straw has not been a selected substrate for energy production because of its complex, lignocellulosic structure that makes it difficult to decompose [3, 23, 25, 31]. But several emerging factors including the abundance of rice straw, discoveries on appropriate inocula and pretreatment strategies, and the depleted tolerance for wasted biomass contributing to greenhouse gas emissions, support the perspective that rice straw can no longer be overlooked as a viable renewable energy source that must be captured and utilized. This review paper seeks to inform the reader about the climate impacts, energy potential, optimal nutrient balance and operational parameters, successful pretreatment strategies, microbiological considerations and data gaps associated with the anaerobic digestion of rice straw.

2.2 Greenhouse Gas Emissions from Rice Fields

Anaerobic digestion of biomass that occurs naturally in the environment is a very substantial source of greenhouse gas emissions [4]. If methane from biomass sources could be captured, then not only could it be used as a clean energy source but it would also reduce greenhouse gas emissions that contribute to global warming. Rice fields are a major line item in the “global methane budget” [26] and expanded rice field cultivation has led to increased tropospheric methane concentrations contributing to global climate change [35]. Flooded rice fields are responsible for 10 to 15% of the worldwide anthropogenic methane emissions [36, 37], and the major contributing factor is rice straw. Several mitigation strategies have been proposed to reduce emissions while sustaining rice production including water management, soil amendments that inhibit methanogenic activity, and applying composted amendments (i.e.

manure with rice straw) rather than fresh organic material [35, 37]. However, the common practices are open burning of rice straw or tilling it back into the soil, both contributing to the problem of greenhouse gas emissions and climate change [1, 8, 26, 28, 33, 38].

Rice straw is commonly tilled back into the soil and used as fertilizer for the crops. Rice straw in the soil has a half-life of two years and approximately 80 to 90% is decomposed within the first year [26]. Rice fields are often flooded in dormant periods creating anoxic conditions, and the decaying organic matter releases gases such as nitrogen, hydrogen, methane, ammonia, and hydrogen sulfide [39]. Thus, the practice of tilling the straw back into the field results in increased methane emissions from rice fields. Methane emissions from rice paddy soils were reduced by 95% for a conventional rice variety and 96% for a high biomass-yielding rice variety when the straw was removed rather than returned to the fields [12]. Methane emission rates increased by a factor of 25 when rice fields in China were fertilized with a mixture of rice straw and ammonium sulfate rather than ammonium sulfate alone; moreover, there were no advantages of using the rice straw mixture since the grain yields (i.e. rice production) were the same under both conditions [37]. In a series of bottle tests, anoxic rice paddy soil was mixed with untreated, chopped rice straw at a soil to straw ratio of 80 to 1, and methane production rates were measured over a 28-day period [26]. By the end of the experiment, the bottles with the added rice straw were producing eight times more methane than the anoxic soil without the straw [26]. In a similar study with a soil to straw ratio of 100 to 1, over 20 times more methane was produced in the anoxic rice soil amended with straw after 30 days of incubation at 32°C when compared to the unamended soil [39]. Some quantity of rice straw will naturally be left in the fields even if some removal technique is used (i.e. cutting and baling); however, the practice of tilling the straw back into the soil results in increased greenhouse gas emissions and has detrimental effects on the environment.

Further compounding the issue is the open burning of crop residues, which releases gases such as carbon dioxide, carbon monoxide, methane, non-methane hydrocarbons, nitrogen compounds, sulfur dioxide and particulate matter into the atmosphere [33]. The significant greenhouse gases that contribute to global warming and are produced during the open burning of rice straw are nitrous oxide and methane [33]. The carbon dioxide released during the process is considered neutral since it is taken up during the growth stage of the crops [33, 36], and it also has very little global warming potential (i.e. 20 times less than methane) [1, 4, 12]. A study was

conducted to quantify the rice straw burned in India, Thailand and the Philippines annually and to calculate the related air pollutant emissions [33]. Using emissions factors specific to rice straw, a combined total of 33,000 megagrams (Mg) of methane (1 Mg = 1 ton) and 2000 Mg of nitrous oxide were estimated to be released into the atmosphere annually from open field burning of rice straw (total of 35 million tons) in these countries [33]. In 2006, China disposed of approximately 115 million tons of rice residues through open-field burning [8].

In a life cycle assessment (LCA) completed for the production of white-milled rice in Northern Italy, the one major contributor to the global warming potential was field emissions (68%), followed by fertilizers (9.2%), transportation (6.1%), refining and packing (4.7%), field operations (3.6%) and other minor contributors [36]. Removing rice straw from the fields following harvest is clearly an effective mitigation strategy for reducing methane emissions from rice cultivation practices and total greenhouse gas emissions from a wide variety of rice cropping systems [12]. The contribution from fertilizers would likely decrease since the digestate could be reapplied to the fields instead of using processed chemicals. The contribution from field operations would increase since the straw would have to be cut, baled and transported or loaded into an on-site digester. Additional contributors such as emissions generated from energy conversion (i.e. use of internal combustion engine to convert methane to electricity) and emissions associated with the initial construction would have to be considered in the LCA. However, if 68% of the global warming potential could be eliminated and a clean biofuel was produced, this would be a substantial improvement for the impact that the rice production industry has on greenhouse gas emissions.

2.3 Biogas Production Potential of Rice Straw

Biogas is the form of energy produced when microorganisms decompose organic matter in the anaerobic digestion process. Biogas is primarily composed of methane and carbon dioxide with trace amounts of hydrogen sulphide, ammonia and water vapor. Methane produced from biomass is a clean, renewable energy source that currently represents approximately 14% of the global energy consumption [1, 40], and is the primary source of energy for over half of the world's population [1]. Fewer air pollutants and less carbon dioxide per unit of energy are released when compared with non-renewable fossil fuels [19].

Europe is the leading producer of biogas with Germany clearly at the forefront [41]. In 2009, the European Union produced 4,340.7 million tons of oil equivalent (amounting to 13,448.3 GWh of electricity) from decentralized agricultural plants, co-digestion plants, and multi-product methanisation plants [41]. Agricultural biogas plants are increasing (especially in Germany), however, the majority of them use food-based or energy crops such as cereals and maize [24, 41, 42]. Using food crops for energy production is controversial because the demand for food is expected to increase in the future and food prices are likely to rise as a result [24]. Food security is a top global priority and using lignocellulosic materials such as rice straw for energy production does not interfere with that priority [6]. The use of agricultural waste products is more desirable because of high availability and reduced greenhouse gases released into the atmosphere when the waste products are utilized rather than left in the field to decompose. When compared with six other lignocellulosic biomasses (wheat straw, oat straw, barley straw, sorghum straw, corn stover and sugar cane bagasse), rice straw was selected as the most favorable feedstock for energy production primarily because of the quantity available [6]. Though the organic matter is not completely converted by the anaerobic digestion of rice straw, the remaining residues can be used as topsoil maintenance or sustainable growth for biomass [19]. When considering factors such as purchase price, potential fuel yields, and environmental concerns, cellulosic biomass can significantly contribute to energy sustainability and security [40].

The methane potential, however, of untreated rice straw is on the lower end when compared to other agricultural biomasses and agro-industrial by-products. Table 2-2 includes methane yields in terms of dry matter that have been determined for various food crops and agricultural by-products. The potential methane production from anaerobic digestion of rice straw has been evaluated under many different conditions in the context of bottle tests, batch reactors and pilot-scale studies [2, 8, 20-22, 24-31, 38, 43]. Several studies have been conducted to determine the ultimate methane yield of rice straw with various inocula, and the results range from 92 to 404 L/kg of VS added at ambient and mesophilic temperatures (see Table 2-3). There is considerable variation in methane yield of straw depending on the type of pretreatment, if any, and the digestion conditions [44].

Table 2-2. Methane yields (in terms of TS) Associated with Various Agricultural Biomasses

	Methane yield (L/kg TS)	Reference
Triticale chopped/squashed	310 / 320	[45]
Rape chopped/squashed	300 / 350	[45]
Oat chopped/squashed	240 / 280	[45]
Jerusalem Artichoke squashed	240	[45]
Sunflower squashed	200	[45]
Wheat squashed	290	[45]
Rye squashed	290	[45]
Maize chopped/squashed/ripped	300/330/300	[45]
Maize drying up residues	378	[24]
Tomato skin and seeds	227	[24]
Barley Straw	239	[24]
Grape stalk	290	[24]
Grape marc	171	[24]
Rice Straw	202	[24]
Rice Straw	240	[8]
Rice Straw	193	[28]

Table 2-3. Methane Yields (in terms of VS) from Anaerobic Digestion of Rice Straw

Methane Yield (L/kgVS added)	Type of Pretreatment	Digestion Temp (°C)	Time period (days)	Reference
46	Cut (1 cm)	35	92	[46]
231	Codigestion with Pig WW	35	189	[47]
302-340	Cut (1 cm) Codigestion with Pig WW & Paper Mill Sludge	35	92	[46]
195	Cut (50-100 mm)	40	40	[24]
280	Cut (3-5 mm)	22	120	[8]
215	Pulverized	35	120	[9]
190	2% NH ₃	35	24	[31]
198	Cut (25 mm) + 2% NH ₃	35	24	[31]
245	Ground (25 mm), 2% NH ₃ , preheated to 110°C	35	24	[31]
273	Cut/pre-digested with biogas sludge for 46 hrs	26-28	146	[29]
224*	Cut/delignified	30	63	[25]
328*	Cut/delignified/white rot fungi	30	63	[25]
296*	Cut/delignified/brown rot fungi	30	63	[25]
240	Cut/white rot fungi	18-28 (Ambient)	89	[28]
92, 93	Milled/white rot fungi	25	59	[28]
120, 124	Milled/white rot fungi	35	59	[28]

* These values are only available in the literature as L/kgTS

2.4 Optimum pH and Buffering Capacity

During anaerobic digestion, organic matter is first converted into sugars, fatty acids, and amino acids in the hydrolysis stage. Acidogenic and acetogenic bacteria further break down these substances and the resulting intermediates are acetic acid, hydrogen and carbon dioxide. The methanogenic bacteria then convert the intermediates into methane and carbon dioxide. The four distinct stages of anaerobic digestion are shown in Figure 2-2. During the anaerobic digestion of rice straw, in particular, approximately 80% of the methane is formed from acetic acid and 20% comes from the conversion of hydrogen and carbon dioxide [26]. The rate-limiting step in biogas production varies depending on the substrate and conditions. In the digestion of lignocellulosic biomass such as rice straw, the rate-limiting step has been defined as the hydrolysis of cellulose [48-51]. With higher (i.e. thermophilic) temperatures, the rate-limiting step is the conversion of acetate to methane by acetoclastic methanogens, which are

known for their slow metabolism and growth rate [28, 52]. Thus, if the system does not have sufficient buffering capacity, methane production will be inhibited by a rapid and over-production of acetic acid [21, 30].

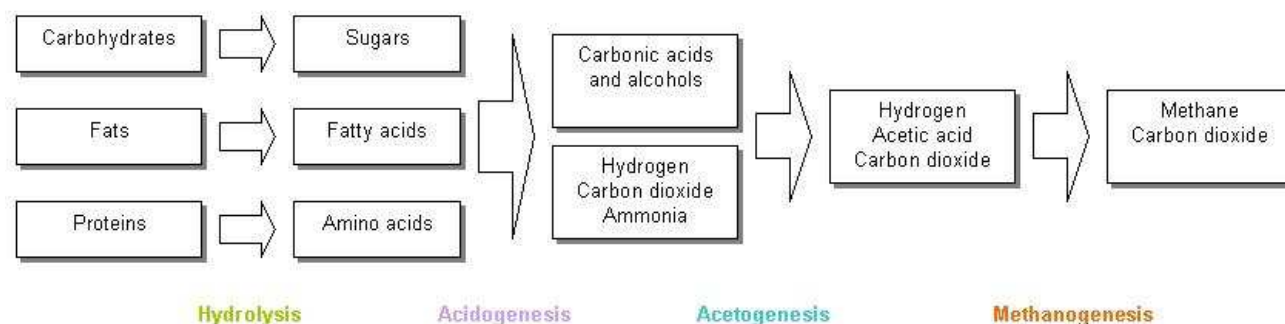


Figure 2-2. Stages of Anaerobic Digestion [53]

Although the acidogenic/acetogenic bacteria can function normally over a broad pH range of 6 to 10 [20], methanogens are far more sensitive to pH [8, 20]. The ideal pH for rice straw digestion was determined by one author to be 7.5 to 8.0 [20], although several batch experiments with rice straw have been successful in pH ranges of 6.5 to 7.3 [2, 8, 26, 28-30]. When acid accumulation created low pH environments (i.e. < 6.0), the methanogens were inhibited and gas production ceased [28, 29]. In a study with grass silage, both the acidogenic bacteria and the methanogens were inhibited when pH was adjusted to 6 using hydrochloric acid [31]. Although the general consensus is that methane inhibition occurs when pH < 6.0, results from a batch fermentation study with glucose as the substrate showed that methanogens became inactive when pH values were between 6.4 to 6.9 [54].

In order to maintain a neutral or slightly alkaline pH during the anaerobic digestion process, the system needs an appropriate buffering agent. Ten different neutralizing compounds and mixtures of compounds were compared in terms of their ability to promote the degradation of rice straw and the best results were obtained using ammonium carbonate for buffering capacity [20]. An alternative option is the co-digestion of rice straw with animal waste to serve as a buffer for the system. There was relatively no difference in pH or biogas yield when rice straw was digested with 0.3% (by volume) of ammonium carbonate compared to digestion with pig manure [29].

2.5 The Appropriate Balance of Nutrients

The appropriate balance of nutrients is a critical factor in the anaerobic digestion process and optimum carbon to nitrogen (C:N) ratios range from 25 to 35 [32, 55]. Untreated rice straw has a very low concentration of total nitrogen (i.e. < 1% on a dry basis) [8, 24, 25, 27], and even less total phosphorus (i.e. 0.044% on dry basis) [8]. A typical C:N ratio for untreated rice straw is approximately 80 [1, 25, 30, 32] and therefore an external source of nitrogen is essential for effective digestion. Rice straw with a C:N (non-lignin carbon to Kjeldahl-nitrogen) ratio of 31 produced 4.5 times more biogas than rice husks with a C:N ratio of 81 [27]. The significantly lower gas yield was attributed to the lower nitrogen concentration and higher lignin content in the rice husks compared to the rice straw [27]. Rice straw digested with cattle manure performed best with a C:N (non-lignin carbon to Kjeldahl-nitrogen) ratio of 25 (versus 12.3, 20, 30, 35 and 40), yielding the highest methane production and lignin reduction [32].

When straw is co-digested with animal manure, appropriate nutrient balance compositions are established and the synergistic effects produce higher methane yields [30, 44, 55, 56]. The biogas production increased by 9% when rice straw was co-digested with cattle dung compared to rice straw alone [27]. Total biogas yield increased by 30% when rice straw was co-digested with pig manure compared to rice straw alone, although the ratio of straw to manure (i.e. 2:1 versus 1:1) made no difference [29]. The degradability of rice husks was increased by 10% when they were used as bedding for pigs and lightly soiled with pig manure compared to unused husks [30]. In a study that compared the methane potential of cattle manure to pig manure, ultimate methane yields were 58% higher in pig manure and the methane plateau phase was reached much faster [44]. Besides recycled nutrients within agricultural waste streams, the benefits of co-digestion with animal manure include enhanced production of a carbon-neutral source of renewal energy and reduced greenhouse gas emissions [56].

Raw piggery wastewater from two full-scale plants in the Emilia-Romagna region of Italy contained influent values ranging from 1500-3500 mg N/kg as total Kjeldahl nitrogen and 350 to 1,000 mg P/kg [57]. During the fermentation process, nitrogen supplied from the pig wastewater is converted to ammonia, which serves as a nitrogen source for the bacteria and also as a weak base to maintain appropriate pH levels for the methanogens and prevent acidification [29, 30]. However, excess ammonia concentrations of approximately 2 g/L are considered inhibitory to unadapted methanogens under mesophilic conditions [56]. If the anaerobic bacteria in wet

digestion systems are given ample time to adapt to higher ammonia concentrations, they can become more resistant to the toxic effects of ammonia [58].

Phosphorus is a macronutrient that is also pertinent to anaerobic digestion. A nitrogen to phosphorus (N:P) ratio of 7 is generally required for anaerobic microbes [59], and untreated rice straw has an N:P ratio of approximately 16 [8]. Since an external nitrogen source is required for the anaerobic digestion of rice straw, the addition of phosphorus would seem to be necessary as well. However, phosphorus supplementation to the anaerobic digestion process of rice straw has not shown favorable results. Batch reactors filled with rice straw and operated under mesophilic conditions with a fixed C:N ratio of 22.24 were supplemented with phosphorus (155, 465 and 775 mg/l P) and potassium (195, 585 and 975 mg/l K), respectively [8]. The addition of phosphorus had very little effect on the anaerobic digestion of rice straw in regards to biogas production, methane concentrations and solids reduction [8]. Another study added 1% phosphorus in the form of K_2HPO_4 to the rice straw and no appreciable differences were observed after 6 months of digestion [20].

Trace elements such as nickel, iron, cobalt, selenium, molybdenum, and tungsten can enhance the growth of methanogens and improve process stability; however, data is very limited regarding the role of these micronutrients in the anaerobic digestion of energy crops, animal wastes and agricultural residues [60]. The addition of iron, cobalt and nickel to a mono-digester with maize silage increased biogas production by 35% when compared to the control [60]. Methane production from Napier grass digestion increased by 40% with the daily addition of nickel, cobalt, selenium and molybdenum [60]. Thus, it is likely that the appropriate addition of micronutrients would enhance the anaerobic digestion of rice straw, but there is need for future work in this area.

2.6 The Effects of Temperature

Temperature is a very important variable to consider in the context of rice straw digestion, not only for efficiency and maximizing methane production but also in regards to economical input. The literature reports that the optimum temperatures for methane production from the anaerobic digestion of straw are in the mesophilic range from 35 to 40°C [28, 61], with one of the earliest discoveries made by Richards and Amoores in 1920 [21]. An historical study was conducted in 1934 to evaluate decomposition of rice straw at different temperatures ranging

from 20 to 45°C [20]. After 6 months, the highest methane production was observed at 35°C, which was 53% higher than the production at 25°C [20]. In a much more recent study, pretreated rice straw was digested using hogger wastewater as the inoculum [28]. When the temperature of the system was increased from 25°C to 35°C, cumulative methane production increased by approximately 25% for both wet and semi-dry conditions [28]. A similar study evaluated eight batch reactors containing barley straw (which is analogous to rice straw in the context of anaerobic digestion [24]) inoculated with pig wastewater and cow manure at both 25°C and 35°C [61]. It should be noted that the 25°C reactors contained nearly double the amount of pig waste (dry weight) than the 35°C reactors and the experiment simulated a dry digestion process. Methane yields increased by 35% (145 to 222 L/kgVS), 18% (171 to 208 L/kgVS), 17% (156 to 188 L/kgVS) and 4% (151 to 158 L/kgVS) with increasing temperature [61]. The difference in these four sets of batch reactors was the amount of cattle manure used. The sets of reactors with higher concentrations of cattle manure resulted in more significant changes with increasing temperature. The average percentage of methane in the biogas also increased from 44% to 53% with the higher temperature [61].

In addition to increased gas production, mesophilic temperatures also promote faster digestion and thus speedier energy recovery and reuse of space. Additional time (ranging from 18 to 89 days) was required to produce the same amount of methane in the barley straw batch reactors at 25°C compared with the reactors that operated at 35°C [61]. In the bottle tests with the rice straw, the methane peaked 3 to 6 days earlier in the 35°C bottles compared to the 25°C bottles and the duration of the experiment was 59 days [28].

Mesophilic temperatures (35 to 40°C) are often preferred over thermophilic temperatures (50 to 55°C) in the anaerobic digestion process because the microorganisms are more robust and less sensitive to changes in their environment [62]. The anaerobic digestion of rice straw inoculated with hogger wastewater was found to be more stable under mesophilic conditions (35°C) rather than thermophilic conditions (55°C) [28]. Animal wastes, which are often co-digested with rice straw or used as an inoculum, have high ammonia concentrations and the anaerobic digestion of these wastes is susceptible to ammonia inhibition [63]. At higher temperatures, pH increases and the ammonia nitrogen becomes unionized and is generally more toxic to the methanogens [58, 64]. Rice straw digestion has been evaluated at thermophilic temperatures, even though it is highly impractical on a full-scale system due to the large amount

of heat input required. Results from bottle tests showed that overall methane yield increased from 120.2 L/kgVS at 35°C to 136.3 L/kgVS at 55°C under wet digestion conditions. However, with semi-dry digestion conditions, the methane yield decreased from 123.5 L/kgVS at 35°C to 76.3 L/kgVS at 55°C [28]. The authors attributed this decrease to acid accumulation in the system, which inhibited the overall gas production [28].

2.7 The Effects of Total Solids Concentration

Anaerobic digestion can take place under wet conditions (i.e. TS concentration less than 15%) or dry conditions (i.e. TS concentrations between 20 to 40%) [65]. Advantages of wet systems include better homogeneity and simpler mechanics (i.e. pumps and piping), but dry systems tend to be more flexible and produce slightly higher biogas yields [65]. Under natural conditions, the TS of rice straw is 30 to 40% immediately following harvest, but once it has been baled it typically contains a TS concentration greater than 75% [3]. In the simplest conditions with rice straw mixed with water, buffering agents and nutrients, the ideal TS concentration was determined to be 10% for optimum methane production [20]. When conditions were dry (i.e. TS = 25%), the methane production decreased by 97% [20].

A few studies have attempted to define the optimum TS concentrations using rice straw inoculated with piggery wastewater [28-30]. Rice straw was co-digested with pig manure (straw to manure ratio of 2:1) and inoculated with a pig manure/sewage sludge mixture (20% by volume) [29]. TS concentrations ranging from 8 to 35% were compared to determine what conditions produced the highest methane yield in terms of volatile solids added. Though biogas volume nearly quadrupled with increasing TS concentrations (8% to 30%), the overall methane yields were essentially the same since the percentage of methane in the biogas decreased with increasing TS concentrations [29]. Longer digestion periods were required as the TS concentration increased in order to achieve the same overall methane yield. For example, an additional 43 days was required when the TS was 30% versus 15% [29]. With a TS concentration of 35%, biogas production ceased after 15 days [29] and the low pH indicated that acidification had occurred.

In bottle tests comparing dry digestion system (TS = 20%) with a wet digestion system (TS = 7.5%), the cumulative methane production over 59 days was essentially the same [28]. It took 3 to 15 days longer to produce methane under dry conditions, however the drier conditions

were more stable and benefits included water savings and higher solids content for disposal [28]. The authors observed that acidification was more likely to occur in the dry digestion process; however, in a follow-up pilot scale study (TS = 19.35%), acidification was avoided through the daily recirculation of leachate [28]. In another bottle study, soiled rice husks from a pig farm were used as the substrate and wet digestion (TS = 8%) was compared with semi-dry digestion (TS = 14%); however, no significant difference was observed in the digestability of the rice husks or the theoretical methane yield under wet and semi-dry conditions [30].

From these studies, it can be concluded that digestion of rice straw under wetter conditions occurs faster, but the overall methane yield is essentially the same in both wet and dry systems. Acidification is more likely to occur in dry digestion systems; however, as these findings are applied to large-scale systems, leachate recirculation can be employed to prevent acidification. Recirculation of leachate also increases the overall degradation processes in high solids reactors by dispersing the nutrients and bacterial populations [66].

2.8 Various Pretreatment Alternatives to Enhance Gas Production

Rice straw is a lignocellulosic biomass that contains a relatively high lignin content (i.e. 10 to 15% dry weight)[1, 3, 25, 67], which makes it very difficult to degrade because the ligno-carbohydrate complexes form strong bonds and the plant cell wall is resistant to microbial attack [3, 23, 25, 49]. Lignin is a highly complex polymer that holds together the polysaccharide fibres and contributes to the structural rigidity of the straw [40]. The structure of rice straw is shown in Figure 2-3. Therefore, a pretreatment step is often used to increase the degradability of the rice straw and accelerate the digestion process. Several approaches for pretreatment of rice straw have been investigated including physical (i.e. size reduction), biological (i.e. fungi), and chemical (i.e. acid and alkali additions).

Mechanical size reduction of the rice straw ruptures the cell walls and makes the organic matter more readily available for the microorganisms to decompose [31]. Milling, or cutting, the biomass exposes more surface area and reduces the polymerization, which leads to an increased hydrolysis of the lignocellulosic biomass [68]. The milling of lignocellulosic materials has resulted in increased methane yields (5 to 25%) and reduced digestion times (23 to 59%) [68]. With all other variables held constant, the methane yield was slightly higher for rice straw that had been cut in 25-mm lengths (198 L/kg VS) versus rice straw that was digested whole (190

L/kg VS) [31]. In a study with wheat straw, the increase observed in the methane yield from 30-mm lengths (145 L/kg VS) compared to 1-mm lengths (161 L/kg VS) was statistically significant after 60 days of digestion [44]. However, the milling process requires high energy inputs and is not economically feasible [68].

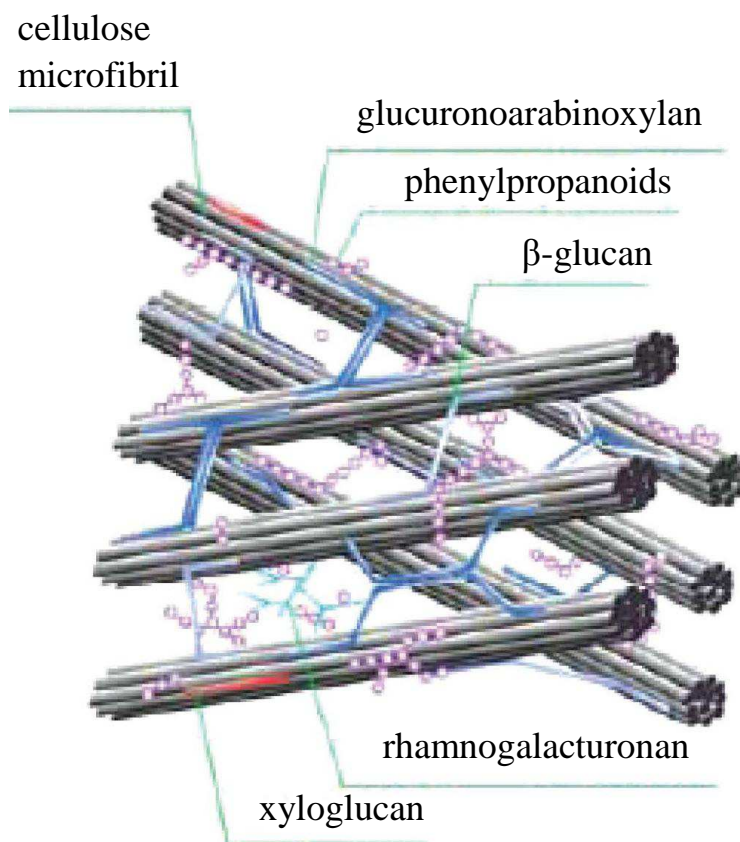


Figure 2-3. The Structure of Rice Straw [40]

Increased biodegradability of rice straw has been demonstrated with aerobic and anaerobic fungi [23, 25]. Rice straw was pretreated with white rot fungus (*Phanerochaete chrysosporium*) and brown rot fungus (*Polyporus ostreiformis*), which are classified as aerobic, mesophilic microorganisms [40], using a straw-to-fungi ratio of 14,285:1 and then digested in batch reactors at 30°C [25]. After a three-week incubation period, the white rot fungi destroyed 47.5% and the brown rot 20% of the initial lignin content, which equated to increased methane productions of 46% and 31%, respectively, when compared to the untreated control [25]. In a

study that utilized sheep rumen as an inoculum, the anaerobic fungi from the rumen consistently increased the digestability of rice straw when compared with fermentors where the fungi were inhibited [23].

Increased biodegradability has also been demonstrated with chemical pretreatment methods for rice straw including both acid and alkali additions [2, 23, 38, 43]. Acid pretreatment is desirable for rice straw digestion not only because it breaks down the lignin component of the straw, but also because the methanogens can acclimate to acidic conditions that will be encountered during the hydrolysis phase of the anaerobic digestion process. However, it requires a substantial energy input (i.e. temperatures ranging from 100 to 200°C depending on the acid concentration) and care must be taken to avoid the development of inhibitors such as carboxylic acids, furans and phenolic compounds that prevent the growth of the methanogenic bacteria [69]. A dilute organic acid pretreatment method (i.e. equal mixture of acetic and propionic acids) demonstrated that the lignin content of the rice straw could be reduced by up to 34% and methane production could be increased by 36% in pretreated versus untreated rice straw [2]. Pretreatment with a sodium chlorite/acetic acid mixture reduced the lignin content by 60%, which increased the biodegradability by 20% when compared with the untreated rice straw [23]. The corresponding increase in methane production was not reported in this study, however, volatile fatty acid production also increased by approximately 20% [23].

Alkaline pretreatment not only breaks down the lignocellulosic material and some nitrogenous materials [43] making them more available for the anaerobes, but it can also remain in the substrate and serve as a necessary buffer for the anaerobic digestion process [38]. Alkali pretreatment of rice straw with 6% sodium hydroxide (dry weight) resulted in increased biogas yields from 27 to 65% (depending on loading rate of straw) when compared with untreated straw [38]. Comparable biogas yields resulted when pretreating with 6% sodium hydroxide (0.45 L/g TS) and 5% sodium hydroxide (0.38 L/g TS) [38, 43]. Pretreatment with alkaline hydrogen peroxide as well as with 3% ammonia both increased the digestability of the rice straw when compared with untreated rice straw; however, these methods were not as effective as pretreatment with the sodium chlorite/acetic acid mixture [23].

2.9 Microbiological Considerations

Due to the complex and rigid structure of rice straw, diverse catalytic activities of several enzymes is necessary for the efficient break down of this material. Synergistic effects have been shown between cellulolytic and noncellulolytic bacteria when they are cultured together for the degradation of cellulose [40, 70]. Microorganisms obtained from four different compost piles consisting of mixtures of rice straw, chicken feces, pig feces, cattle feces and sugar cane dregs were cultured for the specific purpose of digesting rice straw [70]. The resulting microbial community was capable of degrading more than 60% of the rice straw within four days [70]. Environmental genomics, which is the extraction and sequencing of DNA from an environmental sample [40], was used to identify the specific organisms involved in the degradation process. The bacteria that was attached to the straw and most likely responsible for the first step in the degradation process was identified by polymerase chain reaction (PCR) as *Clostridium thermosuccinogenes*, which is a strict anaerobe that has been found in manure, beet pulp, soil and mud [70]. Other bacterium identified included a mixture of aerobes and facultative anaerobes as well as both cellulolytic and noncellulolytic bacteria. The stability of this microbial community, which was evidenced by a tolerance to extreme heating and cooling and an ability to degrade both sterilized and non-sterilized substrates, was attributed to its diversity and complexity [70]. A similar synergist effect was observed during the degradation of rice straw by *Clostridium cellulovorans* involving both cellulosome and non-cellulosome enzymes working under mesophilic conditions [40]. Most of the compounds in the rice straw were degraded within 10 days without any type of pretreatment [40].

2.10 The Move Toward Full-Scale Biogas Plants

Despite the existing literature on this topic, there are currently no documented full-scale biogas plants using rice straw as the primary substrate or co-digestion of rice straw with animal waste. The main deterrents of constructing such a system include the lignocellulosic structure of rice straw that makes it difficult to degrade and the costs associated with start-up and maintenance of a full-scale anaerobic digester. Despite the benefits of using renewable energy and reducing emissions that contribute to climate change, economics are the deciding factor in whether or not this technology will be implemented [32]. In order for an anaerobic digester to be

effective, it needs to function continually and produce enough energy to offset the overall costs of the system (i.e. it must be a net energy producer rather than a net energy consumer) [4].

Studies that define the economics and logistics involved in implementing a full-scale biogas plant utilizing rice straw are lacking. The cost of external heating in order to achieve mesophilic temperatures on a large scale and the potential for using the thermal heat generated in the biogas conversion process are concepts that still need to be defined more clearly. In a study that evaluated barley straw digestion with manure, a simple heat energy balance was conducted to determine if higher temperatures were efficient and economical [61]. The authors found that the heat energy that could be obtained from the increased methane production at 35°C compared to 25°C did not off-set the external heat energy required to maintain the higher temperature for 110 days [61].

Optimum capacities for full-scale digesters, loading strategies and leachate recirculation volumes and frequencies also need to be investigated. Adapting existing biogas plants designed for energy crops such as maize is not feasible due to sizing requirements, slower digestion rates associated with lignocellulosic materials, and the necessity of dry versus wet digestion processes. Sizing of the digester would be dependent upon factors such as available space, the capacity of the conversion equipment, and economic feasibility. A theoretical scenario for the co-digestion of rice straw with animal waste would be a centrally located farm-scale digester equipped with a 100-KW engine. Assuming 35% electrical conversion efficiency and a specific biogas production rate of 2.5 Nm³/ton straw/day, this size engine could accommodate a batch reactor with approximately 600 tons of rice straw codigested with 300 tons of animal manure in 160 days. Therefore two batch reactors could be digested per year. If the straw were stacked in cylindrical bales, the spacing requirement would be approximately 50 m long by 35 m wide by 3 m high. Assuming five tons of rice straw are produced per hectare, which ultimately depends on the rice variety, this size digester and engine could accommodate a 240-hectare rice field.

The majority of pretreatment strategies summarized herein are not necessarily practical when dealing with 1000 Mg of rice straw rather than 10 g (i.e. 1x10⁻⁵ Mg), and therefore economically feasible modifications for pretreatment in full-scale systems should be evaluated with specific emphasis on waste disposal. Dry anaerobic digestion systems for rice straw summarized in the literature rarely exceed 35% TS [20, 28, 29]; however, practical implementation of a full-scale rice straw biogas plant would likely need much higher TS

concentrations to avoid high waste disposal costs of the selected inoculum (i.e. pig or cattle wastewater). Thus, there are several aspects of this topic that need further investigation before full-scale biogas plants utilizing rice straw can become a commercial success.

2.11 Conclusion

As rice production continues to rise in order to provide a stable food source for over half of the world's population, rice straw will continue to be an abundant and accessible agricultural waste. The collection and treatment of rice straw through anaerobic digestion is not only a viable option for producing clean, renewable energy, but it will also eliminate a major source of greenhouse gas emissions from common practices of open burning or tilling the straw back into the fields. The specific methane potential of rice straw ranges from 92 to 404 L/kg VS added, depending on the digestion parameters and pretreatment methods. Rice straw has very low concentrations of nitrogen (<1%) so using inoculums such as pig or cattle manure provide an appropriate nutrient balance for the system and serve as a buffering capacity to prevent acidification in the reactor. Proven, mesophilic temperatures of 35 to 40°C are preferred over ambient (20 to 30°C) or thermophilic (50 to 55°C) conditions for higher methane yields, shorter retention times and more stable systems. Total solids concentrations, as long as they are below 35%, are not a critical factor in the anaerobic digestion of rice straw.

One of the major challenges with the digestion of rice straw is the lignocellulosic structure that makes bacterial decomposition difficult. This challenge can be overcome by pretreatment strategies such as the addition of fungi, acid mixtures, and alkaline solutions that have been successful at increasing biogas potential and accelerating the digestion process of rice straw. A diverse conglomeration of both cellulosome and non-cellulosome enzymes acquired from various compost piles of rice straw, chicken feces, pig feces, cattle feces and sugar cane dregs formed a stable microbial community and were very effective at digesting rice straw under anaerobic conditions (i.e. 60% decomposition in four days). Research on economic inputs, operational parameters, feasible pretreatment methods and the overall logistics of implementing a full-scale biogas plant for rice straw still needs to be conducted before this concept can become the preferred treatment option.

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CHAPTER 3

Design Considerations for a Farm-Scale Biogas Plant Based on Pilot-Scale Anaerobic Digesters Loaded with Rice Straw and Piggery Wastewater

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3.1 Introduction

The anaerobic digestion of rice straw has been studied for nearly a century [1], but the implementation of full-scale biogas plants using rice straw as the primary substrate had not been reported prior to this dissertation research. Rice is the most important staple food for over half the world's population [2], and rice straw is one of the most abundant and renewable energy sources in the world [3]. Common solutions for dealing with rice straw are open-field burning or tilling the straw back into the field, both contributing to increased greenhouse gas emissions [4, 5]. CH₄ emissions from anoxic soils amended with rice straw are much higher than those without straw [5-7], and one mitigation strategy is to collect the biomass and convert it into a clean-burning fuel through anaerobic digestion. The problem is a lack of realistic operational parameters on how to efficiently convert rice straw into energy as a sustainable practice (i.e. net energy producer versus a net energy consumer).

One of the major challenges associated with using rice straw as a substrate in the anaerobic digestion process is the complex, lignocellulosic structure which makes it difficult to decompose [8]. Several biological and chemical pretreatment strategies have proven successful in lab-scale experiments to break down the lignin and accelerate decomposition [3, 9-12]. However, most of these approaches may be inappropriate for a farm-scale application because of large chemical quantities, high energy inputs, excess water required and waste disposal issues associated with the residues, or digestate.

The co-digestion of manure and rice straw can help to overcome this challenge because it provides not only the necessary microorganisms but also the appropriate balance of nutrients to create favorable conditions for the methanogens to thrive. If there is a rapid accumulation of volatile fatty acids in the initial stages of the digestion process and the pH drops below 6.5 or 7.0, then the methanogens will be inhibited [13]. Pig wastewater, with its high ammonia content, can provide a buffering capacity to help maintain a favorable pH as well as trace elements such as iron (Fe), nickel (Ni), molybdenum (Mo) and cobalt (Co) that help to stimulate activity among the methanogens [14]. Biogas production increased approximately 35% and the methane yield increased from approximately 270 to 340 L CH₄/kg volatile solids (VS) when inoculated rice straw was co-digested with pig manure (2:1 ratio dry basis) compared to the digestion of the inoculated rice straw alone [15].

Numerous bench-scale experiments have been published on rice straw digestion, which define optimal parameters such as temperature, nutrient balances, inocula ratios and pretreatment strategies [3, 9-11, 16-21]; however, only a few studies involving rice straw have been conducted in dry digestion conditions (i.e. total solids (TS) concentration $\geq 20\%$) [12, 15, 22, 23]. Data from a pilot-scale system ($\geq 1\text{m}^3$) is limited to one previous study [23]. Though principles can be better understood through bench-scale studies, it is very difficult to design and operate a farm-scale plant from these microcosms. A pilot-scale study designed with similar operational parameters and limitations as a farm-scale system such as leachate recirculation, limited heating capacity and limited mixing will provide comparable results that can be used in the design of a farm-scale plant.

The goal of this research is to define basic operational parameters that are practical for the design and implementation of a farm-scale anaerobic batch reactor using rice straw and pig wastewater. Two pilot-scale batch reactors (1m^3) filled with untreated rice straw and co-digested with pig wastewater were constructed and parameters such as TS concentration, straw to wastewater ratio, digestion temperature and digestion time were evaluated. The rice straw used in these experiments was not pretreated because it is unlikely that a rural, farm-scale system designed for more than 600 tons of straw could implement existing pretreatment strategies. Specific objectives of this study were to compare gas production in varying temperature conditions in order to evaluate heat input considerations for a farm-scale plant, minimize manure to straw ratios and simulate dry digestion to minimize management and disposal issues, and to avoid pretreatment or additional inocula in an attempt to simplify the loading strategy for a farm-scale plant. A scenario analysis for the design of a farm-scale plant using both untreated and pretreated rice straw produced on a 100-hectare rice farm is also included.

3.2 Material and Methods

3.2.1 Experimental Set-up

The pilot-scale digesters (Figure 3-1) are cylindrical tanks constructed of stainless steel with an internal cavity for the substrate and a surrounding compartment that serves as a water jacket and hydraulic seal. The substrate capacity for each digester is approximately 1m^3 with the following dimensions: the inner diameter is 0.945 m; the outer diameter, encompassing the water jacket, is 1.025 m; and the height is 1.500 m. The digesters are equipped with an internal

thermometer, gas flow meter, leachate recirculation pump, and a leachate holding tank. The leachate tank is constructed of HDPE and has a working volume of 1 m³.

The digesters were designed so that leachate could be recirculated on an as-needed basis with a gravity drain and 0.85 kW recirculation pump. A layer of gravel (0.15 m thick) was added to the base of the digesters to promote flow and prevent clogging during leachate recirculation. A vent pipeline connects the digester and the leachate tank in order to equalize the pressure during recirculation.

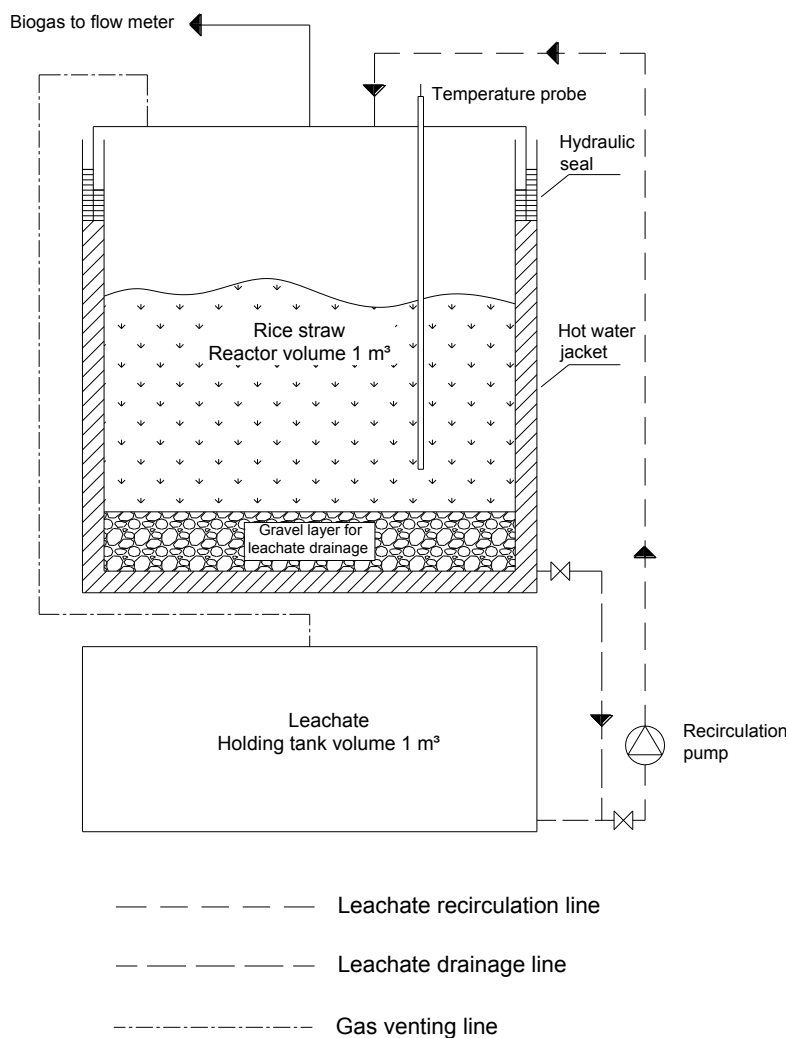


Figure 3-1. Schematic of the Pilot-Scale System

3.2.2 Digester Composition and Operating Conditions

Rice straw was co-digested with piggery wastewater in each of the pilot-scale digesters with no additional nutrients or inocula for a total of 189 days. The rice straw was harvested from

a rice field in the Pavia Province of Italy approximately six months prior and stored in a dry location. No pretreatment, drying, cutting or milling activities were applied to the rice straw. It was introduced into the digesters directly from the bale in lengths ranging from 0.2 to 0.6 m. Raw piggery wastewater was collected from a preliminary holding tank at a local pig farm in the Pavia Province of Italy. Prior to collection, the wastewater was not mechanically aerated but it was in an open tank exposed to the atmosphere. The wastewater was immediately transferred to the digesters and they were closed and sealed under anaerobic conditions. The digesters were operated in a covered area protected from the elements; however, the building was only enclosed on three sides so the digesters were exposed to the atmosphere so they could be influenced by the daily change in ambient temperature.

With the exception of temperature, the digestion conditions and sampling strategies were the same for both digesters. Diluted pig wastewater (i.e. leachate) was recirculated once every three to four days and a sample was collected during each cycle of recirculation. The biogas production volume, gas quality (i.e. methane concentration), and temperature data was recorded at least twice a week. Digester A was designed to achieve a maximum output in terms of methane production. A total of 50kg of dry straw was mixed with 150L of piggery wastewater. Additional water was added to reach a TS concentration of 20%. The water jacket surrounding the digester was heated constantly in order to keep the digestion temperature inside the digester in the mesophilic range between 30 and 40°C.

In Digester B, the heat and wastewater inputs were reduced in an attempt to minimize the energy inputs and determine minimal design parameters for a farm-scale digester. From a farm-scale perspective, adding excess wastewater can be expensive due to transportation costs and too much residual leachate could result in waste disposal issues. Therefore, to assess a minimal wastewater input, a total of 50 kg of dry straw was mixed with 60 L of piggery wastewater. Water was also added to achieve a 20% TS concentration of the mixture. The digester was initiated with ambient conditions in mid-May in the Pavia Province of Northern Italy. Periodical cycles of heating were conducted from mid-August through mid-October and constant heating conditions to maintain mesophilic temperatures were applied through mid-December. The periodic heating cycles simulated a likely scenario in which the recirculated leachate in a farm-scale system could be heated through a simple heat exchange process and returned to the digester. The composition of both digesters is shown in Figure 3-2.

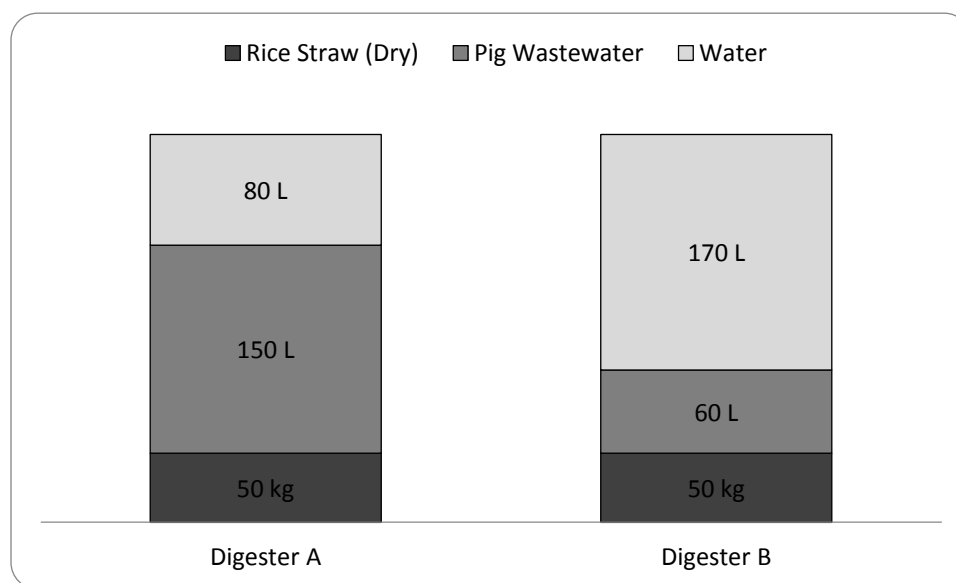


Figure 3-2. Initial Composition of Pilot-Scale Digesters

3.2.3 Analytical Methods

At the beginning of the experiment, samples of rice straw and raw pig wastewater were collected and analyzed for the following parameters: TS, VS, Total Kjeldahl Nitrogen (TKN), Total Phosphorus (TP), Chemical Oxygen Demand (COD), Zinc (Zn), Copper (Cu), and Lead (Pb). All analyses were conducted in triplicate. The solids concentrations were measured according to APHA Standard Method 2540, TKN according to APHA Standard Method 4500-N (C), TP according to 4500-P (B/C), COD according to Standard Method 5220 B, and the metals were analyzed according to APHA Standard Methods 3111B (Zn) and 3111 (Cu, Pb) [24]. The results of the solids, nutrients and metals concentrations of the rice straw and pig wastewater used in the experiments are summarized in Table 3-1. A biochemical methane potential (BMP) assay was also conducted for the pig wastewater in mesophilic conditions to determine if the fraction of methane production coming from the wastewater was significant [25].

During the experiment, leachate samples were collected during recirculation and analyzed for pH, volatile fatty acids (VFA), carbonate alkalinity (C_T), and ammonia nitrogen (NH_3-N). The leachate analyses were completed within two hours of the collection time. The pH was analyzed with a Hamilton Filltrode probe and was conducted in accordance with the APHA Standard Method 423 [26]. VFA and C_T were analyzed by a titration method using 0.1 M hydrochloric acid (HCl) and acidifying the sample to pH of 2.2, while continually recording pH

and using a computer modulation to calculate the results [27]. The total ammonia nitrogen (TAN) concentration (NH_3 and NH_4^+) was analyzed using a spectrophotometer (SPT-500) with a Carlo Erba reagent kit (0800.05405). A sample dilution ratio of approximately 1 to 250 was used throughout the experiment to respect the sensitivity limits of the instrument. Free ammonia (NH_3) was calculated from TAN using an equation from Anthonisen et al. that incorporates pH and temperature [28].

Ambient temperatures were recorded using a Comark N9011 thermometer and permanent temperature probes read with Gefran instruments were situated inside the digesters. The temperature data was collected approximately twice a week for Digester A and more frequently (i.e. five times a week) for Digester B since it was influenced by ambient conditions. Gas quantities were measured with volumetric flow meters and the gas composition (i.e. methane, carbon dioxide and oxygen) was measured with a Geotech biogas analyzer.

Table 3-1. Characteristics of Rice Straw and Pig Wastewater

Analyte	Rice Straw ^a	Pig Wastewater ^a
TS	92.9 %	4.9 %
VS	85.9 % TS	62.4 % TS
TKN	5.0 g/kgTS	4.3 g/L
TP	0.4 g/kgTS	1.7 g/L
COD	1002.2 g/kgTS	60.9 g/L
Zinc	38.2 mg/kgTS	34.1 mg/L
Copper	<1.0 mg/kgTS	14.2 mg/L
Lead	9.6 mg/kgTS	0.6 mg/L

^a The results represent the average of the triplicates

3.3 Results and Discussion

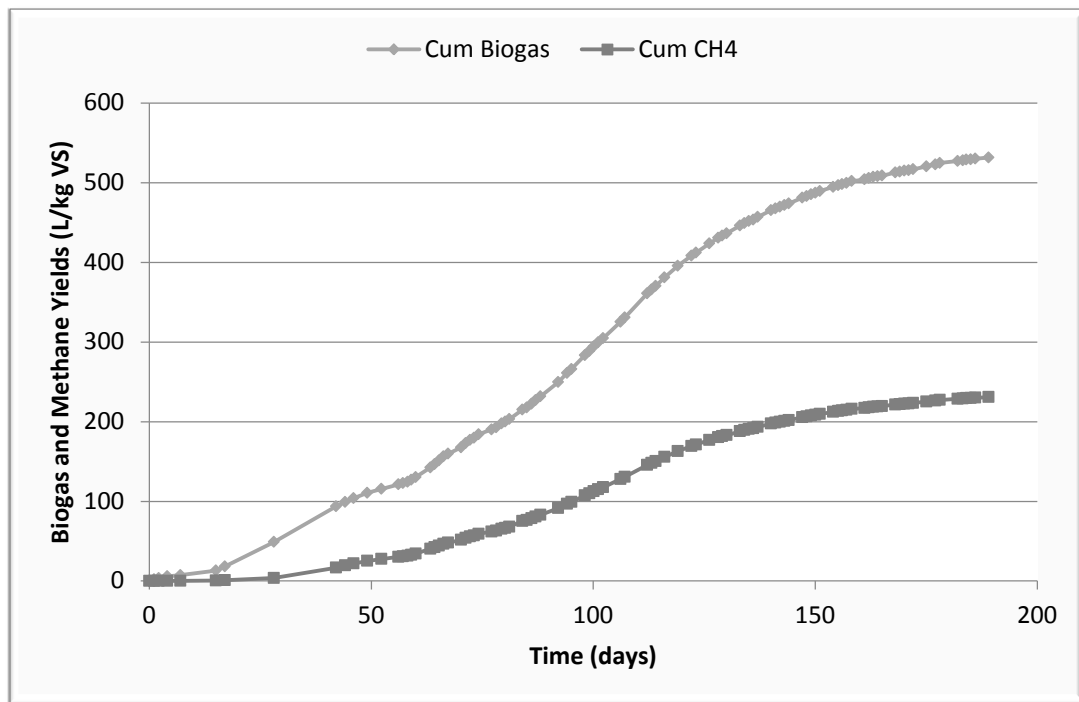
3.3.1 Gas Production

In Digester A, the total quantity of biogas produced was 22,859 L, and the total methane produced during the digestion period was 9,929 L. The methane production rate peaked at 128 L/day on day 112, and the average percentage of methane in the biogas was 49%. The specific biogas and methane yields for Digester A in terms of straw VS added [and straw COD added] were 532 L biogas/kgVS [456 L biogas/kg COD] and 231 L CH_4 /kgVS [198 L CH_4 /kg COD]. Based on the theoretical methane yield of 350 L per kg COD of substrate under anaerobic

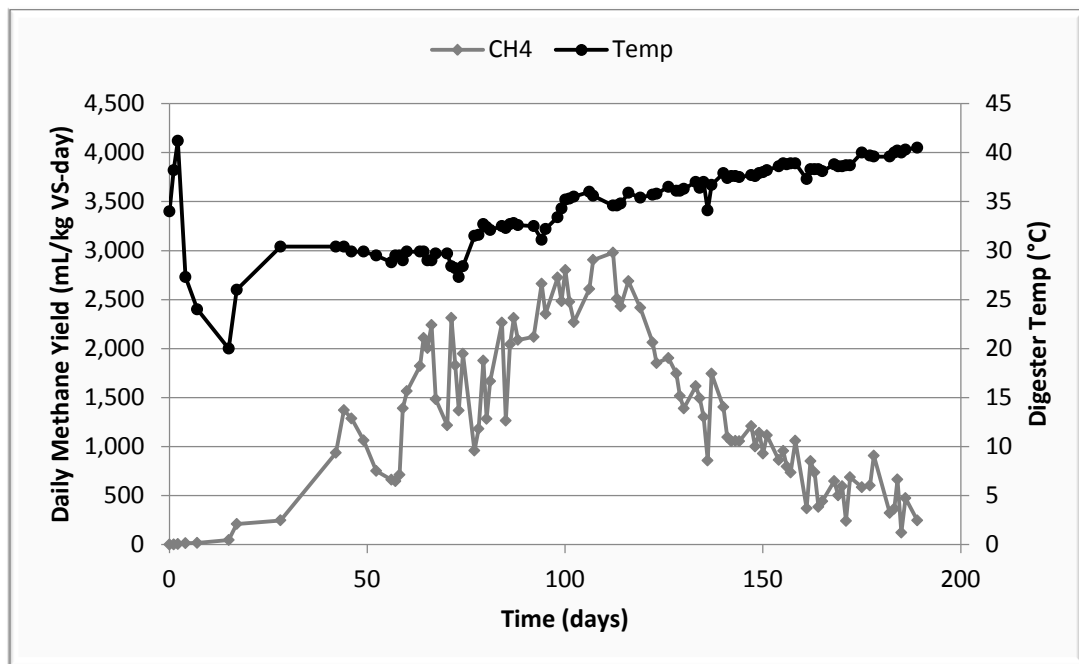
conditions, approximately 57 % of the biomass was converted. The methane produced from the pig wastewater was determined to be negligible (i.e. less than 5%) based on preliminary biochemical methane potential (BMP) assays containing only pig wastewater that resulted in 3.30 L CH₄/L pig wastewater. The biogas yield, methane yield and temperature are shown as a function of time in Figures 3-3(a) and 3-3(b) for Digester A.

The quantity and quality of gas production in Digester B were significantly lower than in Digester A. In Digester B, the total quantity of biogas produced was 1,420 L, and the total methane produced during the digestion period was 533 L (only 5% of the methane produced in Digester A). The methane production never reached a clear peak, and the average percentage of methane in the biogas was 37%. The specific biogas and methane yields for Digester B in terms of straw VS added [and straw COD added] were 33 L/kgVS [28 L/kg COD] and 12 L/kgVS [11 L/kg COD], respectively. The biogas yield, methane yield and temperature are shown as a function of time in Figures 3-4(a) and 3-4(b) for Digester B.

The biogas and methane yields were calculated as the volume of biogas or methane produced per unit weight of rice TS or VS added. The results from both Digester A and B are shown in Table 3-2. Specific methane yields obtained from the anaerobic digestion of rice straw range from 92 to 404 L of CH₄ per kg VS added, depending on the type of pretreatment used, inocula added, and digestion temperatures [11, 15-17, 20, 21, 23]. Lianhua et al. (2010) published results for a pilot-scale system (1m³) using pretreated rice straw digested with hoggerly wastewater, and these results are also shown in Table 3-2 for comparison. The pretreatment method involved cutting the rice straw into 7 to 8-cm lengths and pretreating it with 5% white-rot fungi, as well as an additional 2.5% NaHCO₃ to optimize the non-lignin carbon to nitrogen ratio[23]. The results obtained by Lianhua et al. (2010) are very relevant to this study since they can be used to compare the performance of pretreated to untreated rice straw and describe the differences in design requirements necessary for a farm-scale system. To the best of the authors' knowledge, it is also the only other pilot scale system ($\geq 1\text{m}^3$) discussed in the literature for the anaerobic digestion of rice straw.

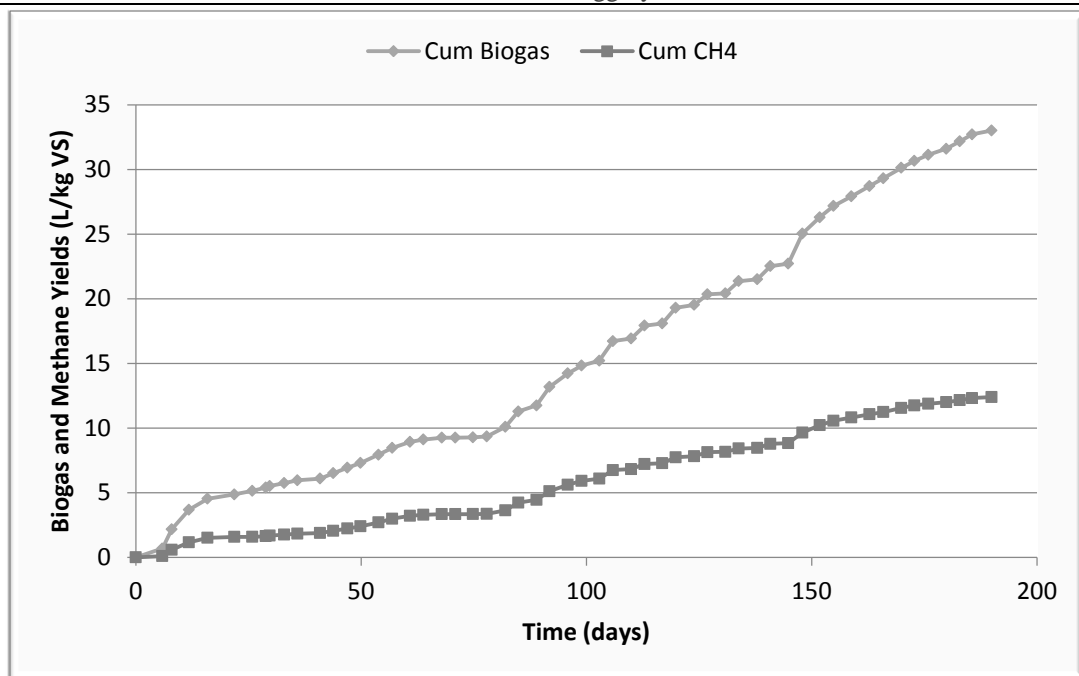


(a)

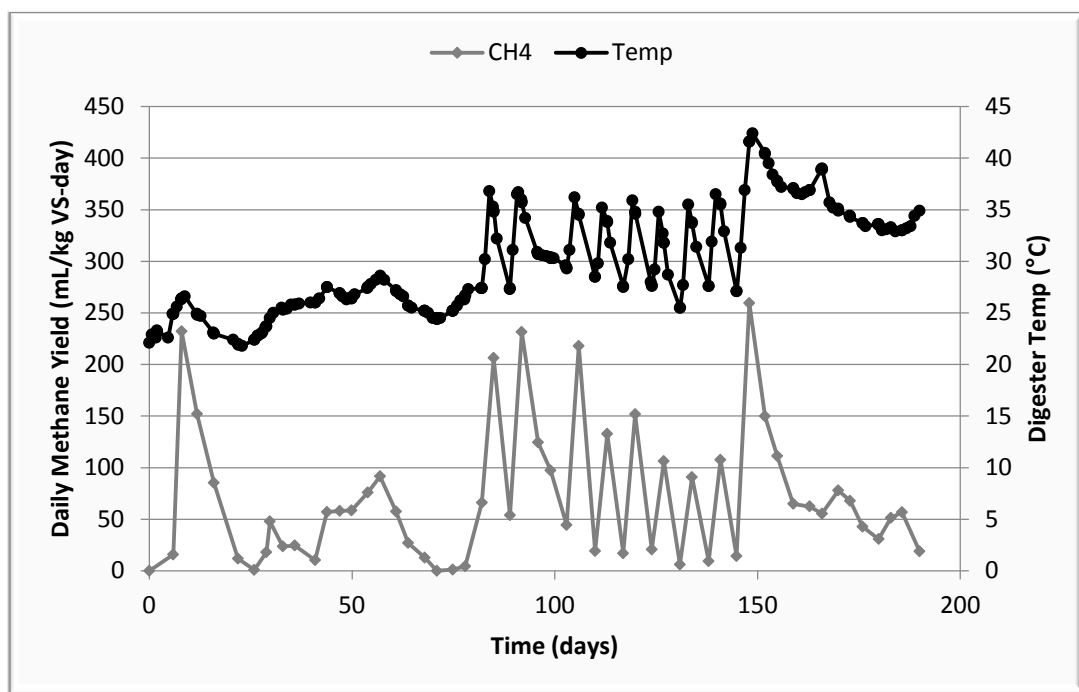


(b)

Figure 3-3. Gas Production in Digester A: (a) Cumulative biogas and methane yield versus time (b) Daily methane yield and digester temperature versus time



(a)



(b)

Figure 3-4. Gas Production in Digester B: (a) Cumulative biogas and methane yield versus time (b) Daily methane yield and digester temperature versus time

Table 3-2. Biogas and Methane yields for Pilot-Scale Digesters (1m³) using Rice Straw and Pig Wastewater

	Digester A	Digester B	Lianhua et al., 2010 [23]
Temperature Conditions	Mesophilic	Ambient/Mesophilic	Ambient
Digestion time [days]	189	189	89
Initial TS Concentration [%]	20.0	20.0	19.5
Straw[kgTS] to Wastewater[L] ratio	1 to 3.0	1 to 1.2	1 to 1.8
Pretreatment Method	None	None	White rot fungi
Total Biogas Production [L]	22,859	1,420	22,586
Total Methane Production [L]	9,929	533	9,501
Specific Biogas Yields:			
[L Biogas/kgTS]	457	28	457
[L Biogas/kgVS]	532	33	570
Specific Methane Yields:			
[L CH ₄ /kgTS]	199	11	192
[L CH ₄ /kgVS]	231	12	240
[L CH ₄ /kg COD)	198	11	Not reported

The total quantity of biogas produced in Digester A over the 189-day digestion period (22,859 L) is essentially the same quantity of biogas produced in 89 days using pretreated rice straw (22,586 L). More importantly, the specific methane yields are very close (within 5%), indicating that either scenario will result in the same degree of energy output of the rice straw. Since the output is essentially the same, a comparison of required inputs such as heat, volume of wastewater, chemical and biological additives, and digestion time can be made for practical design purposes. We can conclude that the physical (cutting) and biological (5% white rot fungi) pretreatment of rice straw digested under ambient conditions will produce nearly the same gas yields as untreated rice straw with 67% more wastewater, digested under mesophilic conditions for an additional 100 days.

3.3.3 The Effect of Temperature

Temperature plays a critical role in the anaerobic digestion process. The ideal temperature range for the anaerobic digestion of rice straw is between 35 and 40°C [1, 23]. The temperatures in Digester A were kept in the mesophilic range for most of the digestion period (the readings from the first month are not verified due to a mechanical problem with the temperature probe), and temperature did not appear to be a limiting factor in Digester A. The

daily gas production had a lag phase at the beginning through day 28, followed by a growth phase from day 28 to day 94, then a stationary phase from day 94 to 116, and eventually a decline phase from day 116 through day 189. Temperature steadily increased from 30 to 40°C over the course of the experiment, reaching approximately 35°C during the stationary phase when gas production was the highest. On days 135/136 the temperature dropped from 37.0 to 34.1°C and, consequently, daily methane yield dropped from 1302 to 857 ml CH₄/kgVS-day (see Figure 3-3b), indicating that sudden temperature changes may affect gas production. Since the temperature remained in the optimum range during this experiment, it was not a limiting factor in gas production.

The pilot-scale results from Lianhua et al. (2010) demonstrated that similar biogas and methane yields as seen in Digester A could be achieved at ambient temperatures if the rice straw was pretreated. Temperatures ranged from 19 to 30°C, with an average of approximately 24°C, but no obvious relationship between temperature and biogas production was observed [23]. Lianhua et al. concluded that the limiting factor in the pilot-scale system was the indigestibility of the organic matter [23].

In Digester B, the temperature did appear to be the most obvious limiting factor in gas production. As shown in Figure 3-4(b), there is a direct correlation between temperature and daily methane yield. Temperatures were variable throughout the digestion period, and the gas production increased and decreased with the rise and fall of digester temperature. Under ambient conditions, digester temperatures ranged from 21.8 to 28.6°C, and daily methane yields generally followed the temperature trend. On day 71, the ambient temperature dropped to 24.4°C and gas production ceased. External heat was added to the digester for approximately 17 hours on a weekly basis starting on day 82 and gas production followed. Digester temperatures cycled from 36°C immediately following the heat addition to approximately 27°C at the end of the week. Methane production always peaked on the same day that the temperature spiked, then decreased as the digester cooled to ambient conditions. Due to increasingly low gas production, continuous heat was added to the system beginning on day 145 through the duration of the experiment. The highest daily methane yield (259 ml CH₄/kgVS-day) corresponded with the highest digestion temperature (42.4°C), and this peak occurred on day 148. Under stable mesophilic conditions, a steady rate of gas production was observed; however, the rate of gas production was minimal compared to Digester A, even with digester temperatures of 35°C. Thus, we conclude that

temperature was not the only limiting factor in Digester B. The overall lack of methanogenic activity and gas production was due to the limited volume of pig wastewater in Digester B, which was not sufficient to establish a stable microbial community.

3.3.4 The Role of Nutrients

Along with temperature, the appropriate balance of nutrients is very important for the anaerobic digestion of rice straw. The advantage of co-digestion with animal manure is that optimum C:N ratios are established without adding chemicals and higher methane yields are the result [29-31]. When rice straw was co-digested with dairy manure, the most efficient methane production per unit COD destroyed occurred when the non-lignin carbon to nitrogen (as TKN) ratio was between 20 and 30 [18]. Untreated rice straw has a low content of nitrogen which results in relatively high C:N ratios, and typical values reported in the literature are shown in Table 3-3.

Table 3-3. Typical C:N Ratios Reported for Rice Straw

	TKN (%TS)	TOC (%TS)	C:N ^a
Present Study	0.50	30.6	61
Hills and Roberts, 1981 [18]	0.42	33.3	79
Lei et al., 2010 [20]	0.69	41.2	60
Ghosh and Bhattacharyya, 1999 [11]	0.51	40.3	79
Arvanitoyannis and Tserkezou, 2008 [2]	0.46	38.8	84
He et al., 2008 [12]	0.8	41.5	52
Zhang and Zhang, 1999 [21]	0.46	34.8	76

^a For clarification, the C:N ratios in the context of this paper refer to TOC and TKN, unless otherwise stated

Total organic carbon (TOC) was calculated based on the measured values of COD for both the rice straw and the pig wastewater. Reported ratios of COD to TOC for rice straw and piggery wastewater were obtained from the literature [18, 32]. Using the estimated TOC values for rice straw (306 g/kgTS) and pig wastewater (17.85 g/L), C:N ratios were calculated. The rice straw used in both digesters had a C:N ratio of 61, which is consistent with the references shown in Table 3-3. Thus, the nutrient balance in the digesters is dependent upon the addition of the pig wastewater, which has a relatively high content of nitrogen. The pig wastewater used in the digesters had a C:N ratio of 4.2, which is close to 4.8 reported by Zhang et al. [14]. The initial mixtures of rice straw and pig wastewater resulted in an overall C:N ratio of 20 in Digester A

and 32 in Digester B. This translates to an overall non-lignin C:N ratio of approximately 17 for Digester A and 28 for Digester B, calculated from the relationship between TOC and non-lignin carbon for both hog manure and rice straw reported by Hills and Roberts [18]. This indicates that the C:N ratios for both digesters is near the optimum range, and there should be no significant difference in gas production attributed to macronutrient imbalances.

Although the role of micronutrients was not evaluated as part of this experiment, certain trace elements are known to improve gas production and stabilize the anaerobic digestion process. Specifically, trace elements such as Fe, Ni, Co and Mo are contained in piggery wastewater and they can enhance the anaerobic digestion of different substrates [14]. Increased methane production was observed during the co-digestion of food waste with piggery wastewater when compared with the food waste alone [14]. As part of the experiment, the piggery wastewater was replaced by a trace element solution containing Fe^{3+} (100 mg/L), Ni^{2+} (10 mg/L), Co^{2+} (2 mg/L), and Mo^{2+} (5 mg/L) and methane production rates remained the same as with the piggery wastewater [14]. Zhang et al. (2011) concluded that the trace elements contained in the piggery wastewater were the key factors for improving the anaerobic digestion of the food waste [14]. Therefore, it is possible that the higher concentration of micronutrients supplied by a greater volume of piggery wastewater in Digester A contributed to better performance than Digester B, especially when both digesters were maintained at mesophilic conditions.

3.3.5 Production of Volatile Fatty Acids (VFA)

Leachate samples were collected throughout the experiment, and pH and VFA data was analyzed to monitor the anaerobic digestion process. VFA's are intermediary products formed during the fermentation of complex organic materials in the hydrolysis stage of anaerobic digestion. In normally operating anaerobic systems, VFA concentrations typically range from 50 to 250 mg/L as acetic acid (HAc) [33]. In Digester A, VFA concentrations increased to approximately 2200 mg HAc/L during high activity of the acidogens, and then decreased to less than 100 mg HAc/L once the VFA's were converted to methane and carbon dioxide by the methanogens. Due to problems with the instrumentation, however, insufficient data was collected to adequately understand and clearly depict the VFA trend for Digester A throughout

the entire digestion period. The presence and routine recirculation of the pig wastewater provided sufficient buffer for the system so that the pH never dropped below 7.

The VFA concentrations were monitored consistently in Digester B. An initial peak (3375 mg HAc/L) occurred at the beginning of the experiment, which likely represented the digestion of the organic matter from the pig wastewater. On day 57, the VFA concentration fell below 200 mg HAc/L and it ranged from 17 to 187 mg HAc/L through day 145. In conjunction with the decrease in VFAs, the pH increased to greater than 8.0 and remained fairly stable for the duration of the experiment. The VFA concentration slightly increased (< 200 mg HAc/L) after mesophilic conditions were established on day 145. Figure 3-5 shows the trend of VFA concentrations as they relate to pH in Digester B. With pretreated rice straw, a similar VFA trend was observed, except that the initial peak was much higher. The initial accumulation of VFAs (7254 mg HAc/L) did not cause acidification in the reactor (pH dropped to 7.1), and Lianhua et al. (2010) concluded that the periodic recirculation of leachate “enhanced” the buffering capacity of the system [23].

VFA accumulations are often the result of some type of microbial imbalance in the reactor caused by factors such as overloading, toxicity, or nutrient deficiency [33]. However, with lignocelluloses such as rice straw as the substrate, these intermediary products are often slow to form and the initial breakdown of the organic matter (i.e. hydrolysis) becomes the rate-limiting step in the digestion process [34, 35]. The VFA/pH trend observed in Digester B suggests that VFA formation from the untreated rice straw was the rate-limiting step, whereas the trend observed by Lianhua et al. (2010) with pretreated rice straw suggests that the methane conversion was the rate-limiting step.

The stability and lack of VFA accumulation in all three pilot-scale systems indicates that pig wastewater is an effective buffer for maintaining pH. When compared with cattle manure, pig manure reaches the methane plateau phase faster and the methane production (in terms of VS added) is twice as much when anaerobically digested under the same conditions [31]. El-Shinnawi et al. [17] co-digested untreated rice straw with enriched cow slurry for 120 days and the methane yield was slightly lower (215 L/kgVS added) than the results of Digester A (231 L/kgVS added). However, these experiments were conducted as wet digestion (TS of 8%) on a much smaller scale (2.5-L bottles) [17], which tends to increase homogeneity and produce more efficient yields that may not be applicable to farm-scale operations. Since pig manure is

produced daily, it could be added for the start-up of a farm-scale digester and also reapplied frequently to the straw to improve stability and increase methane production.

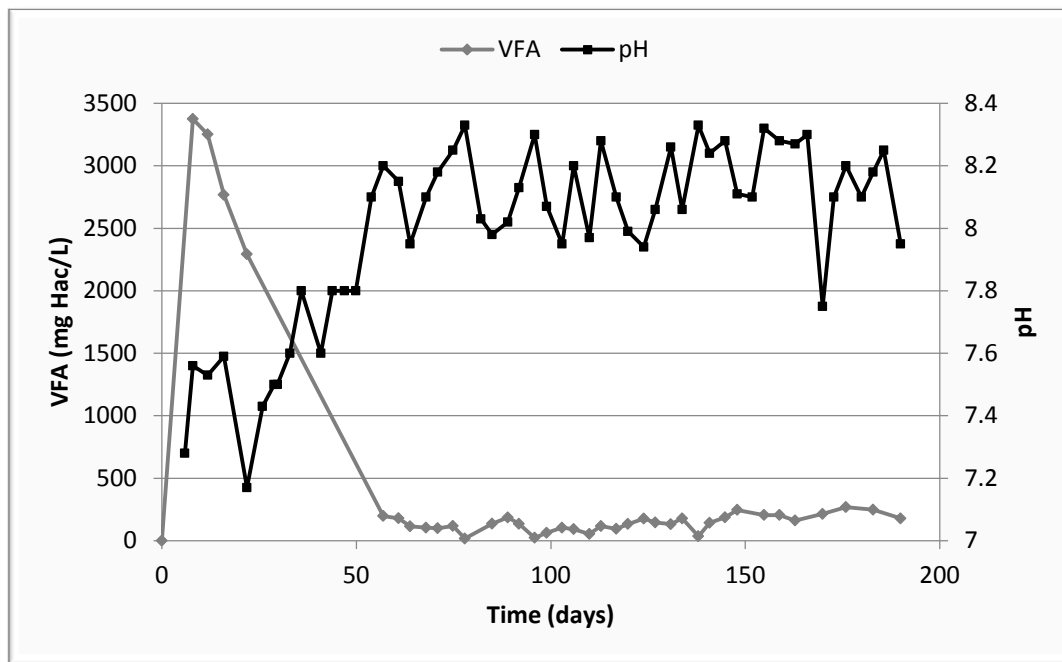


Figure 3-5. pH and VFA concentrations recorded throughout the digestion period in Digester B

3.3.6 Ammonia Inhibition

Ammonia in its toxic form (i.e. free ammonia) can be a source of inhibition for the anaerobic digestion process, and the concentration of NH_3 increases with pH and temperature [13]. The pH in Digester A remained within the ideal range (between 7 and 8) during the digestion period and there was no evidence of ammonia inhibition. In Digester B, however, the hydrolysis process occurred much slower and pH increased to > 8 on Day 54 and remained relatively constant around 8.1 for the duration of the experiment (Figure 3-6). This rise in pH coupled with increased temperatures resulted in increased NH_3 in the system (Figure 3-6) that may have inhibited the microorganisms. A toxicity threshold of 100 mgN/L as NH_3 has been reported [36], and 50% inhibition of methane production was observed with an NH_3 concentration of 215 mgN/L [37]. Since NH_3 concentrations remained above 100 mgN/L for approximately 20 days after mesophilic conditions were established and held constant in Digester B (i.e. day 145), it is possible that some ammonia inhibition occurred.

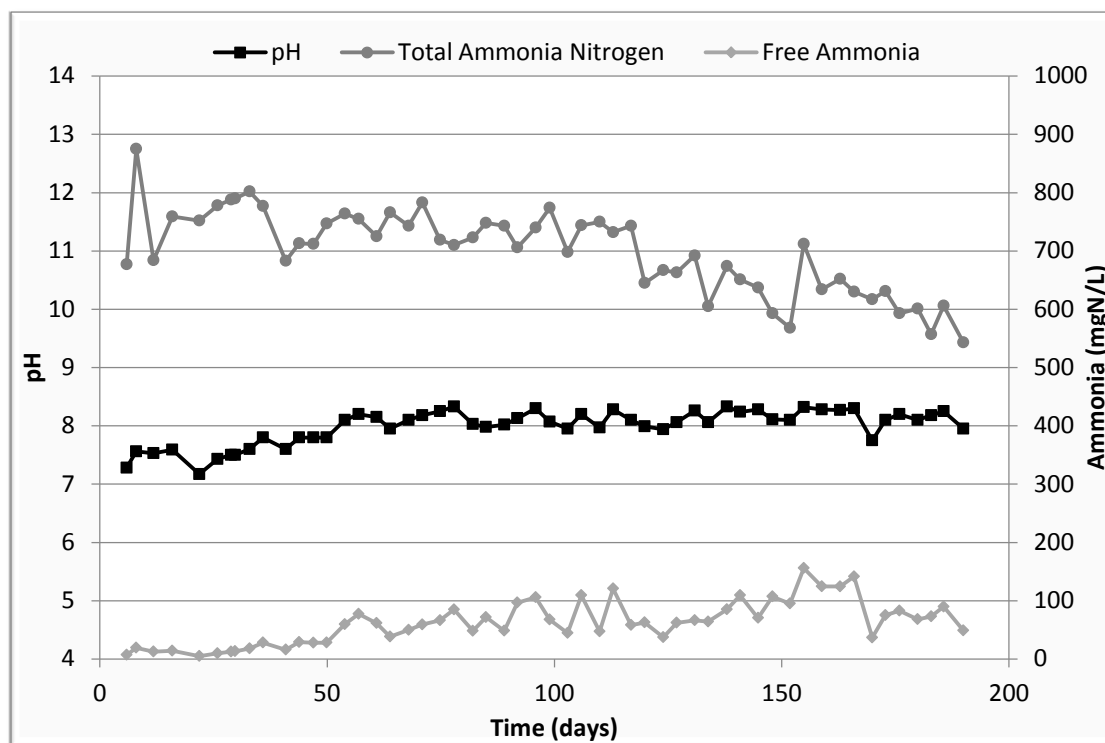


Figure 3-6. TAN, NH₃, and pH recorded throughout the digestion period in Digester B

3.7 Design Scenarios for a Farm-Scale Biogas Plant

The results of these pilot scale experiments can help to establish optimal design parameters for a farm-scale biogas plant using rice straw as a substrate and co-digested with pig wastewater. The first consideration in the design of a farm-scale plant is the space and time requirements. In order for the system to be sustainable, a farmer would need to be able to completely digest the entire waste stock in one year's time so that the waste would be eliminated before the next annual harvest. The sizing of the digester, therefore, would depend on how quickly the digestion occurred. If, for example, a farmer owned a 100-hectare rice field, then he would generate approximately 600 tons of rice straw per year (assuming 6 tons of rice straw are produced per hectare) [8]. If the digestion process required a full year, then the design capacity of the digester would be 600 tons; however, if the digestion time was only 180 days, then the digester capacity could be 300 tons and if only 90 days, then the digester could be designed for only 150 tons of rice straw. Two different scenarios can be correlated with the results discussed herein to define basic operational strategies for two different farm-scale plants that would theoretically produce the same quantity of methane. These two scenarios basically represent untreated and pretreated rice straw showing the advantages and disadvantages of each system

design. Based on the methane yields observed in the pilot-scale studies, either system could produce approximately 100,000 m³CH₄ per year, yielding approximately 328 MWh. This calculation is based on a total of 600 tons of straw (TS of 85%) and a 33% electricity conversion efficiency of the biogas engine.

In the first scenario, the basic considerations for a farm-scale plant are modeled after the pilot-scale study published by Lianhua et al. (2010). Since the digestion time for the pretreated rice straw only took 89 days, the farmer referenced above could digest 150 tons of rice straw 4 times a year. Assuming a maximum specific methane flow rate of 2.40 Nm³CH₄/ton/day (which corresponds to 10.5 Nm³CH₄/hr assuming a 70% straw conversion during the anaerobic digestion process), a gas engine with a minimum power of 34 kW would be necessary for the electrical conversion. Mechanical energy in the form of shredding would have to be added in order to reduce the initial size of the straw to less than 10-cm lengths. The rice straw would be pretreated aerobically using white-rot fungi, and therefore a two-stage digestion system would have to be designed allowing for both aerobic and anaerobic conditions to be achieved. Initially, 150 tons of shredded rice straw would be added to the digester, along with 7.5 tons of white rot fungi and 3.75 tons of NaHCO₃. An additional 450 tons of water would also have to be added to achieve the reported TS concentration (i.e. 26.7% for the pretreated straw [23]). The aerobic pretreatment step would need to accommodate temperatures around 60°C for an unspecified time frame [23]. Once pretreatment was complete, approximately 270 tons of pig wastewater would be added for the digestion and anaerobic conditions would have to be established. The design capacity of the digester should be approximately 1800 m³, allowing for 15% void space. Ambient temperature conditions above 19°C would be acceptable for this scenario, which exists in northern Italy approximately 145 days per year (climatic conditions for the Pavia region of Northern Italy were obtained from the meteorology station for 2011 and used for comparison of heat input for both scenarios [38]). Energy in the form of heat would have to be added to the digester to maintain an average temperature of 19°C on the other 220 days of the year. Additional manpower would also be required since the digester system would need to be reloaded 4 times a year.

The second scenario reflects the results obtained from Digester A, in which 189 days were required for digestion to occur. In this scenario, the farmer would load the digester with 300 tons of rice straw twice a year. Assuming a maximum specific methane flow rate of 1.90

$\text{Nm}^3\text{CH}_4/\text{ton}/\text{day}$ (or $16.6 \text{ Nm}^3\text{CH}_4/\text{hr}$ assuming a 70% straw conversion during the anaerobic digestion process), a gas engine with a minimum power of 55 kW would be needed for the electrical conversion. The straw would have to be disassembled from the bales, but no shredding would be required. Approximately 900 tons of pig wastewater and 490 tons of water would be added to the straw, and the design capacity of the digester would be approximately 3500 m^3 , allowing for a void space of 15%. In northern Italy, heat would have to be added to the system year-round to maintain a digester temperature of 35°C . Approximately six times more heat energy would be required for this scenario, assuming specific heat capacities of 1.7 J/g-K for straw [39] and 4.19 J/g-K for leachate. In order to calculate specific values of heat energy required, the selected construction material and its insulative properties would need to be defined.

Heat is a major energy input required to operate the system, however, a simple heat exchange process can be utilized to recover waste heat from the engine. If this heat can be efficiently transferred to the leachate recirculation cycle then it can be evenly distributed throughout the digester. In the design of a farm-scale system, it is very important to consider the insulative properties of the selected digester material in order to minimize the required heat input. If space is not a limiting factor for the farmer and waste heat could be recovered efficiently from the electrical conversion process, then a single-stage batch reactor without chemical additions would be more feasible in terms of operation. For farm-scale operations, a simpler digestion system is more desirable for a farmer who is not trained and skilled in anaerobic reactor management.

3.4 Conclusions

The co-digestion of untreated rice straw with pig manure was carried out in two pilot-scale experiments with variables of temperature and volume of pig wastewater. The methane yield from Digester A was $231 \text{ LCH}_4/\text{kgVS}$ added and from Digester B was $12 \text{ LCH}_4/\text{kgVS}$ added over a period of 189 days. Temperature was the most obvious limiting factor in Digester B as evidenced by the fluctuation of daily methane production with temperature. However, the overall lack of methanogenic activity and gas production was due to the limited volume of pig wastewater in Digester B, which was not sufficient to establish a stable microbial community. Pig wastewater, without the addition of chemical buffering agents, served as an adequate source

of buffering capacity and ammonia nitrogen to stabilize the anaerobic digestion of rice straw and bring about an appropriate balance of nutrients. Further investigation on the role of micronutrients such as Fe, Ni, Co and Mo are recommended for future studies related the anaerobic digestion of rice straw.

The results obtained in Digester A were compared with results from a pretreated rice straw digestion experiment of the same magnitude published by Lianhua et al. (2010). The untreated rice straw requires a digestion period of 189 days versus only 89 days for the pretreated straw. In addition, approximately 67% more wastewater is required for the untreated straw and mesophilic conditions must be maintained, whereas the pretreated straw can be adequately digested with ambient temperatures ranging from 19 to 30°C. Two theoretical farm-scale scenarios modeled after Digester A and Lianhua's system demonstrate that a typical 100-hectare rice farm could produce approximately 100,000 m³CH₄ per year, yielding approximately 328 MWh. Major differences such as heat input, space requirements, loading frequency, digester volume and type, engine size, wastewater volume, and additives exist depending on whether the rice straw is untreated or pretreated. Although significantly more heat input and space is required for the untreated scenario, less additives and simpler operations may be more appropriate for a farm-scale digester.

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CHAPTER 4

Electrical Energy Production and Operational Strategies from a Farm-Scale Anaerobic Batch Reactor Loaded with Rice Straw and Piggery Wastewater

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4.1 Introduction

Waste-to-energy projects are expanding beyond just landfill gas recovery and into agricultural wastes such as animal manure and cereals silage because of new legislation and governmental subsidies for renewable energy production. In Italy, for example, the government passed a law in July 2009 and agreed to pay €0.28/kWh for electricity generated by agricultural feedstock for farm-scale plants (i.e. <1MW), which was the highest feed-in tariff in Europe [1]. To the authors' knowledge, however, the farm-scale system reported here is the only facility in operation that uses the anaerobic digestion of rice straw for waste-to-energy.

In northern Italy, rice straw is an abundant source of biomass that has the potential to be harvested for renewable energy. Through the anaerobic digestion process, organic matter is microbiologically decomposed to produce biogas that can be incorporated directly into the natural gas grid (after purification) or used for the cogeneration of heat and electricity. Methane yields from untreated rice straw digested in optimum conditions in both lab and pilot-scale studies range from 190 to 280 L CH₄/kgVS added [2-5]. The ideal operational conditions for the anaerobic digestion of rice straw have been defined in numerous lab-scale studies [6] and two pilot-scale batch reactors (i.e. $\geq 1\text{m}^3$)[4, 7].

One specific challenge associated with rice straw is that it is a complex, ligno-cellulosic material, resistant to anaerobic degradation because the lignin component acts as a shield and limits the hydrolysis process [8, 9]. Pretreatment strategies have been effective in overcoming this challenge in laboratory-scale experiments [10-14]; however, these approaches are not practical for farm-scale applications because of design constraints, increased energy inputs, excess chemical and water requirements, and waste disposal issues associated with the digestate. From a farm-scale perspective, co-digestion of straw with animal manure is a practical way to improve gas production as it provides an appropriate balance of nutrients, buffering capacity, and a diverse microbial community to carry out the digestion process [4, 15, 16].

Implementing a farm-scale biogas plant using rice straw co-digested with piggery wastewater offers a sustainable alternative for managing the disposal of agricultural residues, and it reduces a significant portion of methane emissions. Rice paddy fields make up 10 to 13% of the global anthropogenic methane emissions [17], and removing the rice straw from the fields upon harvesting has been shown to reduce total greenhouse gas emissions (Mg CO₂ eq./ha) by 87% [18]. From an economic

perspective, legislative incentives can offer a profit to the user and ultimate increase demand for and efficiency of methanisation units.

The purpose of this research is to define and optimize operational parameters for a farm-scale anaerobic batch reactor loaded with rice straw and pig wastewater. From the existing literature, there are no other farm-scale or full-scale biogas plants currently using rice straw as the primary substrate. The specific objectives of this study are to monitor the operation of a farm-scale co-digestion plant, to recommend optimization strategies in regards to additional energy and wastewater inputs, and to determine if this system is sustainable on a long-term basis. The parameters that will be discussed include cumulative energy production, daily power production, biogas quality, specific methane yields, total solids concentration, straw to wastewater ratios, temperature, leachate recirculation strategies, and analytical monitoring of the leachate.

4.2 Material and Methods

The biogas plant is located on a rice farm in the Pavia region of northern Italy and start-up was initiated in October 2010. An overall scheme of the plant including the digesters, gas collection system, biogas engine, and the leachate recirculation system are shown in Figure 4-1. These components and the typical operational settings of the plant are described below.

4.2.1 Anaerobic Digester Cells

The farm-scale digester consists of two anaerobic cells with a total storage capacity of approximately 13,000 m³, which equates to approximately 1825 tons of rice straw. The digester cells are insulated with a 1-mm PVC-based liner on the top, a 3-mm polyethylene liner on the bottom, and an earthen berm and hydraulic seal around the perimeter. The footprint of each cell, including the earthen berm and hydraulic ditch, is 58.5 m long by 45.5 m wide. The surface area dedicated to the storage of rice straw is 46.5 m by 35.5 m for each cell, and the initial maximum height was 5 m for cell 1 and 4 m for cell 2.

The entire digester is designed as a batch reactor that is ideally loaded with rice straw once a year during the harvest season. During the initial loading event, 3050 bales (1098 tons) of straw were added to cell 1 and 2020 bales (727 tons) of straw were added to cell 2. The rice straw had a TS concentration of 84.3% during the initial loading event. This quantity of rice straw was harvested during one season from a

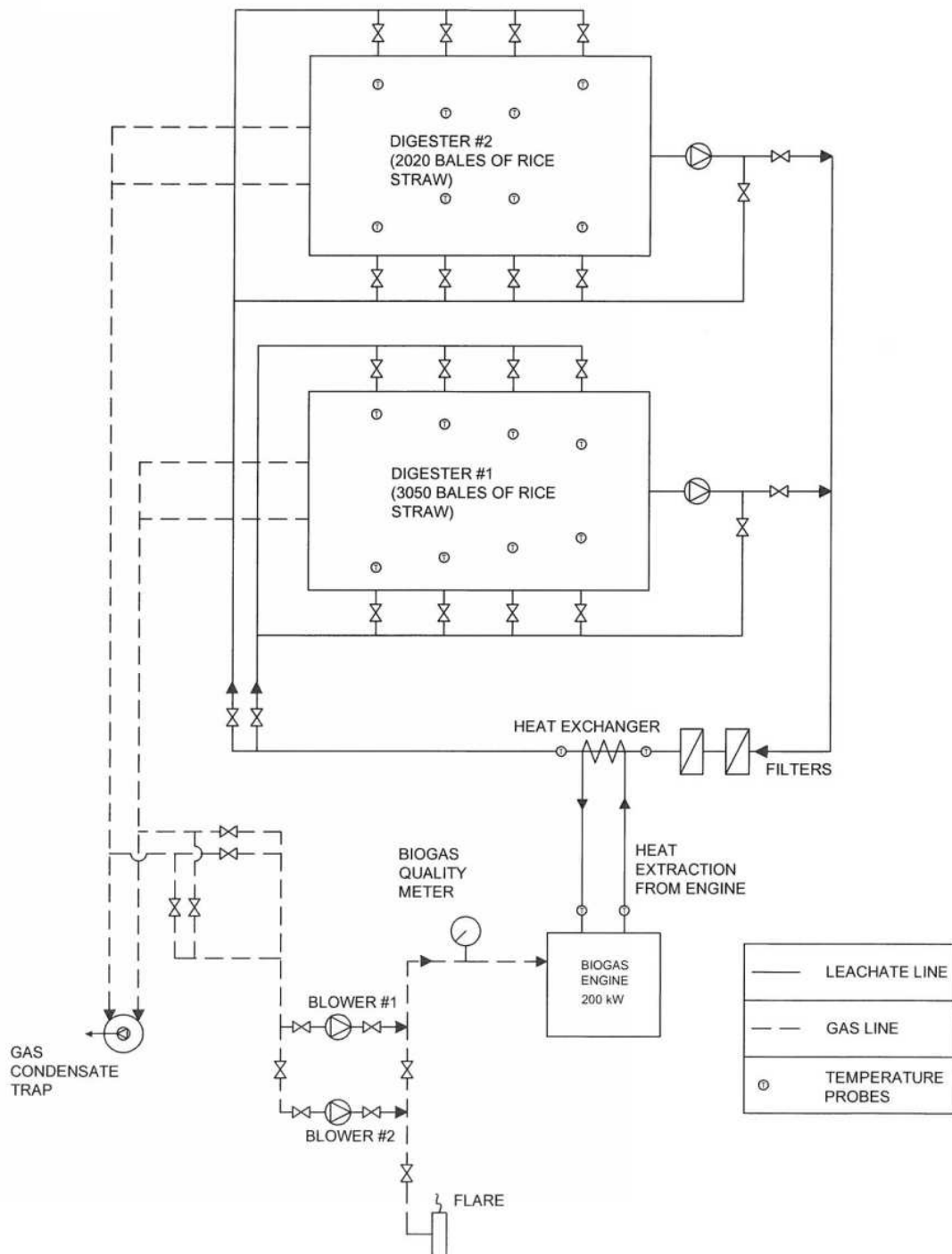


Figure 4-1. Overall process scheme for the farm-scale biogas plant

365-ha (902-acre) rice farm in northern Italy. The digester was initially inoculated with a total of 285 tons (4.9%TS) of piggery wastewater, and approximately 1300 tons of water were added over time during the first year.

The first digestion cycle was initiated in October 2010 and completed in December 2011 and only cell 2 was active during this monitoring period. Therefore, the results and calculations reported herein are based only on the rice straw digested in the active cell over a 422-day digestion period. The overall weight ratio (in tons) of straw to wastewater in the active cell was 2.55 to 1, which equates to a straw (dry wt.) to wastewater (wet wt.) ratio of 2.15 to 1. The overall TS concentration of the entire cell after the addition of water and wastewater was 46%. However, the moisture content is stratified over the vertical profile of the digester, with the bottom third completely saturated and the top two-thirds only partially saturated. Assuming only 30% of the moisture is contained in the top two-thirds where the leachate flows through a repeated pathway, the estimated TS concentration of the bottom third of the digester is 23% and most of the gas production is presumably occurring in this zone.

4.2.2 Gas Collection System

Biogas is transferred from the digester cells to the internal combustion engine by a gas collection system including eight 140-mm polyethylene lines (4 from each digester cell) and four 65-mm polyethylene lines (2 from each digester cell) situated on the west end of the digesters. The digesters are normally kept in negative pressure conditions. When enough biogas has accumulated inside the digester cells and the pressure reaches equilibrium (0 mm H₂O) or becomes slightly positive, the biogas is collected via a blower and used to power the engine for energy conversion. During the monitoring period, the negative pressure generally cycled between 0 and 300 mmH₂O in order to maximize the engine run time while being careful not to cause a breach in the hydraulic seal around the perimeter of the digesters. The primary blower has a variable speed setting with a maximum flow rate of 211.5 m³/h, and under normal operating conditions it is set at 50%. A secondary blower with a maximum flow rate of 171.6 m³/h is available as a precaution in case the primary blower fails. Upon collection, the biogas passes through an internal gas analyzer that displays the composition of the biogas (i.e. %CH₄, %CO₂, and %O₂), and depending on the quality it is either sent through the combustion process or released through the flare system.

4.2.3 Biogas Engine

A 200-kWe IVECO internal combustion engine that has been modified to run solely on biogas (>37% CH₄) produces mechanical energy that in turn powers a generator for electricity production. According to manufacturer specifications, the electrical efficiency ranges from 28 to 36% depending on the energy setting. Energy is produced at a constant rate and the setting can be modified manually through the programmable logic control (PLC). The operational range is generally from 100 kWe (minimum) to 220 kWe (maximum). For optimum heat recovery, however, the minimum setting is used to extend the engine run time and maximize the heat transfer opportunity. The PLC records the cumulative electrical energy production (kWh) on a constant basis during the operation of the engine. Electricity is generated by a three-phase, 400-V Stamford generator and then transformed to 15,000V so it can be fed into the grid of the local power company. The energy produced by this system is currently subsidized by the Italian government, and a feed-in tariff of 0.28 €/kWh is expected for 15 years.

Approximately 66% of the energy produced in the process is in the form of heat energy, of which half is exhausted as steam and the other half is hot water that is sent through a series of heat exchangers. Maximizing the transfer efficiency of this heat energy is a very important part of the process and is carefully monitored throughout the system. A dual heat exchange system consisting of both a plate heat exchanger (engine coolant to intermediate) and a pipe heat exchanger (intermediate to leachate) is employed to safely transfer waste heat to the digestion process. Permanent temperature probes are mounted on the plate heat exchanger to measure the inlet and exit temperatures of the engine coolant. Temperature probes are also situated at the inlet and exit of the pipe heat exchanger to measure the incoming and outgoing leachate temperatures.

There are a total of 8 separate temperature probes evenly distributed throughout the vertical center of the active cell. The probes are embedded in the packed rice straw and are situated two meters beneath the entry points of the leachate recirculation lines on the top surface of the digester cells. Each temperature probe is associated with a particular leachate recirculation zone so that the heat input from the leachate recirculation can be monitored. An RTD portable thermometer is used to manually collect daily temperature readings from each probe. These combined measurements indicate the average temperature of the active cell. Average ambient temperatures

recorded daily were obtained from a weather station located less than one kilometer from the plant.

4.2.4 Leachate Recirculation System

Leachate is recirculated and dispersed throughout the various zones of the active digester cell to maintain moisture levels, homogenize the mixture to help balance microbes and nutrients, and to serve as a conduit for heat energy transfer. Each digester cell is equipped with eight stations, and each station consists of approximately 10 different lines through which leachate is dispersed. The stations are located around the perimeter of the digester cell and each station is equipped with a manual valve that is only opened when that particular zone requires recirculation. The leachate lines extend from the stations to the top of the digester and are spaced in a grid system approximately 4 meters apart across the top of the liner. Each leachate line has a separate valve that can be opened or closed depending on which particular areas inside the zone need additional moisture for improved settlement. The leachate lines provide coverage over the entire the digester and air-tight screw and cap devices keep air from leaking through the entry points.

The rice straw is underlain with a gravel bed that slopes toward a central drain located on the east side of each digester cell. Leachate is collected through the drain system and recirculated by a variable-speed submersible pump. The maximum design flow rate of the pump is 29.5 m³/h, but optimum flow rates are determined by the most efficient heat transfer to the leachate through the heat exchange process. Volumetric flow rates are recorded manually using an Ultraflux Mini Sonic P portable flow meter. The piping system allows leachate to be pumped from the central drain through the heat exchanger and then back to the digester or simply in a closed loop from the central drain back to the top of the digester. Prior to entering the pipe heat exchanger, leachate is pumped through two basket filters in series that are designed to capture particulates. The initial filter captures particulates larger than 6 mm in diameter and the second filter is sized to capture debris larger than 2 mm in diameter.

4.2.5 Analytical Monitoring

Samples of the initial rice straw and the final digestate were analyzed and the analytical results are shown in Table 4-1. During the experiment, leachate samples were collected periodically during recirculation and analyzed for pH, volatile fatty acid

(VFA), carbonate alkalinity (C_T), and total ammonia nitrogen (TAN). The pH was analyzed with a Hamilton Filltrode probe and was conducted in accordance with the APHA Standard Method 423 [19]. VFA (expressed as acetic acid only) and C_T were analyzed by a titration method using 0.1 M hydrochloric acid (HCl) and acidifying the sample to pH of 2.2, while continually recording pH and using a computer modulation to calculate the results [20]. The TAN concentration (NH_3 and NH_4^+ -N) was analyzed using a spectrophotometer (SPT-500) with a Carlo Erba reagent kit (0800.05405).

4.3 Results

4.3.1 Cumulative Energy and Power Production

Biogas production started in the active digester (cell 2) on October 5, 2010 with the addition of piggery wastewater. During the first 55 days, the methane content in the biogas was less than 37%, therefore the engine was not in operation and no energy was produced. During this start-up period, approximately 3500 m³CH₄ was sent through the flare system. On Day 55, the methane content in the biogas had increased to 45%, so energy production was initiated and the engine cycled on and off every few days as determined by the pressure change in the digester. As shown in Figure 4-2, the cumulative energy production (MWh) was relatively constant through Day 200. During this lag phase, the ambient air temperatures and corresponding digester temperature were below 20°C. An exponential increase in energy production started around Day 200 when the digester temperature exceeded 20°C. The digestion cycle for cell 2 was completed after 422 days with a cumulative energy production of 295 MWh.

Figure 4-3 clearly shows the direct correlation between daily electrical energy (i.e. power) production (MWh/d) and digester temperature. The highest ambient temperatures were measured during the month of August 2011, when the digester temperature remained at 35°C for approximately 30 days resulting in a monthly average power production of 2.12 MWh/d. The maximum power production (2.74 MWh/d) occurred on Day 335 (i.e. September 5, 2011), at the end of this 30-day mesophilic period. At the peak of daily gas production, the engine run cycle was 18 hours on and 6 hours off. The gas production rate was never high enough to run the engine continuously for more than a 24-hour period at the minimum engine setting of 100 kWe.

Table 4-1. Characteristics of Rice Straw Before and After Digestion

Analyte	Rice Straw	Digestate
TS [%]	84.3 ± 6	20.9 ± 5
VS [%TS]	87.8 ± 3.2	75.2 ± 0.3
TOC [%TS]	42.8 ± 0.4	41.7 ± 0.2
Humic + Fulvic Acid [%TS]	10.9 ± 0.1	17.9 ± 1.0
TKN [g/kg]	5.0 ± 0.3	3.7 ± 0.3
TAN [g/kg]	0.055 ± 0.001	0.076 ± 0.01
Arsenic [mg/kgTS]	0.21 ± 0.01	0.36 ± 0.02
Cobalt [mg/kgTS]	0.23 ± 0.03	0.22 ± 0.06
Cadmium [mg/kgTS]	0.12 ± 0.02	1.71 ± 0.01
Chromium [mg/kgTS]	6.1 ± 0.3	13.8 ± 0.1
Chromium VI [mg/kgTS]	0.03	0.05
Mercury [mg/kgTS]	0.43 ± 0.02	0.68 ± 0.06
Lead [mg/kgTS]	3.2 ± 0.1	3.9 ± 0.1
Nickel [mg/kgTS]	4.4 ± 0.6	8.9 ± 0.1
Copper [mg/kgTS]	18.3 ± 1.2	15.8 ± 1.4
Zinc [mg/kgTS]	54.3 ± 1.6	124 ± 0.6
Sodium [mg/kgTS]	712 ± 11	1,345 ± 2
Iron [mg/kgTS]	320 ± 24	1,123 ± 14
Calcium [mg/kgTS]	6,669 ± 17	17,205 ± 20
Phosphorus [mg/kgTS]	1,082 ± 118	2,913 ± 133
Potassium [mg/kgTS]	9,676 ± 23	24,893 ± 267
Magnesium [mg/kgTS]	1,572 ± 12	4,276 ± 31

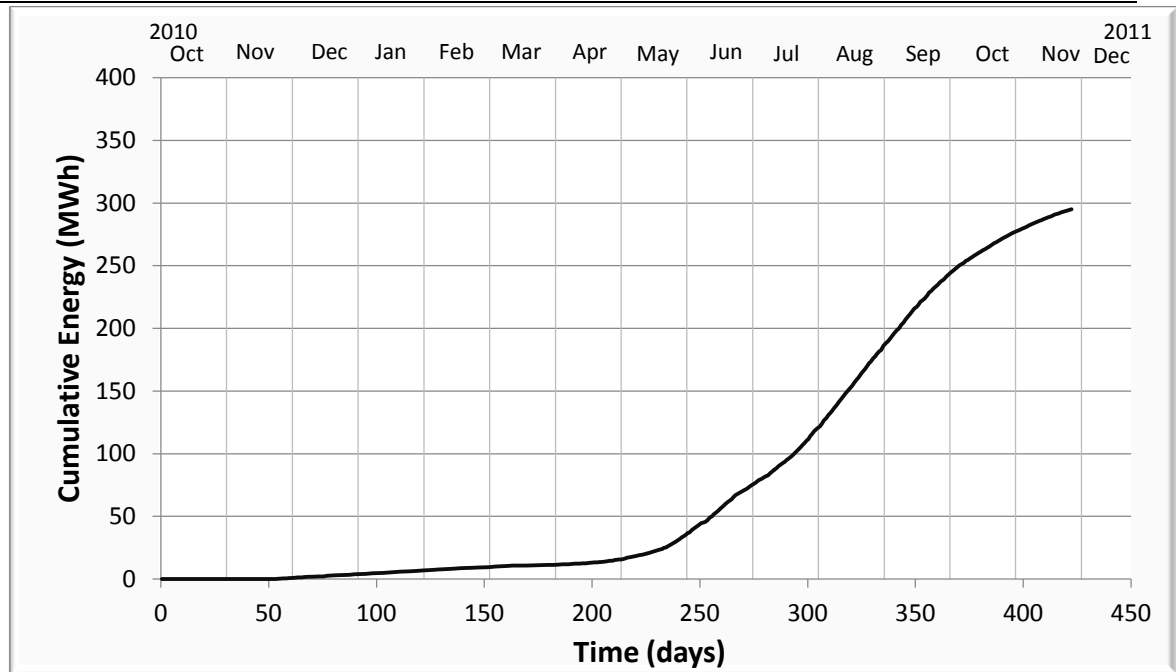


Figure 4-2. Cumulative energy as a function of time

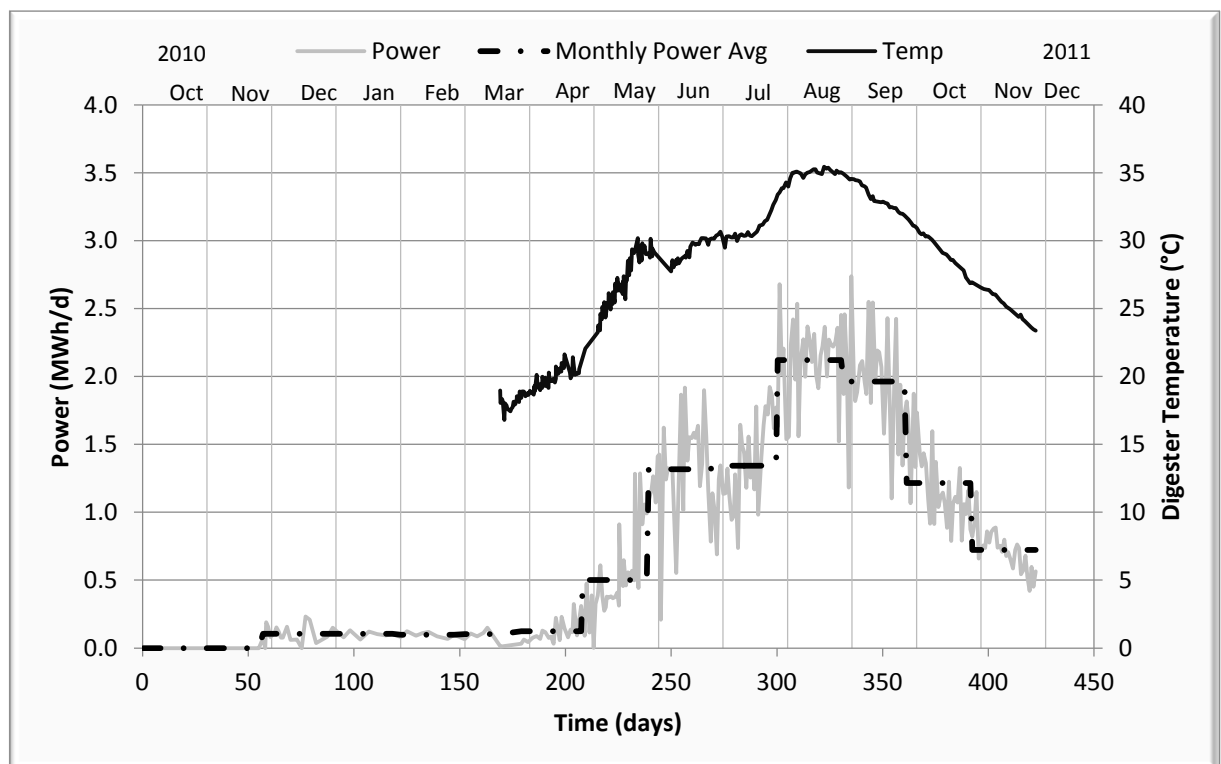


Figure 4-3. Relationship between power production and digester temperature over time

4.3.2 Biogas Quality and Methane Yield

The biogas composition was measured daily to monitor the stability of the anaerobic digestion process and to ensure there was sufficient fuel for the engine (i.e. methane > 37%). Figure 4-4 shows the measured methane and oxygen content in the biogas over time. The methane steadily increased during the first 100 days from 30 to 50%, where it remained fairly stable until Day 200. The quality of biogas dropped on two separate occasions, each characterized by a slight detection of oxygen. On these occasions, the digester was inspected thoroughly to determine the source of the breach. Oxygen was detected over a 20-day period surrounding Day 200, and the source was a leaky metal flange attached to the cover where the gas collection pipe entered the digester. Oxygen entered the system again around Day 275 when leachate spilled over the earthen embankment and compromised the hydraulic seal. The average methane content (excluding the initial start-up period of 55 days) was 48% which is consistent with results from rice straw digested with similar inocula in both lab-scale [2] and pilot-scale studies [4].

The cumulative specific methane yield over time is also shown in Figure 4-4. The specific methane yield is an estimated value since the volume of biogas sent to the engine was not directly measured. The volume of methane used to fuel the engine was calculated from the energy (MWh) assuming a 33% electrical conversion efficiency and using the calorific value of methane (50.1 MJ/kg CH₄). This combined with the measured volume of methane sent to the flare was used to calculate the specific methane yield. The yield is based on the volatile solids of rice straw (526.4 tons VS) and pig wastewater (8.0 tons VS) added to the digester at the beginning of the digestion cycle. After 422 days, a specific methane yield of 181 LCH₄/kgVS added was estimated. The volume reduction of the biomass in the active digester cell was 2550 m³ (i.e. 50% reduction) with a uniform height reduction of 2.23 m. Both the microbiological degradation as well as the changing geotechnical properties of the material throughout the digestion period contributed to the overall reduction.

After the initial saturation at the beginning of the digestion cycle on Day 13, leachate recirculation rates were generally less than 20 m³/d (0.04 m³/m³straw-d) in the winter months and minimal heat was distributed since the engine run times were so short (3 to 4 hours at a time). With ambient temperatures approaching 20°C, leachate recirculation was sharply increased to improve mixing of the biomass during days 174 through 182, with an initial peak of 90 m³/d (0.18 m³/m³ straw-d).

With the exception of periodic maintenance periods, leachate recirculation was employed 24 hours a day in the summer months, even when the engine was not running. A particular zone was first saturated with leachate using high volume flows (engine off), followed by low volume flows of heated leachate during engine operation. The recirculation was rotated on a daily basis so that each of the eight zones were saturated and heated over an eight-day period. The highest recirculation rate was $107 \text{ m}^3/\text{d}$ ($0.21 \text{ m}^3/\text{m}^3 \text{ straw-d}$) and it occurred on day 294.

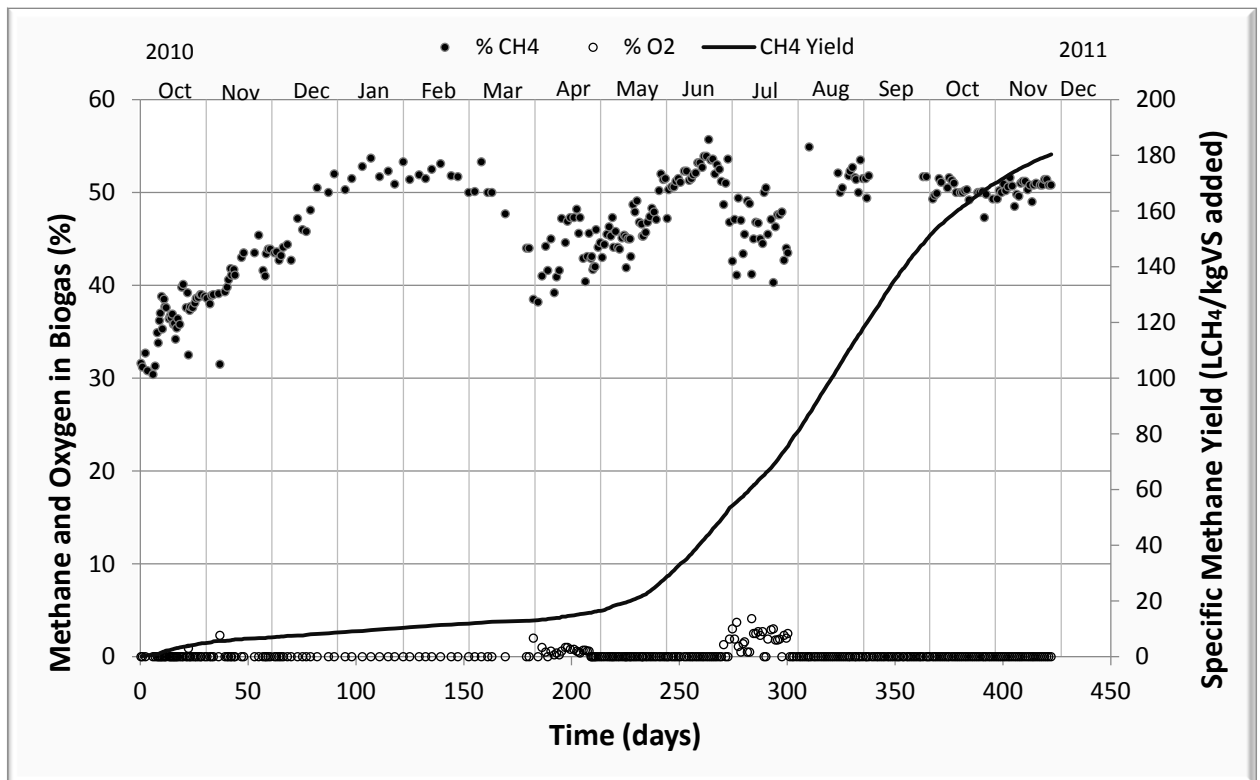


Figure 4-4. Measured biogas quality and estimated specific methane yield over time

4.3.3 Temperature Profile

The average ambient temperature, digester temperature, and maximum leachate temperature exiting the heat exchanger were recorded daily and the trends are shown on Figure 4-5. The ideal operating temperature for the digester is 35 to 40°C [6]; however, this operation condition was only achieved during the month of August 2011 when daily average ambient temperatures ranged from 23 to 30°C . Several factors limited the heat input into the digester such as discontinuous engine operation, limited leachate recirculation volume, and maintenance problems with the heat exchanger. During the winter months, minimal biogas was produced and the engine only operated 10 to 12

hours a week. Therefore, not enough heat was generated to raise the digester temperature. When the ambient temperatures exceeded 20°C in mid-April 2011, digester temperatures increased and biogas production rates also increased. This led to more continuous engine operation, which provided more waste heat recovery for the leachate recirculation system.

Once the ambient temperatures exceeded 20°C, the insufficient capacity of the heat exchanger was the limiting factor in the overall digestion process. In order for sufficient heat exchange to occur (i.e. leachate temperatures exiting the heat exchanger at 35°C or higher), the leachate recirculation volume rate could not exceed 100 m³/day, even though the maximum pump capacity was 700 m³/day. This limitation minimized the leachate recirculation potential and therefore the heat had to be concentrated in only one of the eight zones per day during engine operation. Maintenance problems such as the build-up of solids over time and clogging of the heat exchanger also reduced heat transfer efficiency. The heat exchanger was not functioning properly for approximately 45 days in June/July 2011 as evidenced by the digester temperature trend in Figure 4-5. Maintenance was completed and the heat exchanger was repaired on Day 287, after which the digester temperature rose from 30 to 35°C in 16 days, with no significant change in the ambient temperature.

A major advantage in the system is that heat can be maintained in the digester. Once the entire digester reached a maximum temperature of 35.5°C on Day 322, the digester remained an average of 14°C higher than the daily average temperature for the duration of the digestion cycle. Since the volume of leachate recirculated during September through October 2011 was decreasing (see Figure 4-6), the heat maintained in the digester was primarily a result of the insulation provided by the liner system and the internal heat generated from the biological activity inside the digester.

4.3.4 Leachate recirculation strategy

Leachate recirculation is not only used to achieve a homogenous mixture between the substrate and the inocula, but also to introduce heat into the digester. It is important to maintain the appropriate balance between high volume flows to saturate the media and low volume flows to supply maximum heat input during engine run times. The daily flow rate of leachate recirculated in the active cell is shown in Figure 4-6. The flow rate is depicted in relationship to the straw contained in one zone of the digester, since recirculation was applied to a single zone each day.

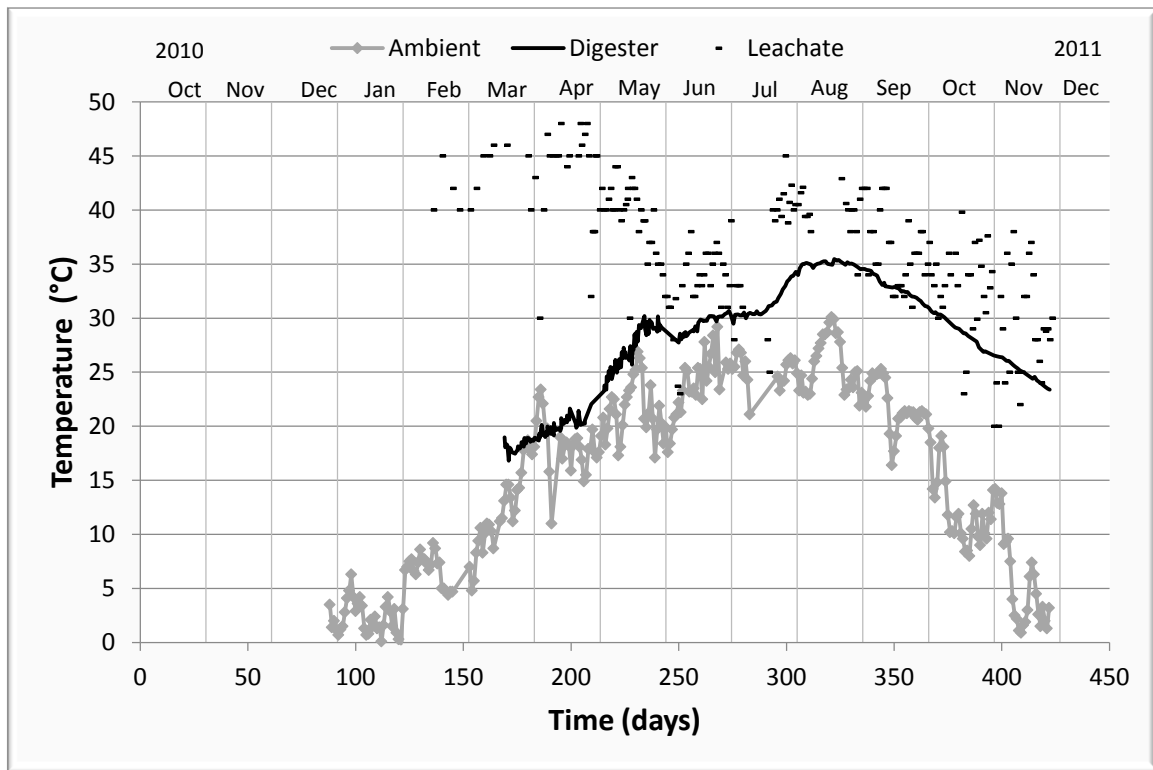


Figure 4-5. Temperature profile over time including ambient temperature, digester temperature and maximum leachate temperature exiting the heat exchanger

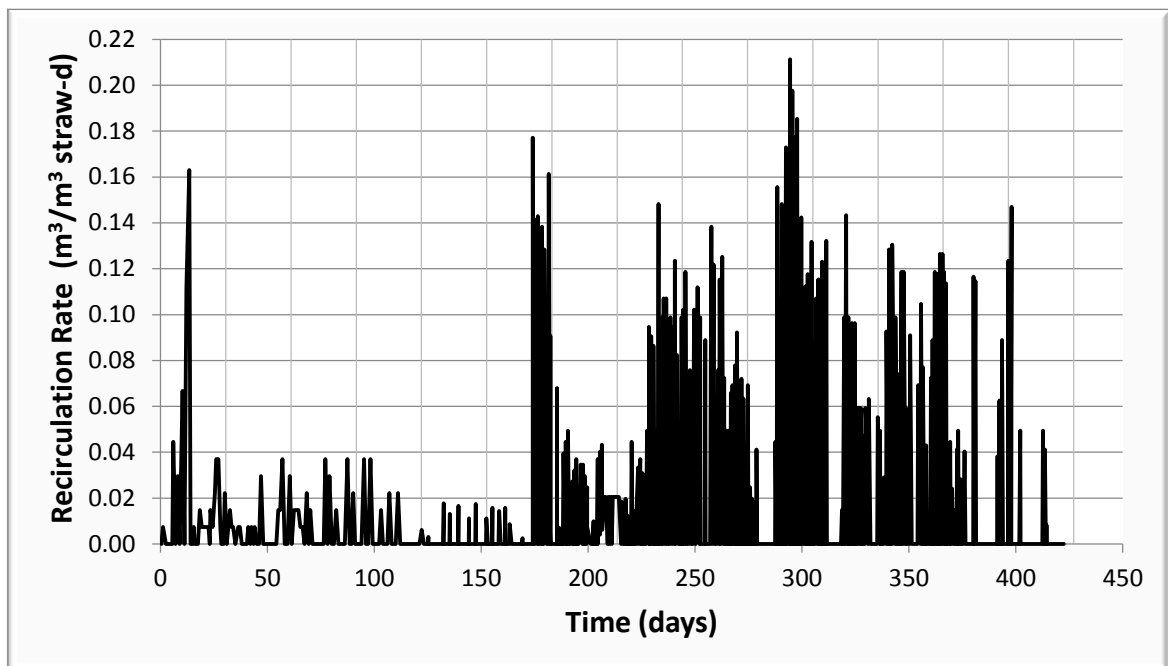


Figure 4-6. Leachate recirculation daily flow rate (based on volume of straw in one zone)

4.3.5 Analytical Monitoring Results of Leachate

Periodic measurements of the leachate pH, alkalinity and VFA concentrations were taken to monitor the stability of the digestion process over time and the results are shown in Figure 4-7. VFAs are intermediates produced during the hydrolysis and acidogenic stages of the digestion process, and VFA accumulation can lead to acidification of the reactor. In the farm-scale digester, a typical evolution of VFA production (expressed as acetic acid) was observed. A sharp increase in acetic acid occurred during the first month following the addition of the pig wastewater, with a maximum concentration of 7185 mgHAc/L occurring on Day 28. This peak in VFA production was likely associated with the degradation of the simple soluble organic matter contained in the pig wastewater since concentrations steadily decreased to 80 mgHAc/L over the next 83 days. A second slower rise in VFAs occurred over the next 71 days with a second peak of 5268 mgHAc/L occurring on Day 182. This peak represents the solubilization and hydrolysis stages of the straw material, as evidenced by the increased energy production that continued through the end of the digestion cycle. VFA and alkalinity concentrations were not measured during May and June 2011 because of problems with the instrumentation.

The pH remained fairly stable within the ideal range for methanogenic bacteria of 6.6 to 7.8 [21] for most of the digestion cycle. Sufficient alkalinity was supplied by the pig wastewater and no additional buffer was necessary to maintain concentrations above 4000 mg CaCO₃/L for most of the digestion cycle. On Day 211 (29 days after the second peak in VFA production) the pH and alkalinity dropped to minimum values of 6.3 and 1025 mg CaCO₃/L, respectively, but quickly recovered.

Nitrogen is an essential nutrient for the anaerobic microorganisms and TAN concentrations below 200 mg/L are considered to be beneficial while concentrations exceeding 1500 mg/L can be moderately inhibitory [22]. The TAN concentration of the leachate is shown in Figure 4-8. The TAN concentration rapidly increased to 1700 mg/L with the addition of the pig wastewater and remained above 1000 mg/L until Day 182. From Day 182 through Day 211, consumption of TAN is evident (i.e. concentration decreases from 1186 to 292 mg/L), and then the concentration remains slightly above 200 mg/L through the end of the digestion cycle.

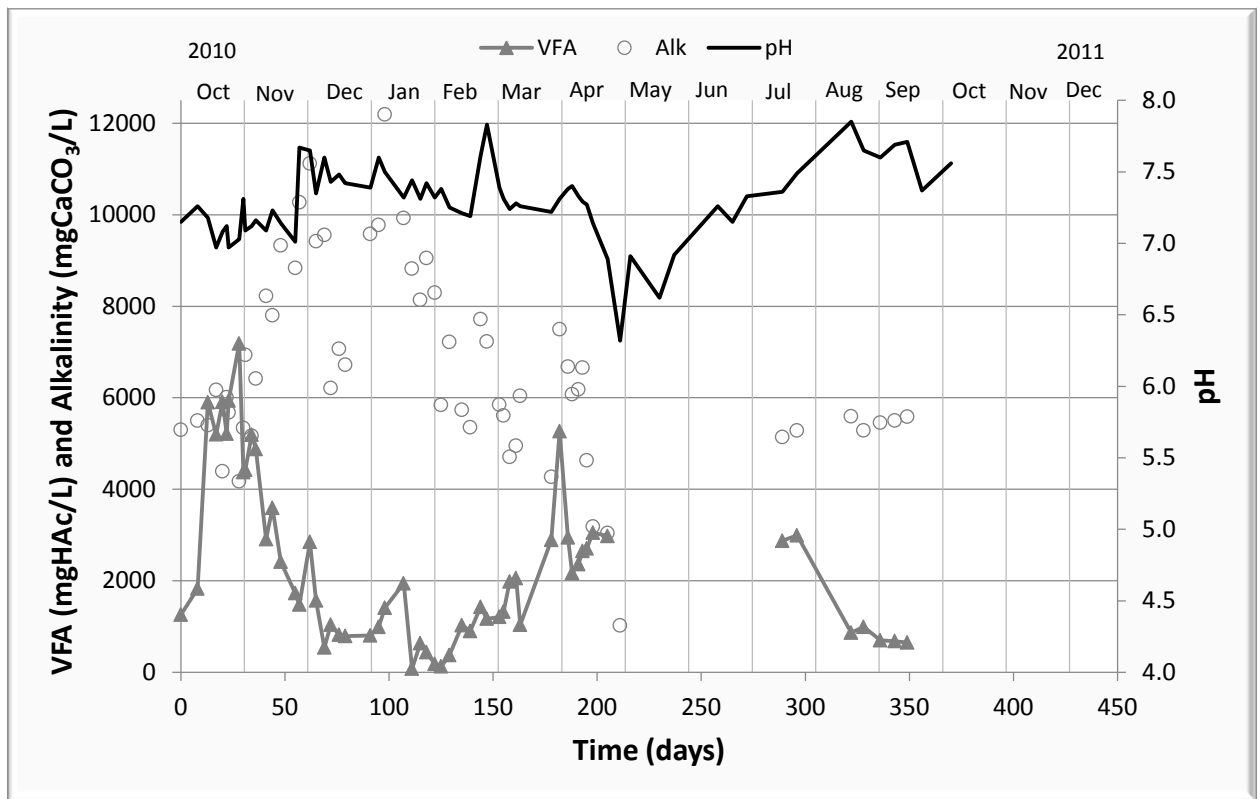


Figure 4-7. VFA (as acetic acid), alkalinity, and pH of leachate

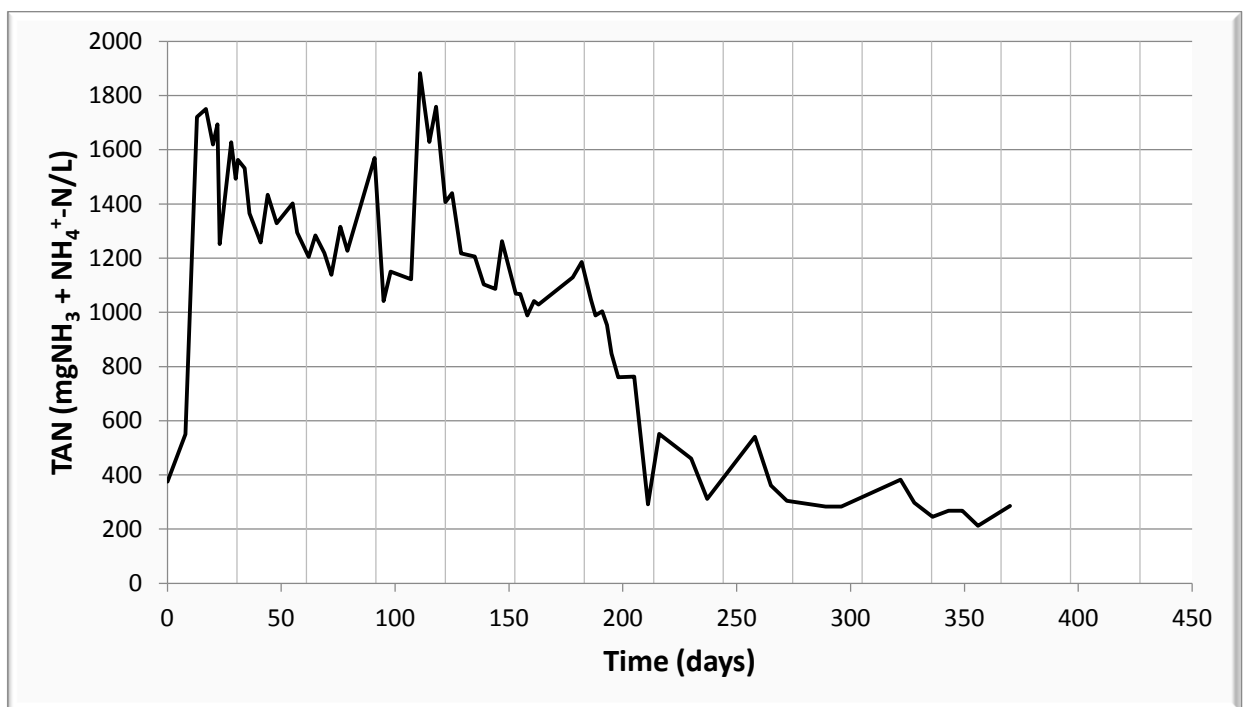


Figure 4-8. TAN measured in leachate

4.4 Discussion

The start-up period or acclimation time for the reactor was approximately 200 days, evidenced by the lack of energy production during this time. One reason for the extended start-up was that only raw pig wastewater was added to stimulate gas production with no digested material or anaerobic sludge from an established reactor. This reduced start-up costs and also ensured that the regulatory guidelines for electricity produced from agricultural residues were met. Other factors also contributed to the slow acclimation period including a high straw to wastewater ratio, low ambient temperatures ($<15^{\circ}\text{C}$), and low leachate recirculation rates ($<0.04 \text{ m}^3/\text{m}^3\text{straw-d}$). In order to critically evaluate the system, it is important to understand which of these factors should be adjusted to make the system more efficient and attain a higher energy output in a shorter digestion cycle.

The appropriate straw to wastewater ratio for a farm-scale system of this magnitude is difficult to predict because the digestion is likely occurring in the saturated zone at the bottom. With limited mixing (*i.e.* only from the leachate recirculation), the most homogenous layer is at the bottom. Once this layer is consumed, more substrate settles into the saturated zone, creating a plug flow scenario in the context of a batch reactor. This natural progression minimizes the volume of wastewater required, which reduces transportation costs and avoids complicated disposal issues. The overall straw (dry wt.) to wastewater (wet wt.) ratio in the active cell, however, was still too high (2.15 to 1) for efficient gas production, which equates to a 1:1 ratio in the saturated zone or bottom one-third of the digester (assuming that 70% of the recirculation volume returns to the bottom). A pilot-scale reactor that operated in ambient conditions with a straw (dry wt.) to pig wastewater (wet wt.) ratio of 1:1 had a long acclimation period (>189 days), while the same reactor operated in mesophilic conditions with a ratio of 1:3 had an acclimation period of approximately 30 days [4]. Therefore, additional wastewater should be added to achieve a straw (dry wt.) to wastewater (wet wt.) ratio of 1:3 in the bottom saturated zone. Coupling this addition with recommended start-up during warmer months could reduce the start up time from 200 days to less than 60 days based on results observed in a pilot-scale reactor [4].

With the initial addition of the soluble material (pig wastewater), VFAs were rapidly formed (by day 28) and the established methanogenic population was capable of converting these intermediate products to methane. The rate-limiting step in the digestion process, however, was the fermentation of the straw material since VFAs were

slow to reappear (i.e. day 182). Fermentation of lignocellulosic material is dependent on enzymes to break the lignin barrier and make the cellulose available for the microbes, thus many have concluded that hydrolysis is the rate-limiting step during the anaerobic digestion of lignocellulosic material [23-25]. Once VFA production from the hydrolysis of the straw material peaked on Day 182, methane production was stimulated. The increased energy production allowed for longer engine run times, resulting in higher heat recovery and increasing digester temperatures. Increasing temperatures improved biogas quality and methane yield, and the productivity of the digester continued to increase. To accelerate the onset of hydrolysis in the following digestion cycle, it is recommended that the leachate remaining from the previous cycle and a remnant of the digestate be kept in the cell and mixed with the new material for growth of acclimated microbes.

The VFA peak that occurred on Day 182 represented the end of the acclimation period and it is important to understand what operational changes contributed to this VFA production. The VFA peak that occurred on Day 182 immediately followed a nine-day period (day 174-182) during which leachate recirculation rates were increased from $0.02 \text{ m}^3/\text{m}^3\text{straw-d}$ to more than $0.14 \text{ m}^3/\text{m}^3\text{straw-d}$. The increase in gas production appeared to be a direct result of the increased recirculation rates, which agrees with previous findings. Veeken and Hamelers (2000) demonstrated that a leachate recirculation rate of $0.1 \text{ m}^3/\text{m}^3\text{biowaste-d}$ applied to dry biowaste (40% TS) more than doubled methane production when compared to the same system without leachate recirculation [26]. Therefore, higher recirculation rates ($>0.14 \text{ m}^3/\text{m}^3\text{straw-d}$) are recommended as well as periodic additions of fresh wastewater to improve gas production and reduce the acclimation period. Leachate recirculation rates cannot, however, be increased unlimitedly because the transport of VFAs from the substrate to the seed may result in VFA accumulation and irreversible acidification of the reactor [26].

The pig wastewater added to the digester provided adequate buffering capacity and sufficient methanogenic activity to prevent prolonged VFA accumulation and acidification of the digester. During the peak VFA production periods, the pH remained stable (7.0) or dropped slightly (6.3) but quickly recovered. Though TAN concentrations in the leachate were adequate, the available nitrogen in the solid phase may have been insufficient. Intermediate sampling of the solid phase was not possible, however, the final digestate sample had a C:N (i.e. TOC to TKN) ratio of 117. One of

the primary benefits of co-digestion is to reduce the average C:N ratio of rice straw (i.e. 70:1) [4] to the ideal range for anaerobic microorganisms (i.e. 25-30:1) [27]. Further analysis of the solid phase throughout the digestion process is necessary to confirm whether or not sufficient nitrogen was supplied to the overall system by the pig wastewater.

Concentrations of micronutrients including cadmium, chromium, nickel, zinc, and iron were also amply supplied by the addition of the pig wastewater, evidenced by the differences shown in Table 4-1 between the rice straw and the digestate. These trace elements, as well as cobalt and molybdenum, typically found in pig wastewater, have been shown to enhance co-digestion by increasing methane yields and stabilizing the system [28]. Macronutrients that were supplied by the pig wastewater included calcium, phosphorus, potassium, magnesium and sodium, as evidenced by the results shown in Table 4-1.

Economic returns are the driving force of the biogas boom in Europe, and a basic economic analysis can help to determine which optimization strategies are profit-gaining. The first digestion cycle lasted 422 days and the gross earnings from the electricity generated were €82,600. Estimating approximately €400,000 for the initial infrastructure of the active cell only and €23,000 per year for maintenance and manpower, the return of the investment could theoretically be achieved in 8.3 years. The estimated cost of the initial infrastructure (€400,000) does not incorporate the second cell since it did not contribute to the energy production during the 422-day digestion cycle. In addition, this raw estimate does not account for the time value of money, nor does it consider the changes in subsidy rates over time. In January 2013, for example, the subsidy rates were reduced by 40% for electricity generation from agricultural residues. Considering these additional factors, the payback time could double, making the current operation economically unsustainable. The system will, however, likely operate more efficiently during the years following the start-up period and specific improvements should be made based on beneficial profit and energy returns.

4.5 Conclusion

A farm-scale biogas plant loaded with rice straw and piggery wastewater was monitored during the start-up period and first digestion cycle. The digestion cycle for the active cell was completed after 422 days with a cumulative energy production of

295 MWh. A direct correlation between daily power production and digester temperature was observed, and the maximum power production (2.74 MWh/d) occurred during mesophilic conditions inside the digester. Waste heat recovery from the engine was capable of heating increasing the digester temperatures to mesophilic conditions when the heat exchange system was functioning properly. After reaching mesophilic conditions, the digester remained 14°C warmer than the ambient conditions primarily as a result of the insulation provided by the liner system and the internal heat generated from the biological activity inside the digester.

The rate-limiting step in the overall process was the hydrolysis of the straw material. The slow acclimation period as well as the volume reduction of the biomass (i.e. 50%) can be improved with several factors including increased leachate recirculation rates ($> 0.14 \text{ m}^3/\text{m}^3\text{straw-d}$), an improved heat exchange system to maintain mesophilic conditions year round, addition of anaerobic sludge acclimated to lignocellulosic material such as the existing digestate, and an increased straw to wastewater ratio. Although sufficient buffering capacity as well as macro- and micronutrients were supplied to the system by the pig wastewater, a straw (dry wt.) to wastewater (wet wt.) ratio of at least 1 to 1.4 is recommended to improve gas production and decrease the acclimation period. This correlates to a 1 to 3 ratio in the bottom third or saturated zone, assuming 30% retention in the upper zones.

A raw economic assessment of the system shows a theoretical investment recovery time of 8.3 years, however, this is based solely on the expenditures and subsidy rate from 2011. For an ideally sustainable system, the digestion cycles for both cells should be completed in a year so that the digester can be reloaded after each harvest. This goal can likely be achieved with simple investments and strategies discussed above in order to increase methane production and improve the overall efficiency of the system.

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CHAPTER 5

Enhanced Methane Production from Lignocellulosic Wastes Co-digested with Anaerobic Sludge from Pulp and Paper Mill Treatment Process in Lab-Scale Digesters

(Rice straw sections previously published: **Mussoline, W., G. Esposito, P. Lens, A. Spagni, and A. Giordano.** 2013. Enhanced methane production from rice straw co-digested with anaerobic sludge from pulp and paper mill treatment process. *Bioresour Technol* **148**:135-143)

5.1 Introduction

Rice straw is an agricultural residue that is available in abundant supply, however, the uses for this material are limited mainly to cooking/heating fuel and animal feedstock [1-3]. Industrial uses are not fully realized and although some attempts are currently being made to capture the energy potential in large-scale applications [4-6], most of the energy potential is lost through common practices such as open burning and tilling the straw back into the fields which both could contribute to methane gas emissions to the atmosphere [1, 7, 8]. Rice straw co-digested with animal manure in anaerobic conditions has been shown to be effective in both lab and pilot-scale experiments [3, 9, 10]. Co-digestion improves substrate treatability since the straw material alone lacks alkalinity and appropriate nutrients to carry out the process [9]. Although the energy recovery by means of anaerobic digestion is an attractive option, the primary obstacle of the process is the microbial degradation of the lignocellulosic material.

The anaerobic digestion of lignocellulosic material has been studied extensively in an attempt to overcome the challenge of degrading this tightly bound material and making it more available for energy conversion. The lignin content in agricultural residues, such as rice straw, make degradation difficult because the ligno-carbohydrate complexes create a barrier for microbial conversion [11]. Lignin is considered the most important factor affecting the hydrolysis of the cellulose component in the lignocellulosic material [12]. Low methane yields and long digestion times are consistently observed with the anaerobic digestion of untreated rice straw, even when co-digested with animal manure [9, 13, 14]. To date, delignification or some other type of pretreatment is used to separate the lignin from the cellulose so the cellulose can be fermented easily and converted to methane for energy recovery. Various studies have demonstrated the effectiveness of thermal, chemical and biological pre-treatment of the straw material [2, 11, 15-18], however, these approaches are not typically energy efficient or practical in terms of design for farm-scale or industrial applications.

A different approach could be the integration of the appropriate microbes and/or enzymes necessary to break down the lignocellulosic material into the anaerobic digestion process via co-digestion. In fact, anaerobic sludge from a pulp and paper mill wastewater treatment facility, for example, may be an ideal candidate for co-digestion with rice straw

since the biomass should be already adapted to lignocellulosic waste residues from the pulping process. By applying this approach, no separate pretreatment step that requires excessive chemicals, high temperatures for thermal pretreatment or aerobic conditions in the case of white-rot fungi is necessary. Although anaerobic reactors are commonly used in Europe for pre-treatment of high-strength wastewater generated by the pulp and paper industry for the combined benefit of COD removal and energy recovery [19], to the best knowledge of the authors, no application of pulp and paper waste and wastewater with other more refractory lignocellulosic materials are available.

The aim of this research is to study the effect of adding sludge collected from an anaerobic digester treating pulp and paper mill waste to lignocellulosic wastes in order to improve hydrolysis and thus methane production. Several anaerobic digesters were prepared using both untreated rice straw and sugar cane bagasse as substrates and varying ratios of piggery wastewater and anaerobic paper mill sludge as inocula to determine optimum conditions for maximum methane yield. The experiments were performed in dry conditions. Advantages of dry digestion include less water input, higher loading rates, more stable digestion conditions, improved efficiency and potentially higher biogas yields [3, 10, 20]. The overall goal is to enhance the methane production by diversifying the microbial community and available nutrients using co-digestion with both agricultural and industrial residues rather than applying pretreatment strategies.

5.2 Methods

5.2.1 Experimental Set-up

Dry batch tests were conducted as described in previous studies [21, 22]. Known quantities of solid material and various fractions of different inocula were mixed thoroughly and added to 1-L glass bottles, flushed with N₂, sealed with metal screw caps and silicone septums, and placed in a thermostatically-controlled room maintained at 35± 2°C. Temperature inside the room and atmospheric pressure conditions were recorded on a daily basis.

Methane gas was measured directly using a liquid-displacement method with 12% NaOH used as a barrier solution and converted to dry gas at 1 atm and 0°C (STP). The digesters were connected to the inverted barrier solution via 21 gauge needles and tygon

tubing (ID = 4.8 mm; OD = 8.0 mm). After the gas production rate stabilized (i.e. three days), the tubing was clamped and gas production was measured periodically for 92 days.

5.2.2 Digester Composition

Rice straw (i.e. substrate) was collected from a field in northern Italy in November 2011, approximately one week after the field was harvested, but prior to baling activities. The straw was sun-dried and stored in a cool, dry environment until the experiments were initiated in June 2012. Sugar cane bagasse was collected from sugar cane fields in Belle Glade, Florida at the end of the harvest season in April 2013. The bagasse was dried and stored in a cool, dry environment until the experiments were initiated in July 2013. Piggery wastewater for the rice straw experiments was collected from a wastewater sump at a farm that raises fattening pigs in Zuid-Holland, the Netherlands. Piggery wastewater used in the bagasse experiments was collected manually from a sow farm in Plant City, Florida. Anaerobic granular sludge was collected from a treatment facility in Eerbeek, the Netherlands, which treats a combined wastewater from five different pulp and paper mill plants. Both the wastewater and the sludge were stored at room temperature for less than a week and degassed before starting the experiments.

For the first experiment, rice straw was cut into 1-cm pieces and added as the substrate to eight experimental digesters (four digesters set up as duplicates) and two control digesters (C2, C3). Experimental digesters 1 through 4 (D1 to D4) contained differing fractions of pig wastewater and paper mill sludge. Control digester C2 contained rice straw mixed with autoclaved paper mill sludge and C3 contained rice straw only. The purpose of C2 was to evaluate the impact of the biomass without the influence of the bacterial conversion processes supplied by the paper mill sludge. C3 was set up as a control to determine the degradation capability of the inherent bacteria contained in the straw itself. For the second experiment, sugar cane bagasse was cut into 1-cm pieces and added as substrate to four experiment digesters (S1 to S4) along with the same fraction of pig wastewater and paper mill sludge used in the first experiments. One control digester (C2) containing bagasse only was also used. The contents of all the digesters (total weight ratios) are shown in Figures 5-1 (a) and 5-1 (b).

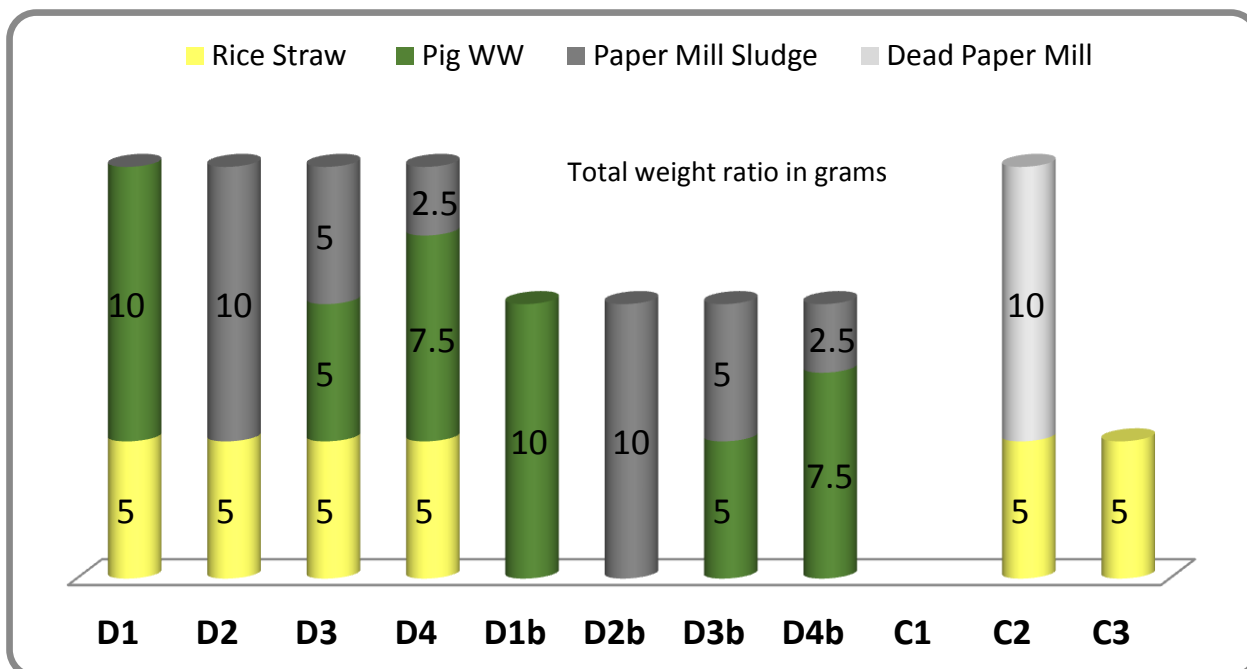
It was assumed that sufficient macro- and micronutrients would be supplied by the inocula mixture, thus no additional nutrients or buffers were supplied to the digesters. Both the experimental and control digesters were adjusted to 20% total solids (TS) by adding demineralized water, and thorough mixing was performed prior to establishing anaerobic conditions. Sludge blanks (D1b to D4b for the rice straw experiment and S1b to S4b for the bagasse experiment) were set up for each experimental digester, which contained the same quantity of inocula mixture, without substrate addition. The sludge blanks consisted of inocula only, which had a lower TS content than 20%. A pressure-control bottle (C1) containing an equivalent amount of de-ionized water was used in each experiment to account for the drip volume created by changing atmospheric conditions. The methane produced by the sludge blanks was subtracted to remove any contribution of gas from the degradable matter in the inocula. The specific methane yields were calculated by dividing the volume of methane produced at standard temperature and pressure (STP) by the weight of volatile solids (VS) of the rice straw added to each digester. The contents and quantities contained in the experimental digesters, control digesters, and sludge blanks are shown in Figures 5-1 (a) and (b).

In the rice straw experiment, a total of 84 replicate sample bottles containing the same quantities of substrate and inocula as the experimental and control digesters were assembled and stored in the same thermostatically-controlled room for the purpose of intermediate chemical analysis. These sacrificial samples were used for analysis purposes only and gas production was not measured. Representative samples (for each digester) were analyzed on day 0, day 3, day 7, day 14, day 28, day 56 and day 92.

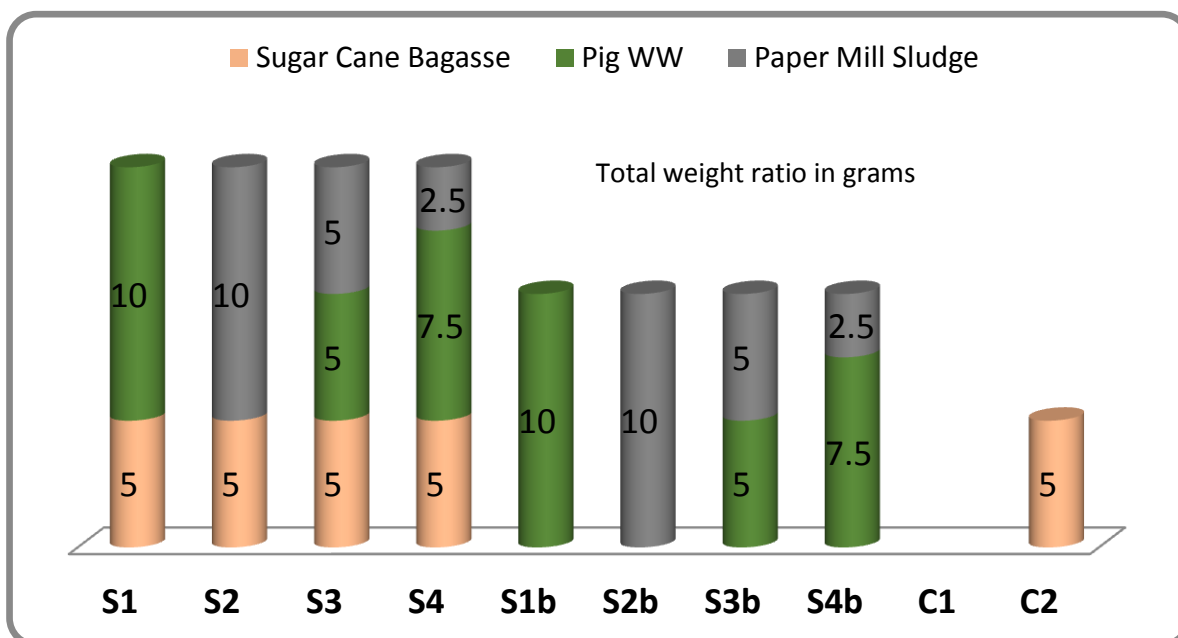
5.2.3 Analytical Methods

TS and VS of the raw materials (rice, pig wastewater and paper mill sludge) were measured in duplicate according to Standard Methods [23] for both the rice straw and bagasse experiments. For the rice straw experiments, TS and VS of the digester mixtures, which included the solid material contained in the replicate sacrificial sample bottles, were also measured and the remaining ash from the volatilization was diluted and preserved with 1% nitric acid for metals analysis. Metals including Ni, Co, Fe, Cu, Zn, and Mn were analyzed for all initial and intermediate samples using a Thermo-Scientific induced coupled

plasma mass spectrometer (ICP-MS). Using this procedure, the minimum detection limit was $2\mu\text{g/L}$ for all metals analysis. In order to report the results in terms of solids present, final concentrations were converted from $\mu\text{g/L}$ to $\mu\text{g/gTS}$. In the cases where the measured



(a)



(b)

Figure 5-1. Composition of experimental digesters, control digesters and sludge blanks for (a) rice straw experiment and (b) sugar cane bagasse experiment (on a wet weight basis)

concentrations were below the detection limit, one-half of the detection limit (1 μ g/L) was assumed and used for calculation purposes.

The solid material from the replicate sacrificial sample bottles was diluted with demineralized water then centrifuged to obtain a representative liquid fraction for pH measurement according to EPA Method 9045D. In addition to pH, volatile fatty acids (VFA), chemical oxygen demand (COD), total ammonia nitrogen (TAN), and alkalinity were measured on this liquid fraction for initial, intermediate and final samples. VFA samples were prepared in 2% formic acid and measured with a gas chromatograph (Varian 430-GC) with a flame ionization detector. The results provided individual concentrations for each of the short chain fatty acids including acetic acid, propionic acid, isobutyric acid, butyric acid, isovaleric acid and valeric acid. COD (colorimetric dichromate closed reflux method), TAN (NH₃ + NH₄⁺), total Kjeldahl nitrogen (TKN) and alkalinity were determined according to Standard Methods [23]. The solids and total nitrogen concentrations of the raw materials used in the experiments are summarized in Tables 5-1 (a) and (b).

Table 5-1(a). Solids and Total Nitrogen Concentrations of Raw Materials for Rice Straw Experiment

	Rice Straw	Piggery Wastewater	Paper Mill Sludge
TS (%)	89.9	11.8	23.0
VS (%TS)	88.4	77.8	62.2
TKN (mg/gTS)	2.9	118.4	27.9

Table 5-1(b). Solids Concentrations of Raw Materials for Bagasse Experiment

	Sugar Cane Bagasse	Piggery Wastewater	Paper Mill Sludge
TS (%)	89.7	10.2	17.3
VS (%TS)	97.6	71.1	63.7

5.3 Results

5.3.1 Methane Production

Methane production and specific methane yields for both the control and experimental digesters for the rice straw experiment are included in Table 5-2 (a). The highest methane production occurred in D2 (340 LCH₄/kgVS straw added), and a direct relationship was observed between paper mill sludge added and methane yields. Higher methane yields were observed with the digesters containing higher proportions of paper mill sludge, and all rice straw digesters containing paper mill sludge exceeded 300 LCH₄/kgVS straw added. Specific methane yields in the control digesters containing rice straw were higher than in D1, but significantly lower than the digesters containing some fraction of active paper mill sludge. As shown in Table 5-2 (a), the methane yield for D1 was 0. The reason is because the sludge blank for D1 (*i.e.* D1-b) produced the same quantity of methane as the digester mixed with straw and pig wastewater. Therefore, the methane produced in D1 was only a result of the inocula mixture, and none of the methane produced in D1 was a result of straw degradation.

Table 5-2 (a). Methane Production and Specific Methane Yields for Rice Straw Digesters

	L _N CH ₄ ^a	L _N CH ₄ /kgTS ^a	L _N CH ₄ /kgVS ^a	L _N CH ₄ /kgCOD ^a
C1	0	0	0	0
C2	0.172	38	43	40
C3	0.183	41	46	43
D1	0	0	0	0
D2	1.351	301	340	314
D3	1.332	296	335	310
D4	1.198	267	302	279

^a The values in this table represent methane produced from rice straw only (gas produced from sludge blanks (*i.e.* D1-b to D4-b) have been subtracted)

Similar results were obtained when the substrate was changed from rice straw to sugar mill bagasse, however the yields were slightly lower with the bagasse substrate. Methane production and specific methane yields obtained using the bagasse are included in Table 5-2 (b). The highest methane production occurred in S2 (326 LCH₄/kgVS straw added), which contained the highest proportion of paper mill sludge. There was no significant

difference between Digesters S3 and S4, and they produced less methane than S2. As in the rice straw experiment, the specific methane yield in the digester containing bagasse and pig wastewater without the paper mill sludge (i.e. S1) was 0. While the VS concentration of the bagasse was slightly higher, the bagasse control digester produced much less (i.e. 10%) methane than the rice straw control.

Table 5-2 (b). Methane Production and Specific Methane Yields for Sugar Cane Bagasse Digesters

	$L_N \text{ CH}_4^a$	$L_N \text{ CH}_4/\text{kgTS}^a$	$L_N \text{ CH}_4/\text{kgVS}^a$
C1	0	0	0
C2	0.016	4	4
S1	0	0	0
S2	1.427	318	326
S3	1.223	273	279
S4	1.267	283	289

^a The values in this table represent methane produced from bagasse only (gas produced from sludge blanks (i.e. S1-b to S4-b) have been subtracted)

5.3.2 Production of Volatile Fatty Acids (VFAs)

Short-chain fatty acids are key intermediate products formed during the anaerobic digestion process and they can indicate process stability, overload or inhibition. Total VFA concentrations in relationship to pH are shown for all the rice straw digesters in Figure 5-2. Typical growth curves were observed for VFA production and peak values were reached at different times depending on the microbial activity within the digester. An accumulation of VFAs and low gas production was observed in C2 (maximum total VFA concentration of 9600 mg/L and minimum pH of 5.95 on Day 28), C3 (maximum total VFA concentration of 7500 mg/L and minimum pH of 4.78 on Day 7), and D1 (maximum total VFA concentration of 16,000 mg/L and minimum pH of 6.15 on Day 14).

The VFA concentrations in the digesters with high gas production were significantly lower, and an inverse relationship between VFA accumulation and paper mill sludge was observed (i.e. the digesters with more paper mill sludge had lower VFA concentrations). In D2, the total VFA concentration never exceeded 70 mg/L after Day 0 and the pH remained fairly constant around 6.65 throughout the digestion period. In D3, VFA production peaked

on Day 7 with a total VFA concentration of 2300 mg/L, while pH remained relatively constant around 6.95. In D4, VFA production peaked on Day 28 with a total VFA concentration of 5900 mg/L and a corresponding drop in pH from 6.94 to 6.72 was observed.

The concentrations of individual organic acids at different stages of the digestion process are shown in Figure 5-3 for each rice straw digester. Acetic acid is always the dominant VFA, however, all short-chain fatty acids are present at some stage in all the digesters except D2. The most even distribution of acetic acid, propionic acid and butyric acid is observed in D4 on Days 3 and 7. Butyric acid also occurs in high concentrations during peak VFA production. In D1, butyric acid comprises about 30% of the total VFA concentration on Days 7 and 14 with corresponding butyric acid concentrations of 3387 mg/L and 4976 mg/L, respectively. In C3, butyric acid comprises 37% of the total VFA production on Day 7, with a corresponding butyric acid concentration of 2749 mg/L.

The VFA production in C2 and C3 is essentially the same during the first 3 days, consisting of similar concentrations of both acetic and butyric acid. On Day 7, however, the total VFA concentrations are comparable (6500 mg/L in C2 and 7500 mg/L in C3), but the proportion of butyric acid is much higher in C3. Following Day 7, the VFA production continues to rise in C2 until Day 56, but it decreases in C3 and is diminished by Day 28.

5.3.3 Total Ammonia Nitrogen and Alkalinity

The TAN trends for each rice straw digester are shown in Figure 5-4. The digesters containing primarily pig wastewater including D1 and D4 start out with the highest values of TAN (1517 and 1118 mg/L, respectively), and it is mostly consumed by the end of the digestion period. The digesters containing only paper mill sludge including D2 and C2 (autoclaved sludge) start with low concentrations (44 and 49 mg/L, respectively) and ultimately produce TAN over the course of the digestion period. The TAN concentration in D3 remains fairly stable, with an initial concentration of 728 mg/L and a final concentration of 880 mg/L. The TAN concentration in C3 (straw only) starts at 11 mg/L and increases slightly to 21 mg/L and then is completely consumed by Day 28. D1, D3, and D4 have relatively high TAN concentrations on Day 0 then an immediate, sharp decrease by Day 3 and rebound by Day 7.

The alkalinity trends for each rice straw digester are shown in Figure 5-5. For all digesters, except D4, there is a net production of alkalinity as CaCO_3 . The highest production of alkalinity was observed in D3 with a starting concentration (Day 0) of 343 mg/L as CaCO_3 , a maximum concentration of 1069 mg/L on Day 56, and a final concentration of 919 mg/L on Day 92. The lowest concentrations are observed in C3 with a starting concentration (Day 0) of 46 mg/L as CaCO_3 , decreasing to 0 mg/L on Day 7, and recovering with a final concentration of 266 mg/L on Day 92. With the exception of C3, a sharp increase in alkalinity is observed in the first three days, during the highest gas production period.

5.3.4 Trace Metals

The concentrations of trace metals including Fe, Co, Ni, Cu, Zn and Mn were analyzed during the digestion process and the reported concentrations ($\mu\text{g/gTS}$) for each metal are shown over time in Figure 5-6. The Fe, Co, and Ni are clearly supplied by the paper mill sludge based on the incremental decrease in concentration comparing D2 to D3 to D4. Zn is primarily supplied by the pig wastewater (based on the reverse trend), and both contribute to the presence of Cu and Mn. There was no addition of trace metals at any stage in the treatment process for the pulp and paper mill waste, nor at the start of the digestion experiments. Since trace metals were not added, the observed concentrations are assumed to be naturally-occurring in the pulp and paper mill waste and the pig wastewater.

The trace metals that potentially stimulated the anaerobic digestion process were Fe, Co, and Ni, since D2 had the highest concentrations of these metals compared to the other experimental digesters. The average Fe concentrations measured in D2, D3, D4 and D1 through Day 56 were 4167 $\mu\text{g/gTS}$, 2581 $\mu\text{g/gTS}$, 1588 $\mu\text{g/gTS}$ and 305 $\mu\text{g/gTS}$, respectively. The same trend was observed in D2, D3, D4, and D1 for both the Co concentrations (3.77 $\mu\text{g/gTS}$, 2.26 $\mu\text{g/gTS}$, 1.39 $\mu\text{g/gTS}$ and below the detection limit for D1) and Ni concentrations (4.84 $\mu\text{g/gTS}$, 3.06 $\mu\text{g/gTS}$, 2.27 $\mu\text{g/gTS}$, and 0.75 $\mu\text{g/gTS}$).

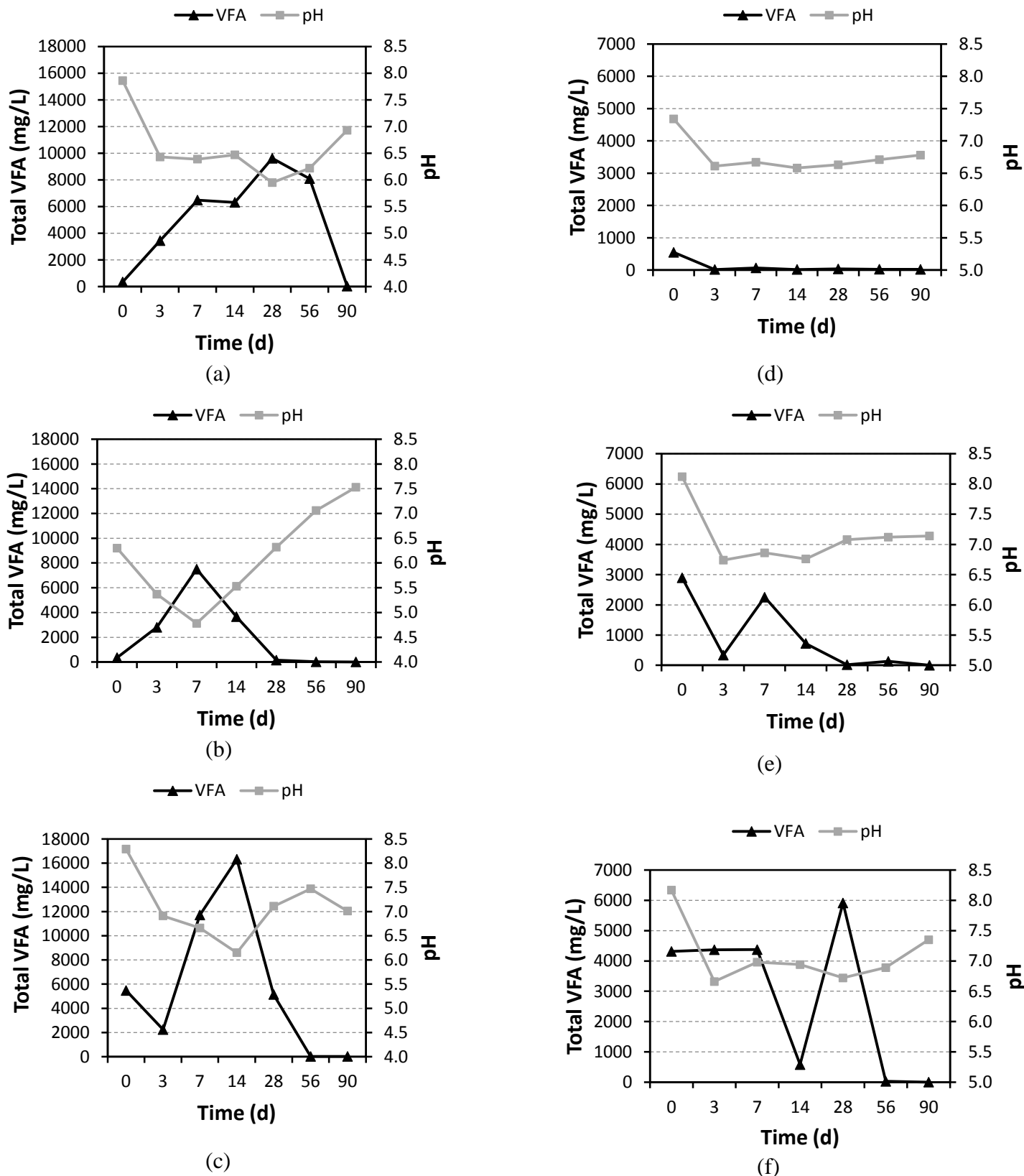


Figure 5-2. Total VFA Concentration and pH as a Function of Time for (a) C2, (b) C3, (c) D1, (d) D2, (e) D3, and (f) D4

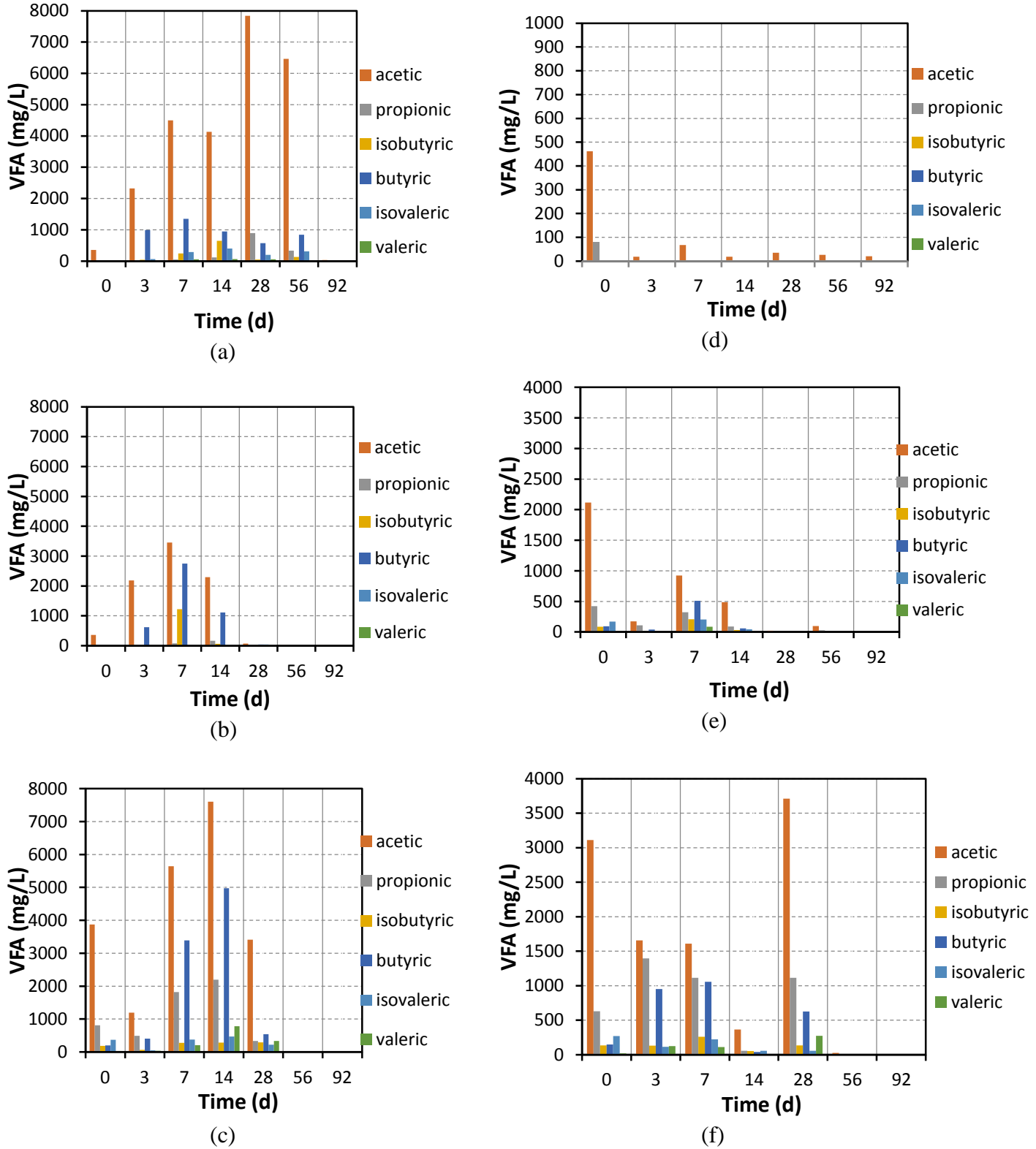


Figure 5-3. Distribution of Short-Chain VFA Concentrations as a Function of Time for (a) C2, (b) C3, (c) D1, (d) D2, (e) D3, and (f) D4

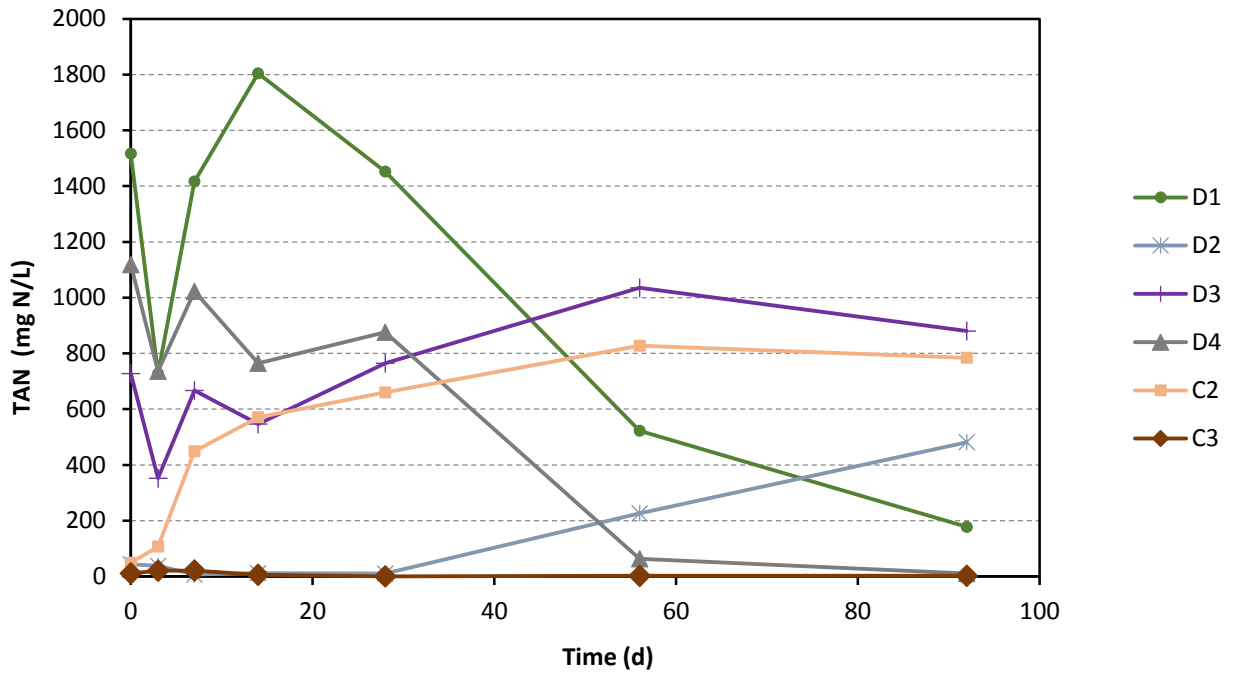


Figure 5-4. TAN ($\text{NH}_3 + \text{NH}_4^+\text{-N}$) Concentrations as a Function of Time for Rice Straw Digesters

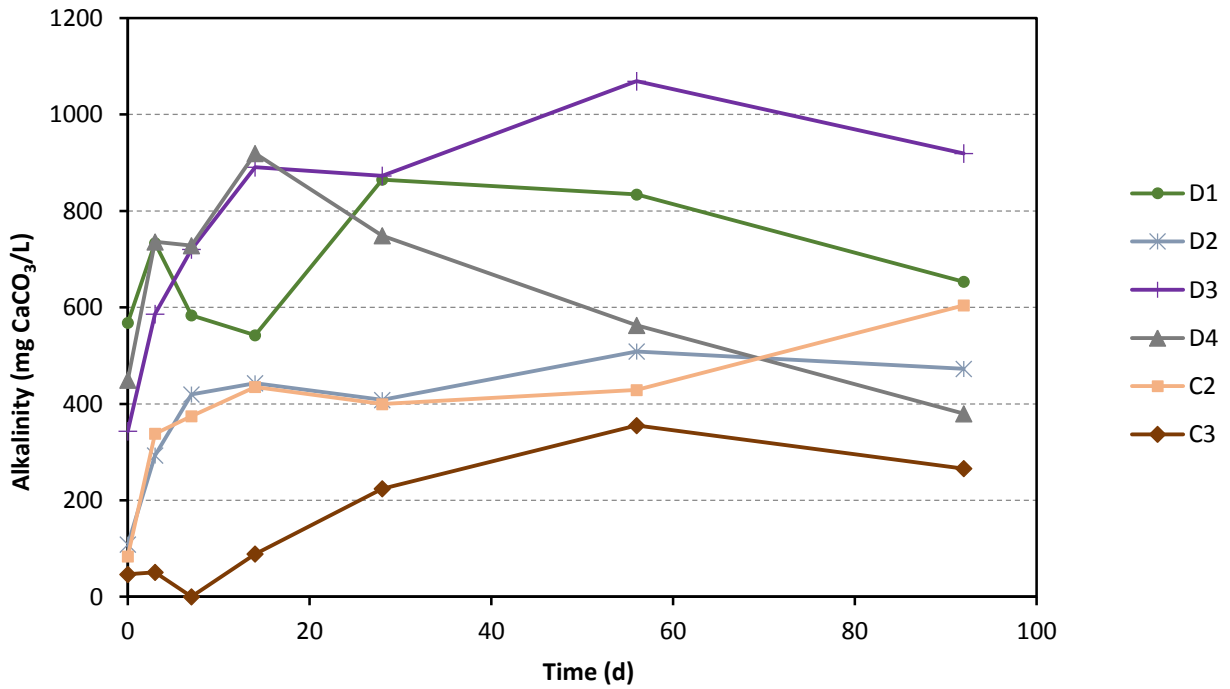


Figure 5-5. Alkalinity Concentrations as a Function of Time for Rice Straw Digesters

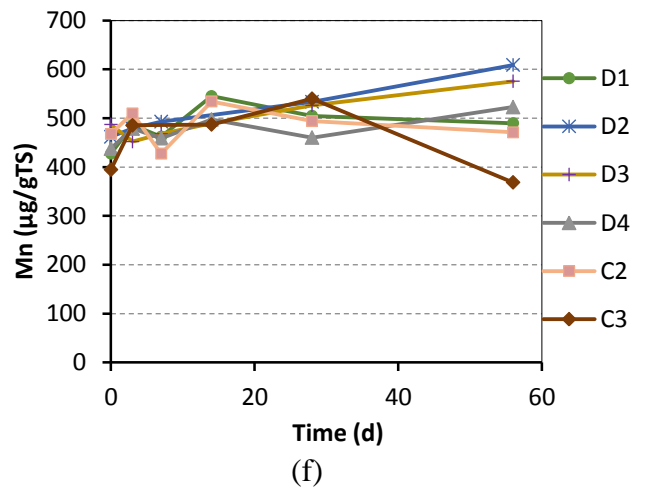
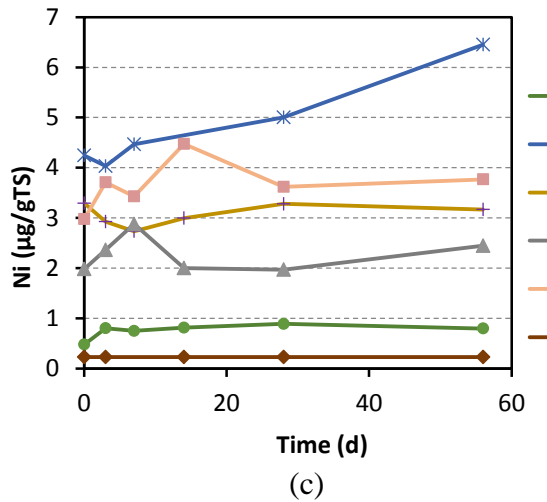
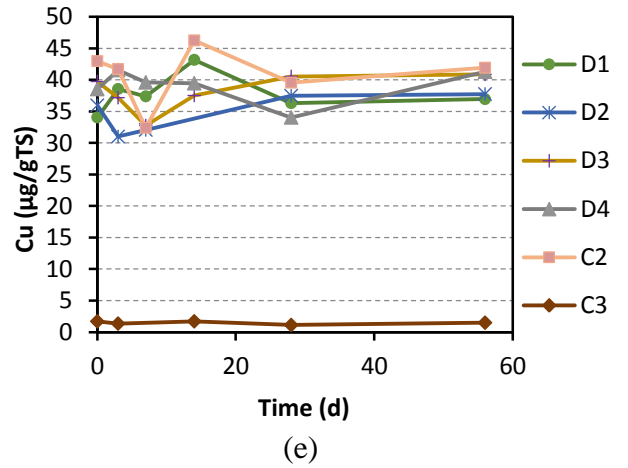
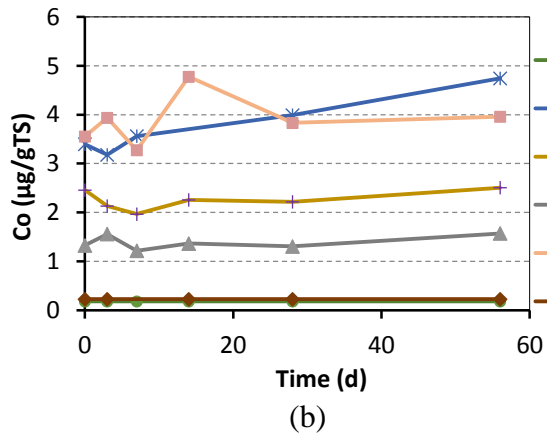
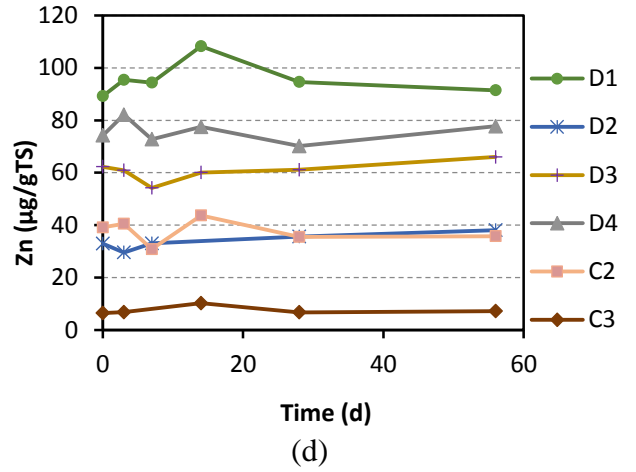
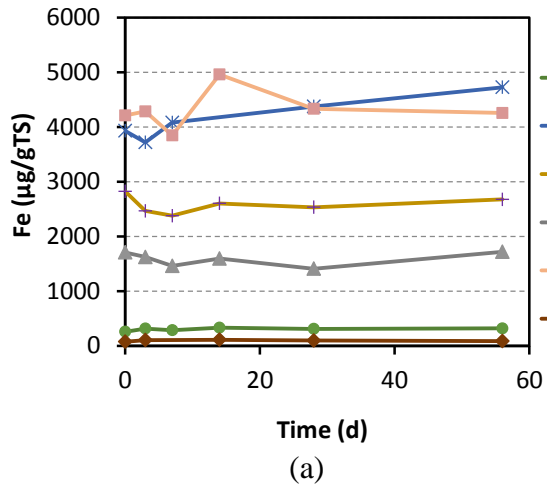


Figure 5-6. Trace Metal Concentrations in All Digesters as a Function of Time for (a) Fe, (b) Co, (c) Ni, (d) Zn, (e) Cu, and (f) Mn

5.4 Discussion

5.4.1 Advantages of Co-digestion over Pretreatment

The theoretical methane yield for rice straw calculated from the stoichiometric formula for rice straw based on TS% ($C_{0.028}H_{0.047}O_{0.029}N_{0.0005}$) is 330 LCH₄/kgVS [1]. However, experimental methane yields for rice straw are often much lower than the theoretical yield due to the difficulty in degrading the tightly-bound lignocellulosic material. Previous studies have shown that anaerobic digestion of untreated rice straw in mesophilic conditions can produce specific methane yields ranging from 195 to 231 LCH₄/kgVS added [1, 9, 13, 14, 16]. Biological, chemical, or thermal pretreatment of the straw is often used to disassociate the polymers and expose the soluble components to improve methane yields. Pretreated and delignified rice straw with white and brown rot fungi resulted in methane yields of 328 and 296 LCH₄/kgTS, respectively (yields are only provided in terms of dry biomass and not VS) [11]. Thermal pretreatment of rice straw at 110°C and 120°C resulted in methane yields of 245 and 261 LCH₄/kgVS, respectively, which were both higher than the untreated controls [16, 18]. Chemical pretreatment of rice straw using a 0.75mol/L acetic-propionic acid solution resulted in a methane yield of 280 LCH₄/kgVS [17].

The higher methane yields observed with the addition of paper mill sludge (i.e. > 300 LCH₄/kgVS added) are comparable and even exceed most of the yields obtained using rice straw that has undergone some type of pretreatment. Although recent studies suggest that lignocellulosic substrates cannot be sufficiently degraded without pretreatment [24-26], this research demonstrates that co-digestion of rice straw with both pig wastewater and paper mill sludge is highly effective and 100% of the theoretical methane yield (i.e. 330 LCH₄/kgVS) can be achieved with the appropriate ratios. This approach eliminates the need for pretreatment, which is often not feasible in the context of farm-scale or full-scale applications because of excessive energy requirements or large quantities of chemicals. Co-digestion is a more practical approach in large-scale systems because it can be easily implemented by simply adding the inocula to the existing leachate recirculation system and no additional infrastructure, chemicals or energy inputs are required. Furthermore, the TKN (Table 5-1(a)) and alkalinity supplied by the piggery wastewater stabilized the digesters as demonstrated in D1 when VFA peaked and pH dropped slightly but quickly recovered.

5.4.2 Evaluation of Intermediate Products

Lignocellulosic material such as rice straw is a poorly-degradable feedstock, and the formation of VFAs (i.e. hydrolysis) is considered to be the rate-limiting step in the anaerobic digestion process [12, 27]. More specifically, the disassociation of the hemicellulose from the lignin is where the bottleneck truly occurs [12]. Studies have shown that hydrolysis is catalyzed by lignin degradation through the combined efforts of extra-cellular enzymes such as cellulases, proteases, lipases [24, 28]. The accumulation of VFAs also presents a problem. VFA production influences pH and if the methanogen population is not well-established, then VFA accumulation can result in inhibition.

In the digesters containing the pig wastewater (*i.e.* D1, D3, and D4) the VFAs detected in the first 3 days are a result of the degradation of the simple soluble organic matter contained in the pig wastewater since concentrations of acetic acid are directly correlated with the quantity of pig wastewater added (Figure 5-3). On day 0, for example, the acetic acid concentrations in D1, D3, and D4 are approximately 4000, 2000, and 3000 mg/L, respectively; this is directly correlated to the quantity of pig wastewater added of 10.0 g, 5.0 g, and 7.5 g, respectively. Since D1 did not contain any paper mill sludge, there were no pre-established methanogens. Therefore, the primary reason for the failure of D1 was likely an accumulation of VFAs that inhibited growth of methanogenic organisms. Even though the pH did not drop below 6.15, total VFA concentrations exceeded 10,000 mg/L, which is considered toxic for methanogens [24]. Although the total VFA production was high, it is unlikely that any of the VFA production was a result of straw degradation since the methane yield was 0 (*i.e.* overall methane production in D1 was the same as in the sludge blank D1-b).

Of the individual VFAs, butyrate and isobutyrate are the best indicators for process instability and stress [29]. An increase in butyric acid was observed in the lowest gas producing digesters (D1 and C3) by Day 7. Although this increase was not the cause of the digesters' poor performances, it clearly and rapidly indicated the process instability in each of these digesters.

The digesters containing active paper mill sludge were much more capable of converting the straw material into methane, evidenced by significantly higher specific methane yields. The specific methane yields in D2, D3, and D4 are similar, but the VFA

trends demonstrate that the methanogenic population is better established in the digesters containing more paper mill sludge. D2 has the most established methanogenic population evidenced by the lack of VFA accumulation, while the microbes in D3 take more time to metabolize the VFAs (i.e. Day 28), and those in D4 even more time (i.e. Day 56). This trend simply demonstrates the advantage of using sludge from an established anaerobic reactor to decrease start-up periods.

A more important factor that can be observed is the accelerated degradation of straw material in the digesters containing higher proportions of paper mill sludge. D3 and D4 clearly show an initial decrease in VFAs from the degradation of the pig wastewater, followed by a peak in VFA production that represents the degradation of the straw material (see Figure 5-3(e) and (f)). The straw degradation in D3 has already started by Day 7, whereas the straw degradation is not initiated in D4 until Day 28. The precise mechanism for faster and more efficient degradation of the straw material with the addition of the paper mill sludge remains speculative, however, it is likely related to the activity of the established microbial population and associated enzymes in the sludge. It is a plausible theory that the paper mill sludge has acclimated over time to develop the appropriate microbes and associated enzymes capable of degrading lignin-containing materials. For example, anaerobic mesophilic bacteria known as *Clostridium cellulovorans* produce both cellulosome and non-cellulosome enzymes that work together in synergy to efficiently degrade the plant cell wall and they are capable of digesting most of the compounds in untreated rice straw in only 10 days [28]. This gram-positive, spore-forming bacteria was isolated from a wood chip pile [30] and thus is likely present in anaerobic sludge used in the treatment of pulp and paper mill waste.

VFA trends observed in C2 and C3 are also interesting. Since there is no active inocula in these digesters, the cellulolytic microorganisms inherent in the straw material is responsible for methane produced. Although the resulting methane yields are essentially the same, the hydrolytic activity is different. C3 is inhibited by acidic conditions on Day 7 (i.e. pH drop to 4.78) and VFA production ceases. The primary reason for the failure of C3 is a lack of buffering capacity, furthermore demonstrating that piggery wastewater can provide sufficient buffering capacity to stabilize the process. The hydrolytic activity in C2 continues through Day 56, and a significant increase in VFA production (i.e. 3300 mg/L)

occurs between Day 14 and Day 28. The increased VFA production in C2 (compared to C3) is still unknown. It may simply be a result of higher pH conditions provided by a slight buffering capacity in the inactivated sludge, or the degradation of the dead organic sludge material, or enzyme activity still present in the autoclaved sludge enhancing the hydrolysis of the straw material (which is less probable due to the inactivating effect of temperature on cellulases). The primary reason for the failure of C2 is a lack of active methanogens.

5.4.3 Nitrogen Supplication From Inocula

TAN and alkalinity both contribute to system stability via nutrient supplication and buffering capacity. Nitrogen, specifically in the form of ammonium (NH_4^+), is considered the most important nutrient for methanogenic bacteria and it is consumed during the anaerobic digestion of fatty acids. Ammonia (NH_3) is a by-product that is produced during anaerobic digestion and it reacts with carbon dioxide (CO_2) to form NH_4^+ and alkalinity. Thus, TAN is both consumed and produced during the anaerobic digestion process and this can be observed in TAN trends shown in Figure 5-4.

The TAN-producing digesters contain paper mill sludge as the primary inocula, while the TAN consuming digesters contain mostly pig wastewater. The microbial communities in each of the digesters utilize the available nitrogen in different ways. The digesters with paper mill sludge and rice straw are clearly deficient in TAN but have sufficient organic nitrogen based on the initial TKN concentrations of the sludge and straw. Thus, the process is carried out using primarily organic nitrogen and the net production of TAN is the result. The digesters with predominately pig wastewater contain a higher percentage of TAN. D3 represents the most stable system where a balance is maintained between TAN consumption and production. The production of TAN observed in C2 may be a result of the degradation of the organic nitrogen by the cellulolytic bacteria originating in the rice straw.

TAN concentrations less than 200 mg/L are considered beneficial, while concentrations above 1500 to 1700 mg/L have been shown to be inhibitory to anaerobic digestion [31]. TAN concentrations remained at or below the inhibitory threshold in all the digesters, except D1 on Day 14 when the TAN concentration peaked at 1804 mg/L. The TAN

concentration (relative to pH) did not appear to be inhibitory or rate-limiting in any of the digesters.

Alkalinity refers to the buffering capacity of a system and in the context of anaerobic digestion, it mitigates pH change during VFA production. Alkalinity is produced (when organic matter is destroyed and ammonia-N reacts with CO₂ to produce ammonium bicarbonate which contributes to alkalinity) and alkalinity is lost during the accumulation of VFAs in the anaerobic digestion process [32] as observed in the trends shown in Figure 5-5. Digester C3 lacks buffering capacity and thus the pH falls to 4.78 and gas production ceases. When comparing the experimental digesters (i.e. D1 to D4), D4, with an overall decline in alkalinity, showed a high pH variability, while D3 with the highest alkalinity of the digesters represents the most stable system in terms of pH.

5.4.4 Effects of Trace Elements

Several trace metals including Fe, Co, Ni, Mo, Se, and W have been shown to enhance methanogenesis. However, information about trace metals required for biogas digesters containing agricultural crops/residues and animal wastes is very limited [33]. In addition, optimum concentrations required for the anaerobic digestion of biomass is difficult to define because measured total concentrations do not represent their availability to the microorganisms. Two previous studies on the anaerobic digestion of agricultural waste residues demonstrate improved gas production with the addition of a trace metal solution. During the anaerobic digestion of maize silage, gas production was increased by 35% by adding a mixture of trace metals containing Fe (205 µg/gCOD), Co (11 µg/gCOD) and Ni (9 µg/gCOD) [34]. In the initial experiment, Co was the most limiting element, however, subsequent experiments demonstrated that the mixture of all three trace elements was necessary for the most efficient interaction between the different enzymes involved in the conversion process. During the anaerobic digestion of napier grass, methane production increased by 40% and VFA concentrations decreased to below detection limits after the addition of a trace metal solution containing Ni (0.25 mg/L), Co (0.19 mg/L), Mo (0.3 mg/L), and Se (0.062 mg/L) [35]. In the present study, higher concentrations of Fe, Co, and Ni were clearly present in D2, D3, and D4 (precisely in that order as shown in Figure 5-6 (a), 5-6 (b) and 5-6 (c)). The presence of Fe, Co and Ni in these digesters appeared to enhance the microbial activity since higher methane production and lower VFA

concentrations were observed. Though C2 had similar trace metal concentrations as D2, the organisms that metabolize VFAs were inactivated and therefore trace metal concentrations in this reactor were not contributory.

5.5 Conclusion

The addition of paper mill sludge to lignocellulosic wastes in dry, anaerobic conditions accelerated VFA formation and increased methane production. Specific methane yields for rice straw reached the theoretical value (*i.e.* 330 L_NCH₄/kgVS), and hydrolysis of the straw occurred faster in the digesters with higher fractions of sludge (immediately for D2, 7 days for D3, and 28 days for D4). The most stable conditions were observed with equal parts of straw, piggery wastewater, and paper mill sludge. The methane yields observed using the sugar mill bagasse were slightly lower than the yields from the rice straw with the same inocula mixtures. For farm-scale systems, this co-digestion approach can digest untreated lignocellulosic materials within three months, with less energy input than pretreatment and higher suitability for existing infrastructure.

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CHAPTER 6

Enhanced Methane Production from a Pilot-Scale Anaerobic Digester Loaded with Rice Straw

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6.1 Introduction

Energy production from lignocellulosic waste is advantageous because there is an abundant supply of agricultural waste residues available, it does not interfere with the provision of valuable food sources, and it offers a potential reduction of greenhouse gas emissions by removing these residues from the field and capturing the methane. Energy from rice straw can be produced from thermochemical processes such as pyrolysis, combustion or gasification [1-4]; however, these processes are energy intensive. Bioethanol production from rice straw via fermentation is also an option but this process is expensive and relatively low yields have been observed [5]. As in anaerobic digestion, the hydrolysis of cellulose is inhibited by lignin and pretreatment is required to enhance ethanol production [5, 6]. Biogas production from rice straw via anaerobic digestion is considered to be one of the most environmentally friendly processes for converting biomass into energy [7, 8].

The challenge associated with the utilization of lignocellulosic wastes for energy recovery is that the lignin acts as a barrier and can inhibit microbial populations that perform hydrolytic conversion of cellulose [9]. Several studies have investigated pretreatment strategies that enhance microbial degradation of lignocellulosic wastes in the context of anaerobic digestion [10-13]. The major goal of this research is to avoid design complications and energy inputs by removing the pretreatment step and use a novel co-digestion approach with waste products. Co-digestion with other wastes has been shown to enhance methane production from lignocellulosic wastes [14], and co-digestion is a simpler and more feasible approach for farm-scale applications. Co-digestion of rice straw with animal manure provides an appropriate balance of nutrients for anaerobic systems [15], and increased biogas yields from rice straw co-digested with both cattle manure and piggery wastewater have been demonstrated [16, 17]. An existing farm-scale system (15,000 m³) in Northern Italy converts rice straw into electricity using piggery wastewater alone to promote microbial fermentation in dry conditions (*i.e.* $\geq 20\%$ total solids (TS) concentration) [18]. However, a long acclimation period (200 days) and slow digestion cycle (422 days) was observed. A practical option for improvement is to add an acclimated microbial population to reduce the start-up cycle and improve methane yields.

Sludge generated in the pulp and paper mill industry likely contains microbial populations that are already acclimated to lignin-containing waste material. *Clostridium cellulovorans*, for example, originate in wood chips [19] and they produce enzymes that are

capable of degrading rice straw in 10 days [20]. To test this hypothesis, sludge was collected from an upflow anaerobic sludge blanket (UASB) reactor that is part of the initial stage of treatment for pulp and paper mill effluent. This effluent is generated from five different pulping facilities that employ different operational practices. An initial laboratory study demonstrated that co-digestion of rice straw with both piggery wastewater and UASB paper mill sludge could accelerate formation of volatile fatty acids (VFA) and produce higher methane yields (302 to 340 $L_NCH_4/kgVS$) than those generated with the piggery wastewater alone [21]. In the present work, a pilot-scale digester (1 m^3) was operated with the same straw-inocula mixture and digestion conditions tested in previous lab-scale experiments to determine if this co-digestion approach could improve methane production and reduce digestion cycles for farm-scale systems. The purpose of this work is to increase the scale of the laboratory experiments in order to simulate farm-scale conditions and determine if this co-digestion approach is an appropriate solution for large-scale applications. The premise of this work is unique because it proposes a co-digestion approach with not only piggery wastewater (tested previously in a pilot-scale system) but also with anaerobic sludge from the pulp and paper mill treatment process to enhance methane production from untreated rice straw.

6.2 Material and Methods

6.2.1 Experimental Set-up

A single pilot-scale digester (1 m^3) equipped with a leachate recirculation system was used for this experiment. The specific components and dimensions of the digester are described in a previous study [22]. The digester was operated as a batch reactor and initially filled with 50 kg of dry straw, 75 kg of piggery wastewater, and 25 kg of anaerobic sludge from the pulp and paper mill treatment process. This substrate to inocula ratio was determined to be the optimum ratio based on methane yields obtained in laboratory-scale digesters [21] and feasible application for a farm-scale system. The digester was operated in dry (20% TS), mesophilic conditions for a total of 153 days. Dry conditions are advantageous since they require significantly less water than wet conditions (*i.e.* $\leq 10\%$ TS) and the farm-scale system is currently operated in dry conditions [18]. Mesophilic temperatures were chosen since optimal gas production from rice straw is within 35 to 40°C [15], and much less energy input is required than thermophilic conditions. The entire volume of excess liquid, or leachate, was recirculated daily. The leachate

recirculation served as the primary means of mixing since there was no mechanical stirrer. On two occasions (Day 62 and Day 99) the digester was opened and the biomass was manually stirred. The digester was flushed with nitrogen gas after each mixing event to reestablish anaerobic conditions. The biogas production volume, biogas quality (*i.e.* % CH₄, %CO₂, and %O₂), and digester temperature was also measured and recorded daily.

The rice straw was harvested from a rice field in the Pavia Province of Italy approximately two weeks prior and stored in a dry location. No pretreatment, drying, cutting or milling activities were applied to the rice straw. The straw was collected directly from the field and added to the digester in lengths ranging from 0.2 to 0.6 m. Raw piggery wastewater was collected from a preliminary holding tank at a pig farm in the Pavia Province of Italy. Anaerobic granular sludge was collected from a treatment facility in Eerbeek, the Netherlands, which processes a combined wastewater from five different pulp and paper mill plants.

6.2.2 Analytical Methods

TS and volatile solid (VS) concentrations were measured on the rice straw, piggery wastewater and paper mill sludge prior to placement in the digester. Solids concentrations were also measured on the digestate at the end of the experiment. These analyses were conducted in triplicate and measured according to APHA Standard Methods 2540 [23]. The results of the solids concentrations are summarized in Table 6-1. Biochemical methane potential (BMP) assays were also conducted for the seed mixture (*i.e.* piggery wastewater and paper mill sludge) in mesophilic conditions to determine if the fraction of methane production coming from the wastewater was significant [24].

Leachate samples were collected daily and analyzed daily for pH, VFA, carbonate alkalinity (C_T), and total ammonia nitrogen (TAN). The leachate analyses were completed within two hours of the collection time. The pH was analyzed with a Hamilton Filltrode probe and was conducted in accordance with the APHA Standard Methods [23]. VFA and C_T were analyzed by a titration method using 0.1 M hydrochloric acid (HCl) and acidifying the sample to pH of 2.2, while continually recording pH and using a computer modulation to calculate the results [25]. The TAN concentration (NH₃-N and NH₄⁺-N) was analyzed using a spectrophotometer (SPT-500) with a Carlo Erba reagent kit (0800.05405). Free ammonia (NH₃-

N) was calculated from TAN using an equation from Anthonisen et al. that incorporates pH and temperature [26].

A permanent temperature probe was placed inside the digester and connected to a Gefran digital meter for temperature readings. Biogas volume was measured with an Elster volumetric flow meter ($Q_{\max} - 4\text{m}^3/\text{h}$, $Q_{\min} - 0.005\text{m}^3/\text{h}$) and the gas composition was measured with a Geotech biogas analyzer.

Table 6-1. Solids Concentrations of Raw Materials

	Rice Straw	Piggery Wastewater	Paper Mill Sludge
TS (%)	64.4	1.1	17.0
VS (%TS)	88.7	40.2	62.4

6.3 Results and Discussion

6.3.1 Gas Production

The specific biogas and methane yields were calculated as the volume of biogas and methane produced per kg of rice straw VS added, and any contribution of gas production from the inocula mixture was subtracted. The methane produced from the inocula mixture was determined during preliminary biochemical methane potential (BMP) assays containing both piggery wastewater and paper mill sludge. Based on these results, approximately 5% of the overall methane produced (*i.e.* 515 L out of 11,325 L) was from the inocula mixture. After subtracting any influence of gas production from the inocula mixture, the specific biogas and methane yields were calculated to be 561 and 252 L/kg VS straw added, respectively, for the 153-day digestion cycle. Cumulative yields are shown as a function of time in Figure 6-1.

The average methane content in the biogas was 51% upon digester stabilization through Day 62. The biogas quality exceeded 50% CH₄ by Day 19 and reached a maximum of 57% on Day 22. The biogas quality was compromised, however, each time the digester was opened for manual stirring (Days 63 and 99), resulting in decreasing CH₄ and increasing N₂. Slight concentrations of O₂ (0.2 to 3.0%) persisted in the digester for 10 days following the second mixing event. The biogas composition (%CH₄, %CO₂, %O₂, and %N₂) is shown as a function of time in Figure 6-2.

The daily biogas production trend shows that 45% of the biogas was produced within the first 35 days, with an initial peak of 660 L/day on Day 22 (see Figure 6-3). Biogas production gradually declined until Day 62. A second peak of 676 L/day occurred on Day 63, immediately following the first mixing event in which the digester was opened and manually stirred. The initial mixing event stimulated biogas production for 10 days (through Day 72) followed by a stable decline. The second mixing event on Day 99 had no stimulating effect on gas production, and the residual oxygen generated from opening the digester may have had a detrimental effect. Though the digestion cycle was carried out for 153 days for data collection purposes, the average daily gas production was less than 40 L/day for the last 50 days. Therefore, 90% of the overall biogas production and 92% of the overall methane production was completed by Day 93. For comparison purposes, a 93-day digestion cycle would have resulted in specific biogas and methane yields of 505 and 231 L/kg VS straw added, respectively.

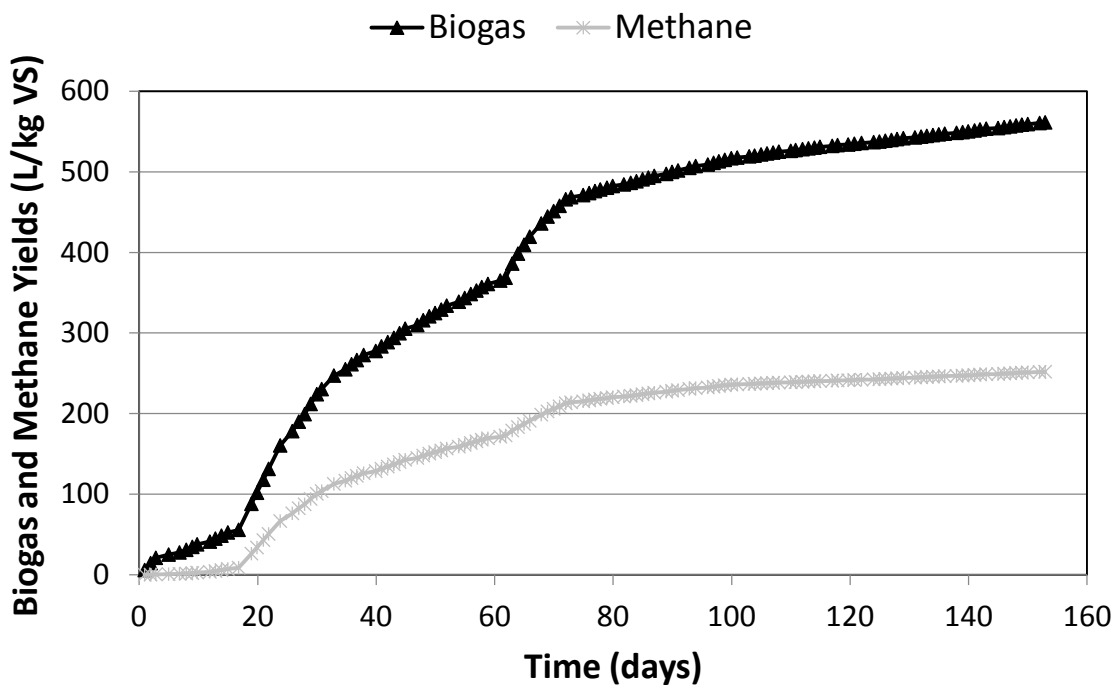


Figure 6-1. Specific Biogas and Methane Yields as a Function of Time

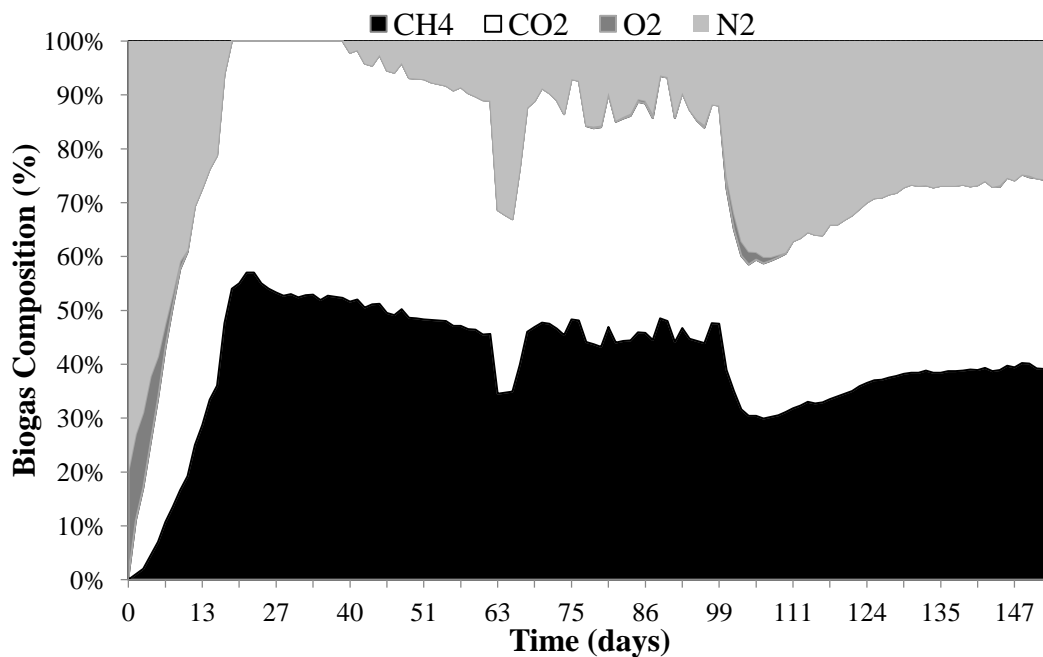


Figure 6-2. Biogas Composition as a Function of Time

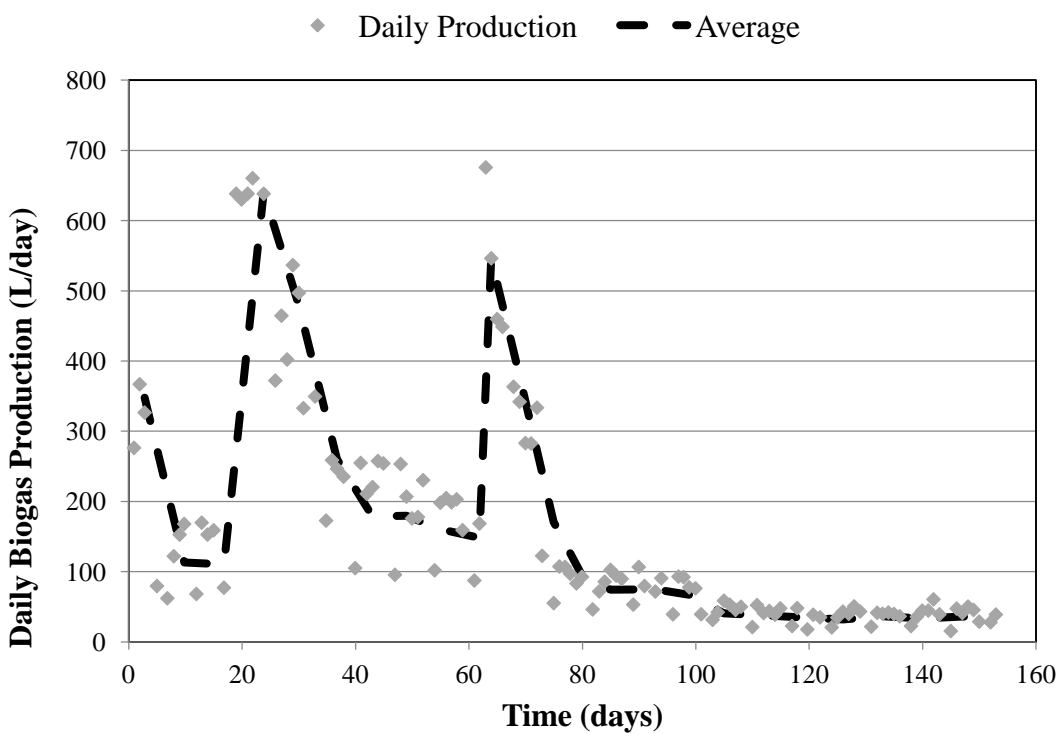


Figure 6-3. Daily Biogas Production

6.3.2 VFA Formation and System Stability

Leachate was recirculated at a rate of $0.2\text{m}^3/\text{m}^3$ straw-day and chemical analysis was performed daily on the leachate samples. The accumulation of VFAs was evident during the first 20 days of the experiment. An initial peak VFA concentration of 6178 mg HAc/L occurred on Day 8, and a second smaller peak of 3387 mg HAc/L was observed on Day 14. By Day 22, the formation of VFAs was moderate with concentrations ranging from 112 to 739 mg HAc/L and averaging 405 mg HAc/L for the remainder of the experiment. Although gas production significantly increased as a result of the mixing event on Day 63, there was no corresponding accumulation of VFAs indicating system stability and microbial acclimation.

The pH and alkalinity were impacted by the initial accumulation of VFAs, but the overall stability of the system was not compromised. During the initial VFA peak on Day 8, the lowest pH value (6.25) and alkalinity concentration (0 mg CaCO_3/L) were observed. The pH completely recovered by Day 12 and remained stable, ranging from 7.38 to 8.08, with an average of 7.78 for the duration of the experiment. The alkalinity also showed signs of recovery through Day 12, but a sudden decrease in alkalinity corresponded with the second VFA peak on Day 14. By Day 22, the alkalinity had completely recovered to 3698 mg CaCO_3/L and it remained above 2000 mg CaCO_3/L for the rest of the digestion cycle. Figure 6-4 shows the trend of VFA concentrations, alkalinity and pH values measured during the digestion cycle.

The ideal temperature range for the anaerobic digestion of rice straw is between 35 and 40°C [15]. Excluding the first 24 hours, the digester temperature ranged from 35.2 to 41.0°C with an average digester temperature of 37.2°C.

6.3.3 Ammonia Nitrogen Concentrations

The TAN ($\text{NH}_3 + \text{NH}_4^+\text{-N}$) and free ammonia (NH_3) trends are shown in Figure 6-5. Upon mixing the straw with the inocula, the leachate from the digester had an initial TAN concentration of 805 mgN/L. TAN was consumed during the peak gas production phase, resulting in a concentration of 613 mgN/L on Day 22. Following Day 22, TAN concentrations remained fairly stable between 525 and 626 mg N/L with an average concentration of 580 mgN/L. Although some variability was observed from day to day, a distinct oscillation (rise-fall-rise-fall) occurred during the 10 days following the initial mixing event (*i.e.* Day 63 to 72).

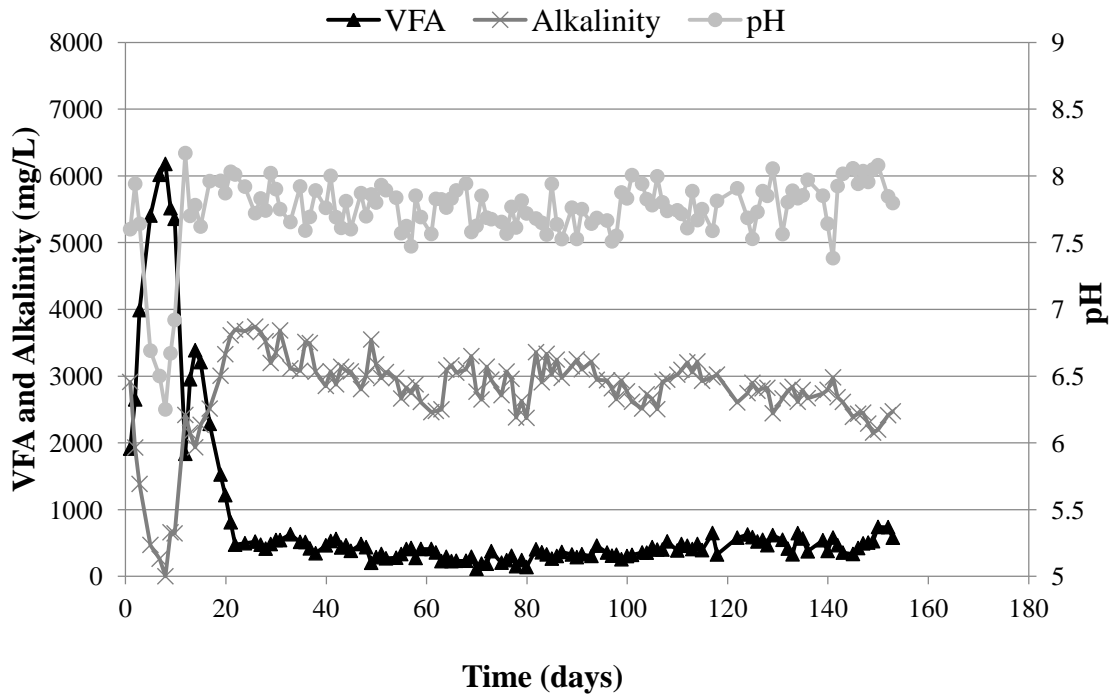


Figure 6-4. VFA Concentration (as mg HAc/L), Alkalinity (as mg CaCO₃/L) and pH as a Function of Time

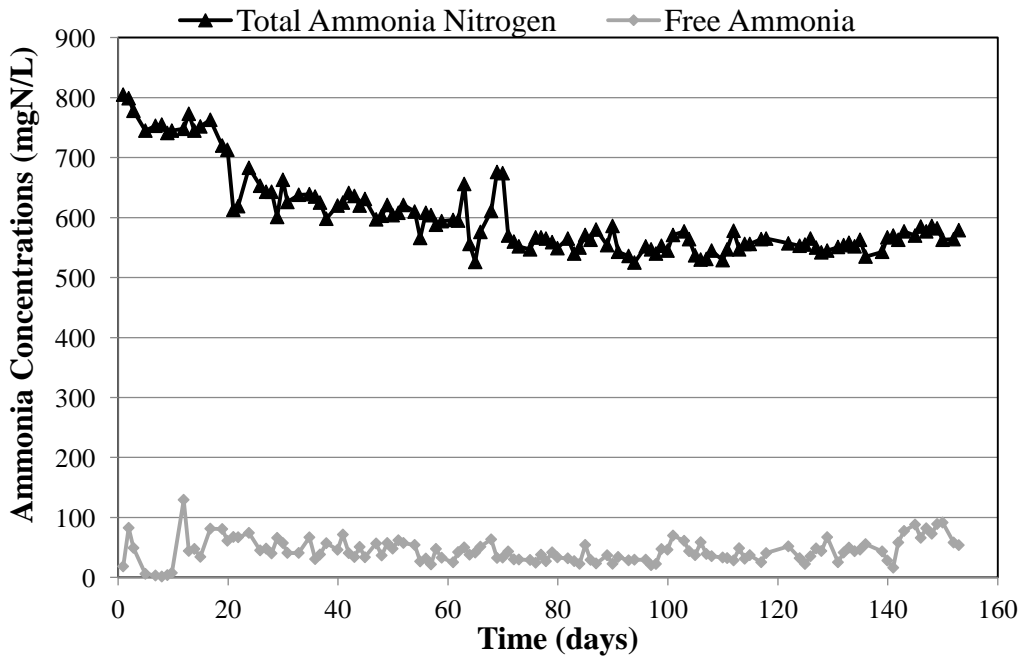


Figure 6-5. TAN and Free Ammonia as a Function of Time

The NH_3 concentration varied based on the slight changes in temperature and pH, but it remained below 100 mgN/L except on one occasion. The maximum NH_3 concentration (129 mgN/L) occurred on Day 12, which corresponded with the minimum pH value.

6.4 Discussion

The experiment was carried out for a total of 153 days resulting in a specific methane yield of 252 L/kgVS. However, after 93 days, over 90% of the methane production was complete and a specific methane yield of 231 L/kgVS was calculated. A comprehensive summary of methane yields obtained for rice straw are reported in the literature review (Chapter 2). For large-scale applications and economic feasibility, it is important to balance maximum energy outputs and minimum biomass retention times. In the current design, a 93-day digestion cycle would be most appropriate to achieve this balance.

The addition of the anaerobic sludge from the pulp and paper mill treatment process resulted in much faster digestion than without sludge. Another pilot-scale system with the same quantity of dry rice straw (50kg), twice as much piggery wastewater (150L) but no paper mill sludge, and the same operational design parameters (20% TS, mesophilic temperature, leachate recirculation, etc.) resulted in the same specific methane yield (231 L/kgVS) in 189 days [22]. The acclimation period for the digester without the sludge was much longer with a daily peak gas production on Day 112 [22] versus Day 22 with the sludge, resulting in an overall digestion time that was twice as long. Another major advantage of using the paper mill sludge is that less volume of inocula is required, which reduces costs for acquiring and transporting wastewater. In the current design, the substrate to inocula weight ratio (dry rice straw to piggery wastewater to paper mill sludge) is 1 to 1.25 to 0.5 while the other pilot-scale digester was 1 to 3 to 0 [22].

The current pilot-scale digester is an upscale of a previous lab-scale digester (1 L) with the same substrate to inocula ratio and operational parameters. The lab-scale digester (D4) had a specific methane yield of 302 L/kgVS in a 92-day digestion cycle [21], which is significantly higher than the results obtained in the pilot-scale digester. The primary reason for this difference is the lack of mixing capacity in the pilot-scale reactor. Lack of mixing typically results in less methane production and incomplete digestion since there is no uniform distribution of substrate, inocula and enzymes [27]. Internal mixing components for dry digestion systems require lots of energy and maintenance since the material is heavy and immobile. In the lab-scale digester, the

contents were stirred by hand to create a homogenous mixture prior to digestion since the volume was manageable and leachate recirculation was not possible. The pilot-scale digester was designed to emulate the farm-scale system in which leachate recirculation is currently the only mechanism used for mixing [18]. The leachate recirculation in the pilot-scale digester, however, did not provide adequate mixing and therefore gas production was hindered. This limitation is evident based on the peak in gas production observed after the digester was manually opened and mixed on Day 62 (see Figure 3) as well as the visual observation when the digester was opened. The black granules of the paper mill sludge were clustered on the top of the straw material, rather than being evenly distributed throughout the digester. Manual mixing to redistribute the sludge material resulted in an immediate increase in gas production in the following days. If the digester contents would have been adequately homogenized at the beginning of the experiment, complete digestion would likely have occurred within a 92-day digestion cycle as observed in the lab-scale digesters [21].

Typically, untreated lignocellulosic material is very difficult to degrade and thus hydrolysis of this material is considered the rate limiting step in the anaerobic digestion process [9, 28]. However, the production of VFAs in the current pilot-scale digester peaked very quickly within the first two weeks of the digestion cycle followed by peak gas production on Day 22. Two definitive VFA peaks were observed on Day 8 and Day 14, representing the hydrolysis of the piggery wastewater followed by the hydrolysis of the rice straw. Similar VFA trends were observed in the lab-scale digesters (specifically in D3 and D4), where the initial VFA concentration began to decrease rapidly followed by a second peak in production [21]. In the lab-scale digesters, the second peak was associated with the hydrolysis of the straw material, which occurred faster in the digesters containing a higher ratio of paper mill sludge [21]. Based on these observations as well as the increased gas production that followed, it is reasonable to assume that the straw degradation in the pilot-scale digester was occurring by Day 14 of the digestion cycle. Following VFA accumulation, concentrations settled around 400 mgHAc/L which is within the optimum range of 50 to 500 mgHAc/L for anaerobic digestion [27], signifying stable digester performance.

The VFA production and specific methane yield observed in this experiment with untreated rice straw in mesophilic conditions are very similar to the results obtained from a pilot-scale digester with pretreated rice straw in ambient conditions [29]. The VFA peak production

for the digester with pretreated rice straw occurred on Day 10 [29], while the VFA peak with untreated rice straw occurred on Day 14. A specific methane yield of 240 LCH₄/kgVS was achieved after 89 days for the pretreated straw, which is comparable to the results for the untreated straw co-digested with piggery wastewater and paper mill sludge (231 LCH₄/kgVS in 93 days).

While the paper mill sludge accelerated VFA formation and gas production, the presence and routine recirculation of the piggery wastewater provided sufficient buffer and nutrients to maintain system stability. The digester leachate was slightly acidic (6.25) during peak VFA production but quickly recovered within a couple days. The alkalinity supplied by the piggery wastewater was sufficient (approximately 3,000 mgCaCO₃/L) to prevent an extreme drop in pH, and the TAN concentrations were adequate but not inhibitory for the anaerobic digestion process [27, 30].

6.5 Conclusion

The addition of paper mill sludge with piggery wastewater in a pilot-scale digester of untreated rice straw operated in dry, mesophilic conditions accelerated VFA formation and gas production. The untreated rice straw with the sludge yielded 231 LCH₄/kg VS within a 93-day digestion cycle compared to 189 days without the sludge. Although the digestion cycle was initially carried out to 153 days, the 93-day digestion cycle was determined to be the optimum time period to balance the maximum energy output with the minimum retention time. Daily leachate recirculation (0.2m³/m³ straw-day) was not adequate for internal mixing and homogenization of the digester material, which is necessary to achieve maximum gas production within the shortest time period. This co-digestion approach is feasible for application to the farm-scale digester, as it would improve methane production, reduce the retention time of the straw, and reduce the quantity of piggery wastewater needed for the optimum digestion conditions.

Future studies should focus on improving the mixing capacity in the existing system as well as the potential for using continuous anaerobic reactor configurations for dry systems such as Dranco, Valorga or Kompogas. To better understand the microbial consortium responsible for the improved digestion with the paper mill sludge, microbiological evaluations should be

conducted on samples collected at the beginning and throughout the digestion process to identify the specific microorganisms present in the mixture.

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CHAPTER 7

Discussion and Conclusions

7.1 Introduction and Objectives

The energy crisis continues to strain global economic markets and world relations and alternative energy solutions must be sought. Europe has been making great strides toward its goal of a 20% renewable share of its energy consumption by 2020. Biomass only constitutes approximately 20% of the overall renewable energy production, while hydraulic power (46%) and wind power (27%) are more widely utilized [1]. A similar trend is present in the US, with biomass representing only 11% of the renewable energy share in 2011, compared to hydropower (62%) and wind power (23%) [2]. The major biomass sources utilized in the US for electricity generation are municipal solid waste that produces landfill gas (31%) and wood (65%), rather than other biomass sources such as energy crops and agricultural residues (4%) [2]. A case study in Northwest China, however, demonstrated that biomass (including crop residues, animal dung, biogas digesters and wood) represented the largest share (55%) of total household energy consumption [3]. Biogas digesters on the household scale (approximately 8 m³) are widely used in China, and crop straw has the lowest cost coefficient on the household scale when compared to other substrates such as firewood, animal dung, coal and electricity [3]. Cost coefficients were calculated by converting the biomass into standard coal equivalents for comparison purposes [3]. Rice straw is widely available as a biomass source in China. In 2012, global rice production was 718 million tons per year, with China (the largest contributor) representing 28.4% of the global production [4]. Optimizing the anaerobic digestion of rice straw for farm-scale or full-scale application will encourage global participation in biomass as a renewable energy source.

Another motivation for continuing research on anaerobic digestion of rice straw is to curb the agricultural impact on global warming. Greenhouse gas emissions are a global concern as they relate to climate change, and rice paddy fields contribute approximately 10 to 13% of the global anthropogenic methane emissions [5]. Methane emissions from rice fields are expected to double by the end of the century because projected increases in atmospheric CO₂ and warming temperatures will intensify greenhouse gas production from rice cultivation [6]. Based on a life cycle assessment, Blengini and Busto (2009) concluded that most (68%) of the global warming potential from rice cultivation is from field emissions [5]. It has also been shown that methane emissions from rice paddy soils can be reduced by 95% if rice straw is removed from the fields [7]. By removing the straw from the fields and digesting it anaerobically, methane is captured and used as a renewable energy source. The CO₂ produced during the anaerobic digestion of the

straw is not detrimental but has a neutral impact on climate change because it is taken up by the crops during the growth stage [5].

The major challenge associated with the anaerobic digestion of rice straw is the resistance of complex, ligno-cellulosic material to anaerobic degradation, because the lignin component limits hydrolysis and methane formation [8, 9]. Chemical, biological and thermal pretreatment strategies have been effective in overcoming this challenge in laboratory-scale experiments [10-15]. However, these approaches are not practical for farm-scale applications because of design constraints, increased energy inputs, excess chemical and water requirements, and waste disposal issues associated with the digestate. Therefore, the focus on co-digestion opposed to pretreatment provides a simple, energy-efficient option for farm-scale operations.

The overall goal of the research was to enhance methane production from the anaerobic digestion of untreated rice straw in dry conditions using a novel co-digestion approach. This research not only included laboratory studies, but also evaluated pilot- and farm-scale systems in dry conditions, which are lacking in the literature. Specific objectives were to: 1) monitor an existing farm-scale system for rice straw digestion; 2) implement pilot-scale digesters with varying temperature conditions and co-digestion approaches for optimization of the farm-scale plant; and 3) study a novel co-digestion strategy that utilizes both pig wastewater and sludge from the pulp and paper mill industry to enhance methane production. Combined results from laboratory, pilot- and farm-scale experiments are included in Figure 7-1.

7.2 Major Research Findings

In Chapter 2, the optimal digestion conditions for rice straw digestion including pH, temperature, moisture and nutrient ratios were defined in a review article [16]. Specific methane yields for rice straw range from 46 to 340 LCH₄/kg VS (Table 2-1) and are widely variable depending on digestion conditions. The overall methane yields and digestability of rice straw were essentially the same in wet systems compared to dry systems [17, 18]. The benefits of dry digestion include a more stable methane content in the biogas, water savings and higher solids content for disposal [17].

In Chapter 3, two initial pilot-scale (1 m³) digesters were designed to represent the optimal and existing conditions of a farm-scale biogas plant in northern Italy [19]. The purpose

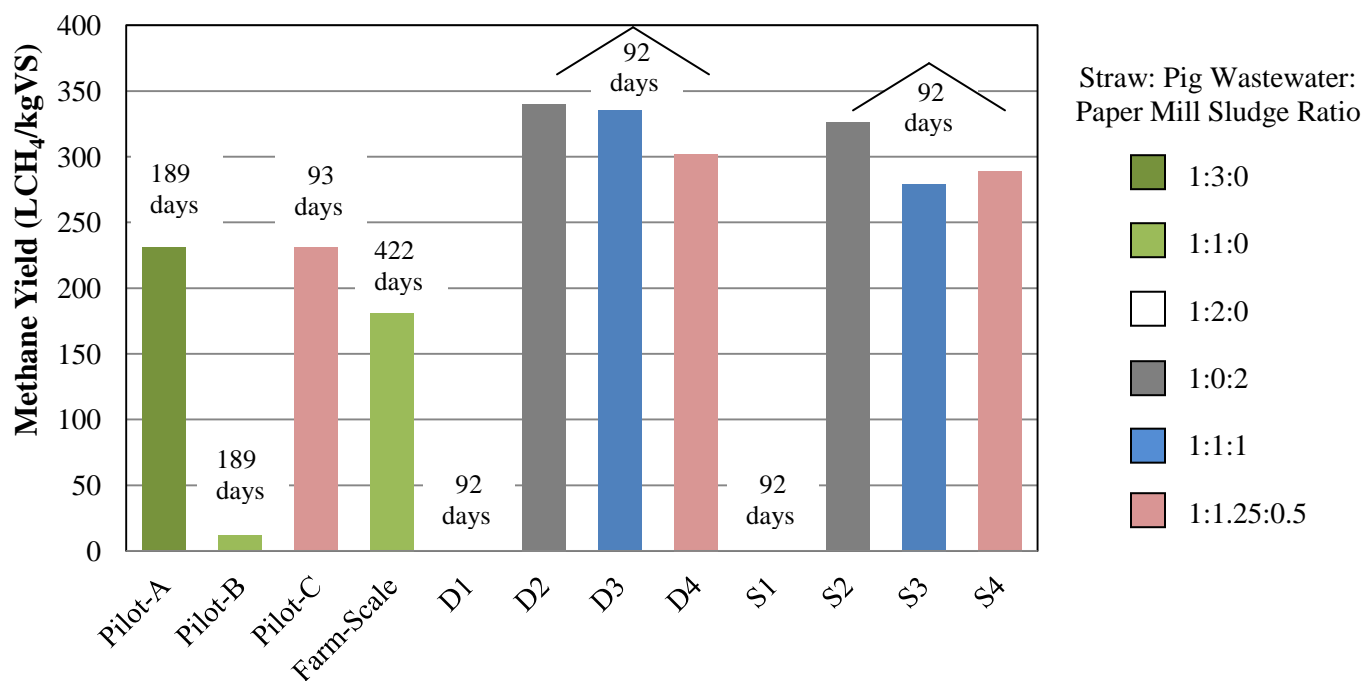


Figure 7-1. Methane Yields from Laboratory-, Pilot- and Farm-Scale Digesters

of the pilot-scale study was to ascertain the potential for improvement of energy recovery by maintaining mesophilic opposed to ambient conditions and by adjusting the substrate to inoculum ratio. The digester designed specifically to model the existing farm-scale biogas plant had essentially the same methane yield as the farm-scale plant on Day 189 (12.4 LCH₄/kgVS in the pilot-scale and 13.6 LCH₄/kgVS in the farm-scale). Although the quantity of pig wastewater in both digesters provided sufficient buffering capacity to maintain a stable pH and appropriate C:N ratio, specific methane yields were vastly different. After 189 days, the digester that was maintained at 35°C and had a substrate to inocula weight ratio of 1 to 3 (versus 1 to 1) produced a specific methane yield that was nearly 20 times higher (231 compared to 12.4 LCH₄/kgVS). The major conclusions of this study were that temperature was the major factor limiting methane production and that the conditions in the pilot-scale digester could be used to predict gas production in the farm-scale system.

Chapter 4 includes a description of a farm-scale batch digester loaded with rice straw and piggery wastewater that was used to produce methane that is converted to electrical energy through an internal combustion engine. Over a 422-day digestion cycle, approximately 700 tons of rice straw and 300 tons of pig wastewater were converted into 295 MWh of energy, with an

estimated specific methane yield of 181 LCH₄/kgVS [20]. This corresponds to a gross earning of €82,600, based on 2011 subsidy rates, or enough electricity to support 64 households in Europe based on an average electricity consumption value of 4000 kWh/yr [3]. A direct correlation between daily power production and digester temperature was observed, with a maximum power production of 2.74 MWh/d when mesophilic conditions were maintained. The major challenge was the slow start-up period of approximately 200 days, which was the result of a combination of factors including a high straw to wastewater ratio, low ambient temperatures (<15°C), low leachate recirculation rates (<0.04 m³/m³straw-d), inefficient heat exchange procedures, and a microbial community that was not acclimated to the feedstock. The introduction of an acclimated microbial community equipped for the rapid degradation of lignocellulosic materials could achieve a more sustainable and profitable system.

In Chapter 5, a novel co-digestion strategy using a mixture of pig wastewater and anaerobic sludge from a mesophilic pulp and paper mill upflow anaerobic sludge blanket (UASB) digestion process was evaluated with both rice straw and sugar mill bagasse in several laboratory digesters [21]. The objective was to increase the methane yield and decrease the retention time by accelerating the hydrolysis of the lignocellulosic material. The experiments were conducted at 35°C for a total of 92 days. Specific methane yields were higher than those attained with several pretreatment approaches (see Table 2-3) and they were comparable to the theoretical value of methane production from rice straw (*i.e.* 330 LCH₄/kgVS) [22], indicating that complete degradation and conversion was accomplished. In addition, the hydrolysis of the straw occurred faster in the digesters with higher fractions of sludge (immediately for D2 with 10.0 g of sludge, 7 days for D3 with 5.0 g of sludge, and 28 days for D4 with 2.5 g of sludge). The most stable conditions in terms of pH, alkalinity and nutrients were observed in D3, which contained equal parts of straw, piggery wastewater, and paper mill UASB sludge. A specific methane yield of 335 LCH₄/kgVS was measured in D3. Based on the farm-scale conversion efficiencies observed at the plant in Northern Italy, a total of 566 MWh could be generated in a 92-day digestion cycle, which is enough electricity to support 561 households in Europe based on an average electricity consumption value of 4000 kWh/yr [3]. Similar methane yields (see Figure 7-2) and gas production trends were observed with the sugar mill bagasse. The overall conclusion from these laboratory experiments is that the microbial community and nutrients in the anaerobic sludge obtained from the pulp and paper mill UASB treatment process are capable

of overcoming the lignocellulosic challenge and accelerating the hydrolysis stage of the anaerobic digestion process of both rice straw and sugar cane bagasse.

Chapter 6 describes a pilot-scale digester loaded with rice straw and co-digested with pig wastewater and paper mill sludge that was operated under mesophilic conditions to determine whether this approach could be feasible for the farm-scale plant. The pilot-scale experiment confirmed that the addition of paper mill UASB sludge accelerated VFA formation and gas production compared to the previous pilot-scale digester operated in mesophilic conditions without sludge addition. The same specific methane yield (231 LCH₄/kg VS) was obtained within a 93-day digestion cycle in the pilot-scale digester containing the sludge compared with 189 days without the sludge. Based on existing farm-scale conversion efficiencies [20], this methane yield would result in 385 MWh, which is enough electricity to support 378 households in Europe [3]. Significantly less inoculum was required (i.e. half the volume of pig wastewater), potentially reducing transportation costs associated with pig wastewater. The daily leachate recirculation rate of 0.2m³/m³ straw per day was not adequate for internal mixing and homogenization of the digester material, which is necessary to achieve maximum gas production within the shortest time period. Therefore, the specific methane yield obtained in the pilot-scale digester after 93 days was less than that obtained in the laboratory-scale digester using the same substrate/inocula mixture (231 and 302 LCH₄/kgVS, respectively), which was homogenized at the beginning of the experiment.

From a farm-scale perspective, the co-digestion of rice straw with pig wastewater and paper mill UASB sludge could have the potential to reduce the retention time to 93 days (versus 422 days) with no changes to the existing infrastructure. Additional costs will be incurred for obtaining and transporting the paper mill sludge to the site for start-up. Further research will be necessary to determine if the microbial community established during the initial digestion cycle can be maintained and stabilized over several digestion cycles. To maximize the efficiency of this approach, an external heat source is needed during start-up as well as an effective heat exchange system to establish and maintain mesophilic conditions since average daily ambient temperatures are below 20°C for nearly half the year in Northern Italy.

The optimization of the anaerobic digestion process, however, is only one aspect of the overall scheme for improved gas production in the farm-scale system. Technical unforeseen challenges were incurred during the initial digestion cycle of the farm-scale plant that must be

addressed for optimum performance. One of the major malfunctions occurred with the heat exchanger in an attempt to transfer the waste heat generated by the engine to the digester via leachate recirculation. The tubular heat exchanger had small diameter tubes that were frequently clogged, despite the preceding filters, and monthly maintenance was necessary to adequately clean the system. Larger diameter tubes could be used to prevent fouling and provide easier cleaning procedures. The leachate distribution system also needs improvement to better disperse the leachate over the straw, since it is a carrier for both heat and nutrients. Higher leachate flows could be accommodated with a more efficient heat exchange process, which would disperse more leachate over the straw and reduce clogging in the tubes.

The central location of the plant within the farm fields also presented challenges. Although it reduced transportation costs, the varmints that inhabited the rice fields were destructive. Nutria interfered with daily operations by chewing through the plastic cover and damaging the leachate distribution pipes. Small field mice interrupted the engine operation by comprising the electrical components of the system, which were adequately enclosed in an elevated, metal housing structure. The top liner deteriorated quickly due to abrasion with the straw material and reparations are costly and labor intensive. Finally, the design of the anaerobic digester cells lined with earthen berms created difficulty with the loading and unloading of the straw bales. This process was labor-intensive and an excavator was required to remove a portion of the berm to load and unload the straw. Therefore, a more secure and functional design of the digester cells is necessary to improve the overall gas production in the system.

7.3 Recommendations for Future Research

7.3.1 Microbiological Evaluations

The addition of anaerobic sludge from the pulp and paper mill treatment process accelerated VFA formation and enhanced methane production in both the lab and pilot-scale digesters. However, the precise mechanism for faster and more efficient degradation of the straw material remains speculative. The hypothesis is that the microbes contained in the paper mill UASB sludge have evolved and acclimated over time to produce enzymes capable of separating the lignin barrier from the cellulose. For example, anaerobic mesophilic bacteria known as *Clostridium cellulovorans*, contained in wood chips [23], produce both cellulosome and non-cellulosome enzymes that work together in synergy to efficiently degrade the plant cell

wall and they are capable of digesting most of the compounds in untreated rice straw in only 10 days [24]. Another closely related microorganism is *Clostridium cellulolyticum* which is often found in rotting grass [24]. A recent study demonstrated that the bioaugmentation with *C. cellulolyticum* can improve the hydrolysis of lignocellulose and result in more efficient digestion and higher methane yields [25]. A methane yield of 326 LCH₄/kgVS was obtained from wheat straw that was bioaugmented with *C. cellulolyticum* [25], which is comparable to the methane yields obtained from digestion of rice straw with the addition of the UASB paper mill sludge (302, 335 and 340 LCH₄/kgVS). To better understand the microbial consortium responsible for the improved digestion with the paper mill sludge, microbiological evaluations should be conducted on samples collected at the beginning and throughout the digestion process to identify the specific microorganisms present in the mixture.

Methodologies that could identify and quantify the specific microorganisms present in the digesters include denaturing gradient gel electrophoresis (DGGE), fluorescence in-situ hybridization (FISH), quantitative polymerase chain reaction (qPCR) or pyrosequencing. Complex anaerobic communities within an anaerobic reactor treating mine drainage were identified and quantified using qPCR [26]. The reactor fed with lignocellulosic material (wood chips, limestone, and corn stover) contained more cellulose-degraders including *Clostridium cellulovorans*, while the reactor fed with ethanol contained 1.5 times more sulfate-reducing bacteria [26]. The primers developed in this study can be used to quantify key functional groups within complex anaerobic communities such as the anaerobic mixture of piggery wastewater and UASB paper mill sludge, while most of the reliable methods for quantification are only suitable for pure cultures [26].

7.3.2 Effect of Trace Metals

The importance of trace metals in the anaerobic digestion process is well documented [27-30]. However, literature reporting trace metal requirements for biogas digesters containing agricultural crops/residues and animal wastes is very limited [31]. In addition, optimum concentrations required for the anaerobic digestion of biomass is difficult to define because total concentrations measured in the biomass do not necessarily represent their availability to the microorganisms.

During the anaerobic digestion of maize silage, gas production was increased by 35% by adding a mixture of trace metals containing Fe (205 $\mu\text{g/gCOD}$), Co (11 $\mu\text{g/gCOD}$) and Ni (9 $\mu\text{g/gCOD}$) [32]. During the anaerobic digestion of napier grass, methane production increased by 40% and VFA concentrations decreased to below detection limits after the addition of a trace metal solution containing Ni (0.25 mg/L), Co (0.19 mg/L), Mo (0.3 mg/L), and Se (0.062 mg/L) [29]. Trace metals originating in the paper mill sludge, specifically Fe, Co and Ni, were present in the lab-scale digesters as discussed in Chapter 5. The higher methane production and lower VFA concentrations observed in these digesters may have been a direct effect of the presence of one or all of these trace metals.

First of all, it is important to determine whether or not the trace metal concentrations contained in the UASB paper mill sludge contributed to the improved methane production. Secondly, it is also important to define precisely which metal or combination of metals contributed to the increased gas production. Finally, optimum concentrations of specific trace metals in the context of dry, anaerobic digestion of lignocellulosic material are not clearly defined in the literature. Therefore, further laboratory studies using untreated rice straw and piggery wastewater with the same concentrations of Fe, Co and Ni contained in the UASB paper mill sludge should be conducted to determine if the enhanced effect is primarily caused by the microbiological inoculation or the presence of trace metals. In addition, a suite of trace metals including Fe, Ni, Co, Mo and Se should be added to dry, anaerobic digesters with rice straw to determine which individual or combination of metals contribute to optimum gas production. Varying concentrations should be used in several series of experiments to clearly define optimum trace metal concentrations. Knowing the optimum trace metal concentration and combination could enhance the biogas production process.

7.3.3 Implications for Anaerobic Digestion Products

A diagram showing the potential pathways for the products and by-products of anaerobic digestion is included as Figure 7-3. The anaerobic digestion of rice straw results in the production of biogas that can be used directly, converted to electricity or upgraded for potentially a more efficient use of energy. Various biogas applications are discussed below in the context of the farm-scale biogas plant. An important by-product from the farm-scale biogas plant discussed in Chapter 4 is the residue, or digestate, remaining from the digestion process that cannot be

converted into methane during the applied residence times. The process described herein is essentially dry, so there is no excess wastewater generated for disposal. Liquid in the form of diluted piggery wastewater is added to the system and recirculated for distribution of nutrients and heat. However, the liquid is absorbed into the dry straw material (80% TS) during the digestion process and the resulting digestate consists of approximately 20% TS. This material contains sufficient moisture and nutrients to be applied as fertilizer on the fields, which is a common practice. Any residual liquid, which is very minimal, would be kept in the digester cell for the next batch of substrate and serve as inoculum, since it likely contains an acclimated microbial community to improve degradation of the straw material.

7.3.3.1 Biogas: The Final Product

Biogas consists of approximately 50% CH₄, which can be used directly, converted to electricity or upgraded to natural gas. In rural communities where infrastructure is lacking, unaltered biogas is commonly used for heating and cooking. In developed countries, government subsidies and economic incentives drive the biogas market toward conversion to electricity. The farm-scale plant described in Chapter 4 uses biogas as a fuel for an internal combustion engine to power a generator that produces electricity for the local grid. This approach is largely motivated by a return of 0.28 €/kWh. The drawbacks for this approach include a relatively low (i.e. 32%) electrical conversion efficiency as well as emissions control requirements for the combustion process. Microbial fuel cells (MFC) can convert biomass substrate directly to electricity through electron transfer, and they tend to have higher conversion efficiencies than anaerobic digestion and no heat input or off-gas treatment is necessary [33]. The feasibility for MFCs to capture energy from domestic and animal wastewaters has been demonstrated [34, 35], however the commercialization of the technology is still unseen [36].

Biogas can also be purified and upgraded to natural gas, which can then be used directly for heating homes or fueling vehicles. The purification process involves the removal of impurities such as CO₂ and H₂S using technologies such as water or polyethylene scrubbing, chemical absorption, pressure swing absorption, bio-filters, cryogenic separation, or membrane separation [37]. Purification technologies result in a high quality fuel with greater than 95% CH₄ content which burns much cleaner than typical fossil fuels. The major disadvantage with biogas upgrading for farm-scale systems is that existing technologies are expensive, ranging from €0.12/Nm³-biogas for membrane separation to €0.44/Nm³-biogas for cryogenic separation [37].

Another obstacle is the difficulty in transporting the upgraded biogas. A pilot-study was conducted to determine the feasibility for converting biogas from a farm-scale anaerobic digester fed with dairy waste in Lynden, Washington (USA) into compressed biomethane that could fuel the nearby airport shuttles [37]. However, the shuttles were unable to fuel directly at the biogas plant and the transport of the compressed biomethane was too complex and costly to carry out the project [37].

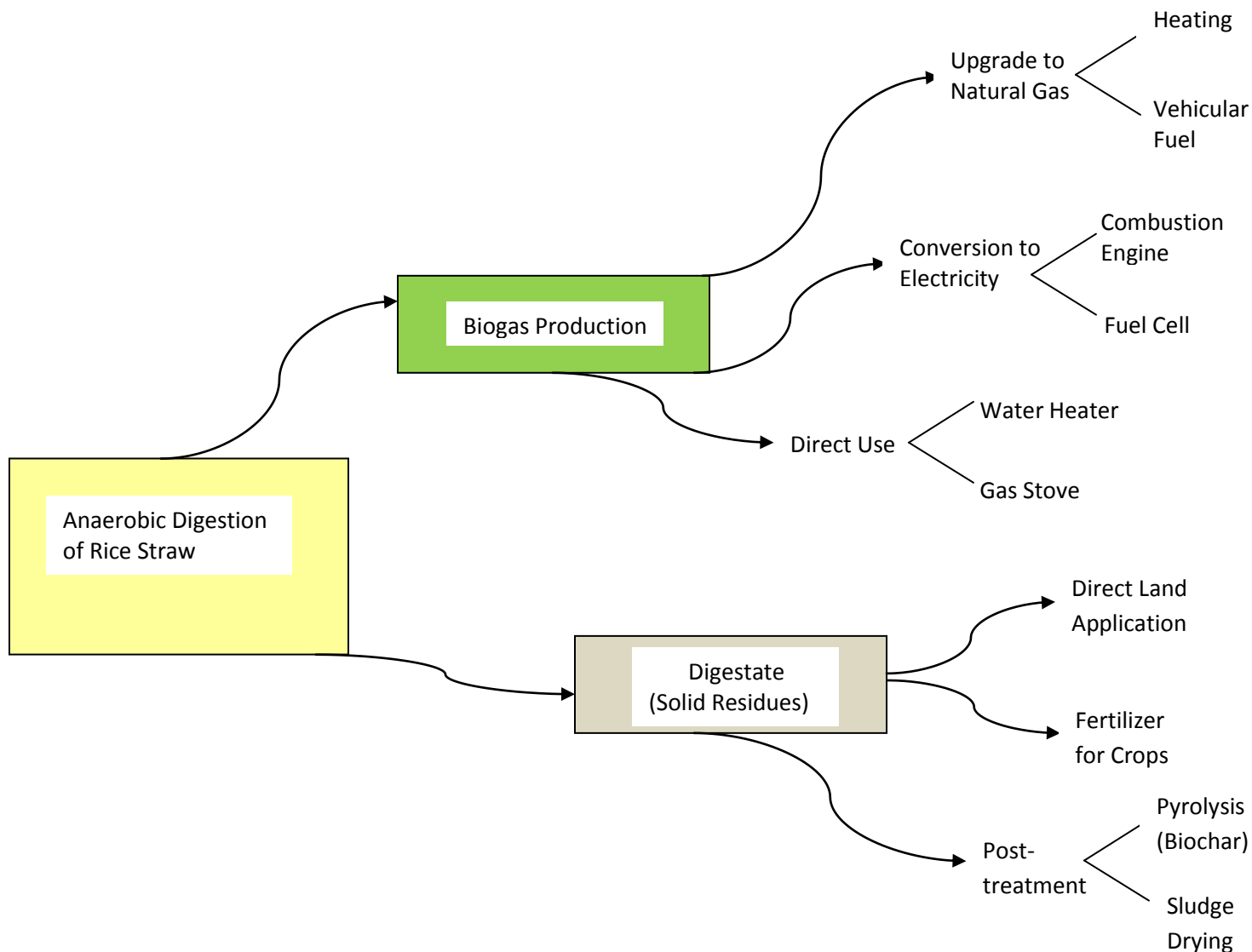


Figure 7-2 Products from the Dry Anaerobic Digestion Process

7.3.3.2 Digestate: The Byproduct

The digestate from anaerobic co-digestion of rice straw and pig wastewater is a nutrient-rich, moist but solid material (>20% TS in dry systems) that can be applied to the fields as organic fertilizer. Macro-nutrients including nitrogen (N), phosphorus (P), and potassium (K) and micronutrients including zinc (Zn), iron (Fe), manganese (Mn) and copper (Cu) are deficient in the rice paddy soils [38] and can be supplied by the digestate material. Application rates vary based on the type of soils, rice varieties, and expected yields; however, approximate doses for upland rice fields (per hectare) are 60 kgN, 40 kgP, 30 kgK, 30 kgZn, 20 kgFe, 20 kgMn and 20 kgCu [38]. Table 4-1 shows the nutrient concentrations for the digestate analyzed from the farm-scale plant after the initial 422-day digestion cycle, which indicates there are sufficient nutrients within the digestate material to serve as a biofertilizer.

Using the digestate on the surrounding rice fields is advantageous in terms of resource recycling and biofertilization; however, there are public health concerns that should be investigated further. The presence of pathogens such as *Salmonella* and helminthes in the digestate material is a concern. The pig wastewater may introduce pathogens that can survive and possibly thrive in an anaerobic environment that ranges from ambient to mesophilic temperatures. The presence of *Salmonella* is of particular concern since it is a common source of food-borne disease outbreaks. The presence of *Salmonella* was detected in 61% (33 out of 54) of the fecal samples collected from 5 different pig farms in Quebec [39] and 45% (27 out of 60) of the samples collected from 10 different pig farms in Mexico [40]. The question remains on whether or not the anaerobic digestion process operated in mesophilic temperatures can safely eliminate the presence of *Salmonella* and other harmful pathogens. Cow manure was co-digested with waste grease in mesophilic conditions for 35 days and there was only a 0.87 log reduction of *Salmonella*, resulting in a final concentration of 8.84×10^3 CFU/100 ml which is considered an infectious dose [41]. *Salmonella spp.* is known for long-term survival and is capable of adapting to various environmental conditions [42]. It is recommended that specific laboratory methods be conducted in the future to enumerate the *Salmonella* contained in both the raw wastewater and the digestate prior to adding it as fertilizer on food crops in the interest of public health.

Another consideration in the context of this research is that the addition of sludge from the pulp and paper mill UASB process could introduce contaminants that may not be appropriate

for fertilizer use on food crops. A variety of toxic pollutants including chlorinated compounds, resin acids, phenols, terpenes and acetone are associated with pulp and paper mill wastewater [43]. Toxic compounds discharged into the environment from pulp and paper mill effluents have had detrimental effects on fish including mutagenicity, liver damage, delayed sexual maturity and lethality [43, 44]. When paper mill effluent was used to irrigate rice paddy fields over a three-year period, adverse effects on the growth and development of rice was observed with high concentrations of the effluent [45]. However, when the effluent was diluted to 30%, the growth performance improved and rice production was higher than in the control soil (without the paper mill effluent) [45]. The waste streams from pulp and paper mill treatment processes are highly variable and studies regarding toxicity in the UASB sludge from the treatment process are lacking. Therefore, further analysis on the digestate from the mixture of straw, pig wastewater, and paper mill sludge should be conducted to determine if the contaminant levels meet the appropriate land application regulations for food crops.

If toxic compounds are discovered or public health concerns regarding pathogens are validated, other options for the digestate should be considered. One option would be to use the digestate as fertilizer on non-food related crops such as sod because the land disposal regulations are less stringent. If necessary, thermo-chemical treatment such as pyrolysis could be applied to the digestate to produce a nutrient-rich biochar. In pyrolysis, biomass is heated to approximately 500°C in the absence of oxygen [46] and the resulting biochar can be applied as a soil amendment. Biochar has been shown to increase water retention, improve soil fertility, improve nutrient retention and increase soil carbon content [47].

7.4 Conclusion

The novel co-digestion approach presented in this research has the potential to accentuate lignocellulosic waste as a viable source of energy recovery through anaerobic digestion without the need for pretreatment. Anaerobic sludge from the pulp and paper UASB treatment process contains the appropriate microorganisms and nutrients to accelerate the degradation of both rice straw and sugar mill bagasse so that hydrolysis is no longer the rate-limiting step in the digestion process. A farm-scale biogas plant in Northern Italy loaded with rice straw and piggery wastewater produced 295 MWh in a 422-day digestion cycle, which is enough electricity to support 64 households in Europe. However, with the addition of paper mill sludge and external

heat, the retention time could be reduced to 93 days, and the specific methane yield could increase from 181 to 231 LCH₄/kgVS based on pilot-scale results using a total weight ratio of dry straw to pig wastewater to UASB paper mill sludge of 1:1.25:0.5. This equates to an energy production of 385 MWh, which could support 378 households in Europe. This co-digestion approach with both pig wastewater and UASB paper mill sludge does not require any changes to the existing infrastructure or any type of pretreatment for the rice straw, but does require additional costs for transportation of the sludge material to the existing plant and external heating to maintain mesophilic conditions. If the infrastructure incorporated a more reliable strategy for homogenization and internal mixing, methane yields could increase to 335 LCH₄/kgVS in 92 days based on the laboratory-scale results with a 1:1:1 mixture. This mixture has the biomethane potential to produce 566 MWh, which could support a total of 561 households in Europe.

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