

**ANAEROBIC DIGESTION OF DAIRY MANURE USING THE MICROWAVE
HYDROGEN PEROXIDE ADVANCED OXIDATION PROCESS**

by

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Abstract

Anaerobic digestion (AD) of dairy manure is an alternative to traditional manure management using land application. There are many benefits of AD but it is currently not an economically viable option in many parts of North America. The focus of this investigation was to determine whether liquid dairy manure treated by microwave hydrogen peroxide advanced oxidation process (MW/H₂O₂-AOP) would increase the anaerobic biodegradability of the substrate, producing higher methane yields. Biochemical methane potential (BMP) tests were conducted to measure biogas generation and the Modified Gompertz equation was used to determine the kinetics of anaerobic biodegradability. The research found that MW/H₂O₂-AOP was effective in solubilizing nutrients and organics in liquid dairy manure, but it does not necessarily translate into enhanced anaerobic biodegradability. The BMP tests revealed that digestion of untreated dairy manure at 35°C had the highest methane yields.

Severe inhibition was observed for the digestion of acidified dairy manure. This was attributed to sulfide inhibition. In addition, ammonia concentrations were suspected as the main reason for severe inhibition of microwave treated dairy manure with no acid addition, but a number of other factors could also be responsible, including sulfide, light metals and lack of temperature acclimation for inoculum. Pretreatment of sewage sludge and BMP tests were also conducted to compare the effect of MW/H₂O₂-AOP with dairy manure. The results showed that the microwave treatment had a positive impact on anaerobic digestibility of sewage sludge, especially for mesophilic digestion. At 35°C, MW alone generated the highest methane yields followed by MW/H₂O₂ and then the control. In terms of methane yield, microwave treatment improved the methane production rate for mesophilic digestion, but not thermophilic digestion.

The positive results for BMP tests of microwave treated sewage sludge suggest that the influence of microwave treatment on anaerobic biodegradability is substrate and solid concentration specific. There was no clear and consistent increase or decrease in soluble metal (Ca, K, Mg, Na) concentrations after MW/H₂O₂-AOP. However, it was found that acidified treatment was effective in releasing calcium and magnesium into solution. After digestion, the soluble calcium, potassium, magnesium and sodium content of dairy manure was reduced.

Preface

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List of Abbreviations

AD: Anaerobic digestion

GHG: Green house gas

WAS: Waste activated sludge

AOP: Advanced oxidation process

MWH₂O₂-AOP: Microwave hydrogen peroxide advanced oxidation process

Ortho-P: Orthophosphate

HRT: Hydraulic retention time

BMP: Biochemical methane potential

EPS- Extra-polymeric substance

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Chapter 1: Introduction

1.1.1 Dairy Industry in British Columbia

In 2010, British Columbia's dairy industry ranked first as the province's top agricultural and aquacultural commodity, generating \$494 million in farm cash receipts (M. o. A. BC, 2011). The dairy industry also ranked first as the main contributor to the organic waste in BC. In 2007, manure consisted of 82% of the organic waste produced in the BC Fraser Valley; out of the manure, 65% comes from cow manure, which equates to approximately 1.75 million tonnes of manure per year (Electrigaz Technologyies Inc., 2007).

1.1.2 Environmental Impact of Dairy Manure

Traditionally, dairy manure is applied on farm land as a source of valuable nutrients for the growth of crops (BC, Ministry of Agriculture Food and Fisheries., 2004; Jin, Hu, & Wen, 2009). However, when the production of manure of a farm exceeds the capacity of the farm land to receive the manure, then it can become a source of pollution. This is especially true for regions of growing population. Modern dairy farms favor high concentration of cows that can produce high volumes of milk. This trend is evident in British Columbia, where dairy production increased 3% from 2001 to 2007, while there was a 14% decrease in the number of dairy farms (PricewaterhouseCoopers, 2009). The increase in the concentration of dairy cows on limited farm stresses the land, which receive the dairy manure and fertilizers for the growth of feed crops. When farmland is oversaturated with nutrients, such as nitrogen (N), phosphorus (P) and potassium (K), it presents both environmental and economic challenges for the farm. Some environmental impacts of dairy manure include green house gas (GHG) emission, eutrophication of waterways and water bodies (Carpenter et al., 1998) and ground water contamination (Jin et

al., 2009; Rico, García, & Rico, 2011). As an example of ground water contamination, in the lower Fraser River Valley in British Columbia, which has the highest density of livestock in Canada, excess application of both manure and commercial fertilizer has led to an increase in the nitrate concentration of the Sumas River watershed (Schindler et al. 2006). The main concern for GHG emissions of dairy manure is the release of methane gas, which has the global warming capacity that is 25 times that of carbon dioxide (Bishop & Shumway, 2009; International Panel on Climate Change, 2007). Other environmental impacts of dairy manure include odor and release of pathogens.

1.1.3 Anaerobic Digestion of Dairy Manure

Anaerobic digestion (AD) of dairy manure is a proven technology that has been used to stabilize organics and capture biogas. It is an alternative to traditional manure management of land application. Benefits of AD include capture of methane gas, which reduces GHG emission and can be used as energy, pathogen reduction, stabilization of organics that generate high COD and BOD, odor reduction, sludge reduction and water quality protection (Bishop & Shumway, 2009; Lo & Liao, 2011; U.S. Environmental Protection Agency, 2002). Further, there can be income from the sales of final solids, while the solid and liquid effluent can still be used as fertilizer (Morse, Guthrie, & Mutters, 1996).

Anaerobic digestion is not a novel technology; it has been around before the turn of the 20th century (Abbasi et al., 2012). The longevity of the technology has not translated to wide acceptance in the United States (Bishop & Shumway, 2009). This is attributed to the high capital investment of conventional anaerobic digesters and the lack of knowledge on performance

(Bishop & Shumway, 2009; Lo et al., 1984). In the United States, AD became popular in the early 1970s due to the energy crisis, and it led to many failures and subsequent development of the technology (Lazarus & Rudstrom, 2007; U.S. Environmental Protection Agency, 2002). Between 1970 and 1990, rising oil prices opened the door for exploration of biogas as a source of renewable energy. During that time, 71 digesters started operation, but more than half of these digesters are no longer in operation (Lazarus & Rudstrom, 2007). In 1996, a survey of six dairy manure digesters operators from California revealed that the lack of cooperation from local utility companies, on top of poor design and technical flaws, led to unsuccessful operation of the digesters. Of the six operators, three of them had stopped using their system, which had a capital cost of \$100,000 to \$950,000 each at that time (Bishop & Shumway, 2009; Morse et al., 1996). A disadvantage of AD includes intense management, because AD is sensitive to changes in pH, temperature, ammonia toxicity and other types of inhibition (Chen, Cheng, & Creamer, 2008). As of July 2010, there were 126 digester projects operating on dairy farms in the United States, while the amount of energy production by AD systems continues to grow each year (U.S. Environmental Protection Agency, 2010).

1.1.4 Economic Feasibility of Anaerobic Digestion

The economic feasibility of AD in a region depends on the electricity prices provided by the local utility company, tipping fees, carbon credits, incentives from government policies, co-digestion with other wastes to increase biogas generation, and fiber sales (Bishop & Shumway, 2009; Electrigaz Technologyies Inc., 2007; Lazarus & Rudstrom, 2007). There is consensus between feasibility studies that the main difficulties for the development of AD is the low electricity pricing by local utilities, and the lack of government policies to encourage the

adoption of AD. Lazarus & Rudstrom (2007) reported that the reasons for the successful operation of an anaerobic digester on a 500-cow farm were due to the high electricity pricing and grants that the demonstration farm received (U.S. Environmental Protection Agency, 2002). Further, Bishop & Shumway (2009) included tipping fees from co-digestion of food waste, along with electricity generation, as key revenue sources for digesters. If AD was to be implemented without the possibility of co-digestion, then AD would not be economically viable in Washington State. Similarly, under the current provincial environment and energy policies, AD is not economically feasible in the BC Fraser Valley (Electrigaz Technologies Inc., 2007). The electricity prices in the Pacific Northwest are simply too low to implement digestion technology (Bishop & Shumway, 2009; Electrigaz Technologies Inc., 2007). In BC, the abundance of rivers and waterways favors low energy prices through hydroelectricity. Currently, BC Hydro has a Standing Offer Program for the feed-in-tariff and it does not favor renewable electricity through biogas generation. In 2007, the total price return is \$81.05/ MWh, but for a small biogas plant (<250 kWe) the average break even cost is \$115/MWh, It is recommended that the AD feed-in tariff should be adjusted to \$150/ MWh to make AD profitable in BC (Electrigaz Technologies Inc., 2007). A local biogas producer in the BC Lower Mainland reported that it cost around \$0.18 to produce one kWe, while the market price was only \$0.05 to \$0.07/ kWe (Canadian Business Executive). At the current electricity pricing by local utilities in BC and elsewhere in North America, AD of dairy manure is simply not a financially viable option as a way of biogas generation and waste management.

One way of making the dairy manure AD financially viable is to improve the digest process so that more biogas can be generated as revenue. In the past, researchers have tried to achieve this

either by reducing the hydraulic retention time (HRT), and hence reduce digester volume and capital cost, or to hydrolyze the anaerobically digestible organics to improve the kinetics of the biogas generation process. The hydrolysis stage of anaerobic digestion has been identified as the rate limiting step of AD of organic particulate slurries (Eskicioglu & Galvagno, 2012; Eskicioglu et al., 2008; Rico et al., 2011). If the rate of the hydrolysis stage can be shortened, then the HRT can be reduced and more biogas can be generated. In particular, slow fiber degradation is identified as a common impediment to AD of dairy manure (Jin et al., 2009). Fiber is a major component of dairy manure and is known for its recalcitrant nature. It is harder to degrade than other lignocelluloses, because the easily hydrolyzed part is already gone when it was previously digested by the cattle (Jin et al., 2009). Therefore, technologies are required for the breakdown of organics in dairy manure, in order for increased biogas generation during the AD process.

1.1.5 Methods for Organic Disintegration

Research focused on the disintegration of organics in dairy manure for the purpose of generating biogas is limited (Angelidaki & Ahring, 2000; Jin et al., 2009). Majority of research involves the study of the microwave hydrogen peroxide advanced oxidation process (MW/H₂O₂-AOP) as a potential pretreatment to release nutrient in dairy manure (Kenge et al., 2009; Lo et al., 2012; Pan et al., 2006; Qureshi et al., 2008; Yawson et al., 2011; Yu et al., 2010) but they do not consider the effect of pretreatment on anaerobic biodegradability. The relationship between MW/H₂O₂-AOP pretreatment and anaerobic biodegradability is an area in need of more research and study.

For municipal sewage sludge, pretreatment and anaerobic biodegradability has been well studied, including thermochemical (Dereix et al., 2006; Eskicioglu et al., 2009; Hong et al., 2006), biological, enzymatic lysis (Barjenbruch & Kopplow, 2003), mechanical disruption using stirred ball mill (Odegaard, 2004), ultrasonic (Nickel & Neis, 2007; Tiehm et al., 2001), physical disintegration using thermal pretreatment (Barjenbruch & Kopplow, 2003; Bougrier et al., 2008), steam explosion (Dereix et al., 2006), chemical base hydrolysis (Chiu et al., 1997) and combined chemical and enzymatic technology, such as wet oxidation (Stendahl & Jafverstrom, 2004; Svanström et al., 2004). This particular study focuses on the microwave treatment technology, combined with hydrogen peroxide addition.

1.1.6 Microwave Hydrogen Peroxide Advanced Oxidation Process

The microwave technology is similar to thermal hydrolysis technology—they both aim to increase the temperature of the sample in order to break apart the cell walls of microorganisms. The benefits of using microwave include less energy usage to increase sample temperatures, no direct contact between heat source and the material being heated, uniform heating throughout the material, selective and concentrated heating, smaller equipment size, instant accurate control, and better process control than conventional heating (Hong et al., 2006; Lo & Liao, 2011). For waste treatment process, such as sludge treatment, microwave treatment can reduce volume, improve dewaterability, enhance digestibility, enhance nutrient release, kill pathogen and stabilize heavy metal (Hong et al., 2006; Liao et al., 2005; Yu et al., 2010).

The microwave hydrogen peroxide advanced oxidation process (MW/H₂O₂-AOP) is a novel technology first developed by researchers in the University of British Columbia (Liao et al.,

2007; Liao et al., 2005; Wong et al., 2006). The technology was first intended to reduce suspended solids, disintegrate organics and solubilize nutrients in municipal sewage sludge; later, the concept was also applied to treat dairy manure (Pan et al., 2006; Qureshi et al., 2008). In addition to the effect of microwave treatment, hydrogen peroxide is a powerful oxidizer of organics. However, its activity is dependent on temperature and a catalyst is required to initiate decomposition of hydrogen peroxide into hydroxyl radicals, according to the Fenton reaction (Imlay et al., 1988). Hydrogen peroxide can be combined with a metal, such as Fe^{3+} , to produce hydroxyl radicals (OH^\cdot) that has an oxidation potential higher than that of H_2O_2 (Liao et al., 2005). Other methods of producing hydroxyl radicals without using metals include using microwave, ultraviolet (UV) ozone (O_3), ultrasound and thermal process (Eskicioglu et al., 2008). The process is called an advanced oxidation process (AOP) because hydroxyl radicals can be generated without the addition of metal catalysts. The addition of hydrogen peroxide in the microwave process helps to oxidize and destroy recalcitrant organic compounds, such as fiber, and convert sensitive organic compounds into CO_2 and water (Eskicioglu et al., 2008).

In the field of sewage sludge treatment, the oxidation power of hydrogen peroxide, along with the synergistic effect of microwave irradiation, is able to rupture cells in order to solubilize nutrients from sludge and disintegrate solids to obtain the high soluble content of COD. In theory, this is a good pretreatment for subsequent processes, such as anaerobic digestion and nutrient crystallization. Other research into the MW/ H_2O_2 -AOP includes acid hydrolysis to investigate the effect on solubilisation of orthophosphate, ammonia and soluble COD (Chan et al., 2007); another research looks at the role of ferrous sulphate addition and its effect (Lo et al., 2008). In the area of anaerobic digestion, work has been done to investigate the effects of

microwave and hydrogen peroxide on anaerobic digestion of municipal sewage (Eskicioglu et al., 2009; Eskicioglu et al., 2008). This study focuses on extending the work to investigate the effects of microwave and hydrogen peroxide on anaerobic digestion of dairy manure.

Chapter 2: Research Objectives

The focus of this investigation was to determine whether liquid dairy manure treated by MW/H₂O₂-AOP would increase the anaerobic biodegradability of the substrate. In the initial stages of this study, it was necessary to confirm that microwave treatment would facilitate nutrient release and organic solubilisation from dairy manure. The microwave treated substrate would undergo biochemical methane potential (BMP) tests to assess the extent and rate of anaerobic biodegradability. In particular, the Modified Gompertz Equation would be employed to assess BMP results and determine the rate of anaerobic digestion (Budiyono et al., 2010). Finally, a mass balance of trace metals (Na, Ca, K, Mg) content would be conducted in order to monitor the change in substrate metals characteristics throughout the microwave-anaerobic digestion process. In summary, the primary research objectives were as follows:

1. Demonstrate that MW/H₂O₂-AOP of liquid dairy manure would result in organic and nutrient solubilisation
2. Demonstrate that the increase in organic solubilisation of dairy manure, through MW/H₂O₂-AOP, would result in greater biogas generation through BMP tests
3. Determine the kinetics of anaerobic biodegradability using the Modified Gompertz Equation
4. Evaluate the effect of MW/H₂O₂-AOP and subsequent batch anaerobic digestion on the mass balance of metals

Chapter 3: Literature Review

3.1 Dairy Manure

Raw dairy manure is rich in nutrients, organic matter and inorganic residue, as it contains much of the feed and nutrient supplements that pass through the digestive system of a dairy cow. At the UBC Dairy Farm and Research Centre, feeds such as hay, grass silage, corn silage and other nutrient supplements are fed to the dairy cows. These feeds contain lignin, cellulose, hemicelluloses inorganic residue, and also carbohydrates, protein and some fats (Lo & Liao, 2011; Rico et al., 2007). The lignocellulosic components from dairy manure can be transformed into fermentable saccharides through enzyme or acid hydrolysis, which can be further transformed into ethanol and other valuable products (Wen et al., 2004). Substantial amounts of volatile fatty acids as acetic, propionic and butyric acids have been found in dairy manure filtrate and supernatants (Liao & Lo, 1987). For screened dairy manure, Rico et al. (2007) reported the concentrations of N-NH₄, TKN, P₂O₅, K₂O, CaO and MgO to be 716± 45, 2198± 117, 707± 70, 1801± 79, 695± 79, and 361± 24 mg/l, respectively. Dairy manure is rich in nutrients, such as phosphorus, nitrogen and minerals. Hence, dairy manure should not be considered a waste, but a valuable biological resource with potential for nutrient and energy recovery, such as through struvite (NH₄MgPO₄·6H₂O) crystallization and anaerobic digestion.

Struvite crystallization is a proven method for phosphorus removal from sewage sludge. However, dairy manure is also found to be quite complex with organic and nutrient content that is hard to solubilise, a condition needed for crystallization. Pan et al. (2006) and Jin et al (2009) attributed the difficulty in releasing phosphorus into solution to the high ratio of dissolved organic matter to microbial cells. Dairy manure contains different forms of phosphorus including

inorganic, residual P (nucleic acid-type), acid-soluble organic p, lipid P (Barnett, 1994; Pan et al., 2006). Most of the phosphorus (about 65%) is not very soluble and not easily hydrolyzed, especially the phosphorus in the structural makeup of manure, or in the suspended, particulate solids (Barnett, 1994; Gerritse & Vriesema, 1984).

For the successful anaerobic digestion of dairy manure, it first requires organics to be hydrolyzed prior to its availability to methane producing bacteria. Jin et al. (2009) report that dairy manure contains much fibrous material that is hard to degrade by anaerobic organisms. Fiber is known for its recalcitrant nature and is harder to degrade than other lignocelluloses, because it has been digested by the cattle and the easily hydrolyzed part is already gone. In literature, the total solids (TS), total chemical oxygen demand (TCOD), and the soluble to total chemical oxygen demand ratio (SCOD/TCOD) of the liquid fraction of separated dairy manure was reported to be 15.2 to 15.7 g/L, 23.6 g/L and 32 to 37%, respectively (Pan et al., 2006; Qureshi et al., 2008; Yu et al., 2010).

3.1.1 MW/H₂O₂-AOP on Dairy Manure

Research on the treatment of liquid dairy manure using the MW/H₂O₂-AOP has been conducted mainly under lab-scale batch studies (Pan et al., 2006; Qureshi et al., 2008). These studies worked with acidified manure, as it is the most effective way to solubilize ortho-P. In the beginning, Pan et al. (2006) focused on the effect of MW temperature (60, 90, 120, 170°C), heating time (5, 10, 15 min) and hydrogen peroxide (fixed, 5% vol/vol) addition on enhancement of phosphorus release; while Qureshi et al. (2008) extended the research to include solids, COD, VFA, ammonia, TP, TKN, Mg, Ca and K. Later, research also encompassed bench-scale,

continuous microwave treatment. Yu et al. (2010) investigated the synergistic effects of MW irradiation with H₂O₂, and the addition of acid dosages (0.2, 0.5 and 1% vol/vol), on solubilisation of nutrient and organic in dairy manure. Overall, the continuous system produced superior performance for increasing ortho-P and SCOD, compared to a batch system. This was attributed to the synergistic effects of MW irradiation and H₂O₂ (Yu et al., 2010). Finally, MW/H₂O₂-AOP treatment with different alkaline and acid chemicals was also investigated (Jin et al., 2009).

3.1.1.1 COD

The main factor for the solubilisation of organics is MW temperature (Yu et al., 2010). At low MW temperatures (60 to 70°C), little solubilisation of organics occurs, regardless of acid concentration or H₂O₂ addition. However, at MW temperatures above 80°C, SCOD/TCOD ratio increases (Yu et al., 2010). Acid dosage is also a factor for solubilisation. High dosages of H₂SO₄ have the highest increase in SCOD/TCOD, even without the use of H₂O₂ (Jin et al., 2009; Yu et al., 2010). With the combination of acid and H₂O₂, it is suggested that acid may help in the solubilisation process by stabilizing hydroxyl radical and inhibiting their degradation (Yu et al., 2010). A comparison between the MW/H₂O₂-AOP, with and without acid for dairy manure, has yet to be done, as most research has been focused on the release of phosphorus with acid addition.

The MW/H₂O₂-AOP has been shown to reduce organics. It was reported that the process reduces glucan/xylan, which is a degradation of cellulose and hemicelluloses in manure fiber (Jin et al., 2009). Reduction of glucan/xylan has been correlated with the increase in SCOD/TCOD. The

highest reduction of organics and increase in SCOD/TCOD were reported for high loading of H_2SO_4 (Jin et al., 2009). Qureshi et al. (2008) and Kenge et al. (2009) also reported a loss of TCOD at temperatures between 80 and 120°C, when H_2O_2 and sulfuric acid was used. The process of MW/ H_2O_2 -AOP on COD has two major reaction processes: breakdown of particulate organic, then further oxidation or gasification of the resulting organic products to VFA and/ or CO_2 (Liao et al., 2007; Shanableh & Shimizu, 2000). It is suggested that at MW temperatures below boiling, there is less oxidation taking place, so VFA are produced as the main end product, while at temperatures above boiling, CO_2 is produced (Qureshi et al., 2008).

3.1.1.2 Ortho-Phosphate

The solubilisation of ortho-phosphate (ortho-P) is most affected by acid addition; the MW/ H_2O_2 -AOP itself cannot release phosphorus into solution and has been shown to actually decrease ortho-P concentrations (Yu et al., 2010). Acid is needed to release P into solution or else little to no hydrolysis of P occurs for microwave treatment of liquid dairy manure (Pan et al., 2006; Qureshi et al., 2008; Yu et al., 2010). However, acid dosage (0.2, 0.5 or 1.0% v/v) was not significant for ortho-P release (Yu et al., 2010). In general, higher MW temperatures improved ortho-P release, although a slight decrease of ortho-P concentrations was observed between the temperatures of 90 to 100°C— this was attributed to greater intermediate products of polyphosphate (Pan et al., 2006). There are mixed results for the role of H_2O_2 in ortho-P release, especially at high MW temperatures of 120°C and above (Jin et al., 2009; Pan et al., 2006; Qureshi et al., 2008). In general, the effect of H_2O_2 is the greatest at low temperatures (60 to 80°C) (Pan et al., 2006; Qureshi et al., 2008), but there is less effect at higher temperatures

(around 120°C) (Jin et al., 2009; Qureshi et al., 2008). This might be attributed to different H₂O₂ dosages used.

3.1.1.3 Ammonia

The main factors for the release of ammonia in liquid dairy manure are H₂SO₄ addition and MW temperature, in that order. The highest ammonia release was at 170°C with H₂SO₄ and H₂O₂, although the addition of H₂O₂ resulted in only a slight increase of soluble ammonia (Qureshi et al., 2008). Without alkaline or acid treatment, MW alone produced low ammonium release (Jin et al., 2009). Also, Yu et al. (2010) found that MW alone decreased ammonia at 90°C. This could be similar to sludge treatment, where higher MW temperature reduced ammonia. This was attributed to polymerization reactions (Milliard reactions), reducing biopolymers containing N (Eskicioglu et al. , 2007). H₂O₂ addition and acid dosage are not significant factors for ammonia release (Qureshi et al., 2008; Yu et al., 2010).

3.1.1.4 VFA

Microwave treatment is able to increase VFA concentration. In general, VFA increased with higher MW heating temperature (Yu et al., 2010). At lower temperatures (between 60 and 80°C), acid and H₂O₂ produced more VFA than MW alone, but at higher temperatures, high acid dosage (1% vol/vol) may have an effect in decreasing VFA through the formation of CO₂ (Yu et al., 2010).

3.1.1.5 Metals

Trace metals such as calcium, magnesium and potassium can be found in substantial quantities in dairy manure (Qureshi et al., 2008). There is some indication that soluble potassium concentration can be increased with temperature, but overall, no consistent increase or decrease in metal concentration was reported after microwave digestion of dairy manure.

3.1.2 Performance of Anaerobic Digestion on Dairy Manure

Conventional digestion of dairy manure involves completely mixed reactors with typical hydraulic retention time (HRT) of 15 days or more (U.S. Environmental Protection Agency, 2004). Early research into AD of dairy manure compared the performance of conventional (suspended-biomass) and fixed-film reactors digesting screened manure (Lo & Liao, 1985). At a digestion temperature of 35°C, the maximum methane productivity of a conventional reactor is $0.63 \text{ L CH}_4 \text{ L}^{-1} \text{ day}^{-1}$ ($0.116 \text{ L CH}_4 / \text{g VS added}$), with a corresponding loading rate of $5.5 \text{ g VS L}^{-1} \text{ day}^{-1}$ and HRT of 6 days; In comparison, the maximum methane productivity of a fixed-film reactor is $6.20 \text{ L CH}_4 \text{ L}^{-1} \text{ day}^{-1}$ ($0.024 \text{ L CH}_4 / \text{g VS added}$), with a corresponding loading rate of $259 \text{ g VS L}^{-1} \text{ day}^{-1}$ and HRT of 3 hours (Lo & Liao, 1985). Fixed-film reactors are superior to conventional reactors because they can be operated at a low HRT without bacterial washout, can retain bacterial biomass, produce more methane gas at a given operating condition, and require less digester volume (Lo & Liao, 1985). More recent studies investigated the digestion of liquid dairy manure using upflow anaerobic sludge blanket (USAB) reactor (Rico et al., 2011). The researchers reported a methane production of $10.3 \text{ L CH}_4 \text{ L}^{-1} \text{ day}^{-1}$, with an organic loading rate of $40.8 \text{ g COD L}^{-1} \text{ day}^{-1}$, COD removal of 74% and HRT of 0.35 days (Rico et al., 2011).

For microwave pretreatment of dairy manure and subsequent anaerobic biodegradability, only one other study was found available. Jin et al. (2009) studied the effects of microwave-based thermochemical pretreatments of dairy manure, using sulfuric acid, hydrochloric acid, sodium hydroxide and calcium oxide. It was found that all pretreatment improved the SCOD/TCOD ratio compared to untreated manure. In particular, the use of sulfuric acid and hydrochloric acids at the highest loading produced the highest SCOD/TCOD ratios. However, methane production did not correlate with COD solubilisation and fiber degradation—the best methane production was for alkaline treatments. When sulfuric acid was used to treat manure, the methane production was lower than the raw manure. For hydrochloric acid addition, biogas release was comparable with alkaline treated biogas release, although a higher dosage of HCl decreased methane production (Jin et al., 2009). Angelidaki and Ahring (2000) also reported an increase in the biogas potential when bases such as sodium hydroxide and ammonium hydroxide were used. The low production of methane, for the sulfuric acid thermochemical microwave treatment, was attributed to sulfide toxicity and possible side reaction, such as Maillard reaction between amino acids and reducing sugars (Jin et al., 2009). Indeed, mesophilic and thermophilic digestion of acidified slurry fractions using sulfuric acid showed signs of sulphate inhibitions in these studies (Sutaryo et al., 2012a; Sutaryo et al., 2012b).

3.2 Sewage Sludge

Sewage sludge is composed of microbial cells and extracellular polymeric substances (EPS). This can include different groups of microorganisms and EPS containing network of organic and inorganic matter and cations (Eskicioglu et al., 2006; Frølund et al., 1996). These networks can be complex, with larger, hard to degrade floc particles made of cell wall fragments, proteins,

lipids, sugars, humic and nucleic acids (Eskicioglu et al., 2007). Microwave treatment usually acts to increase extra-cellular material and reduce the particulate material in sewage sludge.

3.2.1 MW/H₂O₂-AOP on Sewage Sludge

The operational factors for the MW/H₂O₂-AOP on sludge pretreatment include heating temperature, heating time, intensity, hydrogen peroxide dosage and chemical dosage. Their effect on solids disintegration and nutrient solubilisation will be discussed below.

3.2.1.1 COD

Microwave treatment alone of sludge can disintegrate solids and solubilise organics. The efficiency of disintegration is indicated by the soluble chemical oxygen demand (SCOD) and volatile fatty acid (VFA) increase after treatment. Wong et al. (2006) reported that temperature is the most significant factor affecting SCOD, with increased temperatures producing higher SCOD. This is confirmed by further research (Wong et al., 2007). Chan et al. (2007) also reported a higher SCOD at higher temperatures, and also when hydrogen peroxide was used. However, it was found that under high temperatures (above 100°C), the SCOD decreased, possibility due to conversion of organics into final oxidation products, such as CO₂. Other studies also observed a slight reduction in SCOD at MW temperatures between 75 to 96°C. Eskicioglu et al. (2007) attributed the reduction due to caramelization or Maillard reactions that occur between amino acids and reducing sugars, at higher temperatures. During these reactions,

soluble reducing sugars are polymerized and go to the particulate phase (Eskicioglu et al., 2007; Labuza, 1994).

3.2.1.2 VFA

VFA is an important parameter to gauge the level of solids disintegration in the MW-AOP. It is also a product of hydrolysis, and can be fed to methanogens to produce biogas, which can be captured for energy. Liao et al (2007) found that higher hydrogen peroxide dosage provided higher VFA concentration. Also, MW-AOP with acid addition favoured VFA production. The highest VFA concentration was under the highest hydrogen peroxide dosage and after acid addition. Under these two conditions, it was likely that hydrogen peroxide further oxidized organics into intermediate and final oxidation products, such as VFA and CO₂, and acid addition, helped to break down organics. However, acid addition reduced SCOD concentration (Liao et al., 2007). Another variable that improved VFA concentration after MW-AOP treatment is the ramp rate, or intensity of treatment. It was found that a long reaction time, coupled with the highest ramp rate, produced high VFA solubilisation (Lo et al., 2010).

3.2.2 Performance of Anaerobic Digestion on Sewage Sludge

The success of the microwave treatment on enhancing anaerobic biodegradability of sewage sludge is well recognized in research (Coelho et al., 2011; Eskicioglu et al., 2009; Hong et al., 2006; Park et al., 2004). Park et al., (2004) reported that BMP tests of microwave irradiated sludge produced up to 79% more biogas than control system (Park et al., 2004). At MW temperature of 96°C, MW-irradiated waste activated sludge had soluble COD and cumulative biogas productions that were 143± 34% and 211± 2% higher than control, after 23 days of

mesophilic AD, respectively (Eskicioglu et al., 2006). In terms of specific methane production, literature reports a theoretical methane production of 0.35 L/g COD removed (Eskicioglu et al., 2006), while other studies report values of 0.70 L CH₄/ g VS removed for acclimatized digesters and 0.64 L CH₄/ g VS removed for non-acclimatized, at 1 atm and 25± 1°C (Eskicioglu et al., 2009).

Acclimation of inoculum plays an important role in AD of MW pretreated sludge, because it can reduce initial toxicity by reducing lag time and provide higher ultimate biodegradability (Eskicioglu et al., 2009; Eskicioglu et al., 2007). The reason for the initial toxicity is due to the potential toxicity of thermally pretreated sludge. This toxicity can also lead to lower kinetics of microwave irradiated sludge, but the reaction rate and exponential methane production is unaffected (Eskicioglu et al., 2006). Another factor in methane production is the amount of total solids in the pretreated sludge, as a higher total solids content can increase biogas production (Eskicioglu et al., 2007).

Chapter 4: Methods and Materials

4.1 Equipment

The equipment used during the experiment and for chemical analysis are presented below.

Photos of the equipment can be found in Appendix C

Microwave Apparatus

MW pretreatment was performed using an industrial scale MW oven (Enwave Corporation, Annacis Island, BC) (Figure 25, Figure 26, Figure 27, Appendix C). It has a maximum output of 9000W, frequency of 2450 MHz, wavelength of 12.24 cm, and 6.15 L capacity. The microwave chamber houses a stand, holding coils of PETFE Teflon tubing 300 inches (0.350 m) in length and 1.25 inches (3.18 cm) in diameter. There is no pressure control for the MW oven. Hence, it is under atmospheric pressure and cannot exceed boiling temperature of the sample being heated. Temperature is regulated by the flow of influent pumped into the MW oven; given the pump flow, it determines the amount of time that sample is heated. Temperature is measured by a thermowell that is connected to the outlet after the substrate exits the microwave chamber and travels along tubing leading to the outlet, and then into an effluent tank. The flow is controlled by a progressive cavity pump connected to a speed controller.

Hydrogen Peroxide Injection Apparatus

Hydrogen Peroxide (H_2O_2) is directly injected into the influent flow stream after the influent pump. At that point, it is assumed that there is adequate mixing between the sample and H_2O_2 prior to MW pretreatment. The H_2O_2 is pumped using a peristaltic pump (Masterflex) and the flow is regulated by a speed controller.

Sulfuric Acid Injection and pH Control Apparatus

The pH of the influent can be set by a pH meter and controller. The influent tank is completely mixed by a variable mixer and the pH is monitored by a pH meter. The pH controller can pump sulfuric acid into the influent tank until the pH reaches a set-point.

Hach DR 2800™ Spectrophotometer (COD)

The equipment that was used for the experiment was the Hach DR 2800™ Spectrophotometer (Figure 28, Appendix C). A wavelength of 600 nm was used for analysis.

Block Digestion Apparatus (COD)

The block digestion apparatus was used to digest the samples for COD determination by spectrophotometry. The block digester was set at 150°C for two hours for complete reaction of organics into solution (Figure 29, Appendix C).

Lachat QuikChem 8000® Flow Injection Colorimeter (TP, TKN, PO₄³⁻, and NH₃⁺)

The equipment that was used for the experiment was the *QuikChem® 8000 Continuum Series Automated Ion Analyzer* (Figure 30, Appendix C). Calibration standards were prepared by Tim Ma, UBC Civil Engineering, and the concentrations were calculated by fitting the data against the corresponding calibration curve using computer software. This was all done automatically using the analyzer.

Block Digestion Apparatus (TP and TKN)

The block digestion apparatus was used to prepare the samples for determination by flow injection analysis (Figure 31, Appendix C). The temperature and time setting was digestion 135°C for five hours and then 380°C for three hours, in order to completely solubilise phosphorus and Kjeldahl nitrogen into solution.

Gas Chromatography Apparatus (VFA)

The equipment that was used for the experiment was the *Hewlett Packard 6890 Series gas chromatograph* equipped with a flame ionization detector (Figure 32, Appendix C). Calibration using known standards was prepared by Tim Ma, UBC Civil Engineering, using VFA concentrations of 2, 10, 50, 100, 200 and 1000 ppm. The results were calculated by finding the corresponding concentrations from the intensity versus retention time plot using peak area.

Infrared Coupled Plasma (Na, Ca, K, Mg)

The equipment that was used for the experiment was the *Perkin Elmer Optima 7300 DV Optical Emission Spectrometer* (Figure 33, Appendix C). The instrument took three measurements for all samples and reported the average. A calibration curve was also set up to determine the concentrations in the samples.

Block Digestion Apparatus (Na, Ca, K, Mg)

The block digestion apparatus was used to prepare the samples for determination by flow injection analysis (Figure 34, Appendix C). The temperature and time setting was digestion

120°C for two hours and 120°C, until samples were completely dissolved into solution for soluble and total analysis, respectively.

4.2 Microwave Pretreatment of Dairy Liquid Manure

4.2.1 Experimental Design

The dairy manure sampled is from the UBC Dairy Education & Research Centre (UBC Dairy Farm), University of British Columbia, located in Agassiz, BC, Canada. The farm houses approximately 500 animals, including dairy cows and heifers (UBC, 2012). The dairy manure sampled is the liquid portion extracted from the settling pond after liquid-solids separation. Microwave treatment was conducted immediately after manure collection. Four sets of microwave experiments were conducted: microwave only (MW), microwave with hydrogen peroxide (MW/H₂O₂), microwave with acid (MW/H⁺) and microwave with hydrogen peroxide and acid (MW/H⁺/H₂O₂). All sets were treated by a pilot-scale, continuous-flow, microwave set at 6000W, under a flow rate of around 0.6 L/min to achieve an effluent temperature of around 96°C. The effluent tank was open to the atmosphere. The acidified treatment sets (MW/H⁺ and MW/H⁺/H₂O₂) had their pH adjusted to around 2.7 using concentrated (30% vol/vol) H₂SO₄. The continuous flow of diluted hydrogen peroxide (10%) was injected into the microwave influent line at 20 mL/min, to achieve a H₂O₂ dosage of 0.3%.

4.2.2 Sampling and Analysis

4.2.2.1 Sampling Methods

A single batch of 140L of liquid dairy manure was taken from the settling pond and pumped into the microwave influent tank. The influent tank was under constant mixing and no additional volume was added into the tank during the duration of the experiment.

Effluent samples were collected immediately after each treatment set. After the completed treatment was changed over to the next, the effluent tank was rinsed with tap water and then a full hydraulic retention time (~5 minutes) was allowed to pass, in order to flush the microwave and effluent tank. The order of treatment is provided in Table 1.

Table 1. Order of treatment and sampling

Order of treatment	Treatment Set	Approximate sample temperature
1	MW	~92°C
2	MW/H ₂ O ₂	~95°C
3	untreated	~18°C
4	MW/H ⁺	~96°C
5	MW/ H ⁺ /H ₂ O ₂	~96°C
6	H ⁺	~18°C

4.2.2.2 Sample Handling and Preservation

About 10 L of treated samples were stored in 20 L carboys and immediately transported back to UBC. The time for transportation was approximately two hours. Upon arrival at UBC, the samples were stored in 4°C until preservation. Within 72 hours of arrival at UBC, microwave treated samples were divided into three subsamples and analyzed for solids, COD, VFA, PO₄-P,

NH₃-N, TP and TKN. Samples were centrifuged under 24900 rcf for 15 minutes and filtered through 0.45 µm, prior to soluble preservation.

The outline of sample collection, preservation and storage are presented in Table 2. Standard Methods was applied for sample handling and storage (APHA et al., 2005). All soluble samples were filtered through 0.45 µm.

Table 2. Sample volume, preservation and storage

Parameters	Container	Minimum sample liquid	Preservation	Temperature /Storage condition	Holding time before analysis
Ammonia-N	P	5 mL	1 drop 5% H ₂ SO ₄	4 °C, dark	5 to 9 days
Ortho-Phosphate-P	P	5 mL	1 drop of C ₈ H ₈ HgO ₂ polymeric	4 °C, dark	5 to 9 days
VFA	G	2 mL	1 drop of 50% H ₃ PO ₄	4 °C, dark	5 to 9 days
TP, TKN	P	5 mL	-	4 °C, dark	5 to 9 days
Sodium, Calcium, Potassium, Magnesium	P	10 mL	-	20°C, lab	30 days

P= Plastic (polyethylene or equivalent); G= glass

4.2.3 Analytical Methods

The analytical technique and methods for chemical analysis of samples are presented in Table 3.

Standard Methods was consulted for analyses.

Table 3. Summary for analytical techniques

Parameter	Analytical Technique	Reference
Total Solids (TS)	103-105°C	APHA. (2005). #2540 B. Total Solids at 103-105°C
Volatile Solids (VS)	550°C	APHA. (2005). #2540 E. Fixed and Volatile Solids Ignited at 550°C
Chemical Oxygen Demand (COD)	Closed Reflux, Colorimetric	APHA. (2005). #5220 D. Closed Reflux, Colorimetric Method
Ammonia (NH ₃ ⁺)	Flow Injection Analysis (FIA)	APHA. (2005). #4500-NH ₃ ⁻ H. FIA
Ortho-Phosphate (PO ₄ ³⁻)	FIA	APHA. (2005). #4500-P G. FIA for Orthophosphate
Volatile Fatty Acids (VFA)	Gas Chromatography (GC)	APHA. (2005). #5560 D. GC Method
Total Phosphorus (TP)	FIA	APHA. (2005). #4500-P H. Manual Digestion and Flow Injection Analysis for Total Phosphorus
Total Kjeldahl Nitrogen (TKN)	FIA	APHA. (2005). #4500-N _{org} D. Block Digestion and Flow Injection Analysis
Sodium, Calcium, Potassium, Magnesium (Na, Ca, K, Mg)	Infrared Coupled Plasma (ICP)	APHA. (2005). #3120 B. ICP Method
Alkalinity	Titration	APHA. (2005). #2320 B. Titration Method

4.3 Anaerobic Biodegradability of MW Pretreated Dairy Manure

4.3.1 Experimental Design

Anaerobic biodegradability of both untreated and the microwave treated (MW, MW/H₂O₂, MW/H⁺ and MW/H⁺/H₂O₂) liquid dairy manure was tested at mesophilic temperature (35±1°C) and thermophilic temperatures (55± 1°C), in a batch biochemical methane potential (BMP) assays. Five replicates were conducted for each experimental set, with a total of 50 batch digesters. For each replicate, an inoculum of 10 mL was added into 100 mL of sample inside 155 mL serum bottles (Figure 36, Appendix C). pH adjustment was made using either sulfuric acid (30% vol/vol), or sodium hydroxide (12.5M), to bring the pH to approximately 7.6. For the acidified microwave treated sets (MW/H⁺ and MW/H⁺), 2000 mg/L of NaHCO₃ of alkalinity (as CaCO₃) was added to the solution. The air space was sparged with helium before being sealed with a rubber stopper. Serum bottles were kept in an air filled, darkened incubator (35°C) (Figure 35, Appendix C) or a water bath with a cover (55°C) (Figure 36, Appendix C) until they stopped producing biogas. The volume of methane production was measured by inserting a needle attached to a manometer and reported in normal conditions of 293K and 101.3kPa.

4.3.2 Inoculum Acclimation

Mesophilic inoculum used in BMP assays was cultured through anaerobic digestion of untreated liquid dairy manure taken from UBC Dairy Farm. A reactor was created using a 50 L plastic bottle sealed with a rubber stopper. A tube was inserted through the rubber stopper to allow excess biogas to exit the reactor. For acclimation, the reactor was filled with approximately 25 L of liquid dairy manure and fed daily with approximately 500 mL of untreated dairy manure to sustain bacteria biomass. After feeding, the reactor was shaken vigorously to allow for adequate

mixing between the feed and the biomass. Constant biogas production was observed prior to usage of inoculums for BMP tests.

Thermophilic inoculum was cultured in a 1 L capped bottle in a water bath under 55°C for at least 10 days, which is the time required for biogas generation. Biogas was released and approximately 250 mL of untreated dairy manure was fed to reactor daily, except for weekends. Prior to feeding, the reactor substrate was centrifuged under 3000 rpm for 5 minutes and approximately 250 mL of the supernatant liquid was wasted. The characteristics of the inoculums used are reported in Table 4.

Table 4. Characteristics of inoculums used

	35°C ^a	35°C ^b	55°C
TS (%)	1.62± 0.02	4.27± 0.31	7.31± 0.14
VS (%)	1.10± 0.01	2.89± 0.19	4.47± 0.09
VS/TS (%)	68± 1	68± 5	61±1
TCOD (g/L)	18.7±0.47	45.0±3.96	60.3±2.43
SCOD (g/L)	5.07±0.24	6.84±0.59	7.49±0.28
SCOD/TCOD (%)	27±1	15±1	12±1
VFA (mg/L)	1578±99	2874±190	429±17
TP-total (mg/L)	113±17	423±71	792±38
TP-soluble (mg/L)	13±4	8±2	20±2
o-PO ₄ (mg/L)	30±1	16±2	18±1
TKN (mg/L)	1029±170	2032±389	4039±178
NH ₄ ⁺ -N (mg/L)	490±8	703±22	1156±7
pH	7.3	-	7.57
Soluble Na (mg/L)	225± 17	3385± 91	479± 24
Soluble K (mg/L)	2428± 114	3214± 75	2381± 43
Soluble Ca (mg/L)	2± 30	1734± 49	20± 1
Soluble Mg (mg/L)	237± 11	617± 14	263± 4

^a Used for untreated, MW, MW/H₂O₂, and MW/H⁺

^b Used for MW/H⁺/H₂O₂

4.3.3 Sampling and Analysis

The samples were collected immediately before and after digestion. Five replicate samples were analyzed for each treatment set, one for each batch digester. The analytical parameters were the same as that of microwave treatment in Section 4.2.

4.4 Anaerobic Biodegradability of MW Pretreated Sewage Sludge

4.4.1 Experimental Design

Refer to Section 4.3.1.

4.4.2 Inoculum Acclimation

Mesophilic and thermophilic inoculum was cultured in the same manner as the inoculum used for thermophilic digestion of dairy manure in Section 4.3.2, with the exception that the mesophilic inoculum was placed in an incubator under 35°C.

4.4.3 Sampling and Analysis

Refer to Section 4.3.3.

4.5 Mass Balance of Trace Metals (Ca, K, Mg, Na) using the MW/H₂O₂-AOP and Anaerobic Digestion

4.5.1 Experimental Design

The trace metals of calcium, potassium, magnesium and sodium were chosen for analysis because of their abundance in dairy manure, and their importance for nutrient recovery and anaerobic digestion. Total and soluble trace metals were evaluated to observe their changes

during MW/H₂O₂-AOP and subsequent AD. Total concentrations were assumed to be unchanged, except when pH and alkalinity were adjusted using sulfuric acid or sodium hydroxide, and sodium bicarbonate, respectively. Hence, total metals were sampled before and after pH and alkalinity adjustment. Soluble metals were sampled after MW pretreatment, before and after digestion.

4.5.2 Sampling and Analysis

For sample handling and analytical methods, please refer to Sections 4.2.2.2 and 4.2.3.

Chapter 5: Results and Discussion

5.1 Microwave Pretreatment of Liquid Dairy Manure

The full summary of the characteristics of the microwave treated and untreated samples are presented in Table 5.

Table 5. Chemical characteristics of dairy manure after microwave treatment

	Untreated	MW	MW/H ₂ O ₂	H ⁺	MW/H ⁺	MW/H ⁺ /H ₂ O ₂
TS (%)	3.02± 0.25	2.32± 0.03	2.39± 0.11	3.87± 0.12	2.74± 0.12	3.37±0.27
VS (%)	2.00± 0.18	1.54± 0.02	1.59± 0.07	2.46± 0.09	1.52± 0.10	2.06± 0.22
VS/TS (%)	66± 6	66± 1	67± 3	63± 2	55± 4	61± 6
TCOD (g/L)	29.4± 1.65	24.0± 0.60	24.4± 1.35	30.9± 1.56	21.3± 1.71	25.4± 2.91
SCOD (g/L)	9.81± 0.21	10.4± 0.31	11.0± 0.21	7.76± 0.22	8.49± 0.28	10.6± 0.75
SCOD/TCOD (%)	33± 2	43± 1	45± 2	25± 1	40± 3	42± 5
VFA (mg/L)	2853± 141	2759± 124	2762± 59	2523± 66	2596± 36	2418± 44
TP-total (mg/L)	239± 67	233± 2	213± 31	301± 7	278± 5	231± 82
TP-soluble (mg/L)	5± 2	13± 1	16± 1	197± 7	190± 4	200± 5
o-PO ₄ (mg/L)	11± 1	3± 2	3± 2	182± 6	195± 4	182± 1
TKN-total (mg/L)	1636± 476	1792± 26	1528± 203	1984± 45	1617± 100	1437± 547
TKN-soluble (mg/L)	943± 131	762± 149	698± 77	1114± 60	1124± 4	1115± 70
NH ₄ ⁺ -N (mg/L)	904±82	748±27	666±9	1052±50	1080± 15	1116± 22
pH	7.6	9.3	9.4	2.7	2.7	2.7

5.1.1 Soluble to Total COD

The SCOD/TCOD ratios increased for all MW treatment conditions (Table 5, Figure 1). The highest ratio achieved was under MW/ H₂O₂ (45± 2%), with up to 12± 3% increase compared to untreated. All microwave treated substrate achieved SCOD/ TCOD ratios that were in the same range. Compared to MW alone, the use of H₂O₂ appears to provide a slight increase in the solubilisation of organics, but the difference is insignificant. It is speculated that if the H₂O₂ dosage was increased, the difference will be more apparent. Qureshi et al. (2008) and Jin et al. (2009) reported significant increases in SCOD concentrations when H₂O₂ dosages of 0.04% (1:25 vol/vol) and 1% vol/vol were applied on liquid dairy manure, respectively. In addition, Yu et al. (2010) reported that the combine use of acid and H₂O₂ would improve solubilisation

compared to without acid because acid maintains, or perhaps even increases, hydroxyl radical concentration by stabilizing the radicals, thus preventing their degradation.

In the case when higher dosages of H_2O_2 be used, MW/ H^+ / H_2O_2 treatment should provide the highest SCOD/TCOD ratio. Indeed, Jin et al. (2009) reported an increase in organic solubilisation for microwave pretreatment with H_2O_2 addition. However, H^+ addition alone caused a decrease in SCOD, whereas the ratio of VS to TS remained similar to the untreated manure. This may be attributed to the chemical reactions or agglomeration between the soluble portions ($<0.45 \mu m$) in the dairy manure, leading to formation of larger particles ($>0.45 \mu m$). These larger particles would be disintegrated under MW treatment, and hence, SCOD values increased for acidified MW treatments.

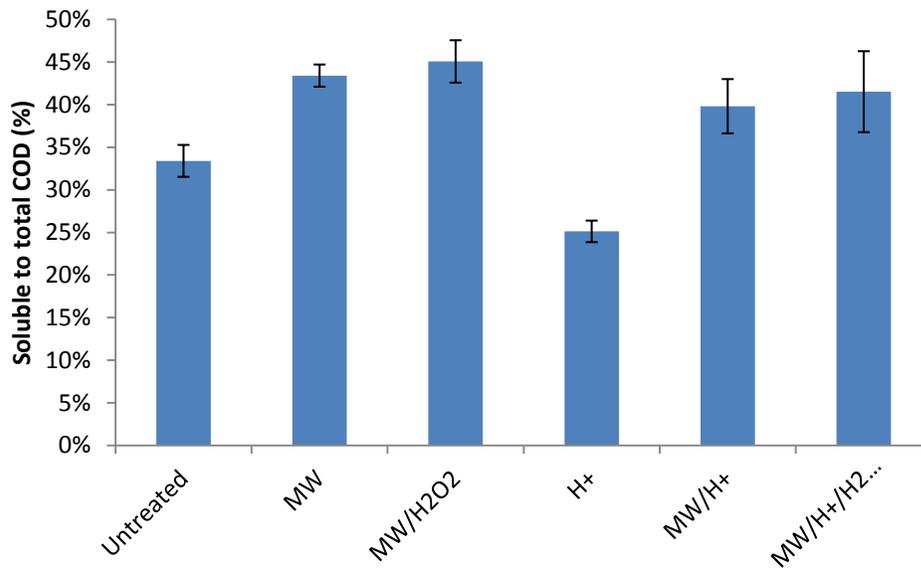


Figure 1. Solubilisation of COD using MW treatment

It should be noted that the SCOD concentration for MW/H⁺ treatment is less than the untreated sample. Nevertheless, the SCOD/TCOD ratio increased and is consistent with the other microwave treatments. It is observed that there is a decrease in TCOD for all microwave treatment conditions. This is attributed to the lack of adequate mixing in the influent tank prior to MW treatment. Another possibility is the decrease of TCOD due to volatilization, but this is not proven. Other studies have observed a decrease in TCOD at microwave temperatures close to 96°C. Qureshi et al. (2008) found that at MW temperatures 80°C and above, there is a reduction of TCOD, especially when the temperature was above 100°C and H₂O₂ was applied. The same phenomenon was observed for waste active sludge under MW temperatures of 80 to 120°C (Eskicioglu et al., 2008). It is likely that at elevated temperatures, the process goes into an oxidation or gasification process where the organics are converted into oxidation products as VFAs and/ or CO₂ (Qureshi et al., 2008; Shanableh & Shimizu, 2000). In this study, the microwave system was continuously operated using open vessels, and volatile organics could have easily escaped from the system. It is speculated that the decrease in TCOD is the combined effect of inadequate mixing and volatilization of organics from the system.

5.1.2 VFA

The total VFA concentrations, as acetic acid, for untreated and MW treated samples are presented in Figure 2. The VFA comprised of various organic acids, such as acetic, propionic, butyric, and valeric acids. Acetic acid accounted for the majority of the VFA. There was no improvement to VFA at any treatment scheme. Instead, VFA remained the same or decreased after pretreatment. The VFA concentrations between the untreated, MW and MW/H₂O₂ were similar, but with acid treatment, there was a slight decrease of VFA. This is different than the

findings by Yu et al. (2010), who reported that MW alone and MW/H⁺/H₂O₂ helped to produce more VFA than untreated manure. However, it was reported that higher acid concentrations (1% vol/vol) may reduce VFA concentrations at high MW temperatures (80 to 90°C) (Yu et al., 2010). This can be attributed to oxidation and gasification processes, similar to the loss of TCOD after MW treatment explained in the previous section. For the treatment of sewage sludge, it was found that microwave treatment did not increase VFA concentration (Eskicioglu et al., 2008).

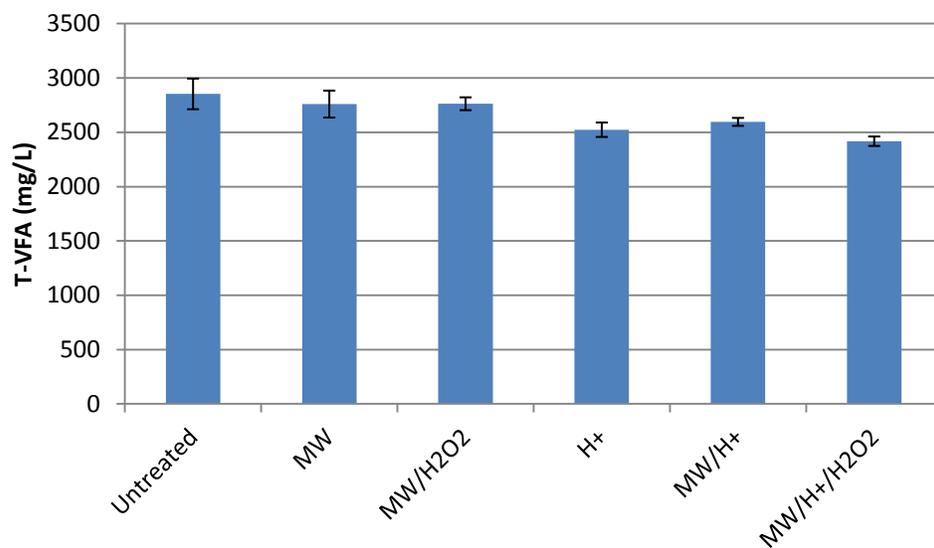


Figure 2. Solubilisation of VFA using MW treatment

5.1.3 Ortho-P

The most important factor for solubilisation of ortho-P was the addition of acid (Figure 3). This is consistent with previous studies (Pan et al., 2006; Yu et al., 2010). Acid alone to pH of 2.7 (0.6% vol/vol) is able to release substantial amounts of ortho-P, from 5± 1% (untreated) to 60± 2% of TP. With the additional effect of MW, the ortho-P release increased to 70± 2% of TP. The results suggest that, in order to release significant amounts of ortho-P from dairy manure into

solution, acid addition is required. MW treatments without acid did not improve the ortho-P concentrations, but instead it decreased ortho-P concentrations. This is consistent with other studies (Kenge et al., 2009; Yu et al., 2010). However, MW and MW/H₂O₂ treatments increased their soluble to total P ratios, from 2± 1% (untreated) to 5± 0% and 7± 1%, respectively (Figure 4). The decrease in ortho-P concentrations may be due to the production of intermediate products of phosphorus, such as polyphosphate, by thermal and oxidative processes. Overall, adding H₂O₂ in the treatment only provided a marginal release of ortho-P. This is consistent with other studies (Jin et al., 2009; Qureshi et al., 2008). The substantially high release of ortho-P reported by Pan et al. (2006) may be attributed to the higher hydrogen dosage used (5% vol/vol).

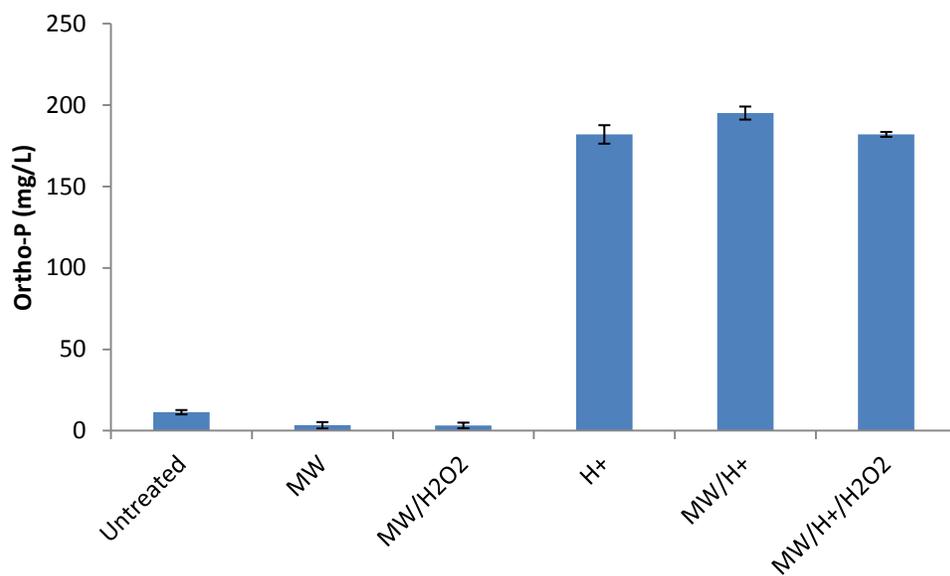


Figure 3. Release of orthophosphate using MW treatment

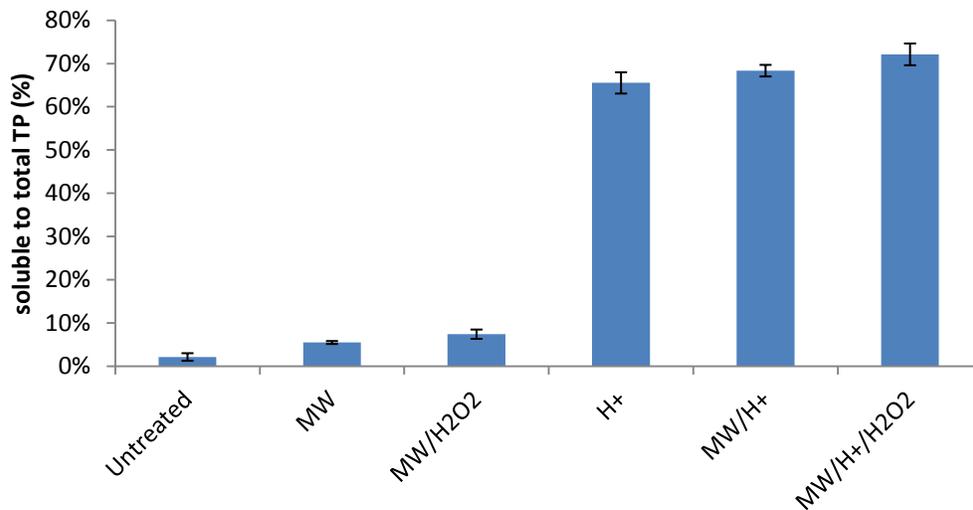


Figure 4. Solubilisation of phosphorus using MW treatment

5.1.4 Ammonia-N

Ammonia increased for acidified MW treatments (Figure 5). The effect of acid addition on ammonia release is similar to that of ortho-P—addition of acid is necessary for ammonia release. With respect to acid treatment, MW/H⁺ and MW/H⁺/H₂O₂ treatment sets provided minimal additional increase in ammonia concentrations. For acid treatment, a decrease of soluble to total TKN for was observed because MW treatment decreased the total TKN concentrations (Figure 6). It is assumed that nitrogen was lost to the atmosphere through volatilization during MW treatment. Hence, the highest ammonia release was observed for MW/H⁺ and MW/H⁺/H₂O₂ treatment sets at 67± 4% and 65± 3%, respectively. Like ortho-P, MW and MW/H₂O₂ treatments decreased ammonia concentrations. This result was also observed in other experiments involving MW treatment of liquid dairy manure (Yu et al., 2010) and waste activated sludge (Eskicioglu et al., 2007). Eskicioglu et al. (2007) attributed the decrease to Maillard reactions between amino acids and reducing sugars at high temperatures (75 to 96°C) leading to polymerization and

reduction of amino acids, proteins and other biopolymers containing nitrogen. H₂O₂ addition is not shown to provide a substantial release of ammonia or solubilisation of nitrogen (Figure 6).

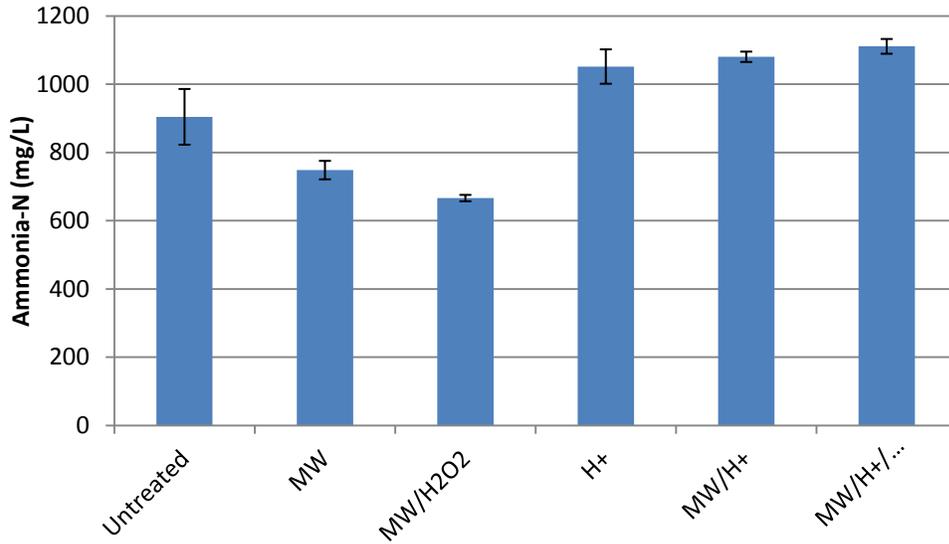


Figure 5. Release of ammonia-N using MW treatment

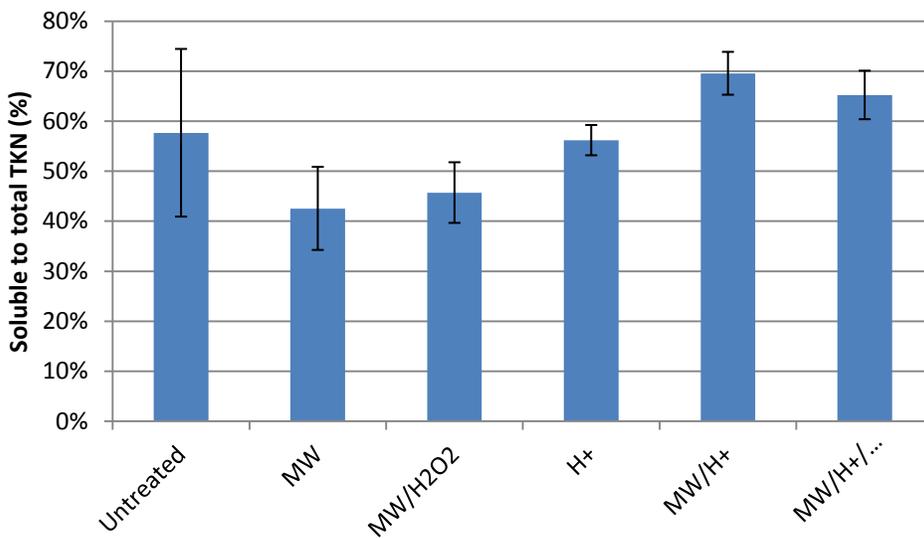


Figure 6. Solubilisation of nitrogen using MW treatment

5.2 Anaerobic Digestion of MW Pretreated Dairy Manure

After microwave pretreatment, the pH of dairy manure was out of optimal range (6.8 to 7.6) for anaerobic digestion. Therefore, an adjustment of pH was required before digestion. The alkalinity addition, prior to anaerobic digestion, was also needed for the MW/H⁺ and MW/H⁺/H₂O₂ sets. The results for the chemical characteristics of substrates before and after mesophilic and thermophilic digestion are given in Table 6 and Table 7, respectively.

Table 6. Chemical characteristics of substrates before and after mesophilic digestion (35°C)

Before	Untreated	MW	MW/H ₂ O ₂	MW/H ⁺	MW/H ⁺ /H ₂ O ₂
TS (%)	3.02 ± 0.09	2.27 ± 0.17	2.28 ± 0.23	4.08 ± 0.08	3.94 ± 0.45
VS (%)	2.00 ± 0.07	1.48 ± 0.116	1.48 ± 0.15	2.10 ± 0.05	1.96 ± 0.25
TCOD (g/L)	31.4 ± 6.33	27.1 ± 2.01	26.5 ± 2.28	33.9 ± 0.87	32.5 ± 5.19
SCOD (g/L)	9.08 ± 0.19	9.20 ± 0.48	9.37 ± 0.67	8.4 ± 0.29	10.7 ± 0.86
VFA (mg/L)	4384 ± 858	3157 ± 431	3427 ± 698	2669 ± 459	2766 ± 295
TP-total (mg/L)	184 ± 10	178 ± 15	167 ± 19	266 ± 21	294 ± 45
TP-soluble (mg/L)	8 ± 1	20 ± 3	24 ± 3	29 ± 4	58 ± 5
PO ₄ (mg/L)	8 ± 2	8 ± 2	10 ± 3	42 ± 2	15 ± 1
TKN- total (mg/L)	1831 ± 440	1796 ± 80	1704 ± 329	1768 ± 167	1757 ± 270
TKN- soluble (mg/L)	738 ± 54	604 ± 195	489 ± 86	1117 ± 40	1116 ± 96
NH ₄ (mg/L)	864 ± 58	689 ± 6	610 ± 47	898 ± 15	798 ± 53
pH	7.6	7.6	7.6	7.6	7.8
After					
TS (%)	2.22 ± 0.08	1.69 ± 0.12	1.66 ± 0.13	3.51 ± 0.08	3.25 ± 0.33
VS (%)	1.18 ± 0.05	0.92 ± 0.07	0.92 ± 0.08	1.85 ± 0.06	1.53 ± 0.16
TCOD (g/L)	20.1 ± 0.98	15.1 ± 1.08	15.4 ± 0.88	30.8 ± 1.32	31.2 ± 4.74
SCOD (g/L)	3.02 ± 0.15	3.89 ± 0.16	3.66 ± 0.34	11.0 ± 1.07	11.3 ± 0.87
VFA	38 ± 10	10 ± 3	0 ± 0	1885 ± 484	1955 ± 192
TP-total (mg/L)	295 ± 19	232 ± 24	241 ± 57	282 ± 6	280 ± 48
TP-soluble (mg/L)	2.0 ± 1	23 ± 1	32 ± 9	8 ± 2	22 ± 2
PO ₄ (mg/L)	10 ± 1	20 ± 1	24 ± 1	19 ± 3	22 ± 1
TKN- total (mg/L)	2057 ± 112	1884 ± 69	1649 ± 170	1770 ± 66	1617 ± 336
TKN- soluble (mg/L)	1102 ± 108	1091 ± 117	1131 ± 32	1368 ± 46	1167 ± 171
NH ₄ (mg/L)	1198 ± 22	1117 ± 16	985 ± 19	1266 ± 29	1050 ± 83
pH	7.5	7.8	7.7	7.6	7.5

Table 7. Chemical characteristics of substrates before and after thermophilic digestion (55°C)

Before	Control	MW	MW/H ₂ O ₂	MW/H ⁺	MW/H ⁺ /H ₂ O ₂
TS (%)	3.72 ± 0.03	3.61 ± 0.14	3.52 ± 0.12	4.60 ± 0.07	4.31 ± 0.46
VS (%)	2.44 ± 0.02	2.42 ± 0.10	2.33 ± 0.09	2.44 ± 0.04	2.18 ± 0.26
TCOD (g/L)	40.3 ± 2.30	38.0 ± 2.77	36.9 ± 0.82	38.1 ± 0.52	34.1 ± 4.58
SCOD (g/L)	10.9 ± 0.21	10.2 ± 0.50	10.6 ± 0.21	9.59 ± 0.09	9.87 ± 1.07
VFA (mg/L)	3802 ± 94	3292 ± 88	3249 ± 66	2755 ± 106	1733 ± 434
TP-total (mg/L)	408 ± 28	373 ± 30	403 ± 19	421 ± 15	380 ± 48
TP-soluble (mg/L)	35 ± 5	21 ± 3	26 ± 10	65 ± 3	68 ± 9
PO ₄ (mg/L)	22 ± 3	6 ± 1	8 ± 2	51 ± 3	39 ± 5
TKN- total (mg/L)	2907 ± 206	2505 ± 201	2638 ± 69	2989 ± 82	2556 ± 297
TKN- soluble (mg/L)	1530 ± 31	1275 ± 28	1155 ± 14	1467 ± 15	1345 ± 92
NH ₄ (mg/L)	1043 ± 15	845 ± 21	728 ± 8	937 ± 44	707 ± 58
pH	7.3	7.4	7.3	7.1	7.3
After					
TS (%)	2.95 ± 0.03	3.09 ± 0.12	3.09 ± 0.10	4.00 ± 0.08	3.81 ± 0.42
VS (%)	1.68 ± 0.02	1.97 ± 0.08	1.96 ± 0.05	2.00 ± 0.05	1.84 ± 0.23
TCOD (g/L)	23.1 ± 0.19	28.9 ± 0.90	27.7 ± 1.20	32.3 ± 1.02	29.6 ± 5.47
SCOD (g/L)	5.30 ± 0.32	10.3 ± 0.24	11.3 ± 0.81	11.9 ± 0.45	11.0 ± 1.53
VFA (mg/L)	271 ± 30	3069 ± 154	3002 ± 188	3073 ± 176	2649 ± 259
TP-total (mg/L)	388 ± 22	339 ± 23	337 ± 22	333 ± 9	311 ± 29
TP-soluble (mg/L)	11 ± 4	38 ± 7	37 ± 2	21 ± 1	29 ± 6
PO ₄ (mg/L)	9 ± 2	23 ± 4	21 ± 1	11 ± 1	17 ± 1
TKN- total (mg/L)	2225 ± 165	2049 ± 41	1893 ± 89	2485 ± 66	2036 ± 229
TKN- soluble (mg/L)	1576 ± 230	1419 ± 43	1199 ± 55	1723 ± 86	1476 ± 155
NH ₄ (mg/L)	1357 ± 22	1209 ± 16	1052 ± 9	1325 ± 9	1118 ± 96
pH	7.8	7.6	7.6	7.5	7.4

5.2.1 Dairy Manure Characteristics Before and After Digestion

5.2.1.1 Soluble to Total COD

Before mesophilic digestion, the SCOD/TCOD ratio for MW and MW/H₂O₂ remained higher than untreated. This should mean that bacteria in subsequent anaerobic digestion should be better able to access the organic matter in the two sets and convert it into biogas. In acidified treatments, the ratios decreased after pH adjustment, and in the case of MW/H⁺ treatment, its ratio was below that of untreated (Figure 7 and Figure 8). This may be attributed to the addition of sodium bicarbonate and sodium hydroxide, which increased TCOD concentrations. It is also

possible that the return of the acidified substrates back to their original pH of 7.6 shifted the soluble organic compounds into insoluble forms. The SCOD/TCOD characteristics prior to digestion are similar for both temperature treatments. However, the decrease in ratio for MW and MW/H₂O₂, before thermophilic digestion, could be attributed to the longer storage time of the substrates before analysis.

After mesophilic digestion, SCOD/TCOD ratios decreased for untreated and microwave treatments without acid, but increased for microwave treatments with acid (Figure 7). The drop in SCOD/TCOD ratios reveals that organic substrate was removed through anaerobic biodegradation. The TCOD removal after mesophilic digestion are 36, 44, 42, 9 and 4% for untreated, MW, MW/H₂O₂, MW/H⁺ and MW/H⁺/H₂O₂, respectively. It is suspected that inhibition played a role for MW/H⁺ and MW/H⁺/H₂O₂, and will be discussed in subsequent sections. Inhibition was also observed for the thermophilic digestion of all MW treatments (Figure 8).

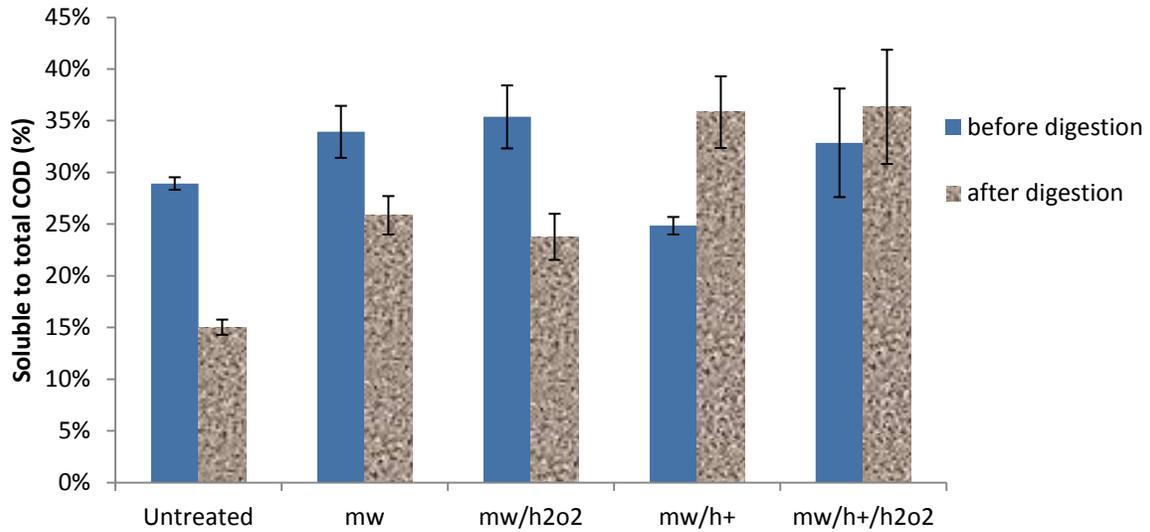


Figure 7. Soluble to total COD before and after mesophilic digestion

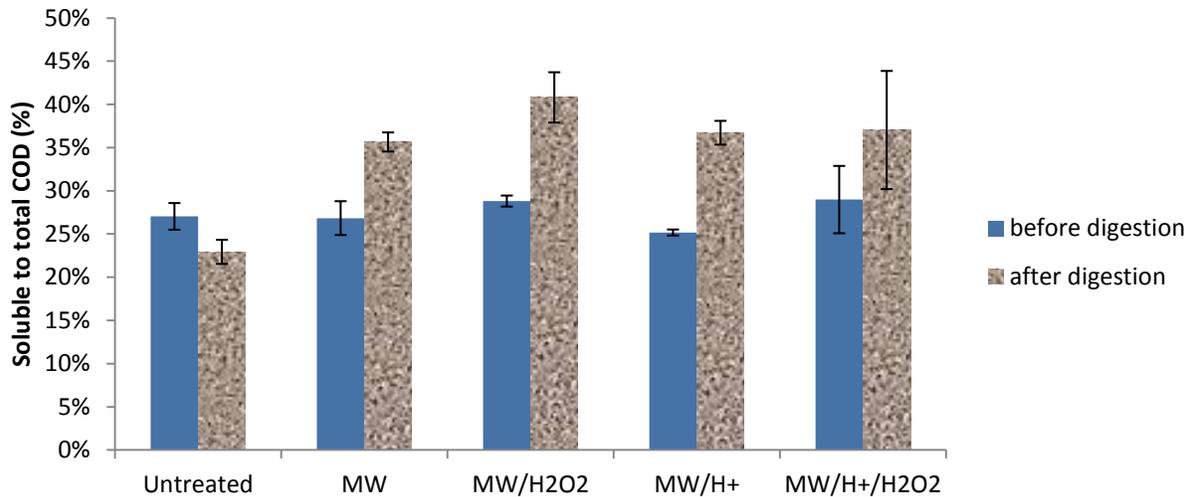


Figure 8. Soluble to total COD before and after thermophilic digestion

5.2.1.2 VFA

Microwave treatment and pH adjustment did not improve the VFA concentrations but rather decreased the concentrations. Some VFA may have been lost to the atmosphere after microwave treatment. After mesophilic anaerobic digestion, VFA concentrations were almost depleted for

untreated, MW and MW/ H₂O₂ (Figure 9). Substantial VFA remained for MW/H⁺ and MW/H⁺/H₂O₂ and for thermophilic digestion of all MW pretreated substrate (Figure 10). This indicates that some type of inhibition occurred.

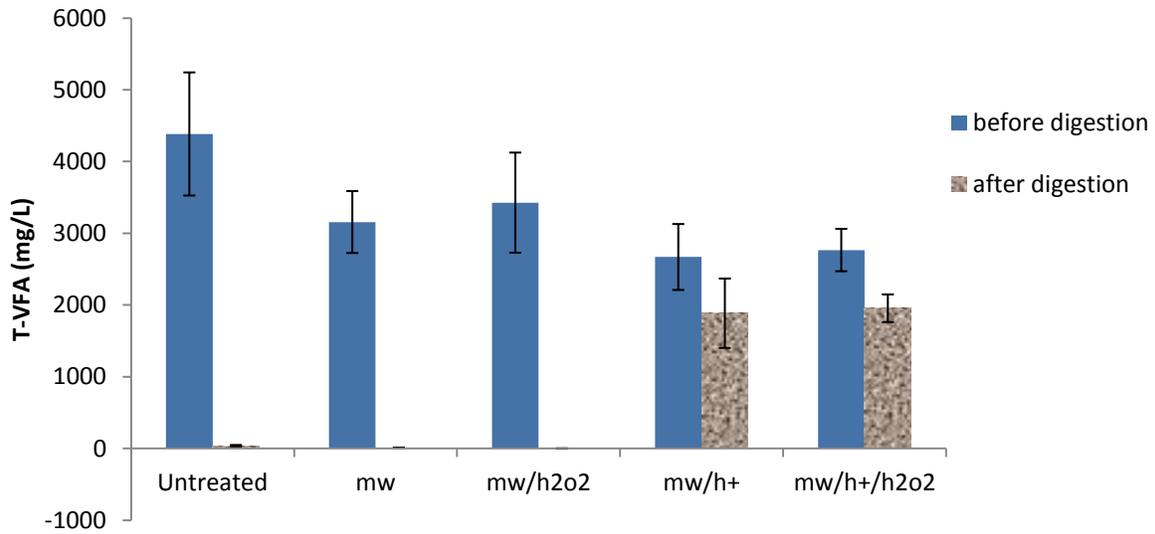


Figure 9. Total VFA concentrations before and after mesophilic digestion

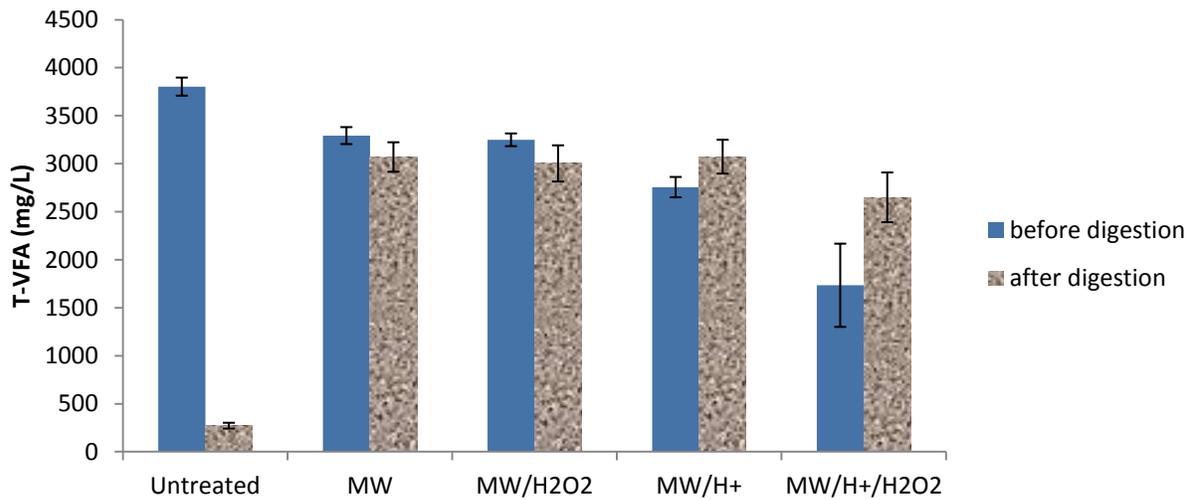


Figure 10. Total VFA concentrations before and after thermophilic digestion

5.2.1.3 Ortho-P

The ortho-P concentration profiles were similar between mesophilic and thermophilic digestion (Figure 11 and Figure 12). Microwave treatment with acid addition had the highest ortho-P concentration. In reality, this ortho-P concentration is still substantially lower than the ortho-P concentration of 195 ± 4 mg/L under low pH conditions. When the released phosphorus in a low pH solution is brought back to the original pH of 7.6, it could have formed struvite or calcium phosphate particles, because the increase in pH lowers the equilibrium solubility of calcium-phosphorus species (Zhang et al., 2010). For MW/H⁺/H₂O₂, due to H₂O₂ residual, the substrate needed to be mixed under room temperature for several days until the H₂O₂ residual disappeared. Unfortunately, during this period, the ortho-P concentration decreased substantially.

After anaerobic digestion, ortho-P increased for microwave treated dairy manure (Figure 11 and Figure 12). This is expected as anaerobic digestion helps to solubilise phosphorus. The majority of phosphorus in manure exists as inorganic phosphorus on the particulate matter of manure (Barnett, 1994). Anaerobic digestion helps to degrade these particles and release calcium-bound P as ortho-P. The reason for the decrease in ortho-P after digestion for the MW/H⁺ and MW/H⁺/H₂O₂ sets is currently unknown, but it is suspected that the phosphates released at low pH mineralized during anaerobic digestion and became associated with particulate-bound solids, forming inorganic P such as dicalcium phosphate (Carliell & Wheatley, 1997; Güngör and Karthikeyan, 2008). Also, the ortho-P concentration of the untreated set decreased after digestion at 55°C.

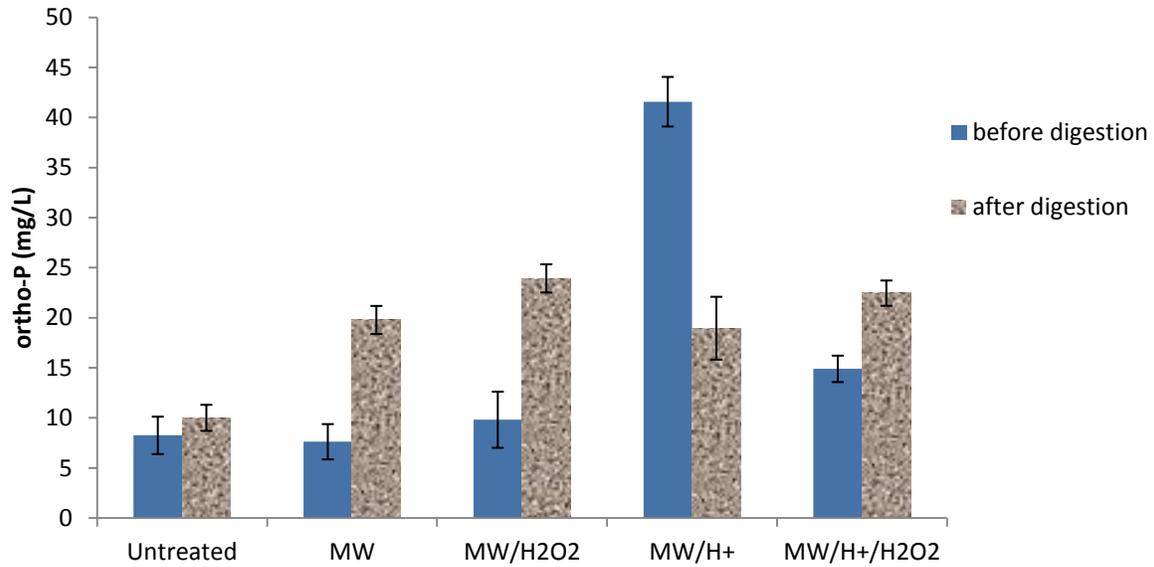


Figure 11. Ortho-P concentrations before and after mesophilic digestion

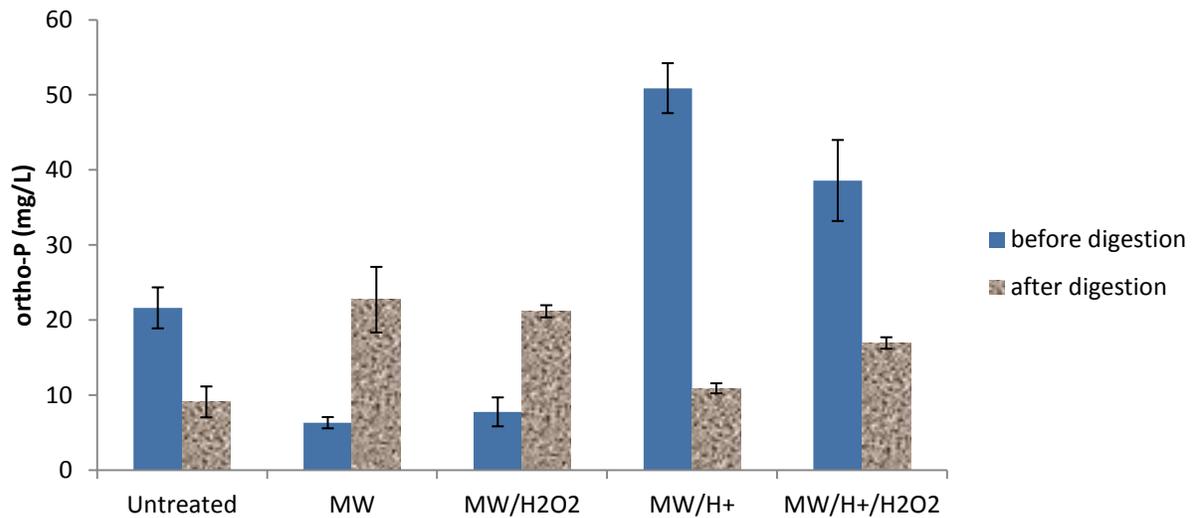


Figure 12. Ortho-P concentrations before and after thermophilic digestion

5.2.1.4 Ammonia-N

The ammonia concentrations were similar and consistent between the mesophilic and thermophilic sets (Figure 13 and Figure 14). Before digestion, pH adjustment and alkalinity

addition to the acidified sets reduced the ammonia concentrations and brought the values to the levels of untreated samples. After digestion, all sample sets increased in ammonia content. The increased solubilisation of nitrogen, as ammonia, through AD is due to the biodegradation of nitrogenous organic compounds (Lo et al., 1984).

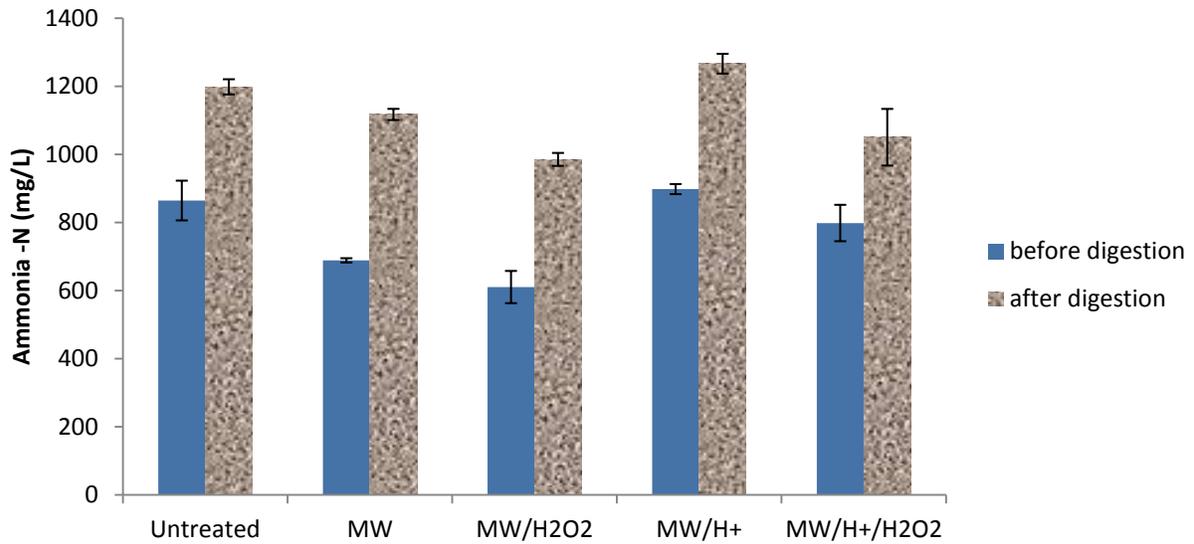


Figure 13. Ammonia-N concentrations before and after mesophilic digestion

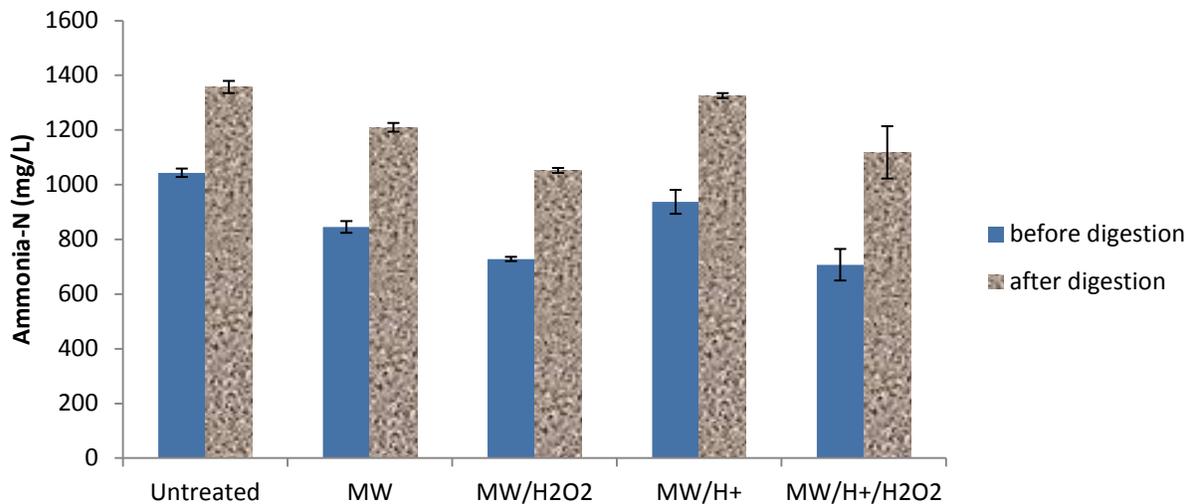


Figure 14. Ammonia-N concentrations before and after thermophilic digestion

5.2.2 BMP Test Results

The cumulative methane production profiles for the BMP tests are graphed in Figure 15, Figure 16 and Figure 17. For this study, due to the differences in TCOD values of initial pretreated substrates before digestion, methane production is provided as the volume of CH₄ per mass of VS added or destroyed. The untreated manure sets (35 and 55°C) achieved the best results for final cumulative methane production (ultimate biodegradability), compared to microwave treated samples. Under mesophilic (60 days) and thermophilic digestion (57 days), untreated manure achieved 265± 13 mL CH₄/ g VS added (34% TCOD removal) and 199± 10 mL CH₄/ g VS added (43% TCOD removal), respectively (**Error! Reference source not found.**). Although the COD/TCOD ratios were higher for the MW and MW/H₂O₂ treatment sets, this did not improve methane production. At 35°C, digestion of MW and MW/H₂O₂ treated dairy manure had similar biogas generation profiles (Figure 16) and methane yields (**Error! Reference source not found.**). This is also true for the treatment sets at 55°C. Hence, the addition of H₂O₂ does not appear to improve the amount of methane generated. Furthermore, acid (H₂SO₄) addition does not help in methane generation. The biodegradation of MW/H⁺ and MW/H⁺/H₂O₂ sets produced lower methane yields than the MW and MW/H₂O₂ treatment sets. H₂O₂ addition appears to play a role in biogas generation when acid is added. The MW/H⁺/H₂O₂ treatment set (35 and 55°C) produced lower ultimate methane yield than MW/H⁺ (Figure 17). This reinforces the assumption that H₂O₂ does not help in methane generation, but might have the adverse effect of inhibiting methane production, due to chemical toxicity.

At 35°C, there was no severe inhibition for the MW and MW/H₂O₂ treatments, while severe inhibition was observed for MW/H⁺ and MW/H⁺/H₂O₂. At 55°C, some degree of inhibition was

observed for all microwave treatment sets— methane yields for the MW and MW/H₂O₂ treatments (55°C) achieved less than half of their counterparts at 35°C. This inhibition was reflected in the exceptionally high SCOD/TCOD and VFA concentrations after digestion (Figure 7, Figure 8, Figure 9, and Figure 10). Inhibition was also reflected in the methane composition of the biogas. For 35°C, after maximum methane composition was observed, the methane composition for untreated, MW, MW/H₂O₂, MW/H⁺, and MW/H⁺/H₂O₂ were 62-74%, 69- 80%, 65- 80%, 35-70% and 41- 70%, respectively (Appendix B). The sets with the lowest methane composition belong to the sets with low methane production, namely, MW/H⁺, and MW/H⁺/H₂O₂. Low methane composition was also observed for all microwave treated sets at 55°C. This means that inhibition is affected by not only sulfuric acid addition, but also the AD temperature. Many different factors could be involved in the inhibition of anaerobic digesters, including ammonia, sulfide, light metal ions, heavy metals and organics (Chen et al., 2008).

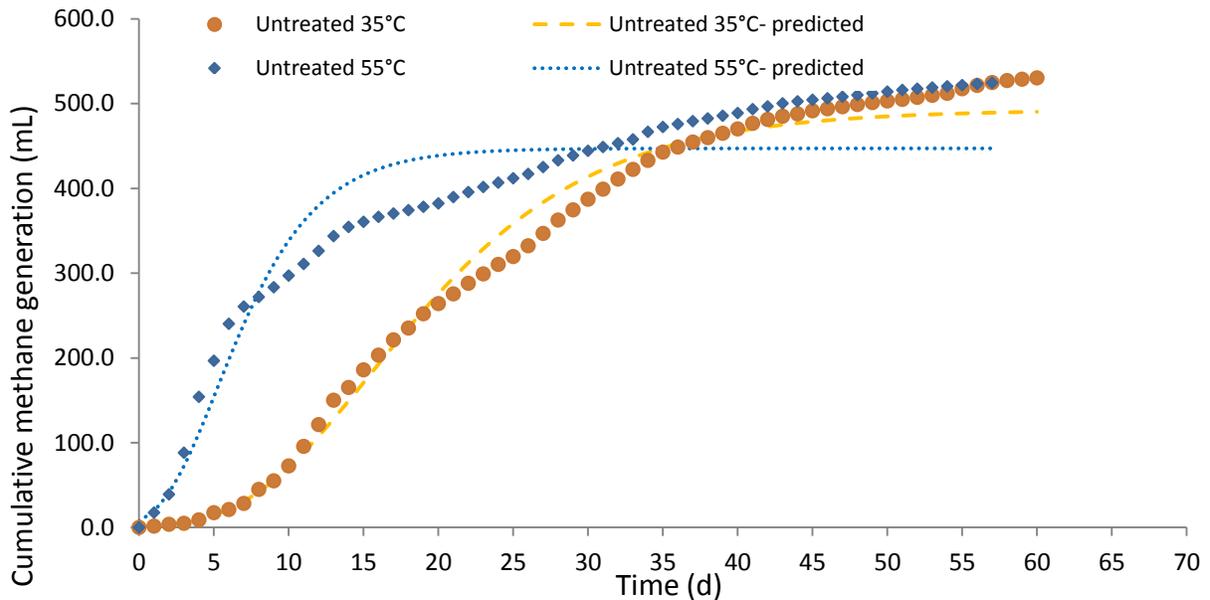


Figure 15. Methane production profile for untreated dairy manure at 35 and 55°C

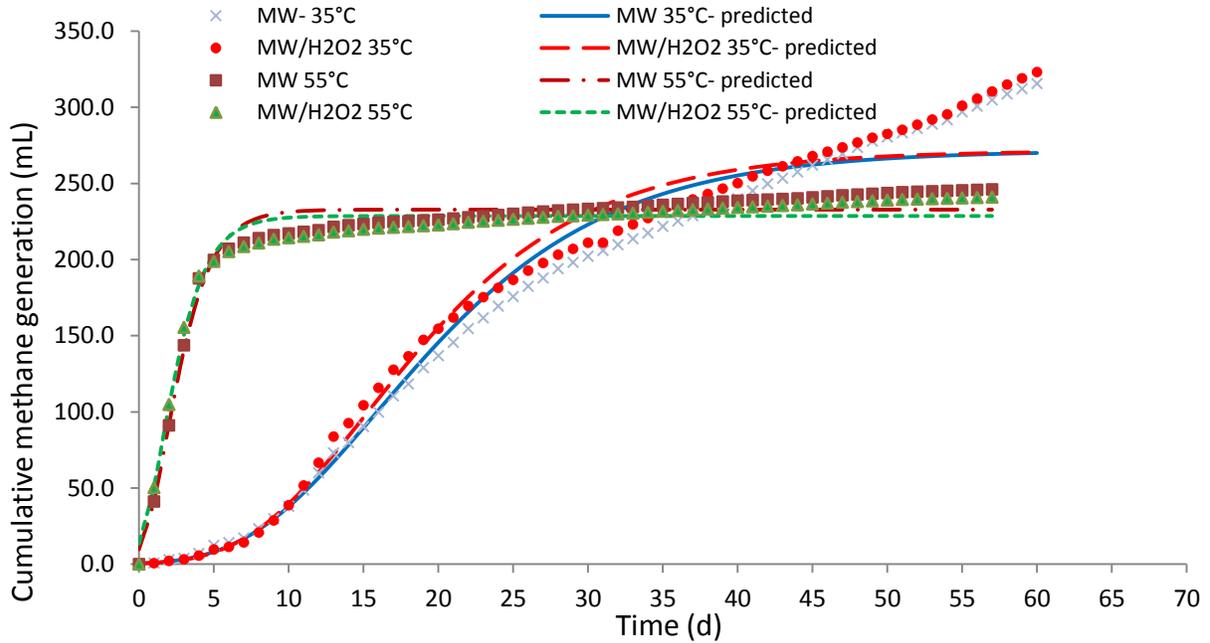


Figure 16. Methane production profile for MW and MW/H₂O₂ dairy manure at 35 and 55°C

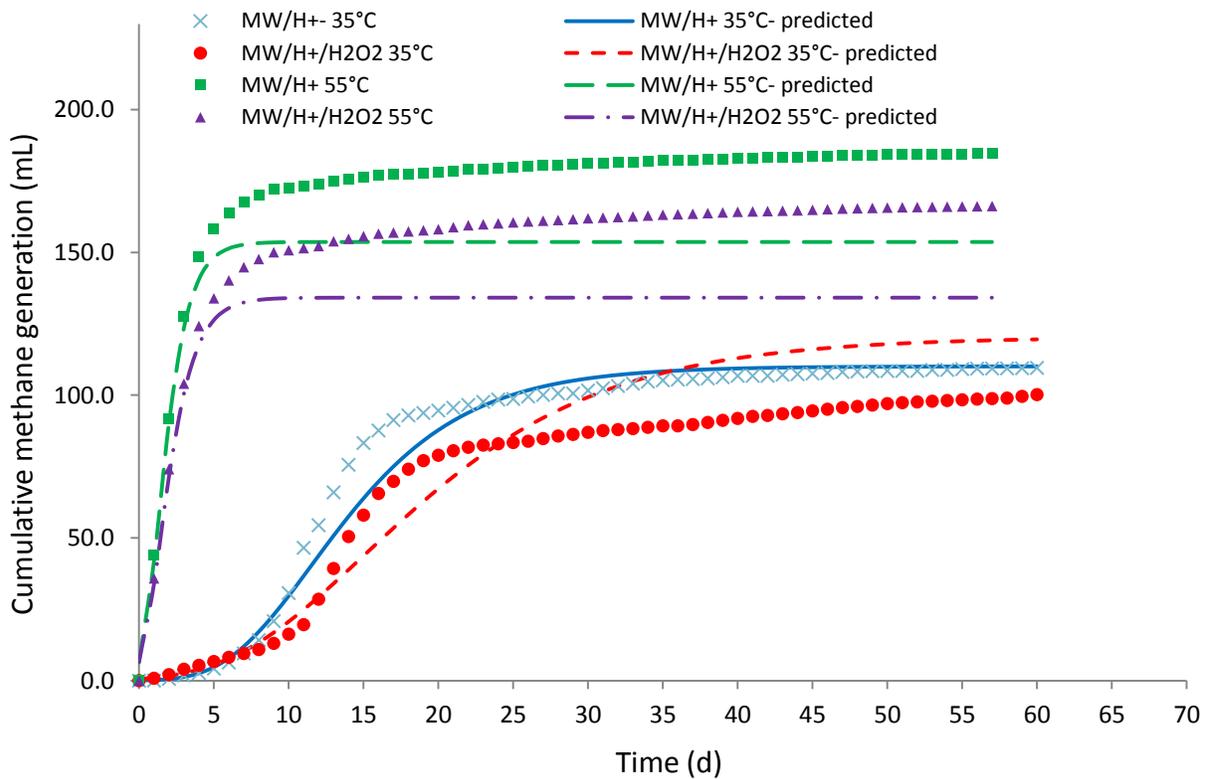


Figure 17. Methane production profile for MW/H⁺ and MW/H⁺/H₂O₂ dairy manure at 35 and 55°C.

5.2.2.1 Inhibition

Dairy manure contains high ammonia concentrations and it may be a contributing factor for inhibition of the batch digesters. High ammonia concentration occurs from the decomposition of organic nitrogen. Free ammonia (NH_3), not ammonium ion (NH_4^+), is the main cause of inhibition (De Baere et al., 1984). Ammonia toxicity has been suggested as the main reason for digester failures of animal wastes (Braun et al., 1981; Hashimoto, 1986). Indeed, the analysis of the substrates before and after BMP tests, reported high ammonia concentrations in the remaining solution, especially for substrates that have gone through acid treatment (Figure 13 and Figure 14). Factors that affect ammonia inhibition include concentration, pH, temperature, presence of other ions and acclimation period (Chen et al., 2008).

There is a significant range of inhibiting ammonia concentrations reported in the literature, due to differences in substrate and inocula, environmental conditions (temperature, pH) and acclimation periods (Angelidaki & Ahring, 1994; Hashimoto, 1986). It has been reported that total ammonia nitrogen (TAN) concentrations causing 50% reduction in methane production range from 1.7 to 14 g/L (Angelidaki & Ahring, 1994; Chen et al., 2008). This would suggest that the batch digesters in this study were at least moderately inhibited by ammonia toxicity. Inhibition caused by high ammonia concentration would be enhanced by an increase in temperature, since anaerobic digestion is more easily inhibited and less stable at thermophilic temperatures than mesophilic temperatures (Braun et al., 1981; Parkin & Miller, 1982). This means that for 55°C, the inhibition observed for the MW and MW/H₂O₂ sets was likely due to ammonia, because the same extent of inhibition was not observed for the same sets at 35°C.

As mentioned before, chemical analysis of the substrates, before and after anaerobic digestion, revealed an increase in ammonia nitrogen after digestion, at both 35 and 55°C (Figure 13 and Figure 14). Analysis also revealed an accumulation of VFA after digestion. Accumulation of acetic, propionic, butyric, iso-butyric and iso-valeric acids was observed for MW/H⁺ and MW/H⁺/H₂O₂ sets at 35°C and all MW treatment sets at 55°C. (Figure 9 and Figure 10). Most of the VFA remaining were in the forms of acetic and propionic acids. It is suspected that, because of the low tolerance to ammoniac levels of digesters at higher temperatures, the VFA accumulation at 55°C would lead to a decrease in pH, thereby reducing ammonia concentration. The balances in pH, ammonia and VFA can lead to an “inhibited steady state”, where digesters are operating steady, but at lower methane production (Angelidaki & Ahring, 1993).

Acclimation was also a factor for ammonia inhibition in the MW and MW/H₂O₂ sets at 55°C. In the initial stages of the study, the inoculum that was used for the thermophilic digestion was adapted to untreated manure at room temperature. When this inoculum was digested at 55°C, it experienced temperature shock and was unable to digest the substrate for the MW and MW/H₂O₂ sets. The untreated manure did experience a longer lag time, but was largely unaffected due to the high amount of bacteria population in untreated dairy manure. However, when the MW and MW/H₂O₂ treated digesters were left alone for over 30 days, three out of the five replicates for the MW set began producing biogas, and the same happened for two out of the five replicates for the MW/H₂O₂ set (Figure 18). This indicates that the initial inoculum either did not survive the temperature change, or they were unable to adapt. Interestingly, for the inoculums that were able to adaption to 55°C, the average biogas generated from the surviving digesters produced more methane gas than its counterpart with temperature acclimated inoculum (Figure 16). This means

that the influence of the acclimation period on biogas generation is not only affected by temperature of acclimation, but also the substrate that is used for acclimation. Indeed, there are more toxic effects and increased concentrations of soluble chemicals when microwave treatment is applied.

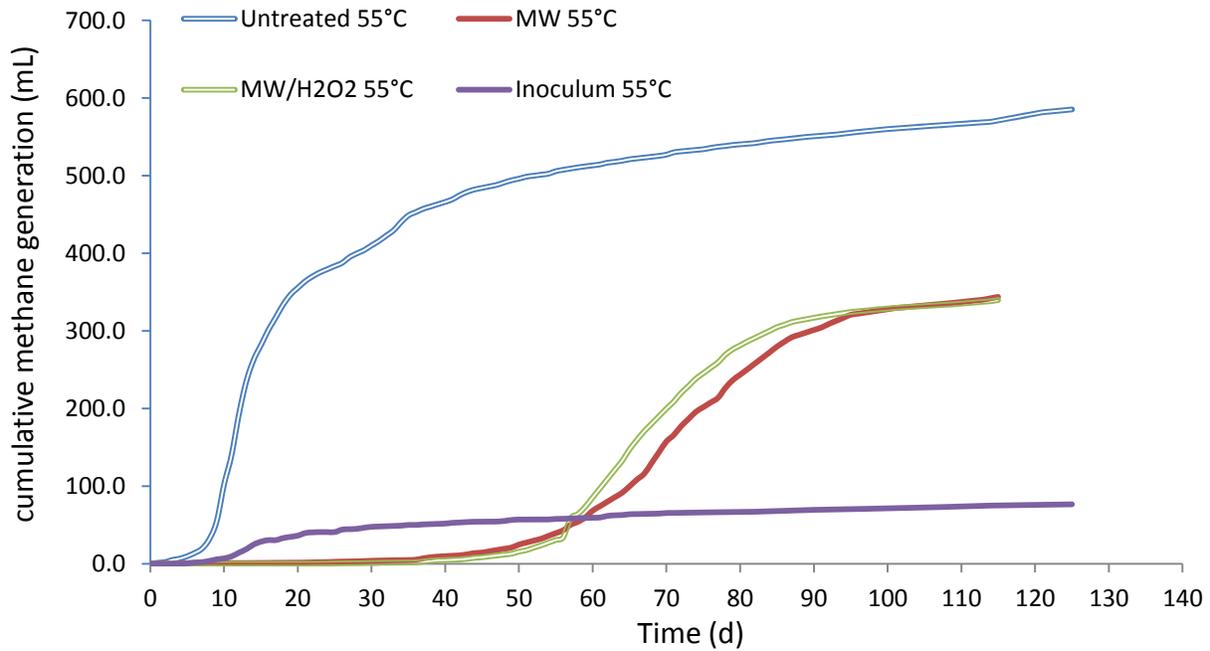


Figure 18. Methane production profile for untreated, MW and MW/H₂O₂ dairy manure at 55°C without temperature acclimation

Another factor influencing inhibition of the batch digesters is sulfate reduction. This is a possible cause for the failure of the MW/H⁺ and MW/H⁺/H₂O₂ sets at 35°C. Sulfate was introduced into the sets, either before microwave treatment or prior to anaerobic digestion, since sulfuric acid was used for acid hydrolysis treatment for MW/H⁺ and MW/H⁺/H₂O₂ and also for pH adjustment for MW and MW/H₂O₂. In anaerobic systems, sulfate is reduced to sulfide by sulfate reducing

bacteria (SRB). There are two ways sulfate reduction inhibits AD. The first and primary inhibition pathway is the competition for organic and inorganic substrate by SRB, thereby reducing methane generation (Harada et al., 1994). SRB may compete with methanogens, acetogens, or fermentative bacteria for free acetate, H_2 , propionate and butyrate in anaerobic reactors (Chen et al., 2008; Colleran et al., 1995). A secondary inhibition pathway is the toxicity of sulfide to various bacteria groups (Colleran et al., 1995). This means that an addition of sulfate into the anaerobic system facilitated the reduction of sulfate and competition of organics, which might otherwise have been used for generating methane. Also, when sulfate was reduced to sulfide in the batch digesters, the increased concentration of sulfide might have inhibited the reactors. Jin et al. (2009) found that the addition of sulfuric acid for acid hydrolysis pretreatment with microwave treatment lead to low methane production, compared to base hydrolysis using NaOH and CaO, and acid hydrolysis using HCl. The researchers correlated the low methane production to the presence of sulfate reducing to sulfide by sulfate reducing bacteria, which also competes for acetate from methanogens. Also, the low methane production could be attributed to the side reaction, such as Maillard reaction between amino acids and reducing sugars (Jin et al., 2009).

Light metal ions, such as Na, K, Mg, Ca and Al, are essential for growth of bacteria in anaerobic systems. However, in excess, these ions can slow down microbial growth and at high concentrations they can even cause server inhibition or toxicity (Chen et al., 2008). In this study, a substantial amount of Na, K, Mg and Ca were found in the substrate used for the batch studies. In particular, sodium was added to MW, MW/ H_2O_2 and MW/ H^+ , MW/ H^+ / H_2O_2 during pH adjustment, using sodium hydroxide (NaOH), and alkalinity adjustment, using sodium

bicarbonate (NaHCO_3), respectively. Indeed, analysis of MW/H^+ and $\text{MW}/\text{H}^+/\text{H}_2\text{O}_2$ treated substrate reported high concentrations of soluble Na^+ ions in the solutions before and after anaerobic digestion (35 and 55°C). Compared to untreated substrate, MW/H^+ and $\text{MW}/\text{H}^+/\text{H}_2\text{O}_2$ treated substrate had around seven times more soluble Na^+ , with average concentrations of 3721 and 4120 mg/L, respectively (Table 11 and Table 12). This is within the range of toxic concentration levels (3500 to 5500 mg/L) to moderately inhibit methanogens at mesophilic temperatures (McCarty, 1964). Also, acclimation of methanogens to high concentrations of sodium was not conducted. This would increase the likelihood of inhibition. Besides sodium, soluble calcium (Ca^{2+}) was also found in higher concentrations in MW, $\text{MW}/\text{H}_2\text{O}_2$, MW/H^+ and $\text{MW}/\text{H}^+/\text{H}_2\text{O}_2$ compared to untreated substrate (Table 11 and Table 12).

Possible organic degradation products from the MW treatment can be toxic to the anaerobic processes. Acid hydrolysis in the MW/H^+ and $\text{MW}/\text{H}^+/\text{H}_2\text{O}_2$ treated substrate can produce byproducts of fufural, hydroxymethyl fufural, formic acid, and levulinic acid, which are potential inhibitors of anaerobic digestion (Chen et al., 2008). For MW treatment of dairy manure, with or without hydrogen peroxide, phenol, aldehydes and ketones are produced, which are known to suppress anaerobic digestion (Chen et al., 2008; Quitain et al., 2002; Sierra-Alvarez & Lettinga, 1991). The extent of inhibition is dependent on concentration of toxicant, acclimation and temperature (Yang & Speece, 1986).

5.2.3 Kinetics for Anaerobic Degradation

In addition to the methane yield and maximum cumulative potential, the kinetics of the anaerobic digestion of untreated and treated dairy manure were also investigated. The modified Gompertz equation was used to examine the fitness of the BMP results to predict for kinetics of the biogas generation (Budiyono et al., 2010). The modified Gompertz equation is stated as following:

$$P = A \exp\{-\exp [Ue/A(\lambda-t) + 1]\}. \quad (1)$$

Where, P is cumulative specific methane production, mL/g VS;

A is methane production potential, mL;

U is maximum methane production rate, mL/g VS day;

λ is lag phase period (minimum time to produce methane), day; and

t is cumulative time for methane production, day.

The methane production kinetic is based on the assumption that the biogas production rate from the batch digesters correspond to the specific growth rate of methanogens in the bioreactor. The kinetic constants A, λ , and U can, therefore, be determined using non-linear regression by MS. Excel.

The projected methane productions from the models were fitted to the actual values in Figure 15, Figure 16 and Figure 17. The estimated values for the kinetic constants are summarized in Table 8. As indicated by the model, the modified Gompertz equation fitted well with the experimental

data from the BMP test results for mesophilic and thermophilic conditions, and the correlation coefficients are over 90%.

For all treatment conditions, the methane production rates (mL CH₄/g VS/day) were higher for 55°C than 35°C. This means that a higher digestion temperature improved the metabolism of the microorganisms in the bioreactors and provided higher methane production rates. Under digestion at 55°C, microwave treatment improves the methane production rate of dairy manure, since the maximum methane production rates of MW and MW/H₂O₂ are both higher, and the lag times are shorter, than untreated manure (Table 8). Under digestion at 35°C, the same increase in methane production rates was not evident. However, mesophilic digestion had higher methane yields (mL CH₄/g VS added) than thermophilic digestion. In cases where the methane yield was lower (MW/H⁺ and MW/H⁺/H₂O₂), it was attributed to inhibition due to changes in pH, ammonia and VFA concentration. Overall, thermophilic digestion provided a faster methane production rate compared to mesophilic digestion, but due to ammonia inhibition, it did not translate into higher methane yield. In general, mesophilic digestion provides higher quantity (methane yield) and quality (methane composition) of methane gas (Figure 21, Figure 22 and Figure 23). Gomez et al. (2011) reported a similar observation.

The methane potentials for this study are generated using non-linear regression using Equation 1 and they are presented in Table 8. Digestion of untreated manure at 35°C and 55°C provided the highest methane potentials of 493 ± 17 and 447 ± 24 mL, respectively. These values are comparable with literature values. Budiyo et al (2010) tested digestion of cattle manure at 38.5°C and reported an A, U and λ of 136.6 mL biogas/g VS, 5.68 mL biogas/ g VS/ d, and 9.07

days, respectively. For digestion of cow manure at 35°C, Adiga et al. (2012) reported an A, U, λ and methane yield of 245.1 mL biogas/g VS, 7.4 mL biogas/ g VS/ d, 10.7 days and 240 mL biogas/ g VS, respectively. The methane yield reported by Gomez et al. (2011) for digestion at 35°C and 55°C were 234 ± 19 and 159 ± 17 ml CH₄/g VS added, respectively; and lag times of 5.7 ± 0.1 and 1.4 ± 0.2 days, respectively. The results for methane potentials and methane yields show that microwave treatment of dairy manure can enhance the solubility of organic content (SCOD/TCOD), but it does not necessarily translate into enhanced anaerobic biodegradability. Further, there is no clear indication that H₂O₂ addition can significantly enhance methane yield. This might be related to the low H₂O₂ dosage used (0.6% vol/vol), however.

The lag times for the digestion of untreated and microwave treatment sets are presented in Table 8. The first week of incubation is the most important because that is the time when most of the substrate is used to produce biogas (Eskicioglu et al., 2007). When there is lag time for biogas generation, it is an indication that there is initial toxicity. In this study, the lag time for digestion under 35°C was more than 55°C. The lag time was shorter for treatment sets under 55°C because thermophilic digestion generates faster metabolism and inoculum was acclimated under the digestion temperature. Mesophilic inoculum was acclimated at room temperature, so the microorganisms needed additional time to adapt to the digestion temperature. Eskicioglu et al., (2007) acknowledged the importance of acclimation in order to reduce initial toxicity and provide higher ultimate biodegradability for digestion of waste activated sludge (WAS). In their study, they found that non acclimated inoculum displayed initial toxicity and much lower biogas production, compared to digestion with acclimated inoculum (Eskicioglu et al., 2007).

Table 8. Methane data for dairy manure

Set	Modified Gompertz Equation (model)				Experimental data		
	Methane production potential, A (mL CH ₄)	Maximum methane production rate, U (mL CH ₄ /g VS/day)	Lag time, λ (d)	Correlation coefficient	Methane production (mL CH ₄)	Methane yield per VS Added (mL CH ₄ /g VS)	Methane yield per VS destroyed (mL CH ₄ /g VS)
35°C							
Untreated	493 ± 17	10.9 ± 0.6	7.18 ± 0.13	0.994	531 ± 18	265 ± 13	584 ± 40
MW	270 ± 23	7.8 ± 0.9	7.19 ± 0.25	0.986	316 ± 25	213 ± 24	517 ± 70
MW/H ₂ O ₂	272 ± 14	8.4 ± 0.9	7.23 ± 0.09	0.985	323 ± 22	218 ± 27	521 ± 79
MW/H ⁺	110 ± 6	3.4 ± 0.2	5.86 ± 0.19	0.987	110 ± 5	52 ± 3	391 ± 24
MW/H ⁺ /H ₂ O ₂	141 ± 57	2.8 ± 0.9	6.52 ± 1.55	0.959	100 ± 17	51 ± 11	213 ± 51
55°C							
Untreated	447 ± 24	16.7 ± 1.1	1.51 ± 0.04	0.938	525 ± 26	199 ± 10	460 ± 24
MW	233 ± 14	20.2 ± 1.6	0.26 ± 0.03	0.983	246 ± 15	94 ± 7	292 ± 25
MW/H ₂ O ₂	229 ± 8	21.3 ± 1.4	0.07 ± 0.01	0.982	241 ± 9	95 ± 6	329 ± 20
MW/H ⁺	154 ± 6	19.2 ± 0.9	0.18 ± 0.02	0.979	185 ± 6	70 ± 3	227 ± 10
MW/H ⁺ /H ₂ O ₂	134 ± 6	16.4 ± 2.0	0.14 ± 0.03	0.980	166 ± 5	70 ± 10	247 ± 44

5.3 Anaerobic Digestion of Microwave Treated Sewage Sludge

The results for the AD of microwave treated dairy manure show that higher soluble organic content does not necessarily mean higher anaerobic biodegradability. This is different than results for AD of sewage sludge. BMP tests from literature reported up to 57% more biogas production for microwave irradiated sludge, compared to conventional mesophilic operation (Park et al., 2004). Also, Eskicioglu et al. (2006) and Park et al. (2004) both reported that MW based pretreatment is effective in enhancing solubility and anaerobic digestibility of sewage sludge. In fact, Eskicioglu et al. (2006) found that cumulative biogas production correlated well with the SCOD/TCOD results. Hence, AD of MW pre-treated sewage sludge (MW and MW/H₂O₂) was conducted to investigate the effect of MW/H₂O₂-AOP and subsequent anaerobic biodegradability on sewage sludge. The laboratory experiments and chemical analyses were conducted in the same manner as that for dairy manure, except a lower H₂O₂ dosage was used (0.1% vol/vol) (to prevent toxicity from excess H₂O₂) and the inoculum used for mesophilic digestion was digested at 35°C, instead of room temperature (20°C).

5.3.1 BMP Tests

The complete chemical characteristics of the substrates before and after anaerobic digestion are presented in - Raw Data

Table 13 of Appendix A. The cumulative methane gas production results are presented in Figure 19 and Figure 20. Overall, microwave had a positive impact on anaerobic digestibility of sludge. At 35°C, MW alone generated the highest methane yields of 125± 3 mL CH₄, or 238± 7 mL/ g VS added, followed by MW/H₂O₂ and then control (untreated). Microwave treatment had a greater impact on mesophilic digestion than thermophilic digestion. At 55°C, the methane yields

were similar for control, MW and MW/H₂O₂ (Table 9). Regardless of digestion temperature, all three sets (control, MW and MW/H₂O₂) generated methane gas profiles that were not significantly different ($t > 0.05$) from each other. However, this can be attributed to the low TS content in the sewage sludge used. If the solids in the sewage sludge were allowed to settle to provide a higher TS content, it is predicted that microwave alone will provide the highest cumulative methane gas generation, compared to control and MW/H₂O₂. The addition of H₂O₂ does not appear to help to improve methane production in BMP tests. Eskicioglu et al., (2008) reported that, although H₂O₂ addition to microwave treatment increases the SCOD content of sewage sludge, MW/H₂O₂ treated samples had lower first-order mesophilic biodegradation rate constants and ultimate methane yields, compared to the control and MW irradiated samples. This means that the soluble organics generated were slower to biodegrade or more refractory than those generated during MW irradiation (Eskicioglu et al., 2008).

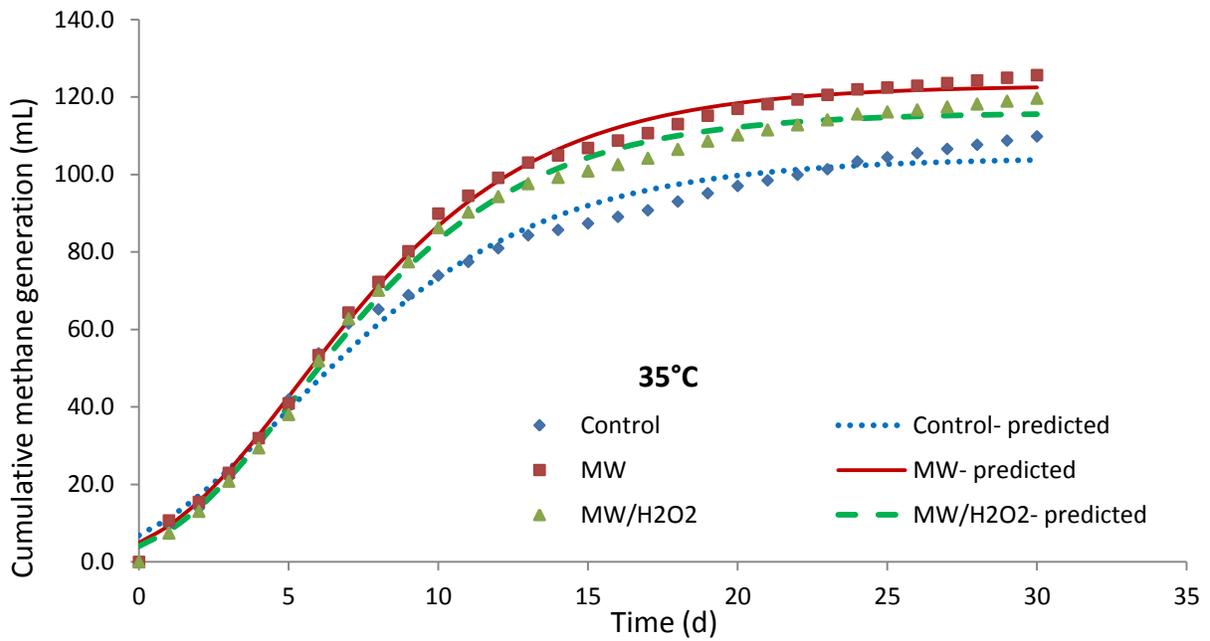


Figure 19. Methane production profile for untreated and MW sewage sludge at 35°C

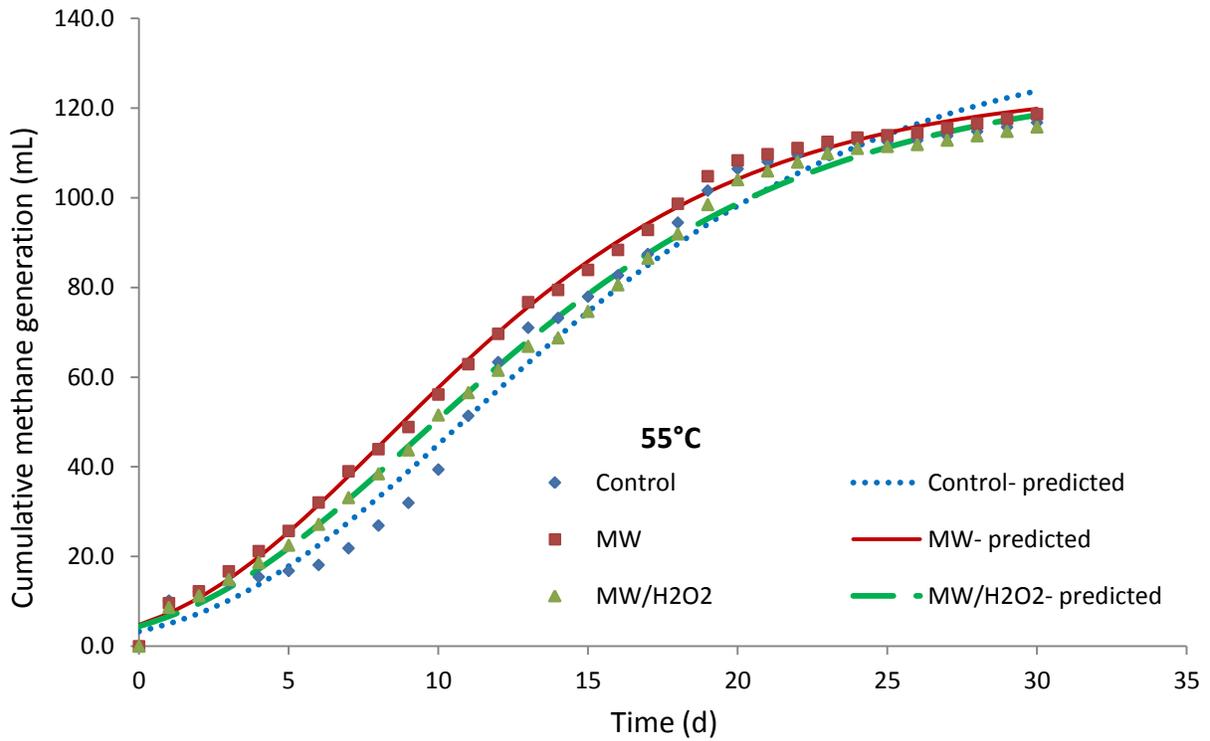


Figure 20. Methane production profile for untreated and MW sewage sludge at 55°C

5.3.2 Kinetics for Anaerobic Degradation

The kinetic data are analyzed using the Modified Gompertz Equation and the results are presented in Table 9. Comparison between digestion temperatures of 35 and 55°C reveals that mesophilic digestion is superior to thermophilic digestion, in terms of methane yield and methane production rate. The methane production rates for 35 and 55°C were 19.1 ± 1.2 and 8.4 ± 0.6 mL CH₄/ VS added/ day. Also, mesophilic digestion had shorter lag times and higher initial methane composition than thermophilic digestion (Figure 24, Appendix B).

These results contradict the findings from anaerobic digestion of dairy manure. One would assume that higher digestion temperature would lead to faster methane production rates. This could be due to lack of acclimation of inoculum to the presence of the MW pretreated substrate under thermophilic condition, leading to mild short term microbial inhibition; this can be more severe for thermophilic digestion (Braun et al., 1981; Parkin & Miller, 1982). However, the exact reason for this is unknown. For 35 °C, control, MW and MW/H₂O₂ have methane yields of 591 ± 16 , 609 ± 18 , and 620 ± 19 mL CH₄/ g VS removed, or 366 ± 19 , 312 ± 17 , and 362 ± 30 mL CH₄/ g COD removed, respectively. These values are comparable with literature values, which suggested methane yields of 0.32 to 0.35 L/ g COD removed at 1 atm, and 25°C, based on methane composition of 70-75% after conventional heating and microwave treatment (Eskicioglu et al., 2006). Eskicioglu et al., (2006) suggested a theoretical methane production of 0.35 L/g COD removed, which matches well with the values from this study and gives confidence to the results.

Overall, microwave treatment (MW and MW/H₂O₂) improved methane production for sewage sludge, although the same effects were not evident for the experiment with dairy manure. Methane yields for digestion of sewage sludge were comparable to that for dairy manure, but the methane production potentials of sewage sludge were lower than that of dairy manure, which can be attributed to the low content of TS in the sewage sludge used. Comparison of the results from the anaerobic digestion study of dairy manure and sewage sludge suggests that the benefits of microwave (MW and MW/H₂O₂) pretreatment on anaerobic digestibility are substrate specific. This is likely the case because dairy manure is much more complex than sewage sludge. Sewage sludge is mostly comprised of microbial cells and extracellular polymeric substances with some metals and nutrients content, whereas dairy manure has a higher content of fiber and more nutrients and metals concentration than sewage sludge. The higher percentage of recalcitrant materials, nutrients and metal contents in dairy manure are sources of possible digestion inhibition and failure.

Table 9. Methane data for sewage sludge

Set	Modified Gompertz Equation (model)				Experimental data		
	Methane production potential, A (mL CH ₄)	Maximum methane production rate, U (mL CH ₄ /g VS/day)	Lag time, λ (d)	Correlation coefficient	Methane production (mL CH ₄)	Methane yield per VS Added (mL CH ₄ /g VS)	Methane yield per VS destroyed (mL CH ₄ /g VS)
35°C							
Control	104 ± 3	15.3 ± 0.4	0 ± 0	0.993	110 ± 3	213 ± 5	591 ± 16
MW	123 ± 3	19.1 ± 1.2	0.76 ± 0.13	0.999	126 ± 3	238 ± 7	609 ± 18
MW/H ₂ O ₂	116 ± 3	19.6 ± 0.4	0.92 ± 0.08	0.998	120 ± 3	238 ± 6	620 ± 19
55°C							
Control	137 ± 11	8.1 ± 1.2	2.60 ± 0.62	0.992	117 ± 10	155 ± 14	555 ± 55
MW	125 ± 14	8.4 ± 0.6	1.29 ± 0.27	0.999	119 ± 12	149 ± 15	373 ± 39
MW/H ₂ O ₂	127 ± 4	8.3 ± 0.8	1.71 ± 0.19	0.998	116 ± 4	156 ± 6	407 ± 34

5.4 Mass Balance of Metals (Ca, K, Mg, Na) using MW/H₂O₂-AOP and BMP Tests

Analysis of the metals content of dairy manure using MW/H₂O₂-AOP is presented in Table 10. It is assumed that total metals content does not change, unless chemicals are added to the samples through pH adjustment, using sodium hydroxide (NaOH) or sulfuric acid (H₂SO₄) or alkalinity addition using sodium bicarbonate (NaHCO₃). In cases where soluble content is more than total, it is attributed to variability in the samples. Dilution was conducted by adding water into the sample tube, up to a designated line. The dilution factors of samples have an average of 20 times dilution. The sampling time can also be a factor in variability of results. The time between analyses of the same sample can be as much as 14 days and can provide variability in the analytical results.

The results reveal that acid treatment helped to solubilise calcium and magnesium in dairy manure. The addition of microwave treatment with acid enhanced the release of both metals into solution. After MW/H⁺ and MW/H₂O₂/H⁺, all calcium and magnesium were in soluble form. This agrees somewhat with the findings by Qureshi et al. (2008), who found that the MW/H⁺ and MW/H₂O₂/H⁺ helped to partially solubilise calcium, but not magnesium. This could be due to the higher sulfuric acid dosage used in this study, which would help to solubilise metals. Their study also reported an increase in potassium by the microwave treatment process. However, no changes to soluble potassium and sodium content by microwave treatment were observed. This is because potassium and sodium already existed in soluble form in the dairy manure analyzed. In general, it is agreed that “no clear and consistent increase or decrease in metal concentrations was reported after microwave digestion” (Qureshi et al., 2008; Wong et al., 2006). For microwave treatment of sewage sludge, Wong et al. (2006) found that the MW/H₂O₂-AOP

process helped to solubilise calcium, magnesium and potassium. However, no clear trend was determined in this study.

Table 10. Metals analysis after MW treatment

	Untreated	MW	MW/H ₂ O ₂	H ⁺	MW/H ⁺	MW/H ⁺ /H ₂ O ₂
Total Ca (mg/L)	1120± 7	905± 143	925± 34	1067± 32	1167± 19	876± 109
Total K (mg/L)	2102± 9	2101± 5	2098± 12	2065± 32	2204± 12	2031± 39
Total Mg (mg/L)	406± 5	329± 44	336± 13	395± 5	425± 4	387± 3
Total Na (mg/L)	417± 8	411± 4	405± 1	395± 6	415± 12	392± 8
Soluble Ca (mg/L)	2± 4	40± 12	50± 2	758± 98	1386± 69	1162± 90
Soluble K (mg/L)	2428± 56	2405± 87	2285± 37	2395± 221	2523± 102	2387± 32
Soluble Mg (mg/L)	237± 13	210± 10	198± 5	460± 20	509± 11	463± 12
Soluble Na (mg/L)	548± 27	539± 18	499± 5	492± 68	438± 36	422± 42

The results for metals analysis before and after digestion at 35°C are presented in Table 11. The soluble content of all the metals decreased after digestion. Soluble calcium decreased the most, while soluble K, Mg and Na decreased to a lesser extent. Alkalinity addition and pH adjustment added substantial amounts of sodium into the acid treated sets (MW/H⁺ and MW/H₂O₂/H⁺), which increased total sodium content from 415± 12 and 392± 8 mg/L to 3097± 83 and 3140± 291 mg/L, respectively. Total calcium also increased after alkalinity addition and pH adjustment, but the exact reason for the increase is unknown (but it is assume to be due to variability in samples). No other sources of calcium were added to the substrate before digestion. Total potassium and magnesium concentrations increased slightly, but this is attributed to variability in the samples—no other sources of K and Mg were added.

The results reveal that digestion at 35°C significantly decreased soluble calcium and also decreased soluble potassium, magnesium and sodium. The decrease in soluble content of metals suggests that they were utilized during the digestion process, likely by microbial biomass.

Calcium is useful for granulation and has influence on enzyme activity and aggregation of microbial biomass (Yu et al., 2001). An optimal Ca^{2+} concentration of 200 mg/L was suggested for methane generation using acetic acid (Kugelman & McCarty, 1965). Like calcium, magnesium is also important for granulation of sludge in anaerobic digesters (Schmidt & Ahring, 1993). For potassium, concentrations of less than 400 mg/L were found to enhance digesters in both the thermophilic and mesophilic ranges (Chen et al., 2008), whereas low concentrations of sodium are essential for methanogens and mesophilic anaerobes (Kugelman & Chin, 1971; McCarty, 1964).

Table 11. Metals analysis before and after digestion at 35°C

Before	Untreated	MW	MW/H ₂ O ₂	MW/H ⁺	MW/H ⁺ /H ₂ O ₂
Total Ca (mg/L)	1252± 317	889± 135	873± 165	1645± 45	1496± 159
Total K (mg/L)	2439± 482	2907± 91	2899± 120	3034± 68	2625± 287
Total Mg (mg/L)	457± 110	427± 35	414± 38	582± 12	525± 55
Total Na (mg/L)	428± 92	467± 18	458± 24	3097± 83	3140± 291
Soluble Ca (mg/L)	97± 44	191± 9	203± 35	1062± 24	836± 62
Soluble K (mg/L)	2386± 40	2593± 76	2637± 117	3647± 80	3323± 382
Soluble Mg (mg/L)	235± 20	224± 11	245± 13	530± 9	458± 62
Soluble Na (mg/L)	545± 170	558± 52	545± 35	3721± 83	4120± 399
After					
Soluble Ca (mg/L)	9± 4	22± 4	27± 4	3± 6	18± 6
Soluble K (mg/L)	1995± 36	1979± 44	1924± 26	2007± 31	1785± 180
Soluble Mg (mg/L)	175± 4	155± 12	156± 13	288± 5	242± 26
Soluble Na (mg/L)	407± 6	404± 5	392± 3	2857± 68	3008± 256

The results for metals analysis, before and after digestion at 55°C, are presented in Table 12. After digestion at 55°C, soluble calcium metals decreased significantly, potassium and sodium decreased slightly and magnesium did not decrease. It appears that there are more soluble metal concentrations after digestion, at higher digestion temperature. This would make sense because solubility increases as temperature increase. The higher concentrations of soluble metals could also contribute to toxicity and inhibition of the digesters. Indeed, thermophilic digesters are more

prone to turn sour than mesophilic digesters, as evident in the previous study on anaerobic digestibility of microwave treated dairy manure.

Similar to mesophilic digestion, digestion at 55°C reduced the soluble content of metals in substrate. This is an indication that soluble metals were consumed by bacteria or precipitated out of solution during the digestion process. For example, different phosphate forms can be produced with soluble calcium and magnesium, including $\text{Ca}_3(\text{PO}_4)_2$, CaHPO_4 , $\text{MgNH}_4\text{PO}_4 \cdot 6\text{H}_2\text{O}$ and $\text{Mg}_3(\text{PO}_4)_2 \cdot 8\text{H}_2\text{O}$ (Carliell & Wheatley, 1997). Depending on the ratios of chemicals and environmental conditions, such as pH and temperature, different phosphate compounds can be formed. Another possible explanation is the formation of organometallic compounds, which could be formed when soluble metals bind to organic substances (Wong et al., 2006).

Table 12. Metals analysis before and after digestion at 55°C

Before	Untreated	MW	MW/H ₂ O ₂	MW/H ⁺	MW/H ⁺ /H ₂ O ₂
Soluble Ca (mg/L)	52± 6	107± 13	241± 8	693± 44	492± 68
Soluble K (mg/L)	2323± 36	2375± 53	2388± 66	2380± 72	2120± 218
Soluble Mg (mg/L)	196± 3	198± 5	208± 5	322± 20	286± 28
Soluble Na (mg/L)	504± 23	503± 12	507± 18	3241± 127	3582± 288
After					
Soluble Ca (mg/L)	9± 5	43± 11	47± 19	83± 6	33± 14
Soluble K (mg/L)	2131± 29	2191± 64	2153± 69	2154± 29	1920± 170
Soluble Mg (mg/L)	203± 16	226± 10	226± 8	312± 4	265± 22
Soluble Na (mg/L)	392± 6	400± 15	393± 12	2587± 49	3304± 244

Chapter 6: Conclusions and Discussion

6.1 MW Pretreatment of Liquid Dairy Manure

The results from this study indicate that MW/H₂O₂-AOP was effective in solubilizing nutrients and organics in liquid dairy manure. In particular, all microwave treatment sets increased soluble COD concentration. Acid alone decreased soluble COD concentrations, but acid addition was necessary for releasing orthophosphate and ammonia into solution. Without acid addition, there was no release of nutrients from dairy manure. Hydrogen peroxide at the dosage applied did not provide any significant effect on nutrient release or organic solubilisation. Missing from this study is the investigation into the factors for optimal nutrient and organic solubilisation. These factors could provide insight into the parameters and conditions under which organic solubilisation and nutrient release is optimized.

All microwave treatment sets resulted in similar SCOD/TCOD ratios. Microwave with hydrogen peroxide treatment released up to 45± 2% of TCOD as SCOD, or a 12± 3% increase with respect to untreated dairy manure. Up to 70± 2% of TP was released as orthophosphate and up to 67± 4% of the TKN as ammonia for MW/H⁺ treatment. Ammonia and orthophosphate released occurred simultaneously upon treatment. The significant increase in nutrients can be useful for subsequent nutrient recovery process, such as struvite crystallization.

6.2 Anaerobic Digestion of MW Pretreated Dairy Manure

The results show that microwave treatment of dairy manure can enhance the solubility of organics, but it does not necessarily translate into enhanced anaerobic biodegradability, including methane yield and production rates. Digestion of untreated dairy manure had the highest methane

yields of 265 ± 13 and 199 ± 10 mL CH₄/g VS added at 35 and 55°C, respectively. Low methane yields observed for the digestion of acidified dairy manure was attributed to sulfide inhibition from the addition of sulfuric acid for acid hydrolysis. For microwave treated dairy manure with no acid addition, severe inhibition was observed at 55°C. Ammonia levels were suspected as the main reason for inhibition, but a number of other factors could also be responsible, including sulfide, light metals and lack of temperature acclimation for inoculum. The exact reasons for the inhibition of microwave treated dairy manure were not investigated, but different types of acid and bases can be used to compare their effects on anaerobic digestion of dairy manure.

Digestion of untreated dairy manure had the highest TCOD removal, with up to 34 and 43% of TCOD removed at 35 and 55°C, respectively. Ammonia concentrations increased for all treatment sets and orthophosphate concentrations also increased for microwave treated dairy manure. The reason for the decrease of orthophosphate for acidified treatment sets is currently unknown. For all treatment conditions, thermophilic digestion of dairy manure produced higher, actual, methane production rates than at mesophilic temperature. However, thermophilic digestion was more susceptible to inhibition and hence, mesophilic digestion had higher methane yields. The highest methane potential was for untreated manure, which provided potentials of 493 ± 17 and 447 ± 24 mL for digestion at 35 and 55°C, respectively. Acclimation appears to play a role in lag time, initial toxicity and methane production.

6.3 Anaerobic Digestion of MW Pretreated Sewage Sludge

The results showed that microwave treatment had a positive impact on anaerobic digestibility of sewage sludge, especially for mesophilic digestion. At 35°C, MW alone generated the highest

methane yields of 125 ± 3 mL CH₄ or 238 ± 7 mL/ g VS added, followed by MW/H₂O₂ and then control. At 55°C, the methane yields were similar for all three sets. The addition of H₂O₂ did not improve the methane production of BMP tests. In terms of methane kinetics, microwave treatment improved the methane production rate for mesophilic digestion, but not thermophilic digestion. The methane production rates for the digestion of MW alone at 35 and 55°C were 19.1 ± 1.2 and 8.4 ± 0.6 mL CH₄/ VS added/ day, respectively. The low methane production rate for thermophilic digestion was attributed to the lack of inoculum acclimation to microwave digested sludge. The positive results for BMP tests of microwave treated sewage sludge suggest that the influence of microwave treatment on anaerobic biodegradability is both substrate and concentration (% solids) specific.

6.4 Mass Balance of Metal (Ca, K, Mg, Na) using the MW/H₂O₂-AOP and BMP Tests

There was no clear and consistent increase or decrease in soluble metal (Ca, K, Mg, Na) concentrations after MW/H₂O₂-AOP. However, it was found that acidified treatment was effective to release calcium and magnesium into solution. The additional effect of microwave treatment enhanced the effect of acid such that all calcium and magnesium were in solution. After alkalinity and pH adjustment using sodium bicarbonate and sodium hydroxide, there were significant increases to total sodium and calcium. The exact reason for the increase in calcium is unknown. For both BMP tests under 35 and 55°C, a significant amount of soluble calcium was removed from solution. Digestion also reduced soluble potassium, magnesium and sodium, although not as much as soluble calcium. Also, the reductions were higher for digestion under 35°C, than for 55°C.

Chapter 7: Recommendations

The results of this study suggest that the MW/H₂O₂-AOP is an effective process for solubilising organics in dairy manure, but it has not proven to be effective for biogas generation, due to probable toxicity and inhibition from high ammonia, sulfur and metal concentrations. It is recommended that a different type of acid or alkaline chemical be used instead of sulfuric acid. Literature shows that chemicals such as hydrochloric acid, calcium oxide and sodium hydroxide can be combined with MW to increase methane yield of dairy manure (Jin et al., 2009). Further, a multivariable analysis can be conducted using different H₂O₂ and chemical dosages, microwave temperatures, and microwave digestion times, to see their combined effects on nutrient and organic solubilisation and anaerobic digestibility. This would isolate the condition to the highest methane yield. It is also possible that a lower microwave (915 MHz) frequency be used, instead of 2450 MHz, since a lower frequency can penetrate deeper into dairy manure. This is now being considered for future microwave research, as part of prototype development.

Another important aspect of BMP tests is the use of inoculum. Acclimatization of inoculum played a role in the biogas production and lag time of BMP tests, especially under thermophilic temperatures. Methanogens and other bacteria in the anaerobic digestion process are sensitive to changes in their environment. Therefore, it is recommended that the inoculum used be cultured under the harshest condition. This means that the inoculum should be fed with substrate pretreated with the highest microwave temperature and with the most chemicals added, such as the MW/H⁺/H₂O₂ treatment condition. Also to consider is the H₂O₂ dosage, since it is toxic to bacterial. Hence, a low H₂O₂ dosage should be used, such as 0.3% vol/vol or less.

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APPENDICES

Appendix A - Raw Data

Table 13. Chemical characteristics of (sewage sludge) substrates before and after digestion

Before	35°C			55°C		
	Control	MW	MW/H ₂ O ₂	Control	MW	MW/H ₂ O ₂
TS (%)	0.64 ± 0.00	0.66 ± 0.00	0.62 ± 0.01	0.63 ± 0.00	0.65 ± 0.00	0.62 ± 0.01
VS (%)	0.52 ± 0.00	0.53 ± 0.00	0.50 ± 0.00	0.51 ± 0.00	0.52 ± 0.00	0.50 ± 0.00
TCOD (g/L)	7.87 ± 0.22	8.28 ± 0.18	7.73 ± 0.54	7.56 ± 0.18	7.97 ± 0.12	7.42 ± 0.52
SCOD (g/L)	0.18 ± 0.01	0.96 ± 0.04	0.95 ± 0.03	0.26 ± 0.01	1.04 ± 0.04	1.03 ± 0.03
VFA (mg/L)	9 ± 1	80 ± 2	28 ± 11	37 ± 1	108 ± 2	56 ± 11
TP-total (mg/L)	221 ± 17	233 ± 4	223 ± 6	221 ± 17	233 ± 4	223 ± 6
TP-soluble (mg/L)	105 ± 6	147 ± 2	140 ± 2	105 ± 6	147 ± 1	140 ± 2
PO ₄ -P (mg/L)	122 ± 2	113 ± 2	78 ± 2	120 ± 0	110 ± 1	76 ± 1
TKN- total (mg/L)	655 ± 51	692 ± 21	666 ± 20	613 ± 50	649 ± 17	623 ± 16
TKN- soluble (mg/L)	50 ± 5	178 ± 7	154 ± 5	59 ± 4	186 ± 6	163 ± 4
NH ₄ -N (mg/L)	39 ± 2	64 ± 2	38 ± 32	44 ± 1	69 ± 1	43 ± 1
pH	6.4	6.5	6.3	6.4	6.4	6.5
After	35°C			55°C		
TS (%)	0.49 ± 0.00	0.48 ± 0.00	0.47 ± 0.01	0.47 ± 0.00	0.47 ± 0.00	0.44 ± 0.01
VS (%)	0.35 ± 0.00	0.34 ± 0.00	0.33 ± 0.01	0.33 ± 0.00	0.32 ± 0.00	0.30 ± 0.00
TCOD (g/L)	5.14 ± 0.19	4.62 ± 0.19	4.72 ± 0.18	5.78 ± 0.23	5.27 ± 0.20	5.00 ± 0.15
SCOD (g/L)	0.11 ± 0.00	0.27 ± 0.01	0.30 ± 0.01	0.70 ± 0.04	0.68 ± 0.04	0.74 ± 0.02
VFA (mg/L)	2 ± 0	7 ± 0	9 ± 1	27 ± 5	27 ± 5	30 ± 2
TP-total (mg/L)	239 ± 3	240 ± 3	221 ± 3	237 ± 22	217 ± 10	211 ± 11
TP-soluble (mg/L)	210 ± 15	192 ± 14	199 ± 8	186 ± 4	177 ± 6	167 ± 10
PO ₄ (mg/L)	201 ± 2	206 ± 2	199 ± 3	185 ± 10	173 ± 13	169 ± 15
TKN- total (mg/L)	345 ± 26	342 ± 33	259 ± 13	586 ± 63	544 ± 32	509 ± 40
TKN- soluble (mg/L)	345 ± 26	342 ± 33	259 ± 13	288 ± 16	356 ± 20	338 ± 25
NH ₄ (mg/L)	269 ± 4	369 ± 2	350 ± 6	287 ± 23	323 ± 4	305 ± 2
pH	6.8	6.8	6.9	6.9	7.0	6.9

Appendix B - Methane Composition

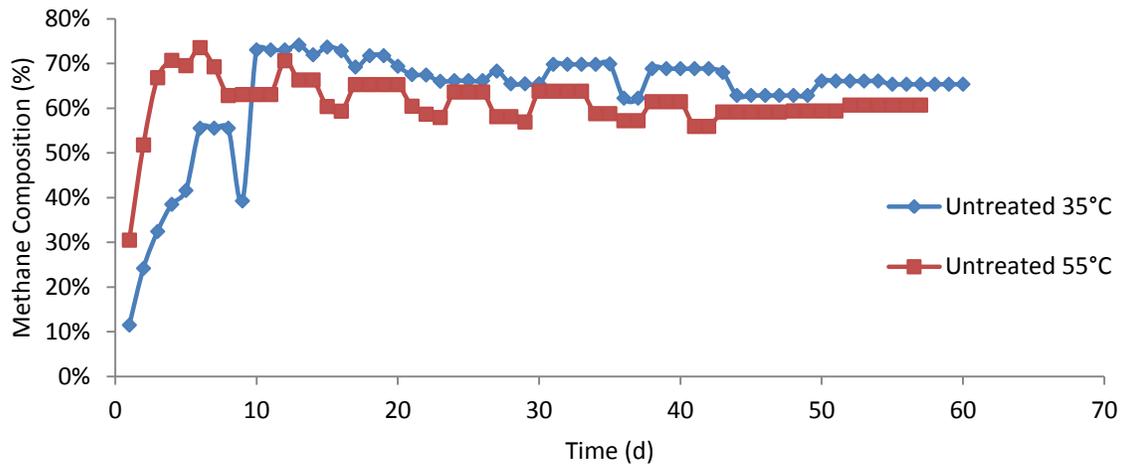


Figure 21. Methane composition profile of untreated dairy manure

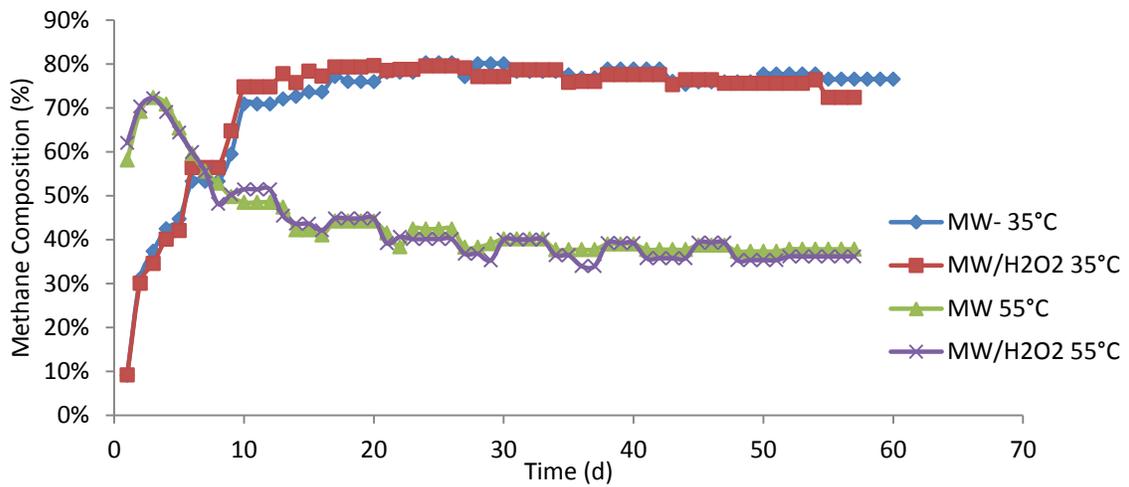


Figure 22. Methane composition profile of MW & MW/H₂O₂ dairy manure

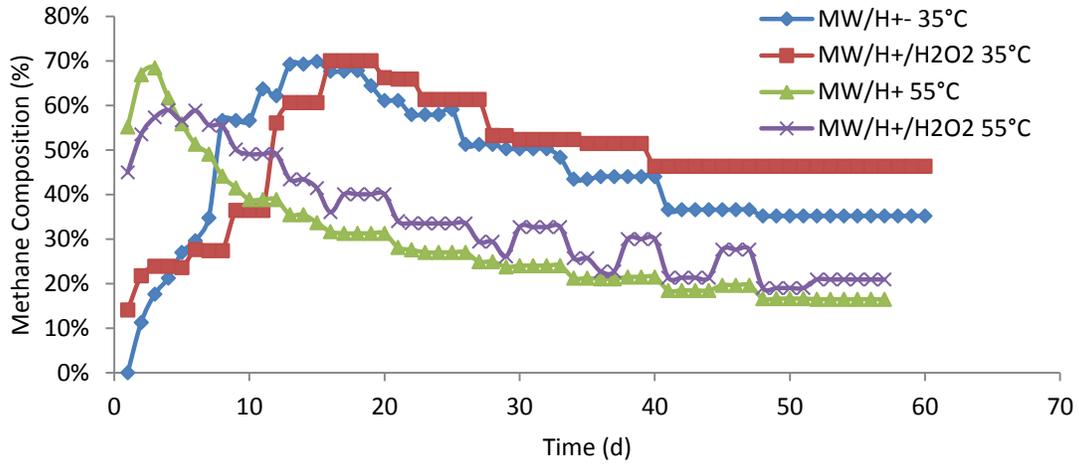


Figure 23. Methane composition profile of MW/H⁺ & MW/H+/H₂O₂ dairy manure

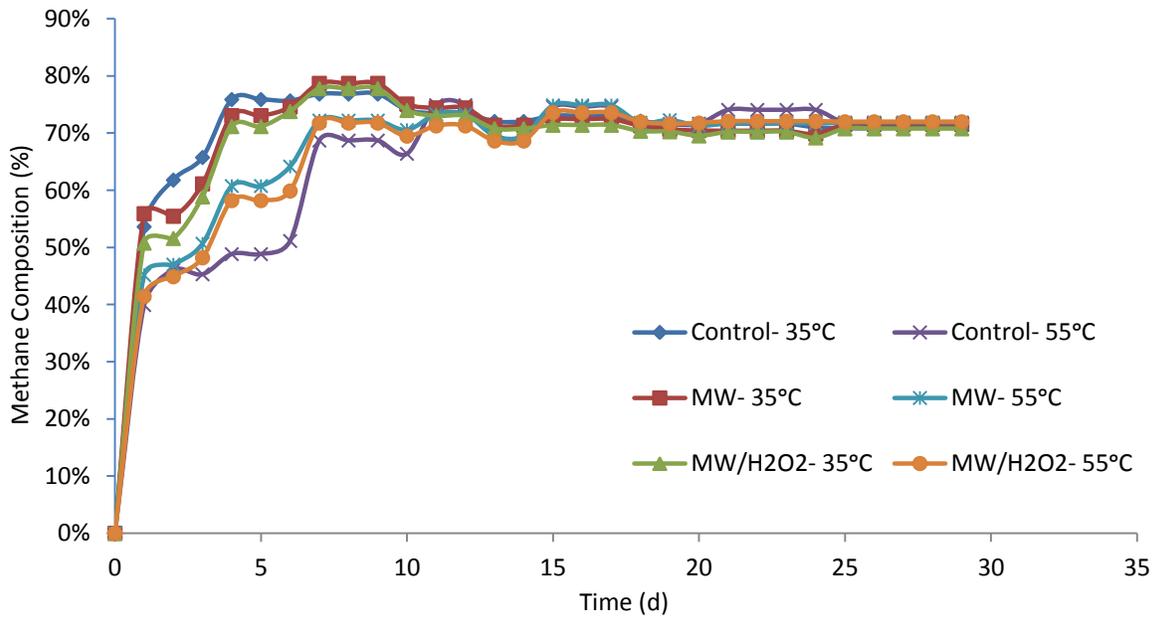


Figure 24. Methane composition profile untreated, MW and MW/H₂O₂ of sewage sludge

Appendix C - Photos



Figure 25. MW generator

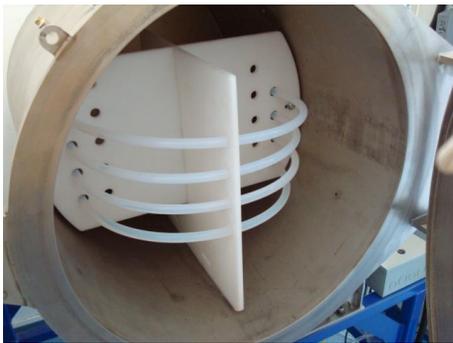


Figure 26. MW chamber



Figure 27. MW influent and effluent tanks



Figure 28. COD spectrophotometer

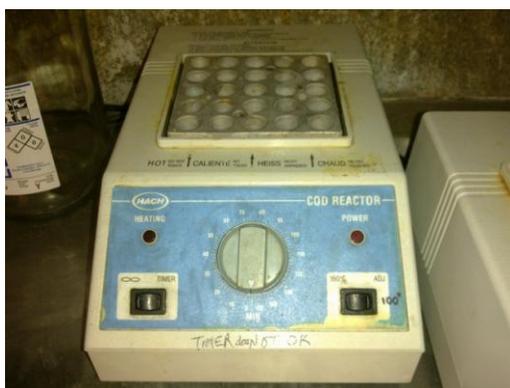


Figure 29. COD digester



Figure 30. Flow injection colorimeter



Figure 31. TP and TKN digester



Figure 32. VFA gas chromatography



Figure 33. ICP



Figure 34. Metals digester



Figure 35. Mesophilic incubator



Figure 36. Thermophilic water bath and batch digesters