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RECONSTITUTION OF DEWATERED FOOD PROCESSING RESIDUALS WITH MANURE TO INCREASE ENERGY PRODUCTION FROM ANAEROBIC DIGESTION

presented by

David M. Wall

has been accepted towards fulfillment
of the requirements for the

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Steven I. Safferman
Major Professor's Signature

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RECONSTITUTION OF DEWATERED FOOD PROCESSING RESIDUALS WITH MANURE TO INCREASE ENERGY PRODUCTION FROM ANAEROBIC DIGESTION

By

David M. Wall

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Submitted to
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ABSTRACT

RECONSTITUTION OF DEWATERED FOOD PROCESSING RESIDUALS WITH MANURE TO INCREASE ENERGY PRODUCTION FROM ANAEROBIC DIGESTION

By

David M. Wall

A potentially effective wastewater treatment strategy for a food processor is to concentrate high carbon solid wastes by segregation and dewatering of bulk flow. This solid-liquid separation process results in a weaker wastewater fraction that can be economically disposed to sewer or potentially land applied with minimum surcharge, while remaining solids are collected and applied to landfill. An alternative to the landfill is to anaerobically digest the waste. This technology involves the degradation of organic matter in the absence of free oxygen. A byproduct is biogas that contains a substantial amount of methane which can be used to generate energy.

The following study evaluates the reconstitution of dewatered food processing waste with manure to gain increases in energy produced per unit volume in a farm digester, and thereby increase profitability for the farmer. Two batch digestion studies were conducted on different blends of food waste and manure to determine if a synergistic relationship existed in gas production. A further semi-continuous study provided a more realistic interpretation of real-life co-digestion. Although gas production appeared additive in the batch studies, the semi-continuous digestion of an optimized food waste and manure blend was found to produce over twice as much methane as manure digested alone in reactors of same working volume.

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Finally I would like to thank my parents, Michael and Aileen, and my sisters, Elaine and Emma, for all their love and support while I have been abroad. There is no doubt that without your encouragement and support I would not be in the position I am today.

Le chroí agus lámh.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

Chapter 1 Introduction and Objective

Anaerobic digestion involves the degradation of organic matter in the absence of free oxygen with a byproduct of biogas that contains a substantial amount of methane. Much time and research has been devoted to improve the performance of anaerobic digestion systems. One technique often considered is the co-digestion of substrates in order to maximize biogas production. The technology has been widely applied for treatment of organic wastes that biodegrade easily.

Food processing sludge waste (FPSW) can have a tremendous amount of imbedded energy and is theoretically an excellent co-substrate when reconstituted with manure at a farm digester. These high carbon wastes are produced at food processing facilities by segregation and dewatering of bulk flow. This solid-liquid separation produces a weaker wastewater that can potentially be cost-effectively disposed in a sewer or irrigated. The collected solids fraction offers a source of feedstock for the digester at the dairy farm that could potentially boost biogas production and profitability for the farmer. Other key benefits include savings in transportation costs and potential tipping fees for the food processor.

If the reconstitution of the FPSW with manure co-digests successfully, then an increase in energy production per unit volume in the digester will be obtained. The production of a lower-strength wastewater at the food processing facility will also result. The majority of previous co-digestion studies have looked at organic wastes with the objective of enhancing biogas production; however the concept of thickening a FPSW and then reconstituting with manure at the digester, is novel.

The evaluation of the FPSW involved the following four distinct stages,

1. An initial respirometer study to determine the FPSW's energy potential, if any synergistic or antagonistic relationships resulted when digested with dairy manure, and if toxicity results.
2. A biogas potential screening assay of the FPSW blended with manure to determine specific optimum blending ratios.
3. The evaluation of the reconstituted FPSW using semi-continuous digester systems based on the most promising conditions established in the previous batch assays. Analysis from this stage provides a more realistic interpretation of real-life co-substrate digestion as no dilutions of the FPSW are required.
4. A report on findings, and recommendations on the potential of the FPSW as a feedstock.

Chapter 2 Literature Review

A number of issues need to be addressed when considering anaerobic digestion. Factors such as the nature of the waste, the biological processes and the critical process parameters all need to be assessed. Any potential benefits or difficulties associated with anaerobic systems must also be considered. This chapter provides some background to these topics while providing a brief outline of previous research undertaken in the anaerobic co-digestion of substrates.

2.1 Solid Food Waste to Landfill

Improper food industry waste management including the collection, treatment, processing and disposal of residuals is a source of concern due to its negative impact on the environment (Dutta and Das, 2010; Mohan et al., 2006). In 2007, the amount of food waste generated in the US alone was estimated at 28.8 million metric tons with only 2.6% of this waste recovered and diverted from landfill (Levis et al., 2010). Methane, a dangerous greenhouse gas, is generated by the degradation of solid waste in landfills. Approximately 23% of the total US methane emissions originate from landfills, the second largest contributor in the country (US-EPA, 2008). An estimated 57 million tons of methane were emitted to the atmosphere as a result of worldwide landfills in 2000 (Themelis and Ulloa, 2007). The hazards associated with solid waste landfill application and the relatively few methane capture initiatives in operation have encouraged the development of alternative technologies. The introduction of carbon credit gains has further reinforced the push for sustainable energy development. Treatment methods such as anaerobic digestion, composting and fluidized bed combustion are attractive alternatives, depending on the food waste of concern (Arvanitoyannis and Varzakas, 2008).

2.2 Anaerobic Digestion for Waste Management

Anaerobic digestion is a mature biological treatment method that can be cost effective, environmentally sound and a source of renewable energy when implemented correctly (Mata-Alvarez et al., 2000). Successful anaerobic digestion results in the production of methane gas which subsequently can be used for power generation (Chynoweth et al., 2001). From a farmer's perspective, correct implementation of an anaerobic digestion system offers advantages such as heat production, electricity generation, reduced odors and increased flexibility in manure management (Tafdrup, 1995). Food processing companies benefit from pollution reduction and revenue gains from carbon credits and renewable energy credits.

Currently in the United States there are estimated to be 151 agricultural digesters in operation with the majority farm owned and having livestock manure as the only feedstock (AgStar, 2010a). The development of the technology in Europe has been more prominent with countries such as Germany, Denmark, Austria and Sweden leading the way in agricultural biogas plant development (Holm-Nielsen et al., 2009). The implementation of government initiatives, for example, the European Union (EU) Landfill Directive (1999/31/EC), and the participation in regulations such as the United Nations Framework Convention on Climate Change (UNFCCC) among EU member states has encouraged the diversion of biodegradable waste from landfill in pursuit of alternative technologies such as anaerobic digestion (Clarke and Alibardi, 2010; Zglobisz et al., 2010).

2.3 Biological Process of Anaerobic Digestion

Anaerobic digestion involves the microbial decomposition of organic waste in the absence of free oxygen. Bacterial hydrolysis occurs first, where the waste's complex organics are broken down into simple sugars, amino acids and peptides. Subsequently, these products of hydrolysis are converted to volatile acids through biological acidogenesis. Acetogen bacteria then convert the fatty acids to acetic acid, carbon dioxide and hydrogen. Finally, methanogenic bacteria convert the products of these reactions in a process known as methanogenesis where as a result methane and carbon dioxide are produced (Grady Jr et al., 1999; Speece, 1996b; US-EPA, 2006).

2.4 Important Parameters in Anaerobic Digestion

There are a number of parameters that must be accounted for to avoid inhibition and provide stability in the digestion process. The microorganisms associated with the acid forming and methane forming stages of the anaerobic digestion process have different requirements in terms of nutrients available and the conditions of their environment. Reactor failure can originate from an unbalanced microbial population. Inhibition of a system is evident through declining rates in methane production and a buildup of organic acids (Chen et al., 2008).

2.4.1 pH

A key variable in the digestion process is the pH of the liquid waste. The suggested optimum pH range of anaerobic digestion is between 6.5 and 8.2 (Liu et al., 2008; Speece, 1996b). Regulation of pH in anaerobic systems leads to process stability. Growth rate of methanogens is significantly diminished below the optimum range while a high pH hinders system stability through the breakup of microbial granules (Ward et al., 2008). By maintaining adequate pH, the likelihood of toxicity due to free ammonia levels is reduced as levels will be lower (Bhattacharya and Parkin, 1989). Hydrolysis of lipids contained in certain food wastes feedstocks can result in the accumulation volatile fatty acids (VFA). The build up of VFA's can cause a reduction in pH and subsequently, inhibition of methanogenesis (Griffin et al., 1998).

2.4.2 Ammonia

When nitrogenous matter is degraded ammonia is released (Chen et al., 2008). Specifically, anaerobic degradation of animal manure proteins into amino acids releases ammonia to the surrounding environment (Uludag-Demirer et al., 2008). Previous literature has suggested that ammonia toxicity levels are in the region of 700 and 1200 mg /L N (Hansen et al., 1998). Sung and Liu, 2003, demonstrated reductions in specific methanogenic activity by 39% and 64% in completely stirred tank reactors (CSTR) of a synthetic wastewater when total ammonia nitrogen concentrations were 4.92 and 5.77 g/L, respectively. Studies relating to the digestion of dairy manure have indicated that small increases in ammonia during digestion can improve biogas production while high increases can result in reductions of approximately 50% (Sterling et al., 2001).

2.4.3 Temperature

The temperature inside the digester is usually operated at a mesophilic or thermophilic range. Mesophilic temperature, corresponding to 35°C, allows for process stability, high methane yields and maximum energy output (Wenxiu and Mengjie, 1989). However, the thermophilic range, corresponding to 55°C, has often shown superior performance in volatile solids (VS) destruction and lower VFA's although a much higher energy input is required (Kim et al., 2002). In the thermophilic range, inhibition by ammonia is more common (Campos et al., 1999). Maintaining the optimum temperature is a key component of manure digestion as indicated in a mesophilic study of swine manure by Chae et al., 2008, where a fall in temperature from 35°C to 30°C decreased methane yield by 3% while a fall to 25°C caused a 17.4% reduction. Approximately 60% of the anaerobic digesters in Europe with capacity for solid waste operate with the mesophilic temperature range. The remaining 40% have systems using a thermophilic process (Mata-Alvarez et al., 2000). Mesophilic digestion is less expensive to maintain than thermophilic as less energy is required (Gerardi, 2003).

2.4.4 Solid Retention Time

The solids retention time (SRT), expressed in days, is average time the solids spend in the digester and a characteristic that can affect the performance of a digester (Appels et al., 2008). Shortening the SRT is sometimes favorable as studies have indicated that a reduction from 30 to 12 days, coinciding with an increase in organic loading rate, can potentially triple the biogas production when dealing with dewatered sewage sludge in CSTR's (Nges and Liu, 2010). The SRT must be sufficient enough to allow the anaerobic bacteria to complete the digestion process. For a digestion system operating at 35°C, it is recommended that the minimum SRT is 10 days to avoid washout of the microorganisms (Appels et al., 2008). Less than 10 days will result in the rate of bacterial loss exceeding the rate of bacterial growth in the system. The recommended SRT for animal wastes is between 10 and 20 days (Keshtkar et al., 2003). Since hydrolysis is the rate-limiting step in the anaerobic digestion, it is essential to optimize the SRT especially when dealing with wastes containing high particulate matter (Burke, 2001).

2.4.5 Mixing

For optimal performance, mixing must ensure that the entire digester volume is being utilized, there is extensive contact between the bacteria and the substrate and that heat is being transferred effectively (Kaparaju et al., 2008). Otherwise methane production may be limited (Keshtkar et al., 2003). For wastes with higher solids content, the implementation of efficient mixing becomes ever more important in terms of producing higher biogas yields (Karim et al., 2005). The cost of mixing for a CSTR digester can be high especially when the feedstock contains materials that must be suspended throughout the digestion period (Burke, 2001).

2.4.6 Nutrients

Methanogenesis is a highly sensitive process and a deficiency in certain nutrients has been shown to result in inefficient substrate removal and lower gas production (Kayhanian and Hardy, 1994). Optimizing the nutrients in anaerobic digestion enables microbial stability resulting in the maximum methane production being achieved (Hills, 1979). The main macronutrients involved are carbon (C), nitrogen (N) and phosphorus (P). Each of the macronutrients are essential in specific quantities. In an anaerobic digestion study of fruit and vegetable wastes, the most suitable ratio of C:N:P for microbial growth was found in the range of 100:4.3:0.9 (Bouallagui et al., 2004). Apart from these macronutrients, a number of micronutrients are also important to the digestion process (Wilkie et al., 1986). Previous literature has shown that the biodegradable organic fraction of municipal solid wastes often requires nutrient supplementation of nitrogen and phosphorus, and that the addition of dairy manure as a nutrient-rich source can substantially improve gas production rates (Kayhanian and Rich, 1995). Likewise, the addition of a high carbon waste to manure digestion can enhance the process by providing a more optimal C:N ratio overall (El-Mashad and Zhang, 2010; Ward et al., 2008). Organic fractions of municipal solid wastes have been successfully blended with manure with the aim of finding optimized co-digestion ratios that produce stable systems with high biogas yields (Hartmann and Ahring, 2005).

2.5 Anaerobic Digestion of Manure

The potential to capture methane from manure comes from the degradation of its organic materials primarily carbohydrates, proteins and lipids (Møller et al., 2004). Previous studies of broiler manure, cattle manure and their mixtures conducted in batch reactors showed that the cattle manure alone led to a highest methane production (Güngör-Demirci and Demirer, 2004). The higher nitrogen content of poultry wastes can lead to ammonia inhibition in a digester. Swine manure and chicken manure have also been investigated as a potential methane sources with some encouraging results comparable to other wastes, however ammonia inhibition has also been detected (Chae et al., 2008; Hansen et al., 1998; Huang and Shih, 1981).

The digestion of livestock wastes is well established with dairy being the most common, representing 82% of all digesters located in the US (AgStar, 2010b). Problems associated with manure-only digesters include the struggle to be economically feasible due to low financial returns for farmers from energy generation and lower methane yields due to inhibition by free ammonia (Hansen et al., 1998; Salminen and Rintala, 2002). However, for large farms, manure still remains a plentiful source of feedstock for anaerobic digestion and its conversion to biogas moderates the quantity of harmful greenhouse gas (GHG) emissions being released to the surrounding environment (Ward et al., 2008).

2.6 Benefits of Technology

The utilization of anaerobic digestion for waste management of livestock manures introduces a number of benefits. The most significant of these advantages are discussed below.

2.6.1 Odor Control and Greenhouse Gas Emissions

Farms producing large quantities of livestock manure are often a source of pollution with regards to offensive odors. In fact, according to Holm-Nielsen et al., 2009, "65% of anthropogenic nitrous oxide and 64% of anthropogenic ammonia emissions originates from the world-wide animal production sector". The implementation of an anaerobic digester ensures significantly less odors than conventional manure management systems and is more favorable on a cost basis compared to other odor reducing alternatives (EPA, 2002).

Harmful GHG emissions can also be cutback by the installation of a digester. This was verified in a study by Kaparaju and Rintala, 2010, where anaerobic systems mitigated GHG emissions on dairy, sow and swine farms. Methane originating from livestock manure is considered a major contributor to agricultural GHG emissions and is deemed 21 times more potent than carbon dioxide on a molecule to molecule basis (Steed Jr and Hashimoto, 1994; Thelen et al., 2010). Anaerobic Digesters capture the harmful gas using it for energy purposes, and subsequently off-set energy that would originate from fossil fuels (EPA, 2002).

2.6.2 Ammonia Control

The largest source of ammonia emissions in the United States originates from livestock with manure storage being one of the most contributing factors (Pinder et al., 2003). The control of ammonia emissions from livestock manure is usually prevented using a storage tank cover. However, the release of ammonia has become ever more serious in recent times through the impact of eutrophication and acidification of the natural environment. Emissions of ammonia are controlled in a digester system. Additionally, technologies such as chemical precipitation and stripping/absorption can further reduce ammonia losses post digestion (Wilkie, 2000).

2.6.3 Environmental Protection

Pathogens, viruses and parasites contained in the feedstock will be eliminated once sufficient digester holding times and temperatures are ensured (Tafdrup, 1995). Operating in the thermophilic range, anaerobic digestion removes microbial pathogens present in the waste (Smith et al., 2005). The destruction of such organisms eradicate the possibility of contamination to surrounding groundwater and hence, human and animal health risks are reduced (EPA, 2002). Mesophilic digestion does not eradicate pathogens directly since the growth and survival of bacteria is in this temperature range. Characteristics such as the retention time are more important for pathogen removal in the mesophilic range (Smith et al., 2005).

2.6.4 Energy Generation

In 2009, it was estimated that approximately 374 million kilowatt-hours (kWh) of energy were produced from on-farm digester systems (AgStar, 2010a). However, the estimation of yields from such biological degradation processes is very much dependent on the particular type of substrate being digested (Mata-Alvarez et al., 2000). Different substrates are often co-digested in order to increase biogas yields and in turn, generate more energy.

2.7 Co-digestion and the Potential to Enhance Biogas Yields

A variety of wastes have been investigated for anaerobic co-digestion purposes. Certain food wastes can be desirable under anaerobic conditions due to high biodegradability characteristics (Zhang et al., 2007). Blending manure with organic wastes has been shown to be beneficial in terms of increasing cumulative biogas yield (Callaghan et al., 1999). This concept of co-digestion is relatively mature. The addition of organic wastes with high carbon content can overcome the problems of digesting activated sludge or manure alone (Habiba et al., 2009). Previous literature concerning organic vegetable wastes co-digested with sewage sludge at four different retention times showed increased methane yields and high degradability of such wastes (Mata-Alvarez et al., 1992). Studies co-digesting cattle slurry with fruit and vegetable waste and chicken manure in a continuously stirred tank reactors revealed that by increasing the fraction of food and vegetable waste from 20% to 50% methane yields could be improved almost two-fold (Callaghan et al., 2002). The co-digestion performance of a fruit-vegetable-municipal solid mixture that included waste from banana, apple, orange, cabbage, potatoes, bread and paper processing have also been tested with a primary sludge. Under different mixing conditions and loading rates the system was found to be stable and produced more biogas than the primary sludge due to higher volatile solids content (Gómez et al., 2006). Alvarez and Lidén, 2008, demonstrated that the mesophilic co-digestion of slaughterhouse waste, fruit vegetable waste and manure gave higher methane yields and productivity as compared to the digestion of the individual wastes alone or mixtures of two wastes. Biogas methane yields of over 60% can be achieved through the co-digestion of food waste and dairy manure (El-Mashad and Zhang, 2010).

2.7.1 Food Waste Segregation

Onsite segregation of food wastes at processing plants facilitates initiatives such as byproduct recovery, recycling and improved waste treatment performance. The idea hinges on technical and economic issues as well as the nature of the waste in terms of the quantity, biodegradability and the location of the processing facility (Zaror, 1992). The concept of thickening food wastes and reconstituting with manure at the digester is a novel co-digestion proposal and so there is only a small amount of relevant literature. For instance, Tsukahara et al., 1999, examined the separated liquid fraction of food waste in an upflow anaerobic sludge blanket (UASB). With the solids removed, the reactor was found to be suitable for efficient digestion of the liquidized food waste.

The recovery of food residuals through energy generation is a primary constituent in any food industry's waste management hierarchy (Bates and Phillips, 1999). By reconstituting concentrated food wastes in a digester, a potential waste-to-energy system is generated. The success demonstrated in the co-digestion of organic materials from food industries with manure has given confidence to the concept of joint biogas plants and centralized digesters on a larger scale (Holm-Nielsen et al., 2009).

2.7.2 Polymer Waste Studies

The onsite addition of polymers for solid-liquid separation of food waste introduces a unique feature when contemplating co-digestion with manure. Literature directly related to the effect of polymers in anaerobic digestion systems has been limited and contrasting. In the past, the addition of organic flocculants to municipal wastewater has resulted in a sludge that was not digestible in anaerobic systems showing indications of reduced methane content, chemical oxygen demand (COD) destruction and VS destruction (Gossett et al., 1978). Chu et al., 2003, investigated the effect of three polyelectrolyte flocculants on the digestion of waste activated sludge in terms of methane generation. Gas production was found to increase in the early stages of digestion but depending on the polymer type, could inhibit digestion at later stages. A more recent study involved the use of a polyacrylamide flocculent for improving separation of solid fractions of pig waste for anaerobic digestion. The polymer was not readily degradable by anaerobic bacteria, however, it was not found to be toxic (Campos et al., 2008).

2.8 Centralized Digesters

Constructing an anaerobic digestion system on every dairy farm is impractical and unrealistic. A centralized digester is a facility that allows for the collection of wastes from small clusters of farms within a certain distance (Ma et al., 2005). Such systems also offer a strategy for areas where food processing wastes are mass produced. Co-digestion of livestock manure and food waste residuals in a centralized digester can generate revenue for the farmer through better biogas production and reduce waste handling costs for the food processor (Dagnall, 1995). Transportation costs are the main obstacle when planning centralized digesters. Effective shipping varies on the distance per unit volume transported (Flotats et al., 2009). Currently there are 9 centralized digestion projects in operation in the US (Roos, 2010).

2.9 Biochemical Methane Potential

A batch biochemical methane potential (BMP) study is an inexpensive technique used in the laboratory to determine how biodegradable substrates are under anaerobic treatment processes (Owen et al., 1979). Literature on the BMP of various fruit and vegetable wastes with the purpose of obtaining ultimate methane yields has been well documented (Gunaseelan, 2004). Likewise, BMP tests have also been demonstrated on various other food wastes and dairy manure (Chen et al., 1988; Cho et al., 1995; El-Mashad and Zhang, 2010). Conducting initial biogas screening assays allows for estimations of the wastes energy potential and general indications of whether further study of the feedstock is warranted. However, the use of a BMP as an indicator to full-scale digestion is challenging and should be avoided as some studies have indicated over prediction of biogas production by as much as 51% (Bishop et al., 2009). The validity of a BMP assay depends on factors such as the inoculum used and the ratio of inoculum-to-waste on a VS basis. The latter parameter will now be looked at in more detail.

2.9.1 Inoculum-to-Waste Ratio

For a BMP study it is essential to begin with the correct amount of acclimated inoculum with respect to quantity of waste being added. Inoculum-to-waste ratios on a VS basis using domestic sewage sludge from an active digester have been examined with different feedstocks such as cellulose, napiergrass and energycane. Various ratios were tested with an inoculum-to-waste ratio of 2:1 showing the maximum conversion rates (Chynoweth et al., 1993). Another study examining inoculum-to-waste importance tested ratios of 2, 1, 0.74 and 0.43 (VS basis) on a restaurant kitchen waste. Production of methane and biodegradability potential decreased significantly for the inoculum-to-waste ratio of 0.43 (Neves et al., 2004). Further literature using ratios of 3, 2, 1.5 and 1 (VS basis) were compared in the study of maize. Digester sludge from a municipal wastewater treatment plant was used as inoculums. The results showed that the percentage of methane in the total biogas volume was very similar irrespective of ratio. This corresponded well with larger batch-scale fermentations (Raposo et al., 2006). In 2009, a similar study was developed examining methane production from the anaerobic digestion of sunflower oil cake. Numerous inoculum-to-feed ratios (3, 2, 1.5, 1, 0.8, and 0.5) were compared using granular sludge inoculum from an anaerobic reactor treating brewery wastewater. High stability was reported for all ratios between 3 and 0.8. The highest concentration of total volatile fatty acids (TVFA) was found at the ratio of 0.5 resulting in an extremely negative effect on methane production (Raposo et al., 2009).

2.10 Semi-Continuous Studies

Semi-continuous operations are often used to analyze wastes in co-digestion. Cuetos et al., 2008, operated 3 L working volume semi-continuous reactors at a mesophilic temperature to examine the blend of slaughterhouse wastes and municipal solid waste. Reactors were fed via side inlet each day and the system was allowed run for two SRT's. A similar study investigating the co-digestion of slaughterhouse waste, fruit-vegetable waste and manure utilized stainless steel semi-continuous digesters with total volume of 2 L at 35°C with gas production measured via a water displacement method (Alvarez and Lidén, 2008). Glass reactors of 5 L total volume and 4 L working volume have been used previously in co-digestion studies of meat processing byproducts and sewage sludge (Luste and Luostarinen, 2010). Magnetic stir bars operating at 300 rpm provided the adequate mixing effect in the reactors. Gas produced was collected in aluminum gas bags. An alternative approach using 15 L glass reactors was conducted in a study examining the digestion of cattle slurry mixed with fruit and vegetable waste (Misi and Forster, 2002). The semi-continuous reactor had an 8.8 L working volume with the remainder comprising as headspace. An external water jacket provided maintained the heat at 35°C. The single stage digester as described by Lafitte-Trouqué and Forster, 2000, consisted of a pyrex bottle with fitted stopper on top of a magnetic stir plate. Fresh feed was pumped into the reactor daily. A wet-tip gas meter was used to provide gas measurement.

Small semi-continuous systems, as mentioned above, represent continuously stirred tank reactors (CSTR). Operations such as this give a more realistic interpretation of full-scale anaerobic digestion than batch BMP studies (Owen et al., 1979). Results obtained from semi-continuous reactors can be used to observe the reaction of different substrates in co-digestion.

Chapter 3 Methods and Materials

This chapter provides a detailed account of the respirometer, serum bottle and semi-continuous studies. Information regarding the origin of each substrate used and the different procedures for each assay are described in detail. Any calculations involved in the experimental setup are also discussed.

3.1 Food Processing Sludge Waste

The FPSW was obtained from a large food processing facility on March 1, 2010. Manufactured foodstuffs at the facility included a variety of sauces (mustard, relish, barbeque), vinegars (white, wine, cider) and pickles (kosher, fresh, sweet). Item production at the plant resulted in leftover food waste, cleaning waste and chemicals which are was washed to drain. The drainage system collects all the waste material from the production line. This wastewater is pumped over screens to remove larger particulates that are collected in a large open-top container outside of the building. A large storage tank (250,000 gallons) is used to hold the remaining wastewater. While stored the wastewater is chlorinated. This effluent is then taken from the storage tank and filtered by means of a cyclone system (Figure 3.1). This system allows for the separation/removal of finer particulates. Calcium hydroxide ($Ca(OH)_2$), otherwise known as slaked lime, is added in order to regulate the pH of the wastewater as it enters the cyclone. Furthermore, polymers (unspecified for proprietary reasons) are added to help enhance solid separation. The final stage of waste treatment involves removing the coagulated solids from the cyclone and utilization of a belt-press filter for further dewatering (Figure 3.2). The solids that emerge at the end of the belt-press filter are known as 'sludge waste' and are ready to be landfilled (Figure 3.3). Remaining wastewater that was generated throughout the waste management process is allowed to be injected to a well in close proximity to the plant.

Figure 3.1 Cyclone System at Food Processing Facility

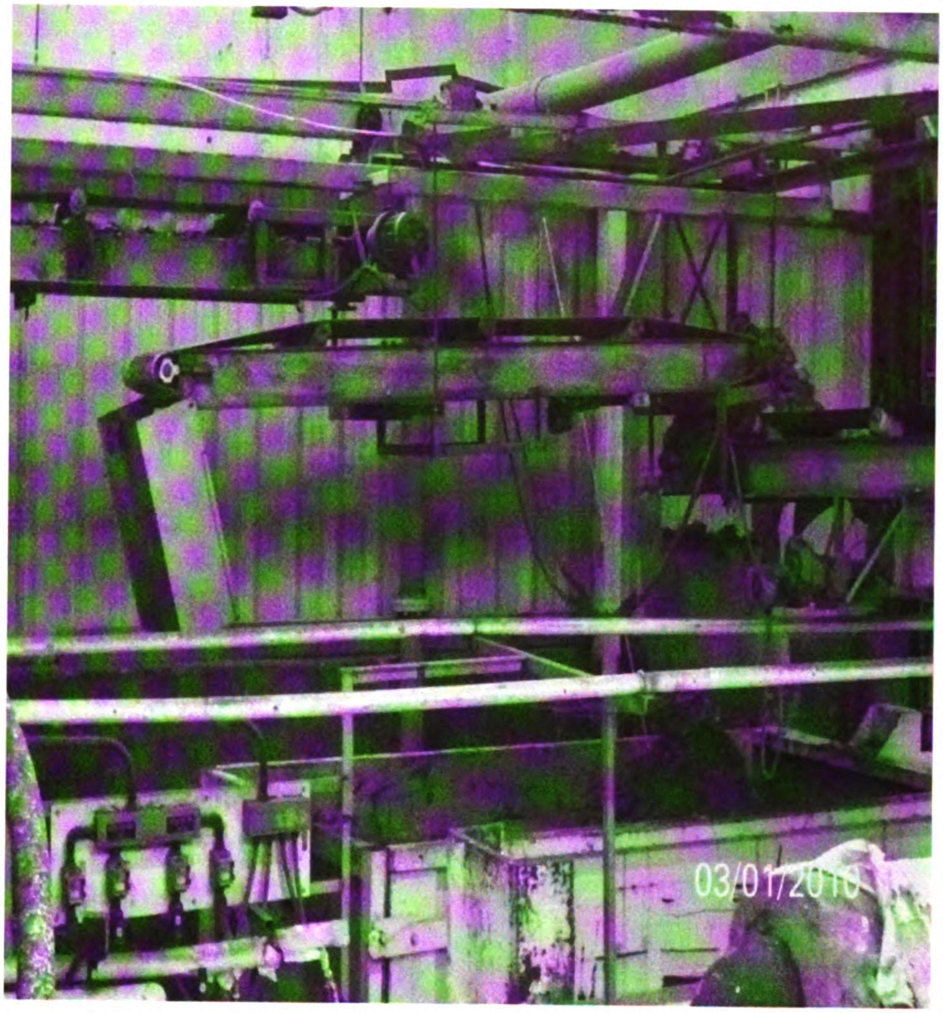


Figure 3.2 Belt-Press Filter

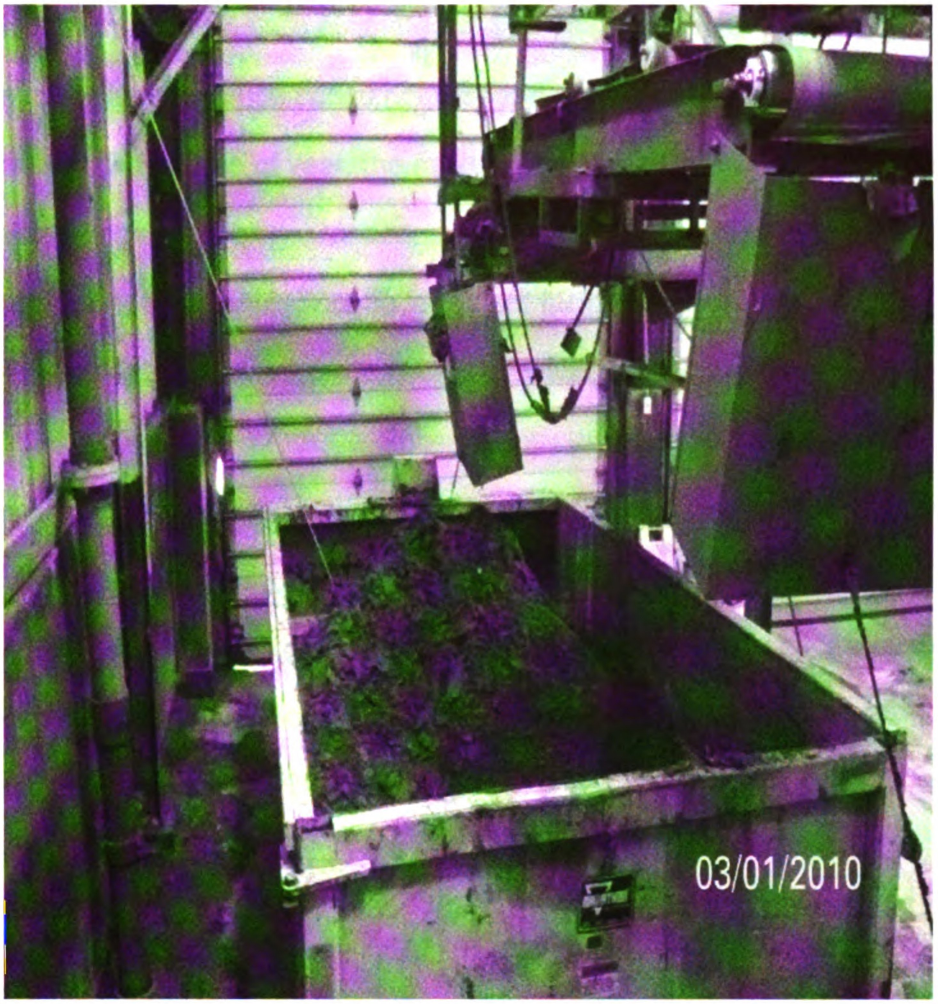


Figure 3.3 Food Processing Sludge Waste

Currently about 1,600 ton/year of FPSW is produced. The costs involved with the waste are estimated at $75-85 per ton, not including hauling costs to the landfill. Since consumer demand on certain foodstuffs varies depending on the time of year, the make-up of the FPSW itself is therefore subject to some variability. Consequently, in researching the FPSW, consistency of the sample was an important factor. The first biogas respirometer assay conducted on the FPSW was carried out on a sample collected on February 17, 2009. Subsequently, the sample for the serum bottle study was collected almost exactly a year later on March 1, 2010. This FPSW sample was used for both the serum bottle and semi-continuous systems. The pH, COD, total solids (TS) and VS of the FPSW were analyzed between studies to ensure characteristics had not changed. Only minimal differences were found in testing.

3.2 Inoculum

The inoculums used for the respirometer and serum bottle assays were collected from a membrane bioreactor (MBR) located on Michigan State University campus, East Lansing, MI. The manure inoculum was digestate from the MBR that was collected in the days preceding start-up of each assay and was stored at 4°C prior to use.

For the semi-continuous systems, the digestate from the batch assay was used as the seed inoculum. All remaining digestate not used for chemical analysis testing were mixed thoroughly and refrigerated until time of use.

3.3 Cow Manure

The manure used for both the respirometer and serum bottle assays was collected from Minnis Farms, Williamston, MI and stored at 4°C prior to use. Any manure held for more than one week was stored in a freezer at -17°C. The same location supplied the manure for the semi-continuous operation. This allowed for consistency between all three studies.

Tables 3.1, 3.2 and 3.3 show the initial characteristics of each constituent for the three studies.

Table 3.1 Respirometer Assay Initial Characteristics

	Seed	Diluted Sludge
pH	NA	4.70
TS (mg/L)	NA	27,612
VS (mg/L)	21,577	20,393
COD (mg/L)	NA	46,725

Table 3.2 Serum Bottle Assay Initial Characteristics

	Seed	Cow Manure	Diluted Sludge
pH	7.44	6.95	5.81
TS (mg/L)	57,600	39,893	35,607
VS (mg/L)	34,300	26,112	20,581
COD (mg/L)	81,100	59,250	38,425

Table 3.3 Semi-Continuous Study Initial Characteristics

	Seed	Cow Manure	FPSW	FPSW/DI Water Feed	FPSW/Manure Feed
pH	7.60	7.34	6.91	7.58	7.50
TS (mg/L)	16,875	27,008	321,690	95,075	112,820
VS (mg/L)	9,613	18,697	176,310	50,580	61,938
COD (mg/L)	17,188	30,763	427,130	93,550	125,550

Seed characteristics between the serum bottle and semi-continuous studies are shown to have significantly different values in terms of TS, VS and COD. This can be attributed to the origin of the inoculums as mentioned in section 3.2. pH values of the diluted sludge samples appeared to have slight differences between studies. The reason for this variation remains unclear.

3.4 Respirometer Assay Design

The initial biogas assay for co-digestion of the FPSW and cow manure was conducted from March 20 – April 17, 2009 with a goal of determining the anaerobic biodegradability and biogas recovery potential of co-digestion. Table 3.4 shows the compositions of the flasks used in the assay.

Table 3.4 Respirometer Flask Compositions

Treatment	Seed (mL)	Cow Manure (mL)	Diluted Sludge (mL)	DI Water (mL)
Seed	136	0	0	514
Seed, Cow Manure	136	72	0	442
Seed, Diluted Sludge	136	0	72	442
Seed, 1:1 Cow Manure: Diluted Sludge	136	72	72	370
Seed, 2:1 Cow Manure: Diluted Sludge	136	144	72	298
Seed, 1:2 Cow Manure: Diluted Sludge	136	36	72	406

The "Seed" treatment was run as a control. "Seed, Cow Manure" and "Seed, Diluted Sludge" treatments were run so that the normalized gas of the individual components (manure and FPSW) could be established by subtracting out the gas produced by the "Seed". This method allowed for the calculation of the individual levels of gas production of each constituent. The sludge used in each treatment was diluted in the ratio of 1 part sludge to 10 parts de-ionized (DI) water so as to allow for a homogenous and consistent product that was suitable for conducting analytical tests. This was achieved by weighing 100 g of FPSW in a beaker on a Scout™ Pro 4000 g scale and diluting the sludge to 1000 g total with DI water. The “diluted sludge” was blended for 1 minute using a Waring® Commercial Blender. This mixture was blended several times until enough substrate was prepared for the assay.

3.4.1 Volume of Diluted Sludge and Cow Manure

The volume of diluted sludge and cow manure was made on a COD basis. According to Speece, 1996, the conversion of 1 g COD destruction is equal to 395 mL CH_4 at 35°C. This represents the maximum theoretical yield of methane. Therefore, to produce at least 1,000 mL methane, 2,531 mg COD was required. Past literature has shown percentage COD reductions to vary substantially depending on the waste (Chinnaraj and Venkoba Rao, 2006; Telles Benatti et al., 2002; Wilkie et al., 2004). A 50% destruction of COD was assumed based on previous BMP’s conducted and hence the amount used was 5,063 mg COD. The total volume of each treatment initially prepared was 1 L. Each respirometer bottle had a working volume of 650 mL while the remaining 350 mL was used to conduct analytical testing. Therefore, 5,063 mg was the desired amount of COD required in the initial 1 L treatment sample prepared. With the COD of the sludge calculated at 46,725 mg/L (Table 3.1), the volume of diluted sludge required for 1 L was obtained using Equation 1. Calculating for a 650 mL bottle, it was determined that 72 mL was needed. The volume of cow manure was based around the volume of diluted sludge required and the chosen blending ratios (discussed in section 3.5.1).

$$\text{Diluted Sludge Volume (mL)} = \frac{\text{Desired mg COD} \times \text{Working Volume (mL)}}{\text{COD of Sludge} \left(\frac{\text{mg}}{\text{L}}\right) \times 1\text{L}}$$

Equation 1.

3.4.2 Volume of Seed

An inoculum-to-waste ratio of 2:1 (VS basis) was chosen for this study (Chapter 2, Section 2.9.1). The diluted sludge volume for 1 L was calculated to be 110 mL. The VS of the diluted sludge and seed were 20,393 mg/mL and 21,577 mg/mL, respectively (Table 3.1). Consequently, the seed volume was calculated by Equation 2 as 208 mL. Accounting for the 650 mL respirometer bottle the amount of seed required was 136 mL.

$$\text{Seed Volume (mL)} = \left(\frac{2 \times \text{Diluted Sludge Volume (mL)} \times \text{VS Sludge} \left(\frac{\text{mg}}{\text{mL}}\right)}{\text{VS Seed} \left(\frac{\text{mg}}{\text{mL}}\right)} \right)$$

Equation 2.

3.4.3 Respirometer Assay Setup

An anaerobic respirometer (Challenge Technology AER-200, Springdale, AR) was used for the biogas assay. The treatment flasks had total capacity of 725 mL of which 650 mL was used for liquid sample while the remaining 75 mL was left as headspace. Using the volumes calculated in section 3.4.2, the constituents of each treatment flask (Table 3.4) were added precisely. All flasks were sealed tightly with a cap and septum. An adhesive was applied to the inside of the cap to ensure a gas tight fit. The headspace in each treatment flask was flushed with 100% N_2 gas for 10 minutes at a flow rate of 0.5 L/min before start-up (Owen et al., 1979). This was performed using a B-D 20 gauge needle that purged the bottle septum introducing the N_2 gas, while a second needle allowed for the initial headspace gases to be flushed out. The flasks were then positioned in a water-bath that was held at mesophilic temperature (35°C). The water-bath sat on a large magnetic stir plate with each flask containing a stir bar that provided adequate mixing (50 rpm). Wires with attached gauge-needles were inserted through the septum of each reaction flask and subsequently connected to individual gas counter cells. Each individual cell was then connected directly to a stand-alone computer. Real-time gas production rates and cumulative gas volumes were measured for each flask using Challenge Technology AER computer software. Biogas from each vessel was analyzed weekly for methane and carbon dioxide using a Shimazdu GC8 gas chromatograph. This was accomplished using a syringe that extracted 2 mL of biogas from the headspace which was injected into the GC column to be analyzed. Once the vessels reach maximum gas production, as indicated by a sharp reduction in the cumulative biogas production curve, the assay is considered complete. However, due to strict time constraints it was not possible for all treatments to reach this stage (discussed in Chapter 4, section 4.1.3). Hydrogen sulfide (H_2S) analysis was conducted once during gas production by collecting gas in a 300 mL gas sampling bag. Concentrations were measured by Gastec tube sampling methods. This is a rapid analysis where 100 mL of gas is pulled from the gas bag and direct measurement of H_2S concentrations is read off the scale on the tube. The H_2S numbers do not represent precise concentrations, but to serve as a general indicator (Appendix A, Table A.8).

3.5 Serum Bottle Assay Design

Table 3.5 shows the constituents of each treatment assessed are in the serum bottle assay. Each treatment was run in triplicate for QA/QC purposes. The "Seed" treatment was run as a control. "Seed, Cow Manure" and "Seed, Diluted Sludge" treatments were run so that the normalized gas of the individual component (manure and diluted sludge) could be established by subtracting out the gas produced by the "Seed". This method allowed for the calculation of the individual levels of gas production of each constituent. The sludge used in each treatment was diluted in the ratio of 1 part sludge to 10 parts water so as to allow for a homogenous and consistent product that was suitable for conducting analytical tests. The diluted sludge was prepared in an identical fashion as to that already discussed in section 3.4.

Table 3.5 Serum Bottle Compositions

Treatment	Seed (mL)	Cow Manure (mL)	Diluted Sludge (mL)	DI Water (mL)
Seed	30	0	0	120
Seed, Cow Manure	30	25	0	95
Seed, Diluted Sludge	30	0	25	95
Cow Manure	0	25	0	125
Diluted Sludge	0	0	25	125
Seed, 2:1 Cow Manure: Diluted Sludge	30	50	25	45
Seed, 1:1 Cow Manure: Diluted Sludge	30	25	25	70
Seed, 1:2 Cow Manure: Diluted Sludge	30	12	25	83
Seed, 1:4 Cow Manure: Diluted Sludge	30	6	25	89
Seed, 1:6 Cow Manure: Diluted Sludge	30	4	25	91

3.5.1 Volume of Diluted Sludge and Cow Manure

The constituents of each bottle were determined in a similar fashion to the respirometer study discussed in section 3.4.1; however, the smaller working volume of the serum bottles were accounted for and a COD destruction of 40% was now assumed based on the results of the previous assay. Using Equation 1 and the values listed in Table 3.6, the volume of diluted sludge for the serum bottles was calculated to be 25 mL. The amount of diluted sludge remained constant for each treatment it appeared in. The blending ratios were developed in relation to the diluted sludge volume of 25 mL. For example, a 2:1 ratio consisted of 50 mL of cow manure to 25 mL diluted sludge. A ratio of 1:4 consisted of 6 mL of cow manure to 25 mL diluted sludge etc. The volume of diluted sludge did not change between treatments.

Table 3.6 Serum Bottle Values for Equation 1

Parameter	Value
Desired COD (mg)	6,239
Amount to be Prepared (L)	1
COD of Sludge (mg/L)	38,425
Serum Bottle volume (mL)	150
COD Destruction (%)	40

3.5.2 Volume of Seed

The determination of seed volume for the serum bottles was calculated similarly to that presented for the respirometer study in section 3.4.2. Using Equation 2 and the values listed in Table 3.7 the volume of diluted sludge for the serum bottles was calculated to be 30 mL.

Table 3.7 Serum Bottle Values for Equation 2

Parameter	Value	Reference
Inoculum-to-Waste (VS basis)	2:1	Section 3.4.2
Sludge Volume (mL)	25	Section 3.5.1
VS Sludge (mg/L)	20,581	Table 3.2
VS Seed (mg/L)	34,300	Table 3.2

3.5.3 Serum Bottle Assay Setup

Borosilicate glass - aluminum seal, Kimble Chase serum bottles of 225 mL liquid capacity were used for all treatments in the assay. The bottles were sealed with septa and covered tightly with septa caps. Once the constituents of each co-digestion treatment were made and the subsequent bottles sealed, the headspace was flushed with 100% nitrogen as described in section 3.4.3. All treatment bottles were placed in a VWR Signature™ Forced Air Safety Oven which was held constantly at 35°C (Figure 3.4). Initially the serum bottles were incubated for two hours in the oven. After two hours, the internal pressure was returned to atmospheric by following the Biogas Production Measurement method (Section 3.5.4) and thus the assay had begun. The assay ran from March 10 – May 25, 2010.

Figure 3.4 Serum Bottle Assay Setup

The wastes were first analyzed for pH, COD, TS and VS to ensure suitability for digestion as characterized in Table 3.8.

Table 3.8 Digestion Parameters (Szczegielniak, 2007)

Parameters	Method	Suggested Range	Source
pH	pH meter	6.5 to 8.2	Speece, 1996
Alkalinity	Hach 8203	2000 to 3000 mg/L $CaCO_3$	Speece, 1996
COD	Hach 8000 (EPA approved)	> 1000 mg/L COD	Speece, 1996
Soluble COD	HACH 8000 after Filtering with TSS Filter		
Total Solids (TS)	Hach 8271 (EPA approved)	< 10% for batch tests	(Carucci et al., 2005)
Volatile Solids (VS)	Hach 8271 (EPA approved)	High Percent of Total Solids	
Ammonia	Hach 10031	200 to 700 mg/L-N	(Hansen et al., 1998)
Nitrogen (N)	Hach 10072		
Phosphorus (P)	Hach 10127		

3.5.4 Biogas Production Measurement

To measure the biogas, the serum bottles were held at a 45° angle. A glass syringe (30 mL or 100 mL capacity) with an attached B-D 20 gauge needle was inserted through the serum bottle septum. Acetone and DI water were applied to the inside of the glass syringe case and plunger before use allowing for the syringe plunger to move freely. Once the needle was inserted the biogas under pressure in the serum bottle came to atmospheric pressure in the syringe. The volume of the biogas could then be measured on the syringe scale. After removing the syringe and biogas, the internal pressure of the serum bottle was back at atmospheric. The contents of the serum bottle were mixed daily by inverting the bottle slowly five times. Measuring the biogas of the serum bottles started on a daily basis. Once the biogas production began to decrease the measurement process was performed on a less regular basis (between 2 – 5 days).

Biogas composition analysis was performed on a weekly basis using an SRI 8610C Gas Chromatograph along with Peaksimple computer software. Biogas was extracted from the headspace of the serum bottles as previously described in section 3.4.3 and analyzed for methane, carbon dioxide, nitrogen and hydrogen sulfide content.

3.6 Semi-Continuous Systems

The final stage of research involved the implementation of five semi-continuous digestion reactors to test the FPSW. The working volume of each reactor was 2 L. Based on previous literature, the SRT of a digester involving animal wastes should be between 10-20 days and so an SRT of 15 days was chosen for this study (section 2.4.4). Using this information, the flow rate to and from the reactors was calculated using Equation 3 as 0.130 L/day.

$$\frac{\text{Working Reactor Volume (L)}}{\text{SRT (days)}} = \text{Flow Rate}\left(\frac{\text{L}}{\text{day}}\right)$$

Equation 3.

This meant that 0.130 L of sample was removed and 0.130 L of fresh sample was fed every day to the reactors.

3.6.1 Evaluating Specific Gravity

An important factor of this study was to test the specific gravity of each substrate used in the semi continuous reactors (i.e. cow manure, FPSW/DI and FPSW/manure). By calculating the specific gravity it could be assumed that 1 mL of cow manure, FPSW/DI water (blended) or FPSW/manure (blended) was equivalent to 1 g in weight allowing for the removal and feeding of substrates to be conducted on both a weight and volume basis. The results of the specific gravity tests are shown in Table 3.9. Tests were conducted by placing a beaker on a Scout™ Pro 4000g scale and zeroing the weight, measuring the weight in grams of 130 mL of each substrate three times, measuring the weight in grams of 130 mL of water once and calculating the specific gravity based on Equation 4.

Table 3.9 Specific Gravity Values for each Substrate

Sample	Weight (g)	Average Weight (g)	Specific Gravity
Cow Manure 0.130 L A	131.6	130.3	1
Cow Manure 0.130 L B	128.8		
Cow Manure 0.130 L C	130.4		
Water 0.130 L	130.0	130.0	
FPSW/DI Water 0.130 L A	132.4	132.5	1.019 ≈ 1
FPSW/DI Water 0.130 L B	132.2		
FPSW/DI Water 0.130 L C	132.9		
Water 0.130 L	130.0	130.0	
FPSW/Manure 0.130 L A	132.8	133.2	1.024 ≈ 1
FPSW/Manure 0.130 L B	132.9		
FPSW/Manure 0.130 L C	133.8		
Water 0.130 L	130.0	130.0	

$$\text{Specific Gravity} = \frac{\text{Weight of the Substance}}{\text{Weight of an Equal Volume of Water}}$$

Equation 4.

3.6.2 Optimization Study

The five semi-continuous reactors contained the following constituents:

1. Seed
2. Seed, Cow Manure
3. Seed, FPSW
4. Seed, Cow Manure, FPSW
5. Seed, Cow Manure, FPSW (Duplicate)

Straight reconstitution of the FPSW with manure was achieved using the semi-continuous operation. No blending ratio seemed ideal from the batch studies as indicated by the purely additive results of combining diluted sludge and manure. A study with optimum COD:N ratio for the FPSW/manure was deemed the best approach (refer to Chapter 4, section 4.2 and Figure 4.5). Chemical analysis tests were run on the cow manure and FPSW so that a COD:N ratio of approximately 20:1 could be obtained. Iterative calculations were made for Equation 5 and Equation 6.

$$\frac{\left(\text{COD Cow Manure}\left(\frac{\text{mg}}{\text{Kg}}\right)\times \text{M(Kg)}\right)(0.45)+\left(\text{COD FPSW}\left(\frac{\text{mg}}{\text{kg}}\right)\times \text{S(Kg)}\right)(0.81)}{(\text{M(Kg)}+\text{S(Kg)})}$$

Equation 5.

$$\frac{\left(\text{Nitrogen Cow Manure}\left(\frac{\text{mg}}{\text{Kg}}\text{N}\right)\times \text{M(Kg)}\right)+\left(\text{Nitrogen FPSW}\left(\frac{\text{mg}}{\text{kg}}\text{N}\right)\times \text{S(Kg)}\right)}{(\text{M(Kg)}+\text{S(Kg)})}$$

Equation 6.

COD of Cow Manure = 27,763 mg/Kg

M = mass of manure in Kg

COD of FPSW = 427,130 mg/Kg

S = mass of FPSW in Kg

0.45 is a representative figure for the percentage of undestroyed COD resulting from the "Manure" treatment in the serum bottle assay. It was calculated by subtracting the percentage COD destroyed (100%-55%). This is the quantity of COD that is relevant to the inside of the semi-continuous reactors to keep the optimum COD:N ratio.

Similarly, *0.81* is a representative figure for the percentage of undestroyed COD resulting from the "Diluted Sludge" treatment in the serum bottle assay. It was calculated by subtracting the percentage COD destroyed (100%-19%). This is the quantity of COD that is relevant to the inside of the semi-continuous reactors to keep the optimum COD:N ratio.

Nitrogen Cow Manure = 1,975 mg/Kg – N

Nitrogen FPSW = 14,910 mg/Kg -N

The COD:N ratio is optimized at approximately 20:1 (Chapter 2, section 2.4.6). Using an iterative process of entering values, coinciding with the already fixed 2 L working volume, the values of M and S were resolved to be 1.1 Kg and 0.5 Kg, respectively, using Equation 7.

$$\frac{\text{Equation 5}}{\text{Equation 6}} = 20$$

Equation 7.

The COD:N:P ratio of the cow manure alone was approximately 140:10:1. The COD:N:P ratio of the FPSW alone was 219:7.5:1. Combining 1,100 g manure and 500 g FPSW was found to give an optimum COD:N ratio of approximately 20:1. For the "Seed, FPSW" treatment, 1,100 g of DI water was used to comprise the full 2 L volume of the reactor. DI water does not contribute to gas production. Therefore the compositions of the semi-continuous reactor flasks could be made as shown in Table 3.10.

Table 3.10 Semi-Continuous Reactor Compositions

Treatment		Seed (g)	Cow Manure (g)	FPSW (g)	DI Water (g)
Reactor 1	Seed	400	0	0	1,600
Reactor 2	Seed, Cow Manure	400	1,600	0	0
Reactor 3	Seed, FPSW	400	0	500	1,100
Reactor 4	Seed, Cow Manure FPSW	400	1,100	500	0
Reactor 5	Seed, Cow Manure FPSW	400	1,100	500	0

Borosil 3 L Erlenmeyer Flasks were used for the five semi-continuous reactors. The flasks were fitted with size 11 rubber stoppers and tied tightly using metal wire. Each reactor was placed on magnetic stir plates with 3.5 inch magnetic stir bars providing the mixing effect (350 rpm). Two holes were drilled through each stopper. Glass tubing (1 cm diameter) was fitted tightly through the holes in the stoppers. The first tube was 18 inches in length and used for removal and feeding of substrate. A valve fitting connected at the top of the glass tubing allowed access for both pumping and shutting off of the line when not in use. The other glass tube was shorter at approximately 6 inches. This tubing was left in the headspace so that the gas generated was free to move to the wet-tip gas meters (Wet Tip Gas Meter Company, Nashville, TN) for volume measurement. Each wet-tip gas meter was calibrated to tip every 100 mL. The reactors were held in a constant temperature room at 35°C. Tubing from the top of the reactors led to the wet-tip gas meters which were held outside of the constant temperature room.

Dual-Valve Tedlar PVF Bags were connected via Y-fitting outside of the constant temperature room on the tubing lines connecting the reactors to the wet-tip gas meters.

3.6.3 Semi-Continuous Start Up

Each reactor was set up one week before the start of the first SRT. Reactor 1 was started with the full amount of seed and DI water and connected to the wet-tip gas meter. No substrate was removed or fed throughout the entire run of Reactor 1.

Reactor 2 was started with full amount of seed. In order to not 'shock' the system, only 500 mL of manure was added initially. The remaining manure was added gradually in the week preceding the start of the first SRT. Until the full 2 L reactor volume was reached, the gas produced was collected in a dual valve Tedlar Gas Bag. This allowed for the reactor to build up an initial amount of biogas that was later used to counteract any negative vacuum created in the system when sample was being extracted via the Masterflex® pump.

Reactor 3 was started with full amount of seed. In order to not 'shock' the system, only 500 mL of FPSW/DI water was added initially. The FPSW/DI water mixture consisted of 500 g of FPSW and 1,100 mL of DI water blended until homogeneous sample was obtained. Reactor 3 was gradually fed to the full 2 L reactor volume in a similar manner as that previously mentioned for Reactor 2.

Reactors 4 and 5 (duplicates) were started with full quantity of seed. In order to not 'shock' the system, only 500 mL of FPSW/manure was added initially. The sludge/manure mixture consisted of 500 g of FPSW and 1,100 mL of manure blended until homogeneous sample was obtained. Reactors 4 and 5 were gradually fed to their full 2 L reactor volumes in a similar manner as that previously mentioned for Reactor 2.

On the first day of the first SRT, reactors 2, 3, 4 and 5 were connected to the wet-tip gas meters. The wet-tip gas meters were continuously connected to the reactors for the remainder of the study. Figure 3.5 shows the semi-continuous reactor setup. Figure 3.6 shows the tubing that connected the reactors to the wet-tip gas meters located outside of the constant temperature room.

3.6.4 Removal and Feeding of Reactors

Reactors 2 – 5 were fed everyday in the following manner:

1. Record number of tips before feeding process,
2. Close gas line to wet-tip gas meter by switching valve off,
3. Open gas line to Tedlar gas bag,
4. Document date and time of feeding,
5. Carefully place reactor on Scout™ Pro 4000 g scale,
6. Attach Masterflex pump lines to the reactor,
7. Record initial weight on the scale,
8. Open stopcock valve on the pump line and switch pump on,
9. Remove 130 g of sample into graduated cylinder (this allowed for removal to be conducted on a weight and volume basis),
10. Close stopcock valve once sample is removed,
11. Record weight of reactor after removal of sample,
12. Pour feed into graduated cylinder (usually over 130 mL to account for sample in the feed lines),

13. Open stopcock valve on the pump line and switch on pump,
14. Feed until weight is equal to the initial weight of the reactor recorded at the start of the removal/feeding process,
15. Close stopcock valve once appropriate weight is achieved,
16. Record weight of reactor after feeding of sample,
17. Carefully place reactor back on to magnetic stir plate and adjust rpm,
18. Record number of tips after feeding process (should be the same),
19. Open gas line to wet-tip gas meter by switching valve on,
20. Close gas line to Tedlar gas bag.

The reactors were fed everyday for the 3 SRTS's as accurately as possible (Appendix C, Table C.1). Stock solutions of each sample (i.e. feed for Reactor 3 and feed for Reactors 4 and 5) were prepared on a weekly basis and stored at 4°C until time of use. The number of tips on the gas meters was accounted for at the end of each day for each reactor (Appendix C, Table C.2). For safety purposes, all gas bags were inspected every day to ensure that maximum capacity had not been reached. If close to maximum capacity, gas was released slowly into the tip meter until the bag was approximately half full. Reactor mixing speeds were also checked every day to ensure it was within the specified optimum range. The gas lines were drained of any water build-up as required. This water build-up occurred due to overflow from the submerged wet-tip gas meter.

Chemical analysis tests were performed on the effluent of each reactor approximately 3 times a week. The tests run comprised of pH, alkalinity, COD and TS/VS, and were carried out using the same methods as discussed earlier for the batch assays (Chapter 3, Table 3.6). The substrate being fed to each reactor was tested for pH, COD and TS/VS at the beginning of the first SRT and was not expected to change throughout. This was confirmed by running additional characteristics tests during the second and third SRT's (Appendix C, Table C.12)

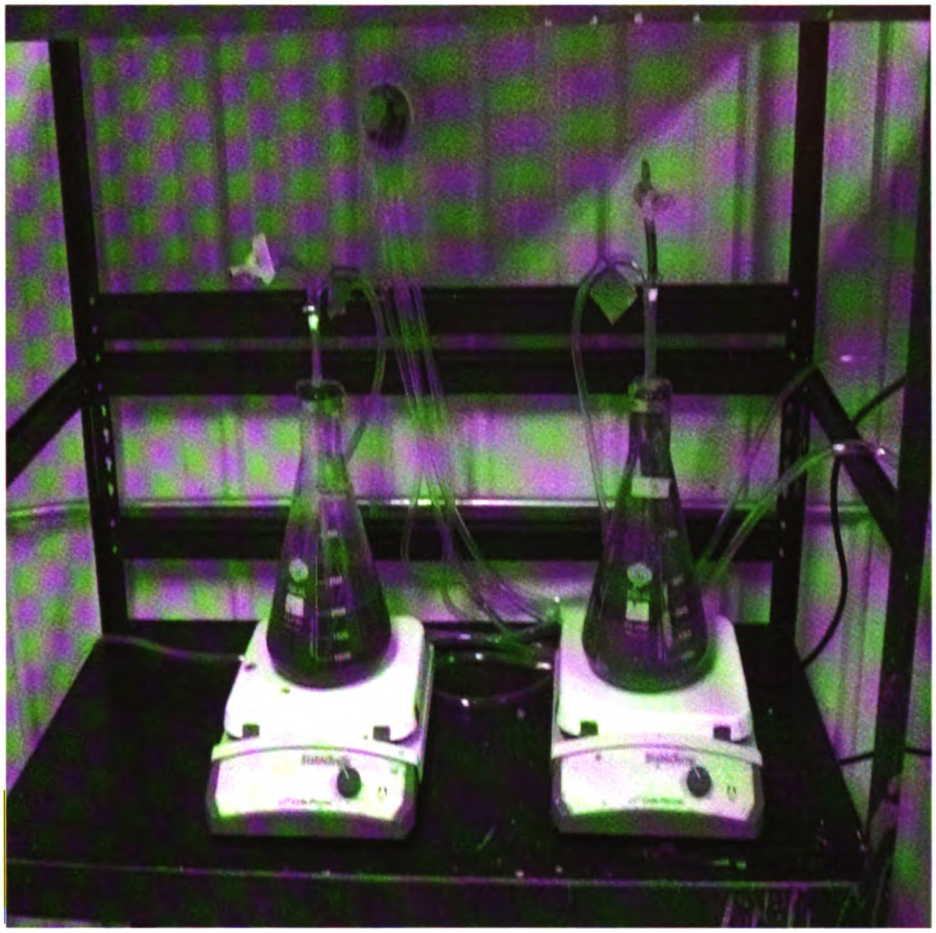


Figure 3.5 Semi-continuous Reactor Setup

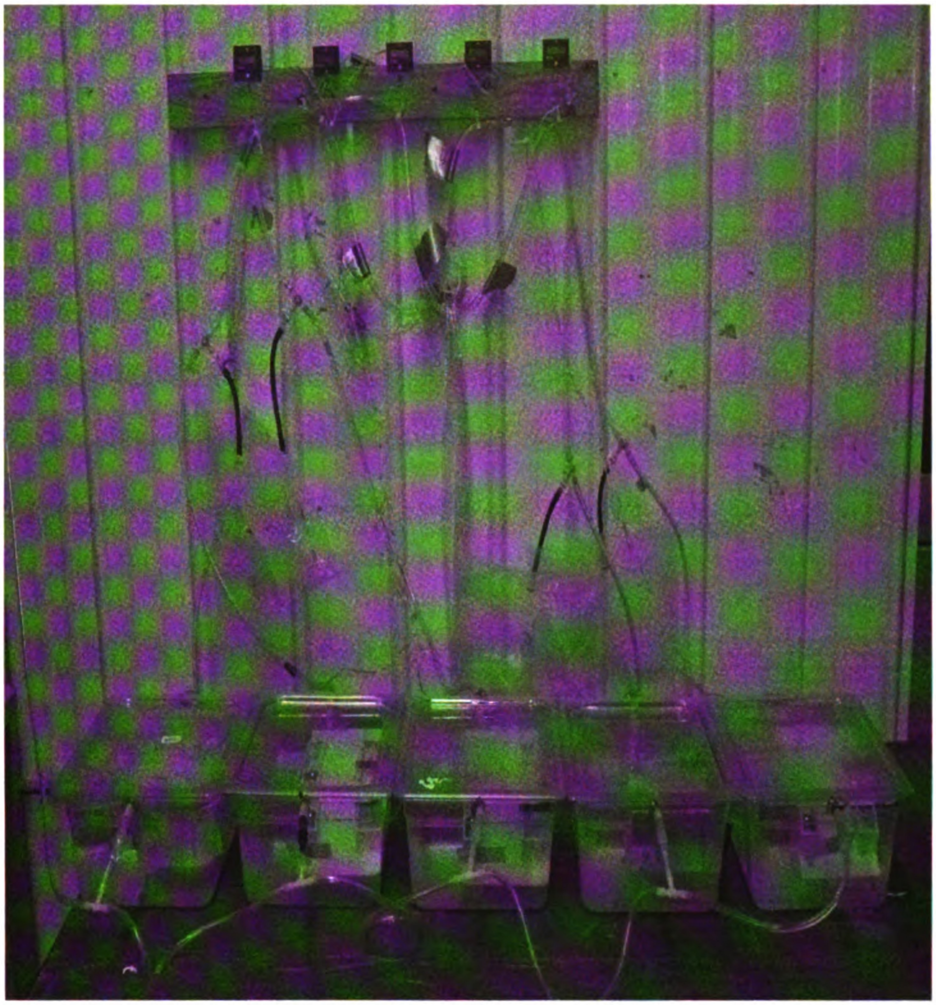


Figure 3.6 Tubing Connections to Wet-Tip Gas Meters

Chapter 4 Results and Discussion

The evaluation of a dewatered FPSW reconstituted with manure in an anaerobic digester was accomplished using three separate studies. The following sections discuss the results obtained from the respirometer, serum bottle and semi-continuous systems. All the data collected throughout the studies are also reported.

4.1 Batch Systems

The respirometer assay was started on March 29, 2009, and ran for 29 days in total. The serum bottle assay was initiated on March 10, 2010, and was discontinued after 77 days.

4.1.1 Respirometer Assay Results

Although pH of the diluted sludge was relatively low (pH 4.7), after mixing with seed and manure, it was adequate at the beginning and end of for all treatments (Appendix A, Table A.3). Alkalinity was also adequate at the beginning and end of digestion (Appendix A, Table A.1, Table A.2, Table A.4).

Ammonia varied between treatments; however, no effect on cumulative biogas yield is expected based on the literature as levels are adequate but not toxic (Table 4.1 and Appendix A, Table A.1, Table A.2). The amount of ammonia in the "Seed, Cow Manure" flask was higher than needed for the COD but not toxic. However, this level was near optimal for several of the manure, diluted sludge blends.

COD and VS destruction ranged from 22-27% when cow manure and sludge were combined and was lower when only manure was digested (Table 4.2). Trends associated with soluble COD destruction were not observed (Appendix A, Table A.9).

Table 4.1 Ammonia and COD/Ammonia Before and After Digestion

Treatment	Ammonia (mg)		COD/Ammonia	
	Initial	Final	Initial	Final
Seed	235	259	20:1	17:1
Seed, Cow Manure	323	390	24:1	16:1
Seed, Diluted Sludge	229	299	31:1	19:1
Seed, 1:1 Cow Manure: Diluted Sludge	340	449	30:1	17:1
Seed, 2:1 Cow Manure: Diluted Sludge	413	544	34:1	19:1
Seed, 1:2 Cow Manure: Diluted Sludge	285	365	30:1	19:1

Table 4.2 Respirometer COD and VS Destruction

Treatment	COD				VS			
	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction
Seed	4,591	4,428	163	4	3,378	2,891	487	14
Seed, Cow Manure	7,613	6,346	1,267	17	5,410	4,212	1,198	22
Seed, Diluted Sludge	7,166	5,753	1,413	20	4,704	3,615	1,089	23
Seed, 1:1 Cow Manure: Diluted Sludge	10,294	7,564	2,730	27	6,728	4,865	1,863	28
Seed, 2:1 Cow Manure: Diluted Sludge	14,235	10,351	3,884	27	8,546	6,280	2,266	27
Seed, 1:2 Cow Manure: Diluted Sludge	8,678	6,809	1,869	22	5,689	4,170	1,519	27

The percentage of methane in the biogas (Figure 4.1) was similar across all treatments (average approximately 68 - 78%). However, the "Seed" and "Seed 2:1 Cow Manure: Diluted Sludge" showed an obvious delay. Table 4.3 shows the biogas production represented as mL of biogas per gram of substrate VS destructed. Cow manure co-digested with diluted sludge at a 1:2 and 2:1 ratio produced more biogas per g VS destroyed than the digestion of manure alone. However, biogas yield from co-digestion of cow manure and diluted sludge at a 1:1 ratio did not produce more biogas compared with the digestion of manure or diluted sludge alone. Figure 4.2 shows a graph of the total biogas production over time.

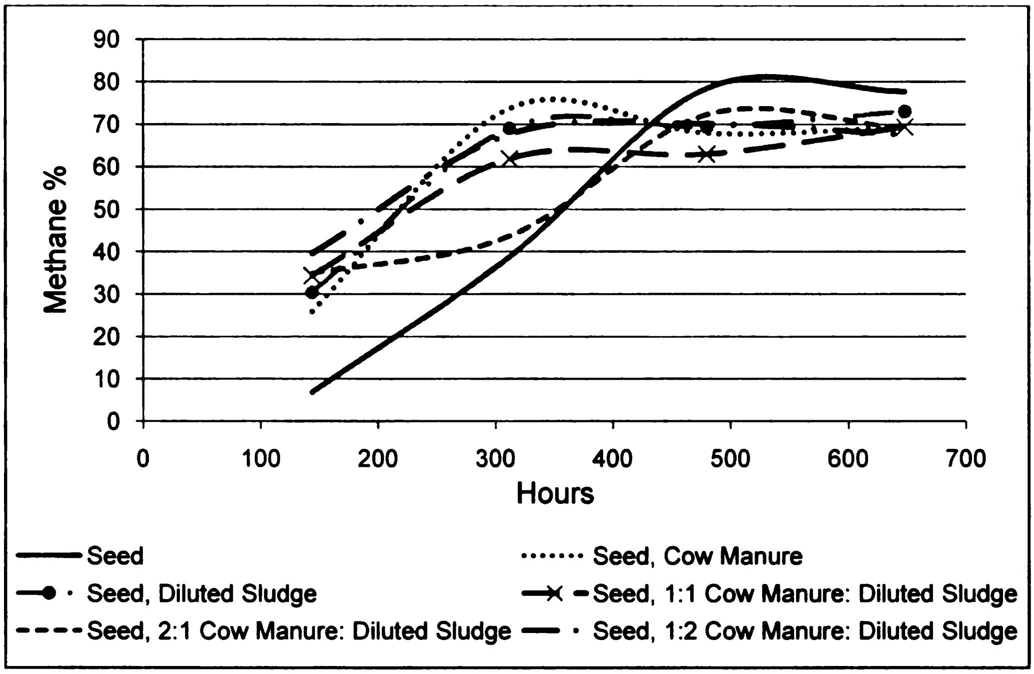


Figure 4.1 Respirometer Biogas Methane Content

Table 4.3 Respirometer Biogas per g COD and g VS Destroyed

Treatment	Total Gas Produced (mL)	COD Destroyed (mg)	Total Gas Produced / g COD Destroyed (mL/g)	VS Destroyed (mg)	Total Gas Produced / g VS Destroyed (mL/g)
Seed	135	163	830	487	280
Seed, Cow Manure	897	1,267	710	1,198	750
Seed, Diluted Sludge	911	1,413	640	1,089	840
Seed, 1:1 Cow Manure: Diluted Sludge	1,291	2,730	470	1,863	690
Seed, 2:1 Cow Manure: Diluted Sludge	1,839	3,884	470	2,266	810
Seed, 1:2 Cow Manure: Diluted Sludge	1,459	1,869	780	1,519	960

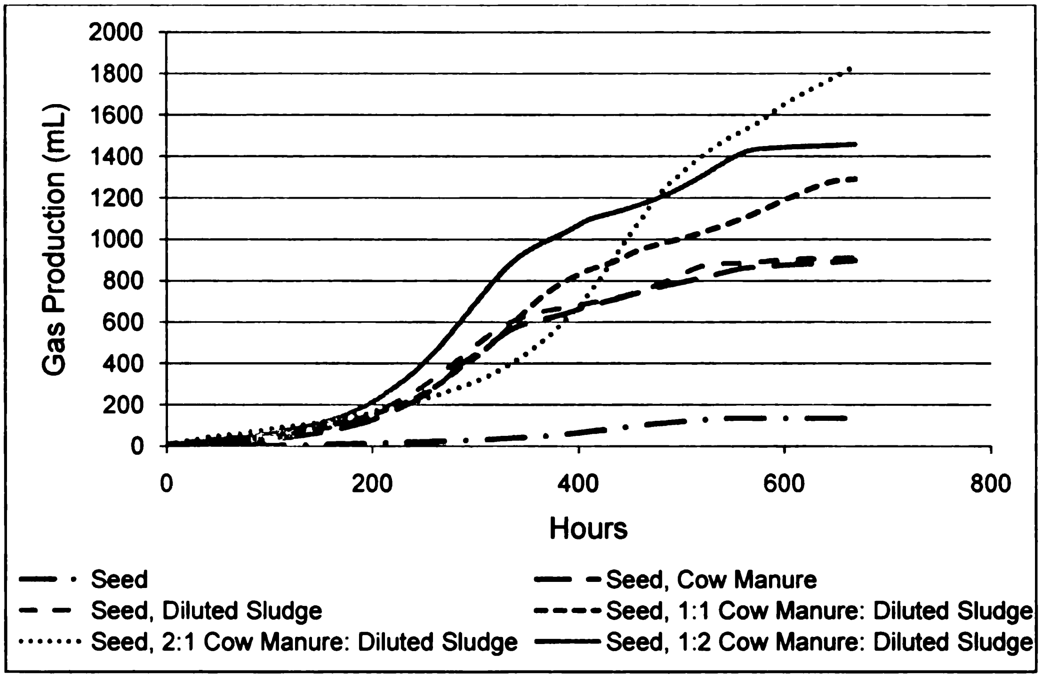


Figure 4.2 Respirometer Cumulative Biogas Volume

Both the diluted sludge and manure showed a very similar amount of normalized gas production (Table 4.4). However, accounting for the 10 fold dilution, the energy potential from the FPSW was an order of magnitude higher than the manure.

In the manure and diluted sludge blended flasks, the "Seed 1:1 Cow Manure: Diluted Sludge" and "Seed 2:1 Cow Manure: Diluted Sludge" flasks produced 23% and 24%, respectively, less biogas than predicted by adding the individual levels of each constituent (seed, cow manure and diluted sludge). This may indicate that the higher level of manure resulted in an antagonistic impact (Table 4.5). However, the "Seed, 1:2 Cow Manure: Diluted Sludge" flask had an actual gas production that was 13% higher than the addition of the individual components indicating a synergistic relationship. Interestingly, this trend matches the quantity of manure. The worst, middle, and best gas production resulted in the flasks with 144, 72, and 36 mL of manure, respectively.

Table 4.4 Normalized Respirometer Energy Potential

Component	Normalized Gas Produced (mL)	Original Volume after Dilution+ (mL)	Biogas/ Dilute Sample (m^3/L)	Dilution Ratio	Biogas/ Original Sample (m^3/L)
Cow Manure	762	72	0.011	1:1	0.011
Diluted Sludge*	776	72	0.011	10:1	0.108

* Based on "Seed, Diluted Sludge" minus "Seed"

+At experimental Temperature (35°C) and Standard Pressure

Table 4.5 Respirometer Biogas Potential

Treatment	Seed (mL)	Manure (mL)	Diluted Sludge (mL)	Expected Gas (mL)	Measured Gas (mL)	% Difference
Seed, 2:1 Blend	135	1,524	776	2,435	1,839	-24
Seed, 1:1 Blend	135	762	776	1,673	1,291	-23
Seed, 1:2 Blend	135	381	776	1,292	1,457	13

Results from this assay indicated that blending FPSW with manure has the potential to significantly increase gas production. However, the ratio of the blend appears to be important.

4.1.2 Serum Bottle Assay Results

Although pH of the sludge was relatively low (pH 5.81), once the treatments were mixed with seed and manure, it was adequate at the beginning and end of digestion (Appendix B, Table B.3). The "Diluted Sludge" treatment had a slightly low pH before and after digestion of 5.76 and 6.25, respectively. Alkalinity was adequate at the beginning and end of digestion (Appendix B, Table B.1, Table B.2, Table B.4).

Ammonia varied between treatments; however, no effect on cumulative biogas yield is expected based on the literature as levels are adequate but not toxic (Appendix B, Table B.1, Table B.2 and Table B.6). The "Seed, 2:1 Cow Manure: Diluted Sludge" treatment had a slightly high post-digestion ammonia value of 813 mg/L-N although no toxicity issues were suspected.

The optimum C to N ratio for anaerobic digestion is 20-30:1 (Bouallagui et al., 2004). Taking COD as a representative value of C, all of the blended treatments were initially in this range apart from the 1:1 blend ratio which was slightly low . However, after digestion, C to N ratio values were all lower than recommended in literature for the blended treatments as shown in Table 4.6.

Table 4.6 COD:N:P Before and After Digestion

Treatment	Nitrogen (mg)		Phosphorus (mg)		COD:N:P	
	Initial	Final	Initial	Final	Initial	Final
Seed	105	106	36	29	75:3:1	57:4:1
Seed, Cow Manure	161	153	42	41	73:4:1	52:4:1
Seed, Diluted Sludge	135	143	36	38	90:4:1	60:4:1
Cow Manure	69	65	6	7	199:12:1	77:9:1
Diluted Sludge	33	20	3	4	309:10:1	232:6:1
Seed, 2:1 Cow Manure: Diluted Sludge	223	255	70	64	76:3:1	50:4:1
Seed, 1:1 Cow Manure: Diluted Sludge	255	190	53	48	79:5:1	56:4:1
Seed, 1:2 Cow Manure: Diluted Sludge	144	176	47	41	77:3:1	64:4:1
Seed, 1:4 Cow Manure: Diluted Sludge	154	150	43	38	80:4:1	62:4:1
Seed, 1:6 Cow Manure: Diluted Sludge	105	145	44	38	NA	66:4:1

COD destruction increased when cow manure and diluted sludge were combined in the 2:1 and 1:1 blended treatments compared to when manure-only was digested. The "Seed, 1:2 Cow Manure: Diluted Sludge" and "Seed, 1:4 Cow Manure: Diluted Sludge" treatments were not statistically different from manure alone. The "Seed, 1:6 Cow Manure: Diluted Sludge" treatment data was not available due to an error in COD testing. It is suspected that an error also occurred for the initial COD of the "Seed" treatment (2,685 mg) as it resulted in a high destruction value in relation to the quantity of gas produced. VS destruction for the "Seed, 2:1 Cow Manure: Diluted Sludge" and "Seed, 1:1 Cow Manure: Diluted Sludge" treatments were higher than when manure was digested alone. The 1:2, 1:4 and 1:6 treatments were not statistically different compared to manure-only digestion (Table 4.7). Trends associated with soluble COD destruction were not observed (Appendix B, Table B.7).

Table 4.7 Serum Bottle COD and VS Destruction

Treatment	COD				VS			
	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction
Seed	2,685	1,624	1,061	40	1,356	979	377	28
Seed, Cow Manure	3,058	2,135	923	30	1,814	1,242	572	32
Seed, Diluted Sludge	3,214	2,266	948	29	1,876	1,210	666	36
Cow Manure	1,193	540	653	55	584	344	240	41
Diluted Sludge	1,044	843	201	19	572	280	292	51
Seed, 2:1 Cow Manure: Diluted Sludge	5,312	3,222	2,090	39	3,085	1,953	1,132	37
Seed, 1:1 Cow Manure: Diluted Sludge	4,179	2,669	1,510	36	2,482	1,517	965	39
Seed, 1:2 Cow Manure: Diluted Sludge	3,606	2,619	987	27	2,084	1,415	669	32
Seed, 1:4 Cow Manure: Diluted Sludge	3,437	2,388	1,049	31	1,915	1,343	572	30
Seed, 1:6 Cow Manure: Diluted Sludge	1,877	2,496	NA	NA	1,977	1,347	630	32

The percentage of methane in the biogas (Figure 4.3) was similar across all blended treatments (average 57 - 59%). The "Seed, Cow Manure", "Seed, Diluted Sludge" and "Cow Manure" treatments had similar biogas methane content. The "Diluted Sludge" treatment had low methane content.

Figure 4.4 shows a graph of the total biogas production over time.

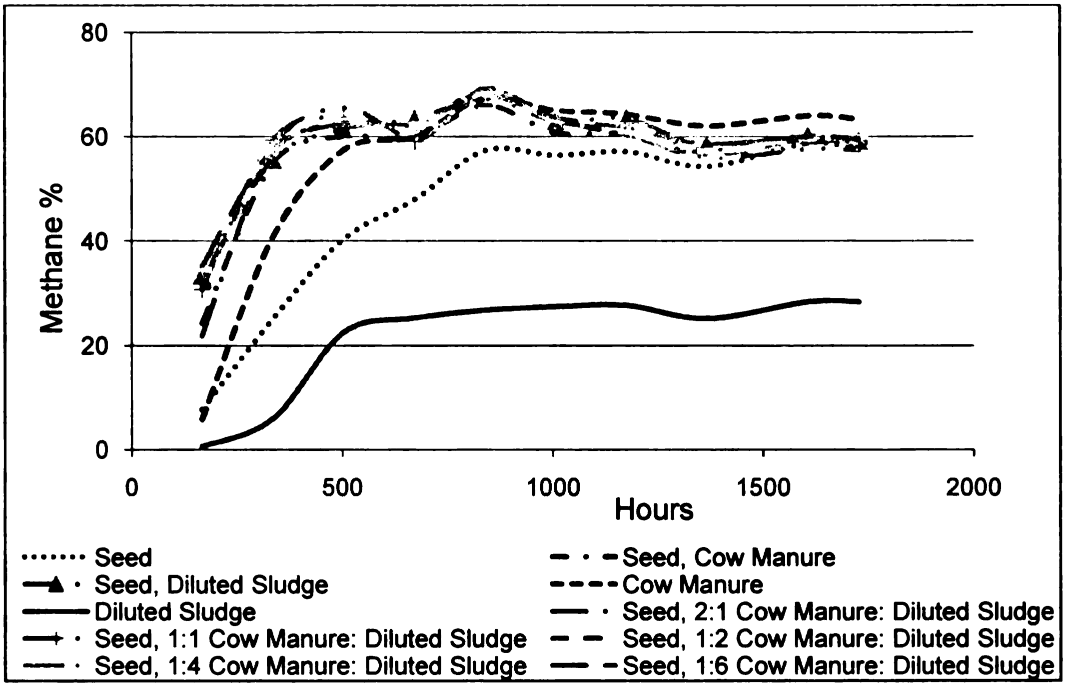


Figure 4.3 Serum Bottle Biogas Methane Content

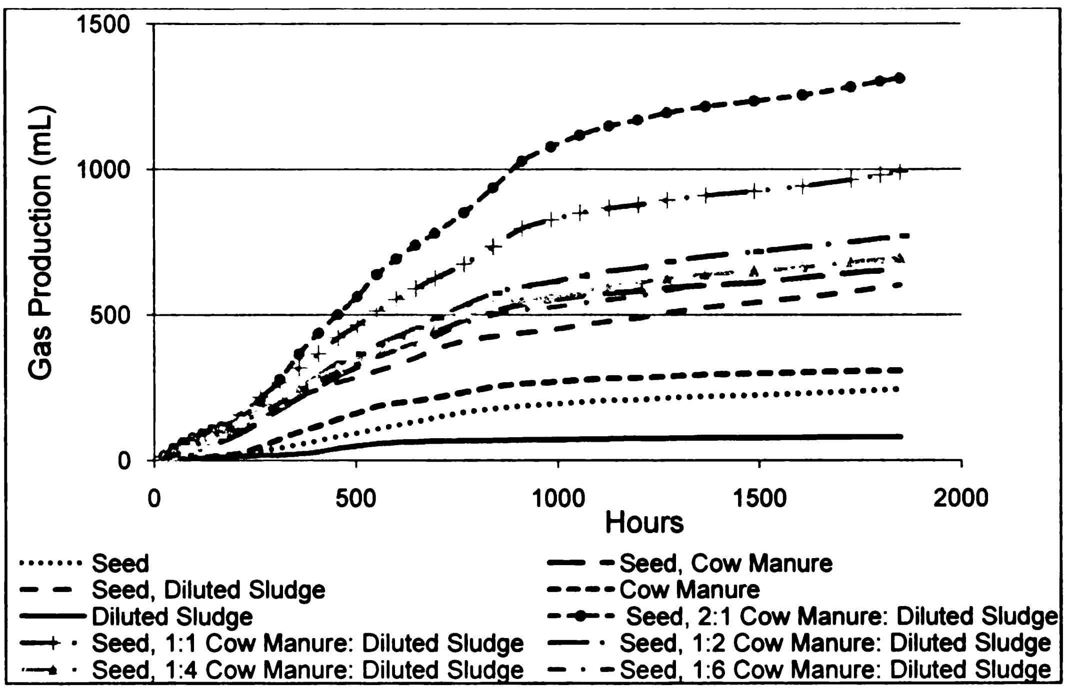


Figure 4.4 Serum Bottle Cumulative Biogas Volume

Total biogas production represented as mL of biogas per gram of substrate VS destructed is shown in Table 4.8. A one-way ANOVA, using Tukey HSD with 95% confidence limits, was run in order to see if there were any significant differences in individual treatment blend ratios and between blend ratios and the digestion of manure alone. The analysis was run on both a total gas produced per g COD destroyed (mL/g) basis and total gas produced per g VS destroyed (mL/g) basis. For both COD and VS parameters, there was no statistical difference found between the different treatment blends and the digestion of manure alone. Similar results were found in the comparison of individual treatment blends with again no statistical difference found. Table 4.9 shows a portion of the statistical output.

Table 4.8 Serum Bottle Biogas per g COD and g VS Destroyed

Treatment	Total Gas Produced (mL)	COD Destroyed (mg)	Total Gas Produced / g COD Destroyed (mL/g)	VS Destroyed (mg)	Total Gas Produced / g VS Destroyed (mL/g)
Seed	246	1,061	230	377	650
Seed, Cow Manure	656	923	710	572	1,150
Seed, Diluted Sludge	602	948	640	666	900
Cow Manure	308	653	470	240	1,280
Diluted Sludge	80	201	400	292	270
Seed, 2:1 Cow Manure: Diluted Sludge	1,312	2,090	630	1,132	1,160
Seed, 1:1 Cow Manure: Diluted Sludge	990	1,510	660	965	1,030
Seed, 1:2 Cow Manure: Diluted Sludge	769	987	780	669	1,150
Seed, 1:4 Cow Manure: Diluted Sludge	695	1,049	660	572	1,220
Seed, 1:6 Cow Manure: Diluted Sludge	660	NA	NA	630	1,050

Table 4.9 Statistical Output

Treatment Blend	Mean g VS	Mean g COD	Std. Dev. g VS	Std. Dev. g COD	Std. Error g VS	Std. Error g COD	Var. g VS	Var. g COD
Average	1.123	0.687	0.093	0.100	0.024	0.029	0.009	0.010
2:1	1.160	0.627	0.036	0.021	0.021	0.012	0.001	0.000
1:1	1.027	0.657	0.045	0.015	0.026	0.009	0.002	0.000
1:2	1.153	0.797	0.070	0.159	0.041	0.092	0.005	0.025
1:4	1.223	0.667	0.107	0.060	0.062	0.035	0.011	0.004
1:6	1.050	.	0.010	.	0.006	.	0.000	.

Once again, both the sludge and manure showed a somewhat similar amount of normalized biogas production (Table 4.10). However, accounting for the 10 fold dilution, the energy potential from the sludge was significantly higher than the manure.

Table 4.10 Normalized Serum Bottle Energy Potential

Component	Normalized Gas Produced (mL)	Original Volume after Dilution+ (mL)	Biogas/ Dilute Sample (m^3/L)	Dilution Ratio	Biogas/ Original Sample (m^3/L)
Cow Manure	410	25	0.062	1:1	0.062
Diluted Sludge*	356	25	0.054	10:1	0.539
Diluted Sludge**	334	25	0.051	10:1	0.506

* Based on "Seed, Sludge" minus "Seed"

**Based on "Seed, 1:1 Cow Manure: Diluted Sludge" minus "Seed, Manure"

+At experimental Temperature (35 °C) and Standard Pressure

Table 4.11 shows the biogas predicted by adding the individual levels of each constituent (seed, cow manure, and diluted sludge) and the actual measured biogas from the study. The co-digestion treatment effects seem to be additive, showing no true synergistic or antagonistic response as the differences between the expected and measured gas are minimal overall.

Table 4.11 Serum Bottle Biogas Potential

Treatment	Seed (mL)	Manure (mL)	Sludge (mL)	Expected Gas (mL)	Measured Gas (mL)	%
Seed, 2:1	246	820	356	1,422	1,312	-8%
Seed, 1:1	246	410	356	1,012	990	-2%
Seed, 1:2	246	205	356	807	769	-5%
Seed, 1:4	246	103	356	704	695	-1%
Seed, 1:6	246	68	356	670	660	-2%

4.1.3 Discussion of Batch Assays

For the respirometer study, further examination of Figure 4.2 deemed that the assay was not run for a sufficient amount of time as gas production was still increasing when the flasks were discontinued. Therefore, the true effect of the diluted sludge in co-digestion was not realized. However the respirometer study did indicate that the reconstitution of FPSW with manure could possibly offer a synergistic relationship at certain blended ratios and so further examination was warranted.

The serum bottle assay ran for over 70 days with the intention of finding the optimum blend ratio of the FPSW with cow manure in co-digestion to give a possible synergistic effect as already indicated by the respirometer assay. Initially, the optimum ratio was thought to be related to the quantity of the manure present in the mixture, with the lowest amount providing the best biogas potential. Therefore, the objective of the serum bottle assay involved investigating even lower manure-to-diluted sludge ratios (1:4, 1:6) while also looking at the conditions already tested (2:1, 1:1 and 1:2).

Gas production results of the serum bottle assay did not show evidence of a synergistic or antagonistic relationship in co-digestion. Results showed that the methane produced from the diluted sludge when mixed with cow manure is additive. However, by volume, the FPSW has approximately

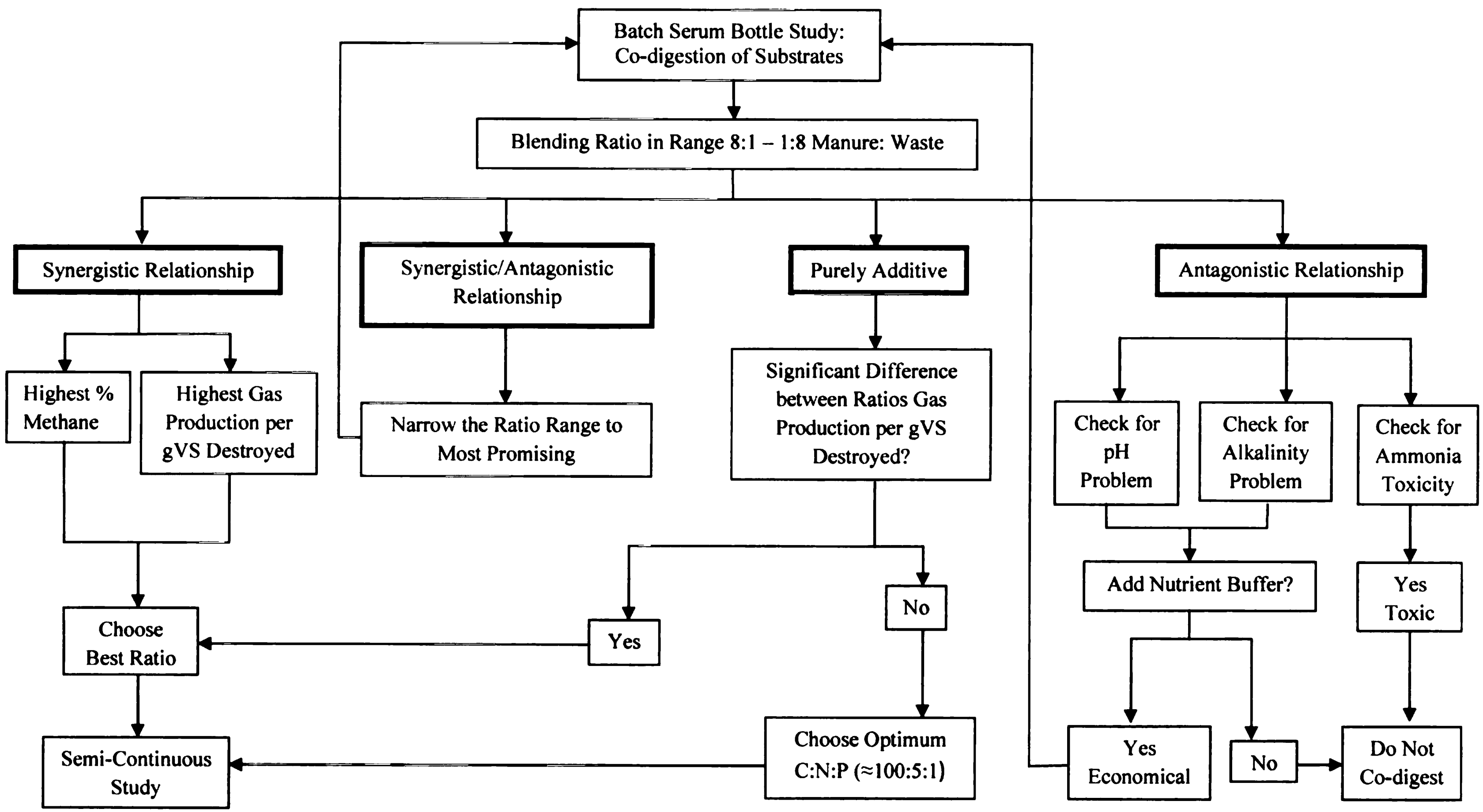
10 times the energy potential of manure. Further, the FPSW alone does not have the addition of nutrients and a buffering system. The addition of manure provides an adequate amount of both.

4.2 Semi-continuous System

The results from the batch assays showed that the FPSW contained a great deal of embedded energy; however no optimum blend in co-digestion was recognized. This was signified by finding no statistical difference between treatment blends in terms of g COD and g VS destruction and also by the relatively small difference between predicted and measured gas production levels. To verify the utility of the FPSW's reconstitution with manure it was important to examine the substrate in a semi-continuous operation that would more accurately represent real-life digestion. The semi-continuous digesters were implemented in the form of continuously stirred tank reactors (CSTR) as might occur on a real-life centralized farm digester.

Since gas production was found to be additive, the best approach to the semi-continuous analysis was optimizing the carbon to nitrogen ratio (C:N). The rationale behind this decision was based on the decision flow chart illustrated in Figure 4.5. This flow chart was carefully created based on past experience of running laboratory-scale batch BMP assays of various wastes. Since no significant statistical difference was found for biogas produced per g VS destroyed for the different blends, it was advocated that the optimization of C:N should be made for a further semi-continuous study. Although the decision flow chart shown in Figure 4.5 was developed for the purposes of this research, it can be used for any potential co-substrate BMP study to make well informed, educated decisions.

Figure 4.5 Decision Flow Chart



4.2.1 Semi-Continuous Study Results and Discussion

The semi-continuous reactors were started on June 17, 2010 and ran for three SRT's, 45 days in all. The study ceased on August 1, 2010.

On day 13 of the first SRT, Reactor 1, containing just seed inoculum, was discontinued as gas production had ceased. This signified that the addition of seed in the start up of the other reactors was negligible in terms of contributing to gas production. The initial and final analysis characteristics of Reactor 1 are shown in Table 4.12.

Table 4.12 Reactor 1 Analysis

Reactor 1	Day 1	Day 13
pH	7.60	7.42
TS (mg)	33,750	6,834
VS (mg)	19,226	3,830
COD (mg)	34,376	7,300

Reactor 2, containing manure with acclimated seed, was replaced with a substitute reactor of same composition on day 13 of the first SRT due to a malfunction. No effect on pH, COD, alkalinity, TS/VS or gas production was evident as a result of the changeover. The reactor was fed on a daily basis without disruption. The COD and VS destruction of the new Reactor 2 returned to normal after the first few days of the new start-up. Reactors 3 and 4, containing FPSW/DI water with seed and FPSW/manure with seed, respectively, ran for the 3 SRT's uninterrupted. Both reactors were fed daily for the entirety of the operational run.

From day 12 of the second SRT and onwards, no sample was removed or fed from Reactor 5 that contained FPSW/manure with seed. This reactor was a duplicate to be run in conjunction with Reactor 4, comprising of the same constituents. Concerns over the quantity of FPSW remaining to complete the study, as it unexpectedly was no longer available, forced the decision to discontinue feeding of the reactor. However, the reactor was left to run for the remainder of the second and third SRT's and gas production was recorded.

The pH of all reactors were within the acceptable range at the beginning and end of their respective digestion run times (Appendix C, Table C.3). However, the pH of reactor 3 was slightly low throughout. This can be attributed to the FPSW's initial pH being low (Chapter 3, Table 3.1, Table 3.2, Table 3.3). The alkalinity of each reactor was also measured for the 3 SRT's. Reactors 2 and 4 showed no imminent problems as values remained at a suitable high range throughout (Appendix C, Table C.4). The possibility of denitrification was suspected for Reactor 3. This was illustrated by increasing alkalinity and a corresponding rise in pH, both of which characterized the denitrification process (Henze et al., 2002). Measurements of COD and TS/VS were made on the effluent approximately three times a week (Appendix C, Table C.5 and Table C.3, C.7, C.8 and C.9).

Figure 4.6 shows the overall gas production for the full 45 days of operation. The graph shows that Reactor 4, containing the optimum blended C:N ratio of manure and FPSW, significantly outperformed the rest of the reactors. In fact, Reactor 4 (106,600 mL) generated more than twice as much biogas as Reactor 2 (45,500 mL) that contained cow manure alone. Reactor 3, containing just FPSW, performed poorly in terms of gas production although this was to be as expected due to deficiencies in nutrient content. As anticipated, only a very small quantity of biogas was produced by Reactor 1 containing just seed inoculum. Reactor 5, a duplicate of Reactor 4 which was discontinued in the second SRT, showed promising signs of high gas production although it did seem to have a longer lag time than its replicate.

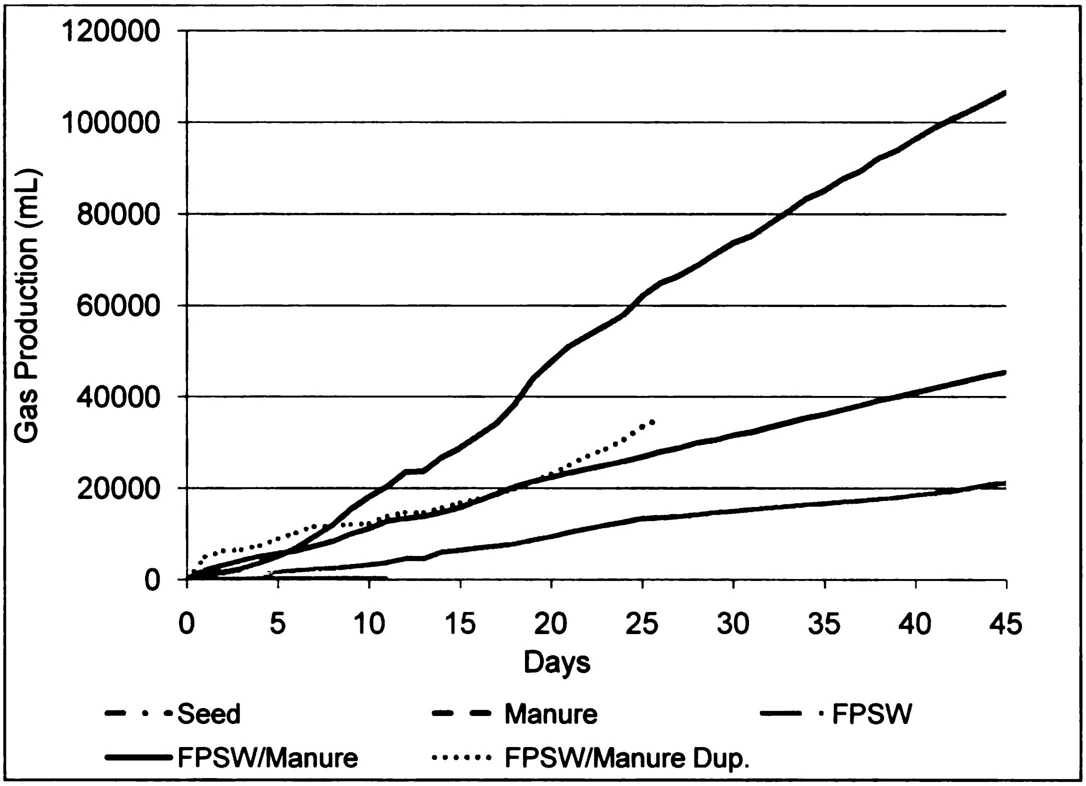


Figure 4.6 Semi-Continuous Cumulative Biogas Volume

Examining the biogas produced per individual SRT, similar results were observed. Figures 4.7, 4.8 and 4.9 show the gas production for the first, second and third SRT's, respectively. With identical 2 L reactor working volumes, the blend of FPSW/manure produced 2.36 times as much biogas as the reactor containing only manure for the third SRT. Again, this third SRT represented a stabilized system. For the first and second SRT's, the FPSW/manure also outperformed the manure digester producing 1.82 and 2.84 times as much biogas. Reactor 3 containing FPSW consistently produced the lowest yield of biogas for each SRT.

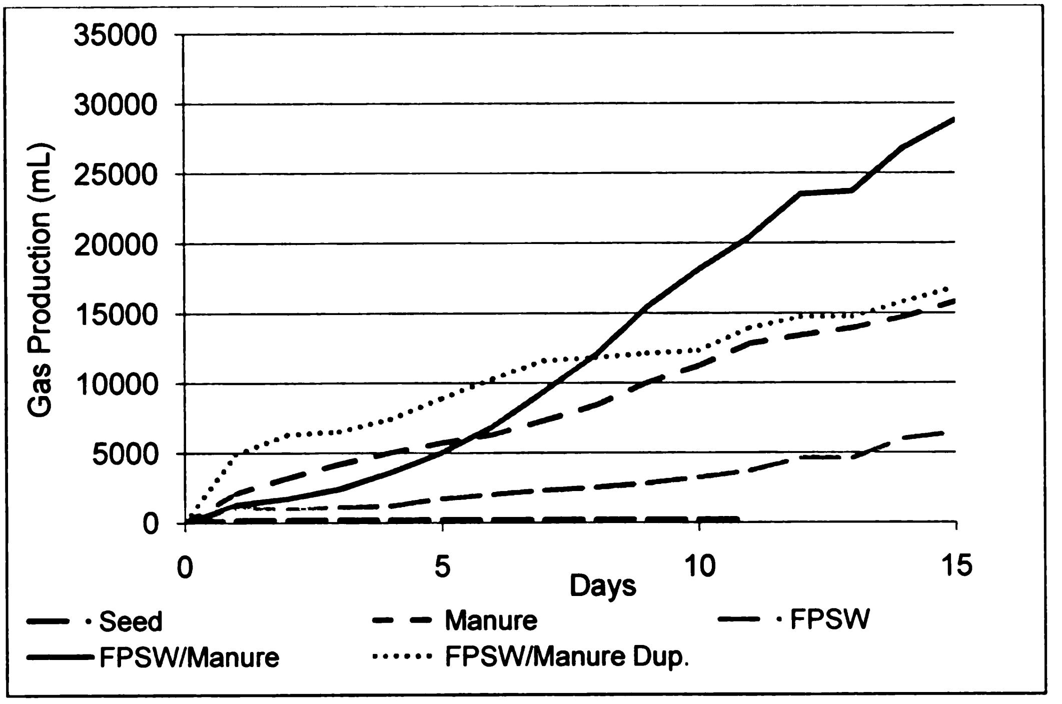


Figure 4.7 SRT 1 Biogas Production

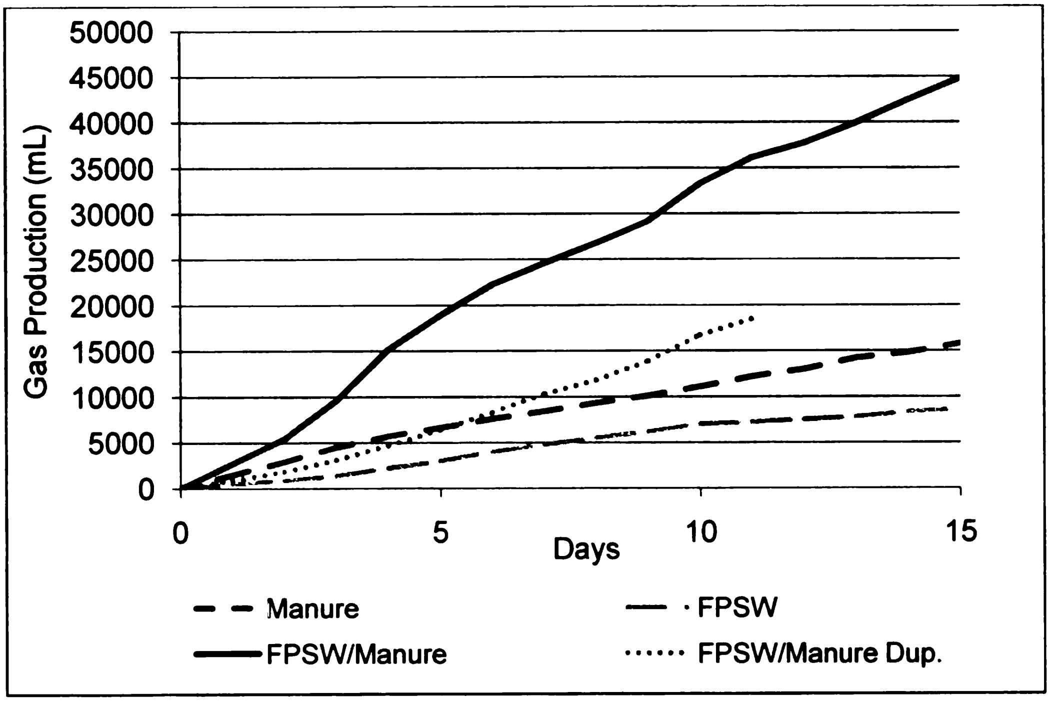


Figure 4.8 SRT 2 Biogas Production

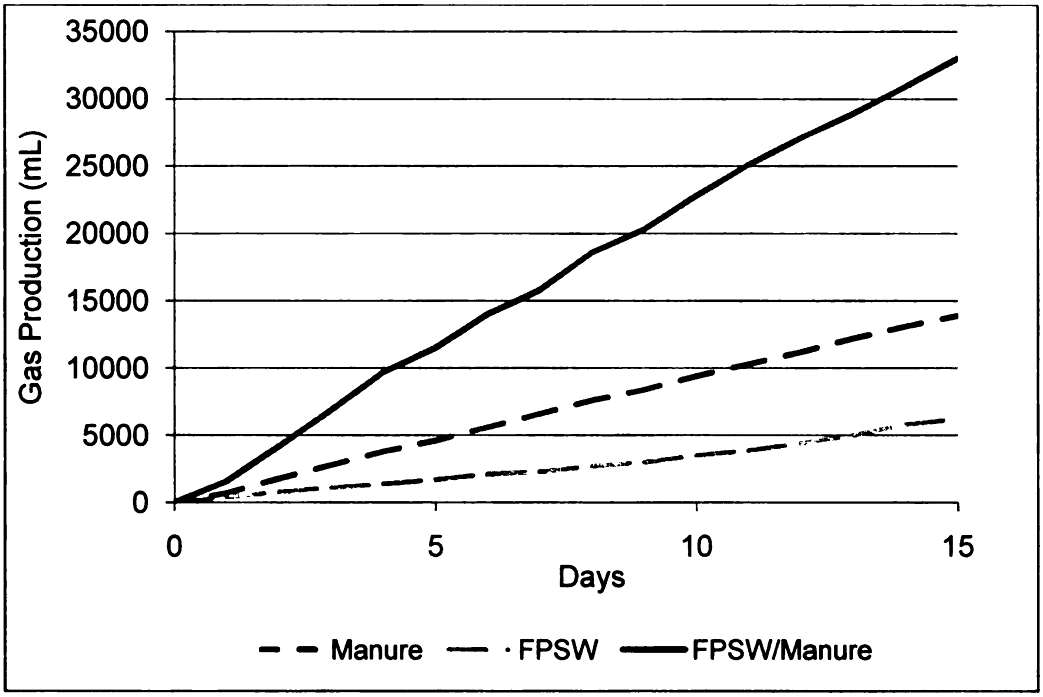


Figure 4.9 SRT 3 Biogas Production

The percentage methane of each reactor from day 26 to day 45 is represented in Figure 4.10. Average values for methane content are determined in Table C.10 in Appendix C. The reactor containing manure-only had the highest percentage methane in the biogas averaging 62%. This was followed closely by the FPSW/manure reactor that contained 58% methane in the biogas. Reactor 3, containing FPSW alone, had substantially lower methane content at approximately 35%, yet again showing the system lacked the sufficient nutrients. Although discontinued at an earlier stage, Reactor 5 had already reached a promising biogas methane content of 58%.

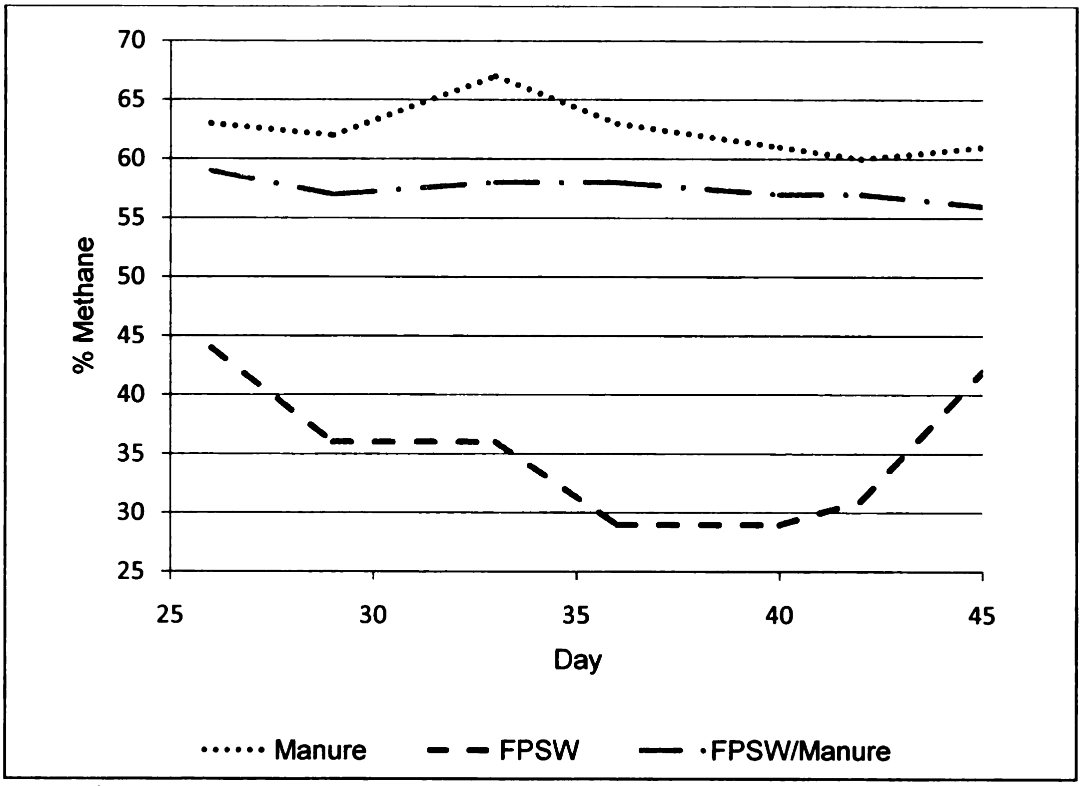


Figure 4.10 Semi-Continuous Biogas Methane Content

COD and VS destruction for the manure reactor (Reactor 2), the FPSW reactor (Reactor 3) and the FPSW/manure reactor (Reactor 4) were examined for all 3 SRT's (Table 4.13, Table 4.14, Table 4.15). Using the third SRT as that representing a steady and stabilized system, the destruction rates could be compared.

For COD destruction, Reactor 2, containing manure and Reactor 4, containing FPSW/manure, were very similar averaging at 33% and 34%, respectively. Reactor 3, containing FPSW alone, was drastically lower at averaging at approximately 7%. Destruction data concerning Reactor 5 is shown in Appendix C, Table C.11, although the relevant percentages are lower as it did not reach the third SRT.

Taking the third SRT as representing a steady and stabilized system, the VS destruction was highest in Reactor 4, containing the optimized FPSW/manure blend, averaging approximately 45%. Reactor 2, containing manure, and Reactor 3, containing FPSW alone, were similar in terms of VS destruction averaging 31 and 33% respectively. VS destruction for Reactor 5 averaged approximately 41% for the duration of its run (Appendix C, Table C.11).

Table 4.13 Reactor 2 COD and VS Destruction

Reactor 2	COD				VS			
Day	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction
2	3,999	3,250	749	19	2,431	1,235	1,195	49
6	3,999	3,663	336	8	2,431	1,932	498	20
8	3,999	3,757	242	6	2,431	1,975	456	19
12	3,999	3,094	905	23	2,431	1,818	612	25
16	3,999	4,212	NA	NA	2,431	2,002	429	18
19	3,999	3,985	15	0	2,431	2,051	380	16
21	3,999	3,549	450	11	2,431	1,827	604	25
23	3,999	3,403	596	15	2,431	1,821	610	25
26	3,999	2,860	1,139	28	2,431	1,698	732	30
28	3,999	2,678	1,321	33	2,431	1,606	824	34
30	3,999	2,782	1,217	30	2,431	1,600	831	34
33	3,999	2,623	1,376	34	2,431	1,541	889	37
35	3,999	2,545	1,454	36	2,431	1,520	910	37
37	3,999	2,603	1,396	35	2,431	2,003	428	18
40	3,999	2,880	1,120	28	2,431	1,894	537	22
42	3,999	2,545	1,454	36	2,431	1,608	823	34
44	3,999	2,730	1,269	32	2,431	1,617	814	33

Table 4.14 Reactor 3 COD and VS Destruction

Reactor 3	COD				VS			
Day	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction
2	12,162	11,141	1,021	8	6,575	3,760	2,816	43
6	12,162	11,564	598	5	6,575	4,074	2,502	38
8	12,162	12,220	NA	NA	6,575	4,264	2,312	35
12	12,162	11,323	839	7	6,575	4,230	2,345	36
16	12,162	11,291	871	7	6,575	4,141	2,434	37
19	12,162	11,388	774	6	6,575	4,449	2,126	32
21	12,162	11,518	644	5	6,575	4,734	1,842	28
23	12,162	12,766	NA	NA	6,575	4,629	1,946	30
26	12,162	12,188	NA	NA	6,575	4,356	2,219	34
28	12,162	11,408	754	6	6,575	4,249	2,327	35
30	12,162	12,162	0	0	6,575	4,470	2,105	32
33	12,162	11,538	624	5	6,575	4,342	2,233	34
35	12,162	11,063	1,099	9	6,575	4,534	2,041	31
37	12,162	11,720	442	4	6,575	4,268	2,308	35
40	12,162	11,499	663	5	6,575	4,389	2,186	33
42	12,162	11,148	1,014	8	6,575	4,335	2,241	34
44	12,162	10,277	1,885	16	6,575	4,435	2,140	33

Table 4.15 Reactor 4 COD and VS Destruction

Reactor 4	COD				VS			
Day	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction
2	16,322	12,233	4,089	25	8,052	4,173	3,879	48
6	16,322	12,747	3,575	22	8,052	4,841	3,211	40
8	16,322	13,176	3,146	19	8,052	4,732	3,320	41
12	16,322	11,863	4,459	27	8,052	4,953	3,099	38
16	16,322	12,428	3,894	24	8,052	4,719	3,333	41
19	16,322	12,877	3,445	21	8,052	4,797	3,255	40
21	16,322	11,778	4,544	28	8,052	4,739	3,313	41
23	16,322	10,849	5,473	34	8,052	4,395	3,657	45
26	16,322	11,869	4,453	27	8,052	4,616	3,436	43
28	16,322	10,738	5,584	34	8,052	4,332	3,720	46
30	16,322	10,355	5,967	37	8,052	4,018	4,034	50
33	16,322	11,518	4,804	29	8,052	4,447	3,605	45
35	16,322	10,712	5,610	34	8,052	4,495	3,557	44
37	16,322	10,446	5,876	36	8,052	4,271	3,781	47
40	16,322	10,810	5,512	34	8,052	4,373	3,679	46
42	16,322	11,362	4,960	30	8,052	5,047	3,005	37
44	16,322	10,602	5,720	35	8,052	4,511	3,541	44

Tables 4.16 and 4.17 show the reactors' biogas produced per g COD and g VS destroyed, respectively. Figures were based on the data collected over the final SRT to ensure analysis was relevant for a stabilized system. Reactor 2, containing only manure, had the highest gas production on both a g COD and g VS destruction basis. Interestingly, the optimized blend of FPSW/manure (Reactor 4) demonstrated the lowest gas production per g COD destroyed. On a g VS destruction basis, Reactor 4 was only half as efficient as Reactor 2 in terms of gas production. However; although less efficient, Reactor 4 was still vastly outperforming Reactor 2 in terms of overall

biogas production. This signifies that hydrolysis of some of the solids associated with the FPSW was not possible in the chosen SRT of 15 days. Though evidence suggests a low rate conversion, Reactor 4 still produced a quantity of biogas that indicated a synergistic relationship.

Table 4.16 Reactor Biogas per g COD Destroyed

SRT 3	Average Total Gas per day over SRT (mL)	Average COD Destroyed per day over SRT (g)	Total Gas Produced per g COD Destroyed (mL/g)
Reactor 2 Manure	868.75	1.33	653
Reactor 3 FPSW	387.50	0.82	473
Reactor 4 FPSW/Manure	2062.50	5.49	376

Table 4.17 Reactor Biogas per g VS Destroyed

SRT 3	Average Total Gas per day over SRT (mL)	Average VS Destroyed per day over SRT (g)	Total Gas Produced per g VS Destroyed (mL/g)
Reactor 2 Manure	868.75	0.75	1,158
Reactor 3 FPSW	387.50	2.18	178
Reactor 4 FPSW/Manure	2062.50	3.60	573

In evaluating the results of the semi-continuous reactors the percentage of the maximum theoretical methane yield was calculated using Equation 8. Equation 8 is based on the principle that 1 gram of COD destruction equals 395 mL CH_4 (Speece, 1996b). Table 4.18 shows the relevant values for each reactor as pertains to Equation 8. All the values presented were based on the third SRT to ensure estimates were made on a stabilized system. Reactor 2, containing manure, had a methane yield minimally greater than the calculated maximum theoretical methane available. Experimental error in COD analysis during digestion was allowed within a 10% range. Accounting for this, the actual methane yield was within range of maximum theoretical yield, however, Reactor 2 showed almost complete efficiency. Reactor 3, containing FPSW-alone, produced a smaller percentage yield of maximum theoretical of approximately 44%. This can be contributed to the higher carbon dioxide content existing in the biogas. Finally, the optimized blend in Reactor 4 was estimated to be yielding approximately 58% of the maximum theoretical methane available. More capacity, for example, increasing reactor SRT, may be required to enhance efficiency.

$$\%\ \text{Yield} = \frac{\text{Total Gas Production (mL)} \times \text{Methane Content (\%)}}{\text{COD Destoyed (g)} \times 395\left(\frac{\text{mL}}{\text{g}}\right)}$$

Equation 8.

Table 4.18 Reactor Values for Equation 8

SRT 3	Average COD Destroyed (g)	Total Gas Production* (mL)	Methane Content (%)
Manure	1.33	907	62
FPSW	0.82	405	35
FPSW/Manure	5.49	2,153	58

* Corrected for standard temperature (35°C) and pressure (STP)

Chapter 5 Conclusions and Future Research

Conducting the two initial batch BMP assays provided the starting point in the assessment of reconstituting the FPSW with manure. Results projected that digestion of substrates was additive and that the FPSW could potentially be an excellent co-substrate if reconstituted with manure at a farm digester. Culmination of the BMP studies resulted in the formation of a decision flow chart, Figure 4.5, to establish optimum co-substrate blending ratios and deciding if further studies of certain wastes are warranted. From this chart, a semi-continuous reactor study was designed based on an optimized C:N ratio.

The FPSW and manure blend with optimized C:N ratio performed exceptionally well in a semi-continuous system. With identical working volumes of 2 L, the blend of FPSW/manure produced approximately 2.19 times more methane than the reactor containing manure alone. The digestion of FPSW alone was found to be unsuccessful without the incorporation of additional nutrients, further advocating the concept of adding manure. Although perceived to be additive in the second BMP study, the reconstitution of the FPSW with manure showed true synergistic signs when studied at the semi-continuous phase. This was signified by the biogas production from the FPSW/manure blend being almost 1.6 times higher than the combination of the manure and FPSW reactors.

Introducing FPSW can generate greater revenue from higher energy production with no alteration to an existing digester's working volume. For the food processor, possible carbon credit gains may be obtained depending on district regulations. Renewable energy certificates can also be attained.

Tipping fees between farmer and food processor may also transpire although this benefit is currently market driven.

The digestion of the FPSW/manure optimized blend was shown to have a low conversion rate, in terms of biogas produced per g COD and g VS destroyed, as compared to that of the manure alone. Three potential reasons were identified as to why this low efficiency occurred. Initially, a problem was suspected in the hydrolysis of certain solids of the FPSW as pertaining to a short SRT of 15 days. With a longer retention time, better conversion rates may have been achieved. Another possibility was toxicity issues arising from the polymers in the FPSW which would have led to poor system performance and inhibition. Finally, sorption of trace nutrients may have occurred, again, through the presence of the polymers. The removal of minerals and micronutrients from the system would ensure lower conversion efficiency.

The research conducted in this study looked at the reconstitution of the FPSW with manure in a very applied manner, focusing on the science and microbiology behind the concept. However, logistical questions still remain unanswered. Future research should focus on an in-depth analysis of the costs associated with dewatering food wastes onsite. Concepts such as the cost benefits associated with shipping dewatered solids as opposed to the transportation of a slurry must be considered. By answering these logistical questions and providing an evaluation of the energy produced per volume of the food waste inserted in the digester, the feasibility of reconstitution at a digester can be determined.

APPENDICES

APPENDIX A

RESPIROMETER STUDY

Table A.1 Respirometer Concentrations before Digestion

Treatment	pH	Alkalinity (mg/L $CaCO_3$)	COD (mg/L)	Soluble COD (mg/L)	TS (mg/L)	TSS (mg/L)	VS (mg/L)	VSS (mg/L)	Ammonia (mg/L N)
Seed	8.15	2,600	7,063	4,750	8,503	6,100	5,197	4,400	361
Seed, Cow Manure	7.82	4,100	11,713	7,275	13,115	7,900	8,323	5,300	496
Seed, Diluted Sludge	7.77	2,950	11,025	4,975	11,173	7,700	7,237	4,100	353
Seed, 1:1 Cow Manure: Diluted Sludge	7.61	4,250	15,838	7,125	15,755	10,100	10,350	6,900	523
Seed, 2:1 Cow Manure: Diluted Sludge	7.51	5,100	21,900	9,388	20,120	12,400	13,147	8,500	635
Seed, 1:2 Cow Manure: Diluted Sludge	7.77	3,450	13,350	6,075	13,467	9,100	8,752	5,800	439

Table A.2 Respirometer Concentrations after Digestion

Treatment	pH	Alkalinity (mg/L $CaCO_3$)	COD (mg/L)	Soluble COD (mg/L)	TS (mg/L)	TSS (mg/L)	VS (mg/L)	VSS (mg/L)	Ammonia (mg/L N)
Seed	7.43	3,100	6,813	5,088	7,403	3,700	4,448	2,300	399
Seed, Cow Manure	7.31	4,850	9,763	6,225	10,752	6,900	6,480	4,400	600
Seed, Diluted Sludge	7.23	3,600	8,850	4,350	9,260	7,700	5,562	5,200	460
Seed, 1:1 Cow Manure: Diluted Sludge	7.40	5,050	11,638	5,188	12,145	8,300	7,485	2,300	690
Seed, 2:1 Cow Manure: Diluted Sludge	7.44	6,350	15,925	8,675	15,833	11,600	9,662	5,900	836
Seed, 1:2 Cow Manure: Diluted Sludge	7.30	4,350	10,475	5,175	10,633	7,700	6,415	4,300	561

Table A.3 Respirometer pH Change during Digestion

Treatment	Initial pH	Final pH	pH Change
Seed	8.15	7.43	-0.72
Seed, Cow Manure	7.82	7.31	-0.51
Seed, Diluted Sludge	7.77	7.23	-0.54
Seed, 1:1 Cow Manure: Diluted Sludge	7.61	7.40	-0.21
Seed, 2:1 Cow Manure: Diluted Sludge	7.51	7.44	-0.07
Seed, 1:2 Cow Manure: Diluted Sludge	7.77	7.30	-0.47

Table A.4 Respirometer Alkalinity Change during Digestion

Treatment	Initial Alkalinity (mg $CaCO_3$)	Final Alkalinity (mg $CaCO_3$)	Alkalinity Change (mg $CaCO_3$)
Seed	1,690	2,015	325
Seed, Cow Manure	2,665	3,153	488
Seed, Diluted Sludge	1,918	2,340	422
Seed, 1:1 Cow Manure: Diluted Sludge	2,763	3,283	520
Seed, 2:1 Cow Manure: Diluted Sludge	3,315	4,128	813
Seed, 1:2 Cow Manure: Diluted Sludge	2,243	2,828	585

Table A.5 Respirometer Total Solids Destruction

Treatment	Initial TS (mg)	Final TS (mg)	TS Destruction (mg)	Destruction (%)
Seed	5,527	4,812	715	13
Seed, Cow Manure	8,525	6,989	1,536	18
Seed, Diluted Sludge	7,263	6,019	1,244	17
Seed, 1:1 Cow Manure: Diluted Sludge	10,241	7,894	2,347	23
Seed, 2:1 Cow Manure: Diluted Sludge	13,078	10,292	2,786	21
Seed, 1:2 Cow Manure: Diluted Sludge	8,753	6,912	1,841	21

Table A.6 Respirometer Total Suspended Solids Destruction

Treatment	Initial TSS (mg)	Final TSS (mg)	TSS Destruction (mg)	Destruction (%)
Seed	3,965	2,405	1,560	39
Seed, Cow Manure	5,135	4,485	650	13
Seed, Diluted Sludge	5,005	5,005	0	0
Seed, 1:1 Cow Manure: Diluted Sludge	6,565	5,395	1,170	18
Seed, 2:1 Cow Manure: Diluted Sludge	8,060	7,540	520	6
Seed, 1:2 Cow Manure: Diluted Sludge	5,915	5,005	910	15

Table A.7 Respirometer Volatile Suspended Solids Destruction

Treatment	Initial VSS (mg)	Final VSS (mg)	VSS Destruction (mg)	Destruction (%)
Seed	2,860	1,495	1,365	48
Seed, Cow Manure	3,445	2,860	585	17
Seed, Diluted Sludge	2,665	3,380	-715	-27
Seed, 1:1 Cow Manure: Diluted Sludge	4,485	1,495	2,990	67
Seed, 2:1 Cow Manure: Diluted Sludge	5,525	3,835	1,690	31
Seed, 1:2 Cow Manure: Diluted Sludge	3,770	2,795	975	26

Table A.8 Respirometer Hydrogen Sulfide Concentrations

Treatment	H_2S Concentration (PPM)
Seed	0
Seed, Cow Manure	1,200
Seed, Diluted Sludge	700
Seed, 1:1 Cow Manure: Diluted Sludge	1,600
Seed, 2:1 Cow Manure: Diluted Sludge	3,200
Seed, 1:2 Cow Manure: Diluted Sludge	1,000

Table A.9 Respirometer Soluble COD Destruction

Treatment	Soluble COD			
	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction
Seed	3,088	3,307	-219	-7
Seed, Cow Manure	4,729	4,046	683	14
Seed, Diluted Sludge	3,234	2,828	406	13
Seed, 1:1 Cow Manure: Diluted Sludge	4,631	3,372	1,259	27
Seed, 2:1 Cow Manure: Diluted Sludge	6,102	5,639	463	8
Seed, 1:2 Cow Manure: Diluted Sludge	3,949	3,364	585	15

Table A.10 Respirometer Normalized Energy Potential per COD

Component	Normalized Gas Produced (mL)	Normalized Initial COD (mg)	Biogas/Initial COD after Dilution+ (m^3/Kg)
Cow Manure	762	3,022	0.252
Diluted Sludge*	776	2,575	0.301

Table A.11 Respirometer Normalized Energy Potential per VS

Component	Normalized Gas Produced (mL)	Normalized Initial VS (mg)	Biogas/Initial VS after Dilution+ (m^3/Kg)
Cow Manure	762	2,032	0.375
Diluted Sludge*	776	1,326	0.585

* Based on "Seed, Diluted Sludge" minus "Seed"

+At experimental Temperature (35°C) and Standard Pressure

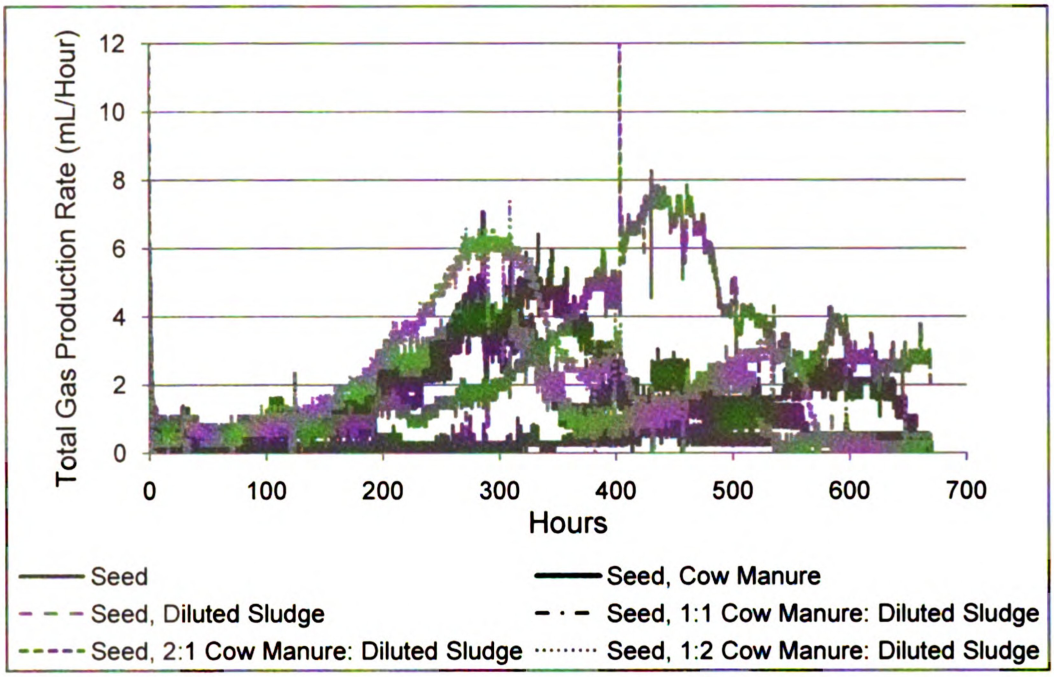


Figure A.1 Respirometer Biogas Production Rate

APPENDIX B

SERUM BOTTLE STUDY

Table B.1 Serum Bottle Concentrations before Digestion

Treatment	pH	Alkalinity (mg/L $CaCO_3$)	COD (mg/L)	Soluble COD (mg/L)	TS (mg/L)	TSS (mg/L)	VS (mg/L)	VSS (mg/L)	Ammonia (mg/L N)
Seed	7.78	2,900	17,900	4,263	12,980	9,042	700	240	277
Seed, Cow Manure	7.54	3,750	20,388	6,950	17,375	12,092	1,075	280	384
Seed, Diluted Sludge	7.61	2,750	21,425	4,913	18,405	12,508	900	238	253
Cow Manure	7.44	1,700	7,950	3,663	5,490	3,892	460	40	168
Diluted Sludge	5.76	100	6,963	875	6,185	3,815	220	23	6
Seed, 2:1 Cow Manure: Diluted Sludge	7.45	5,550	35,413	11,100	30,215	20,567	1,488	465	665
Seed, 1:1 Cow Manure: Diluted Sludge	7.50	4,300	27,863	8,225	24,310	16,545	1,700	353	485
Seed, 1:2 Cow Manure: Diluted Sludge	7.47	3,350	24,038	5,988	20,415	13,895	963	313	346
Seed, 1:4 Cow Manure: Diluted Sludge	7.52	2,900	22,913	5,375	18,768	12,763	1,025	288	313
Seed, 1:6 Cow Manure: Diluted Sludge	7.59	3,000	12,513	4,750	19,342	13,182	700	293	284

Table B.2 Serum Bottle Concentrations after Digestion

Treatment	pH	Alkalinity (mg/L $CaCO_3$)	COD (mg/L)	Soluble COD (mg/L)	TS (mg/L)	TSS (mg/L)	VS (mg/L)	VSS (mg/L)	Ammonia (mg/L N)
Seed	7.11	3,383	10,825	4,158	10,071	6,523	708	191	285
Seed, Cow Manure	7.26	5,383	14,233	4,608	13,636	8,279	1,017	274	572
Seed, Diluted Sludge	7.21	4,150	15,104	3,754	13,726	8,068	950	250	492
Cow Manure	7.15	2,333	3,600	1,129	2,844	2,291	433	47	223
Diluted Sludge	6.25	750	5,621	1,917	3,488	1,867	133	24	130
Seed, 2:1 Cow Manure: Diluted Sludge	7.45	8,100	21,479	4,563	22,424	13,020	1,700	426	813
Seed, 1:1 Cow Manure: Diluted Sludge	7.39	5,600	17,796	4,242	17,311	10,116	1,267	318	637
Seed, 1:2 Cow Manure: Diluted Sludge	7.23	5,050	17,463	4,192	16,052	9,433	1,175	273	533
Seed, 1:4 Cow Manure: Diluted Sludge	7.18	4,433	15,917	3,900	15,137	8,951	1,000	255	460
Seed, 1:6 Cow Manure: Diluted Sludge	7.16	4,467	16,642	4,075	15,142	8,981	967	253	448

Table B.3 Serum Bottle pH Change during Digestion

Treatment	Initial pH	Final pH	pH Change
Seed	7.78	7.11	-0.67
Seed, Cow Manure	7.54	7.26	-0.28
Seed, Diluted Sludge	7.61	7.21	-0.4
Cow Manure	7.44	7.15	-0.29
Diluted Sludge	5.76	6.25	0.49
Seed, 2:1 Cow Manure: Diluted Sludge	7.45	7.45	0
Seed, 1:1 Cow Manure: Diluted Sludge	7.50	7.39	-0.11
Seed, 1:2 Cow Manure: Diluted Sludge	7.47	7.23	-0.24
Seed, 1:4 Cow Manure: Diluted Sludge	7.52	7.18	-0.34
Seed, 1:6 Cow Manure: Diluted Sludge	7.59	7.16	-0.43

Table B.4 Serum Bottle Alkalinity Change during Digestion

Treatment	Initial Alkalinity (mg $CaCO_3$)	Final Alkalinity (mg $CaCO_3$)	Alkalinity Change (mg $CaCO_3$)
Seed	435	508	73
Seed, Cow Manure	563	808	245
Seed, Diluted Sludge	413	623	210
Cow Manure	255	350	95
Diluted Sludge	15	113	98
Seed, 2:1 Cow Manure: Diluted Sludge	833	1,215	382
Seed, 1:1 Cow Manure: Diluted Sludge	645	840	195
Seed, 1:2 Cow Manure: Diluted Sludge	503	758	255
Seed, 1:4 Cow Manure: Diluted Sludge	435	665	230
Seed, 1:6 Cow Manure: Diluted Sludge	450	670	220

Table B.5 Serum Bottle Total Solids Destruction

Treatment	Initial TS (mg)	Final TS (mg)	TS Destruction (mg)	Destruction (%)
Seed	1,947	1,511	436	22
Seed, Cow Manure	2,606	2,045	561	22
Seed, Diluted Sludge	2,761	2,059	702	25
Cow Manure	824	577	247	30
Diluted Sludge	928	523	405	44
Seed, 2:1 Cow Manure: Diluted Sludge	4,532	3,364	1168	26
Seed, 1:1 Cow Manure: Diluted Sludge	3,647	2,597	1050	29
Seed, 1:2 Cow Manure: Diluted Sludge	3,062	2,408	654	21
Seed, 1:4 Cow Manure: Diluted Sludge	2,815	2,271	544	19
Seed, 1:6 Cow Manure: Diluted Sludge	2,901	2,271	630	22

Table B.6 Serum Bottle Ammonia Change during Digestion

Treatment	Initial Ammonia (mg)	Final Ammonia (mg)	Change (mg)
Seed	41	43	2
Seed, Cow Manure	58	86	28
Seed, Diluted Sludge	38	74	36
Cow Manure	25	33	8
Diluted Sludge	1	20	19
Seed, 2:1 Cow Manure: Diluted Sludge	100	122	22
Seed, 1:1 Cow Manure: Diluted Sludge	73	96	23
Seed, 1:2 Cow Manure: Diluted Sludge	52	80	28
Seed, 1:4 Cow Manure: Diluted Sludge	47	69	22
Seed, 1:6 Cow Manure: Diluted Sludge	43	67	24

Table B.7 Serum Bottle Soluble COD Destruction

Treatment	Soluble COD			
	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction
Seed	639	624	15	2
Seed, Cow Manure	1,043	691	352	34
Seed, Diluted Sludge	737	563	174	24
Cow Manure	549	169	380	69
Diluted Sludge	131	288	NA	NA
Seed, 2:1 Cow Manure: Diluted Sludge	1,665	684	981	59
Seed, 1:1 Cow Manure: Diluted Sludge	1,234	636	598	48
Seed, 1:2 Cow Manure: Diluted Sludge	898	629	269	30
Seed, 1:4 Cow Manure: Diluted Sludge	806	585	221	27
Seed, 1:6 Cow Manure: Diluted Sludge	713	611	102	14

Table B.8 Serum Bottle Normalized Energy Potential per COD

Component	Normalized Gas Produced (mL)	Normalized Initial COD (mg)	Biogas/Initial COD after Dilution+ (m^3/Kg)
Cow Manure	410	373	1.1
Diluted Sludge*	356	529	0.67
Diluted Sludge**	334	1,121	0.30

Table B.9 Serum Bottle Normalized Energy Potential per VS

Component	Normalized Gas Produced (mL)	Normalized Initial VS (mg)	Biogas/Initial VS after Dilution+ (m^3/Kg)
Cow Manure	410	458	0.895
Diluted Sludge*	356	520	0.685
Diluted Sludge**	334	668	0.500

* Based on "Seed, Diluted Sludge" minus "Seed"

**Based on "Seed, 1:1 Cow Manure: Diluted Sludge" minus "Seed, Manure"

+At experimental Temperature (35°C) and Standard Pressure

Table B.10 Serum Bottle Hydrogen Sulfide Concentrations

Treatment	Average H_2S Concentration (PPM)
Seed	665
Seed, Cow Manure	1548
Seed, Diluted Sludge	759
Cow Manure	1028
Diluted Sludge	329
Seed, 2:1 Cow Manure: Diluted Sludge	1856
Seed, 1:1 Cow Manure: Diluted Sludge	1303
Seed, 1:2 Cow Manure: Diluted Sludge	1018
Seed, 1:4 Cow Manure: Diluted Sludge	908
Seed, 1:6 Cow Manure: Diluted Sludge	829

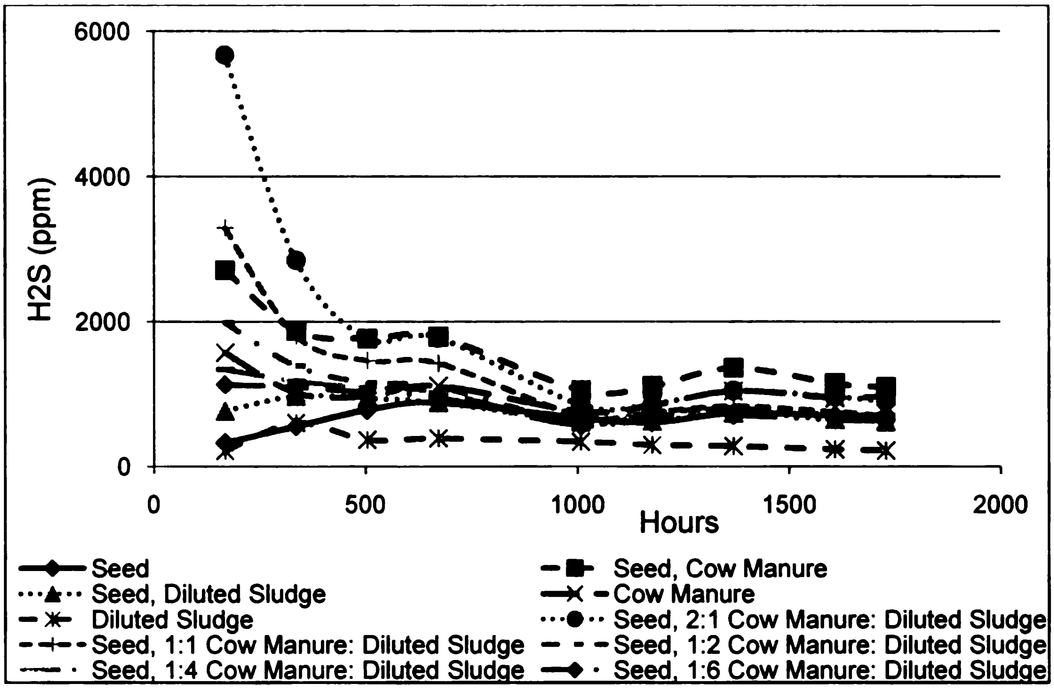


Figure B.1 Serum Bottle H2S Concentrations

Table B.11 Daily Biogas Yields for Controls

Date	Seed			Seed, Cow Manure			Seed, Diluted Sludge			Cow Manure			Diluted Sludge		
	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
3/10/2010	3	4	3	6	5	5	6	5	9	3	3	3	3	2	1
3/11/2010	3	2	2	7	8	8	8	8	8	3	3	2	1	2	2
3/12/2010	2	2	2	10	9	10	10	10	10	2	2	3	2	2	2
3/13/2010	1	1	2	8	9	9	16	16	17	3	3	2	2	2	2
3/14/2010	2	1	2	8	8	8	16	16	17	2	2	2	1	1	1
3/15/2010	3	2	3	11	12	12	16	16	16	3	3	2	0	1	1
3/16/2010	5	3	5	12	13	13	12	13	13	3	2	2	2	3	2
3/18/2010	8	8	10	30	31	32	24	30	25	9	9	10	4	3	5
3/20/2010	9	7	10	38	39	40	34	30	34	21	21	22	3	1	2
3/22/2010	8	7	9	40	41	42	43	37	40	25	26	25	2	1	2
3/24/2010	12	11	14	39	41	40	36	35	35	25	25	24	4	3	4
3/26/2010	15	10	14	35	37	38	23	23	23	20	21	20	6	4	10
3/28/2010	16	11	16	37	38	40	24	23	24	22	24	24	10	9	18
3/30/2010	14	9	12	29	31	30	18	19	18	20	22	22	7	9	9
4/01/2010	16	10	16	33	37	36	23	20	21	21	22	24	5	13	5
4/03/2010	13	10	14	26	28	27	25	18	20	13	11	11	2	8	4
4/05/2010	13	10	14	25	26	27	29	22	23	8	7	7	0	3	1
4/07/2010	15	13	16	30	31	30	30	26	28	10	11	11	2	4	3
4/10/2010	19	14	22	43	43	42	34	26	27	18	20	18	0	2	0
4/13/2010	12	9	14	30	31	30	17	14	14	18	19	19	2	1	2
4/16/2010	10	8	12	24	24	25	14	14	11	10	12	12	3	1	1
4/19/2010	6	4	7	13	14	15	12	11	8	5	5	5	0	0	0
4/22/2010	7	7	9	14	14	16	12	24	12	7	7	8	2	2	2
4/25/2010	5	6	7	10	12	11	9	22	10	4	5	6	1	2	1
4/28/2010	2	1	2	6	6	5	5	17	11	2	2	2	2	0	2
5/01/2010	7	6	8	10	11	11	10	20	25	5	5	5	0	1	0
5/05/2010	5	4	6	8	10	10	12	18	24	6	6	6	2	1	2
5/10/2010	4	3	4	8	9	10	13	14	25	4	4	4	0	0	0
5/15/2010	6	5	7	16	19	17	15	15	22	4	4	4	2	0	2
5/20/2010	6	7	8	14	16	21	16	22	21	4	4	4	2	0	2
5/23/2010	4	6	5	7	7	9	13	14	17	1	1	1	0	0	0
5/25/2010	3	4	4	4	4	4	8	9	8	1	1	1	0	0	0

Table B.12 Daily Biogas Yields for Blends

Date	2:1			1:1			1:2			1:4			1:6		
	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
3/10/2010	13	21	20	14	14	12	7	7	7	6	6	7	6	6	7
3/11/2010	27	22	23	20	19	20	18	17	17	13	13	13	12	13	12
3/12/2010	21	22	21	16	15	16	13	13	13	13	14	14	15	15	15
3/13/2010	15	17	16	13	14	13	12	13	13	13	14	14	14	15	15
3/14/2010	13	15	14	13	12	12	12	11	12	13	13	14	14	13	14
3/15/2010	12	15	14	16	16	17	16	16	16	17	18	18	18	17	18
3/16/2010	10	13	12	16	17	17	16	17	17	19	18	18	18	18	18
3/18/2010	23	36	33	48	51	50	39	41	40	40	35	37	33	35	36
3/20/2010	37	65	58	58	64	57	38	38	38	32	30	31	30	31	32
3/22/2010	68	78	81	51	50	48	35	37	37	36	37	37	35	35	35
3/24/2010	89	87	84	52	54	49	40	39	39	46	45	44	40	41	41
3/26/2010	78	69	68	49	47	48	42	43	44	38	34	34	31	33	31
3/28/2010	66	63	65	56	56	52	43	45	45	34	35	31	30	30	32
3/30/2010	65	58	60	40	47	41	28	30	33	28	30	25	26	25	26
4/01/2010	87	67	73	47	50	51	29	30	35	29	31	26	27	27	29
4/03/2010	60	48	52	36	39	42	25	26	31	24	26	23	23	21	22
4/05/2010	53	47	44	35	38	40	29	30	34	25	28	25	23	22	23
4/07/2010	46	37	40	34	35	37	31	32	36	30	33	32	27	26	27
4/10/2010	63	77	69	48	49	49	44	44	47	50	50	47	44	44	47
4/13/2010	71	100	89	60	63	59	37	40	45	36	30	26	27	28	31
4/16/2010	105	79	89	60	63	62	20	24	27	20	18	17	16	17	19
4/19/2010	55	50	41	28	32	27	14	14	15	10	10	11	8	9	9
4/22/2010	36	52	34	23	26	23	20	21	23	15	16	22	13	13	15
4/25/2010	22	38	34	16	18	16	17	17	17	18	18	24	12	12	14
4/28/2010	10	23	28	12	14	9	12	16	7	9	10	16	9	13	15
5/01/2010	16	25	33	20	15	12	21	25	13	14	12	19	16	18	27
5/05/2010	16	22	26	20	15	14	20	25	10	10	11	16	10	15	24
5/10/2010	16	18	24	18	15	13	17	22	10	9	11	15	8	10	18
5/15/2010	21	18	23	17	17	18	18	20	13	11	15	15	10	12	17
5/20/2010	28	24	30	21	19	27	16	19	15	12	24	15	12	18	18
5/23/2010	21	20	18	19	17	13	11	11	14	7	12	9	6	15	10
5/25/2010	6	13	11	9	12	8	7	10	5	6	6	5	5	12	7

Table B.13 Individual Treatment Gas Production and Averages

Treatment	Total Gas Production (mL)	Average Gas Production (mL)
Seed a	254	
Seed b	205	246
Seed c	279	
Seed, Cow Manure a	631	
Seed, Cow Manure b	664	656
Seed, Cow Manure c	673	
Seed, Diluted Sludge a	583	
Seed, Diluted Sludge b	607	602
Seed, Diluted Sludge c	616	
Cow Manure a	302	
Cow Manure b	312	308
Cow Manure c	311	
Diluted Sludge a	72	
Diluted Sludge b	81	80
Diluted Sludge c	88	
2:1 a	1,269	
2:1 b	1,339	1,312
2:1 c	1,327	
1:1 a	985	
1:1 b	1,013	990
1:1 c	972	
1:2 a	747	
1:2 b	793	769
1:2 c	768	
1:4 a	683	
1:4 b	703	695
1:4 c	700	
1:6 a	618	
1:6 b	659	660
1:6 c	704	

APPENDIX C

SEMI-CONTINUOUS STUDY

Table C.1 Removal and Feeding of Substrate Data

SRT:	1
Day:	1
Time:	1.45 pm
Date:	6/17/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	9	3040	130	2901	3052	9
Reactor 3	7	3116	130	2984	3120	7
Reactor 4	8	3174	130	3040	3174	8
Reactor 5	31	3098	130	2928	3100	31

SRT:	1
Day:	2
Time:	10.30 am
Date:	6/18/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	19	3098	130	2954	3101	19
Reactor 3	9	3077	130	2938	3075	9
Reactor 4	12	3176	130	3038	3174	12
Reactor 5	47	3101	130	2970	3109	47

SRT:	1
Day:	3
Time:	4 pm
Date:	6/19/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	32	3107	130	2976	3104	32
Reactor 3	10	3081	130	2963	3088	10
Reactor 4	17	3106	130	2974	3106	17
Reactor 5	63	3085	130	2945	3080	63

Table C.1 cont'd.

SRT:	1
Day:	4
Time:	2 pm
Date:	6/20/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	42	3085	130	2945	3084	42
Reactor 3	11	3065	130	2918	3071	11
Reactor 4	24	3099	130	2971	3101	24
Reactor 5	65	3090	130	2959	3083	65

SRT:	1
Day:	5
Time:	9.30 am
Date:	6/21/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	47	3084	130	2954	3084	47
Reactor 3	12	3077	130	2934	3084	12
Reactor 4	32	3100	130	2970	3098	32
Reactor 5	69	3096	130	2960	3098	69

SRT:	1
Day:	6
Time:	11 am
Date:	6/22/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	56	3084	130	2953	3081	56
Reactor 3	15	3080	130	2947	3080	15
Reactor 4	44	3095	130	2950	3102	44
Reactor 5	84	3071	130	2940	3070	84

Table C.1 cont'd.

SRT:	1
Day:	7
Time:	11 am
Date:	6/23/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	61	3065	130	2932	3066	61
Reactor 3	19	3022	130	2892	3020	19
Reactor 4	63	3091	130	2959	3089	63
Reactor 5	99	3070	130	2941	3064	99

SRT:	1
Day:	8
Time:	11 am
Date:	6/24/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	71	3066	130	2948	3069	71
Reactor 3	22	3014	130	2880	3013	22
Reactor 4	88	3080	130	2920	3079	88
Reactor 5	113	3048	130	2915	3051	113

SRT:	1
Day:	9
Time:	10.30 am
Date:	6/25/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	84	3054	130	2925	3056	84
Reactor 3	25	3035	130	2913	3033	25
Reactor 4	120	3061	130	2930	3060	120
Reactor 5	118	3038	130	2908	3034	118

Table C.1 cont'd.

SRT:	1
Day:	10
Time:	11 am
Date:	6/26/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	100	3133	130	3004	3130	100
Reactor 3	28	2977	130	2848	2985	28
Reactor 4	154	3058	130	2924	3056	154
Reactor 5	121	3026	130	2986	3027	121

SRT:	1
Day:	11
Time:	1 pm
Date:	6/27/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	112	3089	130	2955	3089	112
Reactor 3	32	2969	130	2838	2974	32
Reactor 4	181	3055	130	2924	3055	181
Reactor 5	123	3015	130	2885	3012	123

SRT:	1
Day:	12
Time:	9.30 am
Date:	6/28/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	12	3126	130	2995	3125	12
Reactor 3	35	2956	130	2824	2962	35
Reactor 4	198	3065	130	2934	3068	198
Reactor 5	135	3040	130	2910	3040	135

Table C.1 cont'd.

SRT:	1
Day:	13
Time:	9.00 am
Date:	6/29/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	134	NA	130	NA	NA	-
Reactor 3	41	NA	130	NA	NA	41
Reactor 4	218	NA	130	NA	NA	218
Reactor 5	139	NA	130	NA	NA	139

SRT:	1
Day:	14
Time:	1.30 pm
Date:	6/30/2010

	Tips Before	Weight In (g)	Weight Out (g)	Tips After
Reactor 2	139	146	130	139
Reactor 3	46	140	130	46
Reactor 4	237	130	115	237
Reactor 5	147	130	130	147

SRT:	1
Day:	15
Time:	2.15 pm
Date:	7/1/2010

	Tips Before	Weight In (g)	Weight Out (g)	Tips After
Reactor 2	147	160	170	147
Reactor 3	59	132	145	59
Reactor 4	266	130	130	266
Reactor 5	157	136	140	157

Table C.1 cont'd.

SRT:	2
Day:	1
Time:	12.30 pm
Date:	7/2/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	157	NA	130	NA	NA	157
Reactor 3	63	NA	130	NA	NA	63
Reactor 4	286	NA	130	NA	NA	286
Reactor 5	167	NA	130	NA	NA	167

SRT:	2
Day:	2
Time:	2 pm
Date:	7/3/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	173	3087	130	2961	3088	173
Reactor 3	69	3046	130	2914	3045	69
Reactor 4	316	3042	130	2910	3040	316
Reactor 5	179	3107	130	2968	3106	179

SRT:	2
Day:	3
Time:	8 am
Date:	7/4/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	187	3078	130	2948	3076	187
Reactor 3	73	3031	130	2901	3032	73
Reactor 4	342	3017	130	2890	3016	342
Reactor 5	186	3090	130	2960	3088	186

Table C.1 cont'd.

SRT:	2
Day:	4
Time:	12 pm
Date:	7/5/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	203	3103	130	2974	3104	203
Reactor 3	78	3005	130	2875	3005	78
Reactor 4	385	3022	130	2890	3021	385
Reactor 5	199	3057	130	2927	3055	199

SRT:	2
Day:	5
Time:	1.30 pm
Date:	7/6/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	212	3092	130	2958	3091	212
Reactor 3	84	2933	130	2804	3074*	84
Reactor 4	431	2961	130	2829	2961	431
Reactor 5	212	3039	130	2907	3037	212

*Added to ensure 2L volume

SRT:	2
Day:	6
Time:	11.50 am
Date:	7/7/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	221	3105	130	2975	3108	221
Reactor 3	91	3083	130	2953	3084	91
Reactor 4	468	2949	130	2816	2947	468
Reactor 5	227	3026	130	2895	3026	227

Table C.1 cont'd.

SRT:	2
Day:	7
Time:	11.30 am
Date:	7/8/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	231	3108	130	2971	3108	231
Reactor 3	101	2984	130	2953	2984	101
Reactor 4	502	2907	130	2773	2909	502
Reactor 5	245	2994	130	2863	2996	245

SRT:	2
Day:	8
Time:	2 pm
Date:	7/9/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	242	3068	130	2938	3070	242
Reactor 3	111	3037	130	2900	3039	111
Reactor 4	533	2940	130	2810	2942	533
Reactor 5	270	2988	130	2856	2985	270

SRT:	2
Day:	9
Time:	2 pm
Date:	7/10/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	251	3138	130	3006	3136	251
Reactor 3	120	3014	130	2880	3017	120
Reactor 4	556	2911	130	2780	2938	556
Reactor 5	286	2963	130	2834	2957	286

Table C.1 cont'd.

SRT:	2
Day:	10
Time:	8 am
Date:	7/11/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	259	3076	130	2947	3076	259
Reactor 3	126	3000	130	2873	3003	126
Reactor 4	580	2917	130	2790	2921	580
Reactor 5	307	2953	130	2825	2959	307

SRT:	2
Day:	11
Time:	11 am
Date:	7/12/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	267	3097	130	2966	3105	267
Reactor 3	133	2889	130	2758	2885	133
Reactor 4	615	2902	130	2774	2914	615
Reactor 5	329	2955	130	2825	2950	329

SRT:	2
Day:	12
Time:	3.30 pm
Date:	7/12/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	279	3094	130	2965	3095	279
Reactor 3	136	2883	130	2756	2880	136
Reactor 4	648	2846	130	2721	2850	648
Reactor 5	-	-	130	-	-	-

Table C.1 cont'd.

SRT:	2
Day:	13
Time:	11.30 am
Date:	7/13/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	288	3072	130	2946	3078	288
Reactor 3	139	2864	130	2735	2856	139
Reactor 4	667	2851	130	2722	2849	667
Reactor 5	-	-	130	-	-	-

SRT:	2
Day:	14
Time:	11.30am
Date:	7/15/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	298	3061	130	2933	3062	298
Reactor 3	143	2839	130	2709	2835	143
Reactor 4	689	2831	130	2706	2827	689
Reactor 5	-	-	130	-	-	-

SRT:	2
Day:	15
Time:	11.30 am
Date:	7/16/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	306	3069	130	2941	3069	306
Reactor 3	147	2844	130	2716	2839	147
Reactor 4	712	2831	130	2701	2827	712
Reactor 5	-	-	130	-	-	-

Table C.1 cont'd.

SRT:	3
Day:	1
Time:	5 pm
Date:	7/17/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	316	3072	130	2945	3072	316
Reactor 3	150	2835	130	2703	2831	150
Reactor 4	736	2837	130	2703	2840	736
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	2
Time:	1 pm
Date:	7/18/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	323	3061	130	2930	3064	323
Reactor 3	154	2835	130	2707	2837	154
Reactor 4	752	2838	130	2708	2840	752
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	3
Time:	2.15 pm
Date:	7/19/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	333	3067	130	2936	3066	333
Reactor 3	157	2857	130	2732	2854	157
Reactor 4	776	2845	130	2717	2845	776
Reactor 5	-	-	130	-	-	-

Table C.1 cont'd.

SRT:	3
Day:	4
Time:	2.30 PM
Date:	7/20/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	343	3068	130	2938	3066	343
Reactor 3	160	2833	130	2697	2844	160
Reactor 4	803	2810	130	2681	2809	803
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	5
Time:	12 PM
Date:	7/21/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	352	3063	130	2933	3064	352
Reactor 3	163	2831	130	2701	2828	163
Reactor 4	828	2772	130	2640	2774	828
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	6
Time:	11.30 am
Date:	7/22/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	363	3032	130	2903	3031	363
Reactor 3	167	2780	130	2650	2780	167
Reactor 4	852	2758	130	2625	2759	852
Reactor 5	-	-	130	-	-	-

Table C.1 cont'd.

SRT:	3
Day:	7
Time:	10.30 am
Date:	7/23/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	373	3015	130	2886	3015	373
Reactor 3	170	2754	130	2624	2754	170
Reactor 4	876	2765	130	2634	2765	876
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	8
Time:	11.30 am
Date:	7/24/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	382	3012	130	2884	3011	382
Reactor 3	173	2775	130	2643	2775	173
Reactor 4	894	2770	130	2641	2772	894
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	9
Time:	3 pm
Date:	7/25/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	392	3008	130	2880	3006	392
Reactor 3	177	2755	130	2624	2755	177
Reactor 4	922	2766	130	2631	2768	922
Reactor 5	-	-	130	-	-	-

Table C.1 cont'd.

SRT:	3
Day:	10
Time:	12 pm
Date:	7/26/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	400	3012	130	2880	3015	400
Reactor 3	180	2760	130	2629	2768	180
Reactor 4	940	2750	130	2620	2749	940
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	11
Time:	2.30 pm
Date:	7/27/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	409	2992	130	2864	2993	409
Reactor 3	184	2756	130	2626	2757	184
Reactor 4	963	2737	130	2600	2727	963
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	12
Time:	1.30 pm
Date:	7/28/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	417	2981	130	2850	2982	417
Reactor 3	188	2716	130	2587	2714	188
Reactor 4	983	2734	130	2602	2736	983
Reactor 5	-	-	130	-	-	-

Table C.1 cont'd.

SRT:	3
Day:	13
Time:	2 pm
Date:	7/29/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	427	2994	130	2864	2994	427
Reactor 3	193	2689	130	2559	2690	193
Reactor 4	1004	2698	130	2566	2697	1004
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	14
Time:	2 pm
Date:	7/30/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	436	2959	130	2829	2962	436
Reactor 3	199	2651	130	2522	2651	199
Reactor 4	1023	2701	130	2571	2701	1023
Reactor 5	-	-	130	-	-	-

SRT:	3
Day:	15
Time:	11.30 am
Date:	7/31/2010

	Tips Before	Initial Weight (g)	Amount (g)	Weight after Removal (g)	Weight after Feed (g)	Tips After
Reactor 2	446	2967	130	2739	2972	446
Reactor 3	206	2639	130	2507	2640	206
Reactor 4	1042	2682	130	2552	2682	1042
Reactor 5	-	-	130	-	-	-

Table C.2 Wet-Tip Gas Meter Data

Date	SRT	R1	R2	R3	R4	R5
06/18/2010	1	2	21	10	13	49
06/19/2010	1	2	32	10	17	63
06/20/2010	1	2	42	11	24	65
06/21/2010	1	2	50	12	36	74
06/22/2010	1	2	57	17	50	89
06/23/2010	1	2	63	20	69	103
06/24/2010	1	2	73	23	94	116
06/25/2010	1	2	84	25	120	118
06/26/2010	1	2	100	28	154	121
06/27/2010	1	2	112	32	181	123
06/28/2010	1	2	128	37	204	139
06/29/2010	1	-	134	46	235	147
06/30/2010	1	-	139	46	237	147
07/01/2010	1	-	147	60	268	158
07/02/2010	2	-	158	64	288	168
07/03/2010	2	-	173	69	315	177
07/04/2010	2	-	187	73	342	186
07/05/2010	2	-	203	78	384	199
07/06/2010	2	-	215	86	440	215
07/07/2010	2	-	224	94	477	232
07/08/2010	2	-	234	104	511	251
07/09/2010	2	-	242	112	534	271
07/10/2010	2	-	251	119	556	286
07/11/2010	2	-	259	126	580	307
07/12/2010	2	-	269	134	621	335
07/13/2010	2	-	280	136	649	355
07/14/2010	2	-	288	139	665	357
07/15/2010	2	-	300	144	695	358
07/16/2010	2	-	306	147	712	358
07/17/2010	3	-	316	150	736	359
07/18/2010	3	-	323	154	752	364
07/19/2010	3	-	334	158	778	372
07/20/2010	3	-	344	161	805	372
07/21/2010	3	-	354	164	833	374
07/22/2010	3	-	362	167	851	374
07/23/2010	3	-	376	171	876	379
07/24/2010	3	-	382	173	894	380
07/25/2010	3	-	392	177	922	384
07/26/2010	3	-	400	180	939	386
07/27/2010	3	-	410	185	964	389
07/28/2010	3	-	419	189	987	392
07/29/2010	3	-	428	194	1007	394
07/30/2010	3	-	438	200	1025	396
07/31/2010	3	-	447	208	1045	397
08/01/2010	3	-	455	212	1066	398

Table C.3 pH for 3 SRT's

SRT	Day	Date	Reactor 2	Reactor 3	Reactor 4	Reactor 5
1	2	6/18/2010	7.62	6.09	7.37	7.36
1	5	6/21/2010	7.50	6.25	7.39	7.48
1	6	6/22/2010	7.46	6.27	7.33	7.38
1	8	6/24/2010	7.43	6.23	7.51	7.36
1	12	6/28/2010	7.58	6.16	7.58	7.29
2	16	7/02/2010	7.41	6.26	7.50	7.20
2	19	7/05/2010	7.79	6.32	7.84	7.16
2	21	7/07/2010	7.62	6.38	7.73	7.29
2	23	7/09/2010	7.65	6.60	7.75	7.47
2	26	7/12/2010	7.57	6.79	7.77	7.60
2	28	7/14/2010	7.63	6.69	7.76	-
3	30	7/16/2010	7.92	6.63	8.06	-
3	33	7/19/2010	7.69	6.49	7.68	-
3	35	7/21/2010	7.64	6.38	7.63	-
3	37	7/23/2010	7.72	6.39	7.75	-
3	40	7/26/2010	7.67	6.38	7.76	-
3	42	7/28/2010	7.64	6.43	7.75	-
3	44	7/30/2010	7.59	6.60	7.72	-

Table C.4 Alkalinity (mg/L $CaCO_3$) for 3 SRT's

SRT	Day	Date	Reactor 2	Reactor 3	Reactor 4	Reactor 5
1	2	6/18/2010	6,300	4,800	9,100	10,000
1	5	6/21/2010	7,100	4,800	8,600	10,600
1	8	6/24/2010	6,900	6,800	10,200	11,800
1	12	6/28/2010	7,300	4,000	11,800	11,800
2	16	7/2/2010	6,400	5,200	11,200	9,400
2	19	7/5/2010	7,700	4,400	13,800	10,200
2	21	7/7/2010	7,100	5,200	12,200	9,800
2	23	7/09/2010	7,600	5,600	12,200	10,800
2	26	7/12/2010	7,900	6,800	13,800	12,200
2	28	7/14/2010	7,000	4,800	12,800	-
3	30	7/16/2010	7,900	6,200	13,800	-
3	33	7/19/2010	8,200	6,600	13,400	-
3	35	7/21/2010	7,900	6,000	13,000	-
3	37	7/23/2010	8,300	5,800	13,800	-
3	40	7/26/2010	8,600	7,400	14,800	-
3	42	7/28/2010	8,700	7,400	15,200	-
3	44	7/30/2010	8,200	7,400	15,000	-

Table C.5 COD (mg/L) for 3 SRT's

SRT	Day	Date	Reactor 2	Reactor 3	Reactor 4	Reactor 5
1	2	6/18/2010	25,000	85,700	94,100	91,600
1	5	6/21/2010	28,175	NA	119,250	133,350
1	6	6/22/2010	28,550	88,950	98,050	104,150
1	8	6/24/2010	28,900	94,000	101,350	99,050
1	12	6/28/2010	23,800	87,100	91,250	101,450
2	16	7/02/2010	32,400	86,850	95,600	88,400
2	19	7/05/2010	30,650	87,600	99,050	108,300
2	21	7/07/2010	27,300	88,600	90,600	103,400
2	23	7/09/2010	26,175	98,200	83,450	102,450
2	26	7/12/2010	22,000	93,750	91,300	-
2	28	7/14/2010	20,600	87,750	82,600	-
3	30	7/16/2010	21,400	93,550	79,650	-
3	33	7/19/2010	20,175	88,750	88,600	-
3	35	7/21/2010	19,575	85,100	82,400	-
3	37	7/23/2010	20,025	90,150	80,350	-
3	40	7/26/2010	22,150	88,450	83,150	-
3	42	7/28/2010	19,575	85,750	87,400	-
3	44	7/30/2010	21,000	79,050	81,550	-

Table C.6 Reactor 2 TS/VS for 3 SRT's

SRT	Day	Date	TS (mg/L)	TS %	VS (mg/L)	VS %
1	2	6/18/2010	15,877	1.62	9,503	0.97
1	5	6/21/2010	22,680	2.27	14,865	1.49
1	8	6/24/2010	23,460	2.33	15,193	1.49
1	12	6/28/2010	21,797	2.18	13,987	1.40
2	16	7/02/2010	25,290	2.54	15,400	1.55
2	19	7/05/2010	25,758	2.59	15,777	1.59
2	21	7/07/2010	23,363	2.32	14,053	1.40
2	23	7/09/2010	23,003	2.37	14,007	1.45
2	26	7/12/2010	21,970	2.17	13,065	1.29
2	28	7/14/2010	20,530	2.03	12,355	1.22
3	30	7/16/2010	21,130	2.09	12,308	1.22
3	33	7/19/2010	20,225	2.01	11,855	1.18
3	35	7/21/2010	20,280	2.00	11,693	1.15
3	37	7/23/2010	26,007	2.60	15,407	1.55
3	40	7/26/2010	24,402	2.47	14,570	1.47
3	42	7/28/2010	20,828	2.09	12,368	1.24
3	44	7/30/2010	21,045	2.09	12,440	1.23

Table C.7 Reactor 3 TS/VS for 3 SRT's

SRT	Day	Date	TS (mg/L)	TS %	VS (mg/L)	VS %
1	2	6/18/2010	64,400	6.64	28,922	2.98
1	5	6/21/2010	70,153	6.99	31,337	3.12
1	8	6/24/2010	72,835	7.02	31,337	3.13
1	12	6/28/2010	72,240	7.19	32,540	3.24
2	16	7/02/2010	69,857	7.02	31,855	3.20
2	19	7/05/2010	76,190	7.58	34,223	3.40
2	21	7/07/2010	78,483	7.74	36,412	3.59
2	23	7/09/2010	76,915	7.55	35,608	3.50
2	26	7/12/2010	73,047	7.14	33,508	3.28
2	28	7/14/2010	70,362	7.06	32,682	3.28
3	30	7/16/2010	74,862	7.47	34,385	3.43
3	33	7/19/2010	73,622	7.35	33,403	3.34
3	35	7/21/2010	78,547	7.59	34,877	3.37
3	37	7/23/2010	74,503	7.50	32,830	3.31
3	40	7/26/2010	77,652	7.73	33,762	3.36
3	42	7/28/2010	79,577	7.97	33,343	3.34
3	44	7/30/2010	83,048	8.14	34,112	3.35

Table C.8 Reactor 4 TS/VS for 3 SRT's

SRT	Day	Date	TS (mg/L)	TS %	VS (mg/L)	VS %
1	2	6/18/2010	70,500	7.14	32,097	3.25
1	5	6/21/2010	80,825	7.96	37,238	3.67
1	8	6/24/2010	80,508	8.19	36,398	3.70
1	12	6/28/2010	82,628	8.23	38,097	3.90
2	16	7/02/2010	80,395	8.02	36,297	3.62
2	19	7/05/2010	81,740	8.19	36,900	3.70
2	21	7/07/2010	80,447	7.96	36,452	3.61
2	23	7/09/2010	77,905	7.64	33,805	3.32
2	26	7/12/2010	80,542	8.13	35,508	3.58
2	28	7/14/2010	77,347	7.76	33,322	3.35
3	30	7/16/2010	75,145	7.49	30,910	3.09
3	33	7/19/2010	78,967	8.13	34,208	3.52
3	35	7/21/2010	82,862	8.17	34,578	3.41
3	37	7/23/2010	81,268	8.02	32,855	3.24
3	40	7/26/2010	78,067	7.68	33,637	3.31
3	42	7/28/2010	88,932	8.78	38,826	3.84
3	44	7/30/2010	84,842	8.28	34,700	3.39

Table C.9 Reactor 5 TS/VS for 3 SRT's

SRT	Day	Date	TS (mg/L)	TS %	VS (mg/L)	VS %
1	2	6/18/2010	73,882	7.42	32,440	3.26
1	5	6/21/2010	74,503	7.42	32,807	3.27
1	8	6/24/2010	79,318	7.88	34,353	3.41
1	12	6/28/2010	83,200	8.32	37,295	3.73
2	16	7/02/2010	77,383	7.68	34,735	3.45
2	19	7/05/2010	84,853	8.67	39,173	4.00
2	21	7/07/2010	87,043	8.86	39,942	4.07
2	23	7/09/2010	89,462	8.83	40,258	3.98
2	26	7/12/2010	80,557	8.30	36,330	3.74
2	28	7/14/2010	-	-	-	-
3	30	7/16/2010	-	-	-	-
3	33	7/19/2010	-	-	-	-
3	35	7/21/2010	-	-	-	-
3	37	7/23/2010	-	-	-	-
3	40	7/26/2010	-	-	-	-
3	42	7/28/2010	-	-	-	-
3	44	7/30/2010	-	-	-	-

Table C.10 Percentage (%) Methane Content

Date	Day	SRT	Reactor 1	Reactor 2	Reactor 3	Reactor 4	Reactor 5
6/17/2010	1	1	NA	54	16	18	44
6/18/2010	2	1	3	49	15	24	40
6/21/2010	5	1	3	46	16	34	41
6/23/2010	7	1	1	50	26	47	41
6/28/2010	12	1	1	63	29	52	46
7/01/2010	15	1	-	34	14	39	29
7/02/2010	16	2	-	42	16	45	31
7/04/2010	18	2	-	45	25	56	38
7/06/2010	20	2	-	65	31	61	44
7/08/2010	22	2	-	60	37	59	50
7/12/2010	26	2	-	63	44	59	58
7/15/2010	29	2	-	62	36	57	-
7/19/2010	33	3	-	67	36	58	-
7/22/2010	36	3	-	63	29	58	-
7/26/2010	40	3	-	61	29	57	-
7/28/2010	42	3	-	60	31	57	-
7/31/2010	45	3	-	61	42	57	-

Table C.11 Reactor 5 COD and VS Destruction

Reactor 5	COD				VS			
DAY	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction	Initial (mg)	Final (mg)	Destroyed (mg)	% Reduction
2	16,322	11,908	4,414	27	8,052	4,217	3,835	48
6	16,322	13,540	2,782	17	8,052	4,265	3,787	47
8	16,322	12,877	3,445	21	8,052	4,466	3,586	45
12	16,322	13,189	3,133	19	8,052	4,848	3,204	40
16	16,322	11,492	4,830	30	8,052	4,516	3,536	44
19	16,322	14,079	2,243	14	8,052	5,092	2,959	37
21	16,322	13,442	2,880	18	8,052	5,192	2,859	36
23	16,322	13,319	3,003	18	8,052	5,234	2,818	35
26	16,322	12,324	3,998	24	8,052	4,723	3,329	41
28	-	-	-	-	-	-	-	-
30	-	-	-	-	-	-	-	-
33	-	-	-	-	-	-	-	-
35	-	-	-	-	-	-	-	-
37	-	-	-	-	-	-	-	-
40	-	-	-	-	-	-	-	-
42	-	-	-	-	-	-	-	-
45	-	-	-	-	-	-	-	-

Table C.12 Characteristics of Substrate Feeds between SRT's

SRT 1	Reactor 2 Feed	Reactor 3 Feed	Reactor 4 Feed
COD (mg)	61,525	187,100	251,100
VS (mg)	37,394	101,160	123,876
SRT 2	**Reactor 2 Feed**	**Reactor 3 Feed**	**Reactor 4 Feed**
COD (mg)	62,675	192,400	246,900
VS (mg)	30,700	99,386	117,910
SRT 3	**Reactor 2 Feed**	**Reactor 3 Feed**	**Reactor 4 Feed**
COD (mg)	61,150	200,400	257,200
VS (mg)	41,340	91,686	107,396

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