

**Increased Energy Recovery and Enhanced Pathogen Inactivation  
Through Anaerobic Digestion of Thickened Wastewater Sludge**

By  
Bartłomiej Maciej Puchajda

A Thesis submitted to the Faculty of Graduate Studies of  
The University of Manitoba  
in partial fulfilment of the requirements of the degree of

DOCTOR OF PHILOSOPHY

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## Abstract

This study had two objectives: to investigate the possibility of increased energy recovery from anaerobic digestion of sewage sludge and to inactivate pathogens in wastewater sludge by means other than thermophilic temperature. All research was done through numerous lab experiments using wastewater sludge from the North End Water Pollution Control Centre in Winnipeg.

It was proposed that toxicity from un-ionized form of VFA could be used in acid mesophilic (38°C) and low-mesophilic (24°C) conditions. The toxicity from the un-ionized form of VFA is based on cytoplasm acidification from a proton released after acid ionization inside the bacterial cell and anion accumulation inside the cell which may lead to osmotic stress. Batch acid digesters were able to achieve Class A biosolids standards with respect to fecal coliforms concentrations under both operating conditions. When acid digesters were operated in semi-continuous mode, three acid digesters operated in series were needed to achieve Class A level. This was possible in mesophilic conditions only. Effluent from the three mesophilic acid digesters operated in series also showed highest inactivation of *Ascaris suum* ova from all systems tested, achieving complete inactivation of eggs enclosed in sentinels and 2.8% viability of free floating eggs.

Sludge thickening, which results in sludge flow reduction, was chosen as a method to achieve enhanced energy recovery. To establish optimum solids content in sludge, a series of hydrolysis tests were performed. The tests were to assess the impact of environmental conditions, such as operating temperature and sludge thickness on digestion. Optimum sludge solids content was defined as solids content that does not

inhibit hydrolysis rates at any given temperature. This should result in uninhibited VS destruction and biogas production. Increase in solids content in raw sludge (from initial concentration of 3.6% TS) that would be accompanied by proportional increase in hydrolysis rate was found to be approximately 50% or up to 5.4% TS at 24°C, and 70% or up to 6.1% TS at 38°C.

Sludge volume reduction after thickening could be used for extension of SRT in existing digestion system for additional VS destruction and biogas production or operational and construction savings at smaller digestion volumes. Both scenarios were investigated and it was concluded that additional digestion time would be the most cost effective solution where infrastructure is available. Extension of SRT depends mainly on the degree of sludge thickening. For instance, by thickening the sludge from 3.9% TS (long term average at NEWPCC) at 22 days SRT, it is possible to achieve at 5.7% TS 32.2 days SRT. Increasing the solids content by 0.9 %TS (from 4.8% TS to 5.7% TS – laboratory experiment) or 1.7% TS (from 3.9% TS to 5.7% TS – simulated data) would increase digestion SRT by an additional 4 to 10 days, and it could potentially increase methane production by 7% to 15%, respectively.

A new proposed digestion system that would enable enhanced pathogen inactivation and increased energy recovery consisted of three mesophilic acid digesters followed by a mesophilic gas digester. Depending on the initial solids content in raw, un-thickened sludge, the new digestion system could provide an additional 13% energy for recovery (including costs of gravity thickening) over conventional mesophilic anaerobic digestion (currently employed in Winnipeg). Also, the quality of biogas from this multi-phase



digestion system showed up to 72% methane content in biogas, which was statistically higher than from a conventional digestion system.

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## Abbreviations and Symbols

1TPAD – 1<sup>st</sup> digester (thermophilic) of two-phase digestion system

2TPAD – 2<sup>nd</sup> digester (mesophilic) of two-phase digestion system

AD – anaerobic digestion

AHAB – anaerobic hydrolytic-acidogenic bacteria

C - concentration

COD – chemical oxygen demand

COD<sub>inf</sub> – influent chemical oxygen demand, mgCOD/l

COD<sub>eff</sub> – effluent chemical oxygen demand, mgCOD/l

D – diffusion coefficient

F – concentration of degradable particulate COD, mgCOD/l

F<sub>o</sub> – initial concentration of degradable particulate COD, mgCOD/l

G – loss of COD attributed to biogas production, mgCOD/l

J - flux

k<sub>d</sub> – decay coefficient, d<sup>-1</sup>

K<sub>H</sub> – Henry's constant

k<sub>h</sub> - first-order hydrolysis rate constant, d<sup>-1</sup>

K<sub>s</sub> – saturation constant, mg/l

MAD – mesophilic anaerobic digestion

MPN – most probable number

μ – net specific growth rate, d<sup>-1</sup>

μ<sub>max</sub> – maximum specific growth rate, d<sup>-1</sup>

P – acid phase products, mgCOD/l



$P_p$  – partial pressure

$PCOD_{eff}$  – particulate COD in effluent, mg COD/l

$PCOD_{inf}$  – particulate COD in influent, mg COD/l

PS – Primary Sludge

$Q$  – flow, l/d or  $m^3/d$

$P_{sludge}$  – specific gravity of sludge,  $kg/m^3$

SCOD – soluble chemical oxygen demand

SRT – solids retention time

STP – standard conditions,  $0^\circ C$  and 1 atmosphere pressure

$T$  – temperature

TAD – thermophilic anaerobic digestion

$T_{air}$  – air temperature,  $^\circ C$

TCOD – total chemical oxygen demand

$T_{digester}$  – temperature in digester,  $^\circ C$

$T_{earth}$  – earth temperature,  $^\circ C$

$T_{exchangerCold}$  – temperature on the cold side of the heat exchanger,  $^\circ C$

$T_{exchangerHot}$  – temperature on the hot side of the heat exchanger,  $^\circ C$

$T_{incoming\ sludge}$  – temperature of incoming sludge,  $^\circ C$

TPAD – temperature-phased anaerobic digestion

TS – total solids

U-VFA – un-ionized form of VFA

$V$  – volume

VAR – vector attraction reduction

$V_{CH_4}$  – volume of methane produced,  $m^3 CH_4/d$

VFA – volatile fatty acids

$VFA_{eff}$  – concentration of volatile fatty acids in effluent, mg COD/l

$VFA_{inf}$  – concentration of volatile fatty acids in influent, mg COD/l

VS – volatile solids

$VS_{in}$  – influent mass of volatile solids, kg VS/d

$VS_{out}$  – effluent mass of volatile solids, kg VS/d

VSR – volatile solids reduction, %

WAS – Waste Activated Sludge

$X_{ACID}$  – concentration of acidogenic population in digester, mgCOD/l

$X_{CH_4}$  – concentration of methanogenic population in digester, mgCOD/l

$Xd_{ACID}$  – decrease in concentration of acidogenic population in digester due to bacterial decay, mgCOD/l

$Xd_{CH_4}$  – decrease in concentration of methanogenic population in digester due to bacterial decay, mgCOD/l

$Xg_{ACID}$  – increase in concentration of acidogenic population in digester due to bacterial growth, mgCOD/l

$Xg_{CH_4}$  – increase in concentration of methanogenic population in digester due to bacterial growth, mgCOD/l

$X_o$  – initial concentration of viable acidogenic population in raw sludge, mg COD/l

Y – true yield, mg biomass / mg COD

$Y_{obs}$  – observed yield, mg biomass / mg COD

## Theme of the thesis

According to “Thesis/Practicum Information and Guidelines”, provided by the Faculty of Graduate Studies at the University of Manitoba, two thesis formats are acceptable: the standard format and manuscripts within a thesis (sandwich thesis format). This thesis is composed as a sandwich thesis, which means it comprises a collection of papers which have been published or have been submitted for publication. Additional chapters include: introduction and objectives, which contains a concise overview of existing literature leading to the study presented in thesis; literature review, which contains a critical review of the papers germane to the study; objectives section, which contains the main hypothesis and specific objectives and goals with references to specific papers/chapters providing detailed descriptions of methods, results and conclusions; and final conclusions which summarize all findings in a concise form but also include engineering significance of the work, and suggest future research.

## QA/QC assurance

The goal of QA/QC was to guarantee the generation of precise and accurate data. The QA/QC procedures have two components: Quality Assurance (QA) – the part of the program used to verify that the entire analytical process is operating within acceptable standards and procedures; and Quality Control (QC) – the part of the program used to verify the analytical results for precision and accuracy.

QA included the following components:

- Standard Operating Procedures – SOPs were used whenever possible. The sources for SOPs included Standard Methods operating procedures, manufacturers procedures, procedures described or devised by other laboratories and/or researchers. In case where the use of SOPs was not possible and a novel method was used, the method was described in the thesis and results were compared with results presented in literature (for example: the AHAB procedure).
- Proper training – tests were performed only by properly trained individuals including laboratory technicians or trained, summer students. Training included general sample handling issues, equipment operating and equipment calibration.
- Equipment – each instrument was well maintained and operated only by trained staff members. To achieve reliability, logbooks were kept for many instruments (such as gas chromatographs, autoclave), and calibration was

carried out according to manufacturer guidelines or in accordance to the relevant SOP. Balances, pipettes, spectrophotometers, and other measuring devices were calibrated frequently (very often before each set of measurements was taken).

QC included the following components:

- blanks – reagent blanks were prepared with many set of samples (including fecal coliforms, VFA or COD). The blank samples included all reagents that were used in the analytical process and were carried through the entire process, which would include extraction, filtering, digestion or incubation.
- Duplicates/triplicates – many samples were routinely collected and tested in duplicates at all times, including TS and VS. Batch experiments were conducted in duplicates or triplicates to assess variability of experimental design and system behaviour.
- Long-term, steady-state sampling – in order to assess reproducibility of obtained results (and stability of the systems tested) many experiments were carried out in steady-state conditions over long periods of time (weeks and months). Measurements were taken periodically and average values and variability was assessed.

# 1 Introduction & Objectives

## 1.1 Research background

Wastewater sludge, a by-product of wastewater treatment process, constitutes about 1% of incoming wastewater by volume and contributes to about 50% of the overall costs of a wastewater treatment plant (WWTP). It is a suspension of both organic and inorganic solids with concentration between 1% and 5% by weight. Ultimate sludge disposal include land application in agricultural areas as fertilizer or soil amendment, forests, parks (preferred options) or storage in lagoons, landfills (less preferred option). Sludge treatment is chosen depending on the type of ultimate disposal required.

Among the many options of sludge treatment technologies, the most commonly used are: anaerobic or aerobic digestion, alkaline stabilization, composting, drying, incineration or combination of processes. In the past, and at present day, anaerobic digestion is probably most often used (or is part of the sludge process) because agricultural sludge utilization has been considered as the most beneficial and sustainable use of sludge. However, before sludge can be accepted for agricultural use it has to comply with regulations.

Under US. EPA regulations (most commonly used or referred to throughout the world) sludge is divided into Class A (unrestricted land application) and Class B (restricted land application). Two parameters are taken into account for sludge classification: vector attraction reduction, which in practice means achieving certain volatile solids (VS) reduction, and pathogen concentrations in sludge. Pathogen classes found in sewage sludge, including bacteria, protozoa, viruses and helminthes, levels of *Escherichia coli*, *Salmonella sp.* and viable helminth ova are monitored under US. EPA requirements. Anaerobic digestion has to comply with time-temperature requirements, which means

thermophilic treatment (50°C or more) is required to produce a Class A product. This requires an additional energy input for sludge heating (compared to conventional anaerobic digestion at mesophilic temperature).

Sewage sludge is a material rich in organic matter. One of the key assets of anaerobic digestion is the fact that energy can be recovered in the form of biogas. Very often anaerobic digestion systems are self sustainable (provide enough energy to heat digesters) or even produce net energy that can be used within the plant or sold to grid if converted to electricity. Biogas production is proportional to VS reduction in sludge and much attention has been paid in the past years to increase sludge digestibility. The commonly accepted three step model of anaerobic digestion includes hydrolysis of particulate matter, acidification and conversion of organic acids to methane. An increase in sludge digestibility and methane production, necessitates an increase in sludge hydrolysis, which has been found to be a bottleneck in the process. Many methods are used to increase sludge hydrolysis such as thermal pretreatment, ultrasound, ozonation, chemical additions (acids, enzymes) or phase digestion. The most commonly applied processes in practice include thermal treatment, and/or thermal treatment combined with phase digestion. Phase digestion is based on the fact that there are three distinguished groups of bacteria: acidogens (those that create organic acids), syntrophs (those that convert solvents and higher organic acids to acetate, hydrogen and carbon dioxide) and methanogens (those that convert organic acids to biogas) that have different growth rates. Fast growing acidogens are separated from slow growing syntrophs and methanogens with the use of short SRT in the first reactor of a two-stage system (acid-gas system). It has been shown that acid digestion contributes to increase in sludge hydrolysis (Ghosh, 1981, Ghosh et

al., 1975, Ghosh et al., 1987, Perot and Amar, 1989). Phase digestion, combined with thermal treatment of either phase, may provide Class A biosolids and partially offset costs related to energy in the form of heat that must be provided. Usually, two-phase digestion and/or temperature-phase digestion has been reported to contribute to an additional 10 to 15% increase in VS destruction (Wilson and Streicher, 2001). However, there are conflicting reports as to the accompanying increase in gas production. Very often little or no improvement is achieved compared to conventional mesophilic digestion. Besides, there are no guidelines or optimization parameter for retention time of acid-digester except for minimum time to achieve time-temperature compliance for Class A biosolids. That leads to considerable differences in the operational regime of existing plants and may contribute to unnecessary energy spending.

Much energy involved in sludge treatment is associated with sludge volume, since the bulk of the volume is taken by the water. Doubling solids content reduces water amount by half, and therefore sludge volume decreases roughly by 50%. Drawing some part of water from solids (sludge thickening) could result in overall improvement in energy balance of sludge treatment and decrease investment costs related to the size of the facility. It is unknown what level of sludge thickness could be applied in practical situations. Sludge at approximately 9% solids level begins to behave as non-Newtonian fluid and problems with pumping and digesters mixing must be taken into account. Also, the impact of sludge thickness on sludge hydrolysis and effectiveness of methane production (as per mass VS added) is not fully known.



## 1.2 Objectives

One of the aspects of phase digestion (especially at higher solids concentration) is the ability of achieving high concentrations of organic acids in the acid-digester. Organic acids in un-ionized form have been proven to have antibacterial properties and are used in the food industry as disinfectants. If it is possible to achieve the level of toxicity that would enable effective inactivation of pathogenic organisms then thermal treatment would be unnecessary. This could improve the overall energy balance for the digestion system. However, this has not yet been demonstrated before during anaerobic sludge digestion and factors such as organic acids concentration, pH, retention time, and temperature for successful pathogen inactivation in sludge are not known at this moment. Reviewing the current state of anaerobic digestion technology, the following questions were asked:

- Is thermal treatment of sludge beneficial from an energy view point or can it be justified only for inactivation of pathogens found in sewage?
- How does sludge thickness affect efficiency of anaerobic digestion, specifically hydrolysis rate (considered the major bottleneck of the process)?
- Could toxicity of organic acids be used instead of temperature to effectively inactivate pathogens in sludge?
- Could an anaerobic digestion system that uses organic acids toxicity to inactivate pathogens and, at the same, offers increased energy recovery compared to conventional digestion systems, be developed?

## 2 Literature Review

### 2.1 Energy from anaerobic digestion

The process of anaerobic digestion (AD) stabilizes the organics in wastewater sludge and leads to the production of significant amounts of biogas which contains methane.

Therefore, during the process of sludge stabilization through AD, the amount of energy, which is proportional to the amount of biogas produced, is created. Besides that energy there is also a certain amount of energy that needs to be delivered for the process (such as heat). A simplified overview of energy flow during AD process is presented in Figure 1-

1.

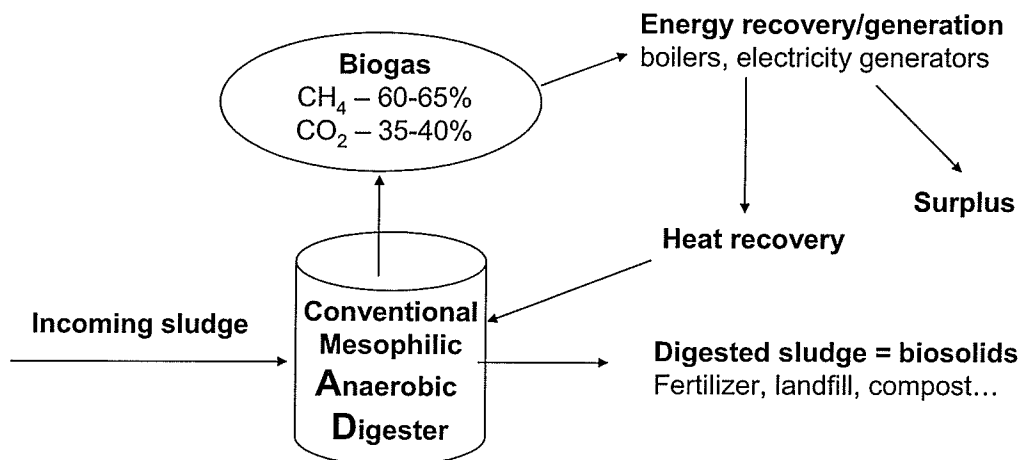


Figure 1- 1. General overview of mass and energy during conventional mesophilic anaerobic digestion.

### 2.2 Biochemistry of AD

Anaerobic biological decomposition is generally considered to be a three step process: hydrolysis and acidogenesis, acetogenesis and methanogenesis (Schieder et al., 2001) as presented in Figure 1-2. In the first step, solid reactants have to be rendered soluble because it is only in a water-soluble state that they can be transported into the cells of

microorganisms to be degraded further. From a chemical point of view, hydrolysis means breakdown of long-chain bio-polymers by the reaction with water. The resulting oligomers and polymers (which are soluble), can easily be fermented into organic acids and solvents by anaerobic microorganisms. Biologically, hydrolysis works through the influence of enzymes. As a result of hydrolysis and acidogenesis volatile fatty acids are produced as well as intermediary metabolites (lactate, succinate, pyruvate), some end products (acetate, ethanol, acetone, propionate, etc.), substrates that can be utilized by various reducing bacteria (such as sulfate reducing bacteria or iron reducing bacteria), and finally hydrogen and carbon dioxide.

In the acetogenic stage, acetic acid, carbon dioxide and hydrogen is produced. These are converted in the third stage to the main components of final biogas which are methane and carbon dioxide.

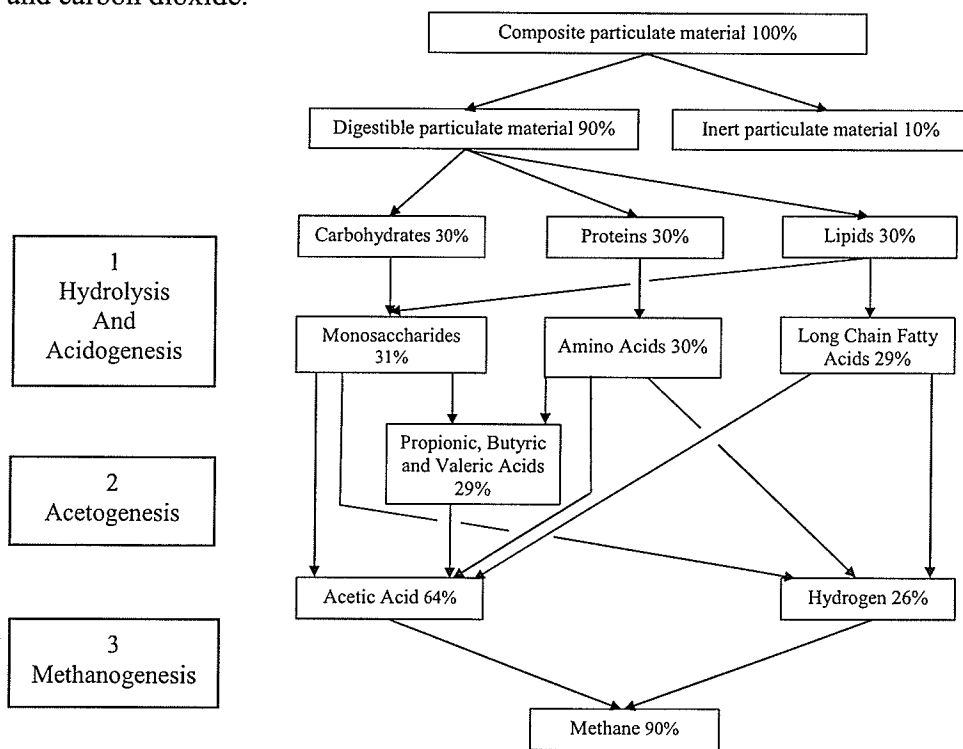


Figure 1- 2. Main stages of anaerobic digestion – COD flux (based on Batstone et al. 2002).

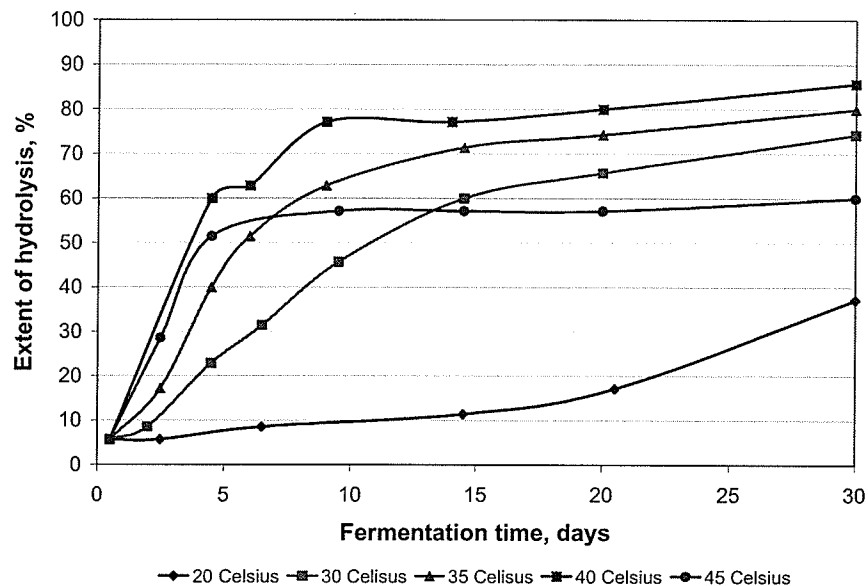
### **2.3 Hydrolysis as a key to improvement of digestibility**

Hydrolysis of complex, insoluble substrates into simpler, more soluble intermediates has been reported as the rate limiting step of AD (Fox and Pohland, 1994, Ghosh, 1981, Mino et al., 1995, Zeeman and Sanders, 2001). The most commonly applied engineering solution to improve hydrolysis is an increase in temperature of the process. Conventional MAD is applied in mesophilic temperature range (commonly between 35 and 38 Celsius). By increasing the temperature to thermophilic range (between 50 and 60 Celsius) many researchers claim that hydrolysis rates increase (Ghosh et al., 1999, Ghosh et al., 2001, Hasegawa et al., 2000, Kiyohara et al., 2000, Mitsdörffer et al., 1990, Moen et al., 2001). Temperatures higher than thermophilic have also been applied but they remain outside the scope of this description.

#### **2.3.1 Impact of temperature on hydrolysis rates**

A study conducted by Kiyohara et al., (2000) showed that the protein, carbohydrate and lipid removal efficiencies under thermophilic conditions were 2.4, 1.7 and 1.1 times higher than those under mesophilic conditions. Bonmanti et al., (2001) showed that the soluble COD (SCOD) concentration was much higher (almost double) at a temperature of 80<sup>0</sup>C than in 60<sup>0</sup>C of thermally treated pig slurry. Moen et al., (2001) found SCOD concentrations in the thermophilic reactor at 55<sup>0</sup>C from 1.6 times to 5 times higher compared to mesophilic reactor at 35<sup>0</sup>C over all SRTs. Ghosh et al., (2001) showed that VS reduction was from 10 to 20% greater in a thermophilic reactor than in a mesophilic reactor which was accompanied by overall greater reduction of proteins, carbohydrates and lipids in thermophilic conditions. When anaerobic treatment of slurry of

lignocellulosic material was studied the rate of hydrolysis was found to be temperature dependent as shown in Figure 1-3.

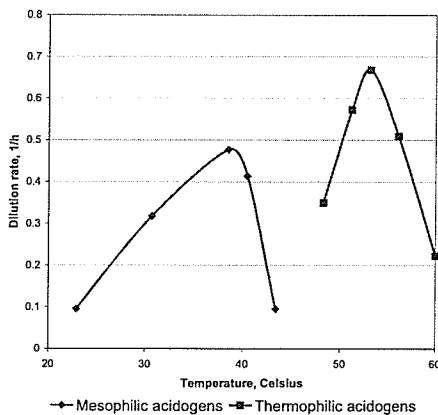


**Figure 1- 3. Effect of temperature and time on the hydrolysis of cellulose and hemicellulose by an anaerobic mixed culture (adapted from Rittmann and McCarty, 2001 after Tong and McCarty, 1991).**

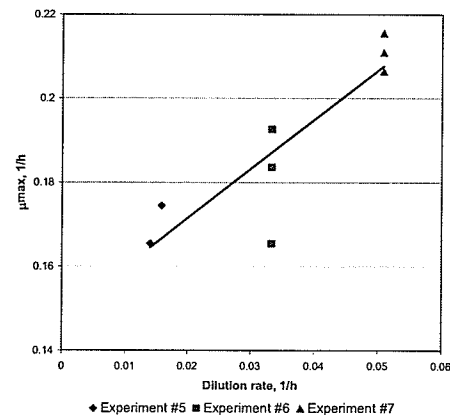
Zoetemeyer et al., (1982a) studied the influence of temperature on the anaerobic fermentation of glucose (1% solution of glucose monohydrate). Optimum temperatures for mesophilic (37<sup>0</sup>C) and thermophilic (52<sup>0</sup>C) phases were found by measuring maximum dilution rate of glucose as shown in Figure 1-4. A study conducted by Templeton and Grady (1988) revealed that dilution rate corresponded to the maximum specific growth of pure bacterial cultures (Figure 1-5).

Hydrolysis (or dilution) rate is directly associated with growth rates of microbial mass. Thermophilic biomass has greater maximum growth rates than mesophilic biomass using biological polymers because higher hydrolysis rates lead to greater monomer availability for growth. Zoetemeyer et al., (1982a) found that maximum specific growth rate of thermophilic biomass was found to be 0.72 h<sup>-1</sup> and for mesophilic biomass 0.51 h<sup>-1</sup>.

Ghosh et al., (2001) found maximum specific growth rate of thermophilic and mesophilic acetogens to be  $0.7 \text{ day}^{-1}$  and  $0.4 \text{ day}^{-1}$ , respectively. In another study conducted by Ghosh et al., (1975) it was established that the maximum specific growth rate of acidogenic microorganisms under mesophilic conditions fed with activated sludge and glucose corresponded to  $0.16 \text{ hour}^{-1}$  and  $1.25 \text{ hour}^{-1}$  respectively. The lower growth rates of acid formers grown on sludge were attributed to “the rate-controlling nature of the sludge hydrolysis step” (Ghosh, 1981). At SRT of 20 days the activities (defined as VFA production rate) of acidogenic bacteria under thermophilic conditions were about 1.8 times higher than those under mesophilic conditions (Kiyohara et al., 2000).



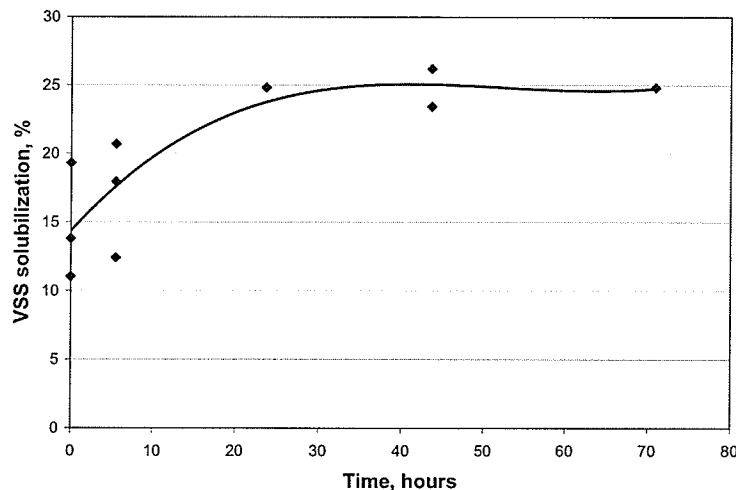
**Figure 1- 4. Maximum dilution rate as a function of temperature (adapted from Zoetemeyer et al., 1982a).**



**Figure 1- 5. Effect of dilution rate on the  $\mu_{\max}$  determined with the batch procedures (adapted from Templeton and Leslie Grady, 1988).**

Hasegawa et al., (2000) conducted an experiment to distinguish solubilization (measured as a VSS removal) by either heat ( $70^{\circ}\text{C}$ ) or by thermophilic bacteria. The use of sterilized sludge results showed that 20 to 25% of the organic sludge was solubilized in 1 day as shown in Figure 1-6. That means that higher temperature affect hydrolysis rates not only through its influence on microbial growth rates but also by other factors.

Generally, an increase in temperature creates an increase in biochemical reaction rates as predicted by Arrhenius equation. Temperature has strong influence on reaction pathways due to thermodynamic yields and microbial populations (Batstone et al., 2002). Temperature is therefore a critical factor controlling hydrolysis and acidogenesis of particulate matter. Thermophilic reactors have been shown to perform better than mesophilic reactors in terms of hydrolysis of organic matter which is strongly correlated with higher microbial growth rates in thermophilic temperatures.



**Figure 1- 6. Sludge solubilization by heat (70°C) treatment (adapted from Hasegawa et al., 2000).**

### **2.3.2 Hydrolysis rates constants**

Kinetic parameters for sludge hydrolysis were collected and are presented in Table 1-1. The importance of these parameters is growing since, with the recent development of modeling programs, hydrolysis is a key parameter needed to predict the process behaviour correctly. While they are very sensitive variables, they are also the weakest part of mathematical models.

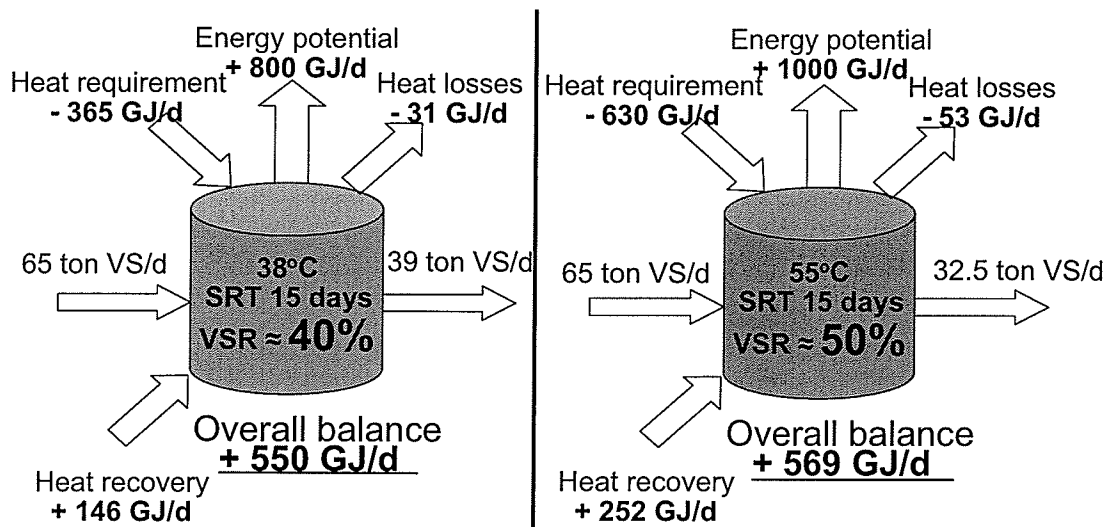
**Table 1- 1. Hydrolysis kinetic parameter values – summary.**

| Study   | First-order hydrolysis constant, $day^{-1}$ | Temperature, °C | Reference                   |
|---|---|-----------------|-----------------------------|
| Domestic primary sludge   | 3.0   | 35              | Eastman and Ferguson, 1981  |
| Hydrolysis of starch by pure culture of <i>Bacillus amyloliquefaciens</i> in aerobic, anoxic and anaerobic conditions | 3.5, 3.32 and 2.88                          | 28              | Mino et al., 1995           |
| Hydrolysis of starch by pure culture of <i>Aeromonas hydrophila</i> in aerobic, anoxic and anaerobic conditions       | 6.07, 5.44, 5.98                            | 28              | Mino et al., 1995           |
| Hydrolysis rate of nitrogenous compound under aerobic, anaerobic and anoxic conditions                                | 0.12, 0.06, 0.03                            | 20              | Henze and Mladenovski, 1991 |
| Hydrolysis of MSW   | $0.8 \times 10^{-3}$                        |                 | Borzacconi et al., 1997     |
| Hydrolysis of MSW; OLR = 4.1, 6.8 and 2.1 kgTVS/m <sup>3</sup> d  | 0.37; 0.43; 0.29                            | 37              | Cecchi et al., 1988         |
| Raw sludge  | 0.15-0.004                                  | 25              | Haug, 1993                  |
| Digested sludge   | 0.0282-0.0037                               | 25              | Haug, 1993                  |
| Limed raw sludge  | 0.0293-0.0045                               | 25              | Haug, 1993                  |
| Primary papermill sludge  | 0.0033                                      | 25              | Haug, 1993                  |
| Pulpmill sludge   | 0.0095                                      | 25              | Haug, 1993                  |
| Kraft papermill sludge  | 0.0015                                      | 25              | Haug, 1993                  |
| Hydrolysis of lipids  | 0.010-0.005                                 | 55              | Christ et al., 2000         |
| Hydrolysis of proteins  | 0.075-0.015                                 | 55              | Christ et al., 2000         |
| Hydrolysis of carbohydrates   | 0.200-0.025                                 | 55              | Christ et al., 2000         |
| Thermophilic food digestion   | 0.079-0.036                                 | 55              | Kim et al., 2000            |
| Hydrolysis of primary sludge under mesophilic conditions IWA, 2002 for ADM No. 1                                      | 0.25  | 35              | Batstone et al., 2002       |
| Hydrolysis of primary sludge under thermophilic conditions IWA, 2002 for ADM No. 1                                    | 0.40  | 55              | Batstone et al., 2002       |



## 2.4 Impact of increased temperature and hydrolysis rates on energy recovery

An energy balance was performed for single phase thermophilic and mesophilic digester as shown in Figure 1-7. Energy balance included the following components: energy associated with methane content of biogas, heat requirement, losses and recovery for the digester. It is also assumed that thermophilic digestion due to higher hydrolysis rates, results in 50% volatile solids (VS) destruction and mesophilic in 40% VS destruction. It should be noted that it is the most favorable scenario for thermophilic digestion. It has been shown that thermophilic digestion may not result in greater biogas production and VS destruction compared to conventional MAD (Garber, 1982, Kugelman and Guida, 1989).



**Figure 1- 7. Energy balance for single thermophilic and single mesophilic anaerobic digester.**

Explanation of terms used in Figure 1-7:

$$\text{Energy potential} = V_{\text{CH}_4} \times 37 \text{ GJ/m}^3$$

Where: methane production  $V_{\text{CH}_4} = 0.35 \times (\text{COD}_{\text{in}} - \text{COD}_{\text{out}}) \times Q$

Additionally: 1.0 g TS  $\approx$  1.42 g COD, VS/TS = 0.65, and sludge flow is approx. 3333 m<sup>3</sup>/d at 3% solids content

$$\text{Heat requirement} = Q \times (T_{\text{Digester}} - T_{\text{incoming sludge}}) \times \rho_{\text{sludge}} \times c_{\text{sludge}}$$

Where: Q – sludge flow, m<sup>3</sup>/d, T<sub>digester</sub> – temperature in digester, °C, T<sub>incoming sludge</sub> – temperature of incoming sludge, °C,  $\rho_{\text{sludge}}$  – specific gravity of sludge, kg/m<sup>3</sup>,  $c_{\text{sludge}}$  – specific heat of sludge, J/kg °C.

$$\text{Heat recovery} = Q \times (T_{\text{exchangerHot}} - T_{\text{exchangerCold}}) \times c_{\text{sludge}} \times 0.8$$

Where: T<sub>exchangerHot</sub> – temperature on the hot side of heat exchanger, °C, T<sub>exchangerCold</sub> – temperature on the cold side of the heat exchanger, °C; 0.8 – efficiency of heat exchanger

Heat losses = Heat losses through walls + Heat losses through floor + Heat losses through cover

Heat losses through walls = Area of walls  $\times$  (T<sub>Digester</sub> – T<sub>air</sub>)  $\times$  Heat transfer coefficient for wall and insulation

Heat losses through floor = Area of floor  $\times$  (T<sub>Digester</sub> – T<sub>earth</sub>)  $\times$  Heat transfer coefficient for floor

Heat losses through cover = Area of cover  $\times$  (T<sub>Digester</sub> – T<sub>air</sub>)  $\times$  Heat transfer coefficient for cover

Where: T<sub>air</sub> – temperature of the air, assumed 10°C, T<sub>earth</sub> – temperature of the earth, assumed 10°C, Heat transfer coefficient for wall and insulation = 0.054 MJ/(m<sup>2</sup> °C), Heat transfer coefficient for floor = 0.025 MJ/(m<sup>2</sup> °C), Heat transfer coefficient for cover = 0.103 MJ/(m<sup>2</sup> °C).

Calculations of heat requirements, recovery and losses were based on Tchobanoglous and Burton, 1991.

Using common logic and economic rationale, additional spending of energy to elevate temperature of digestion from mesophilic to thermophilic temperature should at least pay for itself or bring surplus of energy. Even though, it was assumed that thermophilic digestion destroyed more VS and produced more biogas, overall energy balance for thermophilic digestion is only slightly better (approximately 1% more) than mesophilic digestion.

Therefore, from an energy recovery view point, increasing temperature of digestion from mesophilic temperatures (commonly between 35°C to 38°C) to thermophilic temperatures (usually between 50°C and 60°C) does not seem beneficial (even, including 10% more VS destruction at thermophilic AD accompanied by additional biogas production). Besides, effluents from thermophilic digesters have been reported to contain higher contents of soluble organic compounds, which contribute to poor dewaterability and odour problems.

## **2.5 Development of acid-gas digestion**

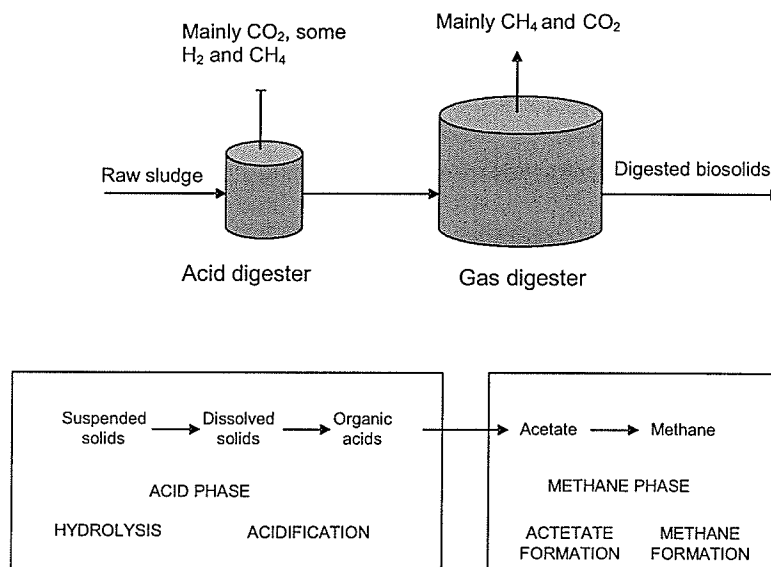
The importance of the hydrolysis and fermentation steps of AD was investigated in the mid 70's and the idea of a two-phase digestion was developed (Ghosh et al., 1975). The idea had its basis in findings that the full metabolic potential of the organic acid and methane producers may not be reached in a single-phase process. When these two groups of microorganisms are separated it is possible to stimulate them and enhance process efficiency and reliability by creating the proper environment. In the first reactor where hydrolysis and acidification of sewage sludge takes place the environment is controlled to promote the growth and proliferation of the acid formers. The second reactor receives the products from the first and is designed to provide an optimum environment for the methane formers -

Figure 1-8. The sizing of digesters could be optimized by the use of the kinetic controls that govern growth rates of these two important bacteria groups separately. In completely mixed reactors, the organisms' specific growth is equal to the reciprocal of the HRT. Because the digester is optimized for methanogens, the prevalent specific growth rate of acidogens in a high-rate digester would be between 0.06 and 0.025 day<sup>-1</sup>. This is the specific overall growth rate that can sustain methanogens, (Ghosh and Buoy, 1993) while in the acid-phase digester it could be as high as 3.84 day<sup>-1</sup> (Ghosh, 1981). This fact alone allows for the design of

digestion at shorter HRT in the acid phase part and/or to increase the load on this digester. As it has been demonstrated during many studies (Ghosh, 1981, Ghosh et al., 1975, Ghosh et al., 1987, Perot and Amar, 1989) that phase separation offers:

- enrichment in cultures of each group of digester organisms, thus optimizing the overall digestion process
- a substantial reduction in total reactor volume and consequent savings in capital and operating costs
- higher rates of solids stabilization and methane production
- much higher content of methane (up to 85 mol %) in the final product gas
- decreased heat requirements and increased net energy production
- suitability for incorporation into existing treatment plants with minimum capital investment
- increased process stability and faster start-up period
- increased stabilization of WAS which is more difficult to degrade when compared to

PS



**Figure 1- 8. Two phase AD, also known as acid-gas digestion (based on Wilson, 2001).**

Separate hydrolysis-acidification step offers:

- the kinetics of overall process of AD (acidogenic process) to be determined primarily by the rate of hydrolysis and not by growth kinetics of methanogenic stage of the process. If favorable conditions are provided for bacterial growth, hydrolysis may be efficient due to enzymatic reactions (Eastman and Ferguson, 1981)
- separation of the two limiting steps of AD – hydrolysis and methanogenesis. It improves the reaction kinetics of the whole AD process by improving reaction kinetics of hydrolysis and methanogenesis separately.
- when ambient temperatures prevail or influent has high TS concentration, a better removal of solids is achieved in first-step of AD (Miron et al., 2000)
- the presence of about 20 % of COD in hydrolysed form (Miron et al., 2000, Perot and Amar, 1989)
- the strongest increase in hydrolysis and acidification of total COD occurs between first 3 days of the process (Miron et al., 2000)
- promotion of the production of soluble TOC at the level of 1.5 g/l d by hydrolysis-acidification step (Perot and Amar, 1989)
- the production of soluble COD during acid-phase digestion which consists of 85 to 95% VFA content (Chyi and Dague, 1994, Eastman and Ferguson, 1981)
- acid-production which is not affected by feed concentration over a wide range. Some acid-forming bacteria can grow well at pH 5. Therefore, the acid phase digester should be highly resistant to variations in feed strength and be able to convert about 60 to 90% of the degradable particulates to VFA and biomass (Eastman and Ferguson, 1981)

- possibility to increase loading on acid-phase digester up to 16 kg VS/m<sup>3</sup>day as compared with conventional high rate digester at loading of 0.64 kg VS/m<sup>3</sup> day (Ghosh et al., 1987).

In summary, two-phase digestion separates and isolates the two-limiting steps – hydrolysis and methanogenesis – and thereby enhances them by improving reaction kinetics and stability of each separately – see Figure 1-9. In the first reactor, hydrolysis is enhanced by more intimate contact between intracellular enzymes and complex substrates. The improvement of hydrolysis is due to the fact that usually a smaller reactor volume contains primarily hydrolyzing and acidifying biomass which creates a high concentration of enzymes. In the second reactor, acetogenic and methanogenic bacteria proliferate at the same time also allowing a syntrophic relationship to take place (Fox and Pohland, 1994).

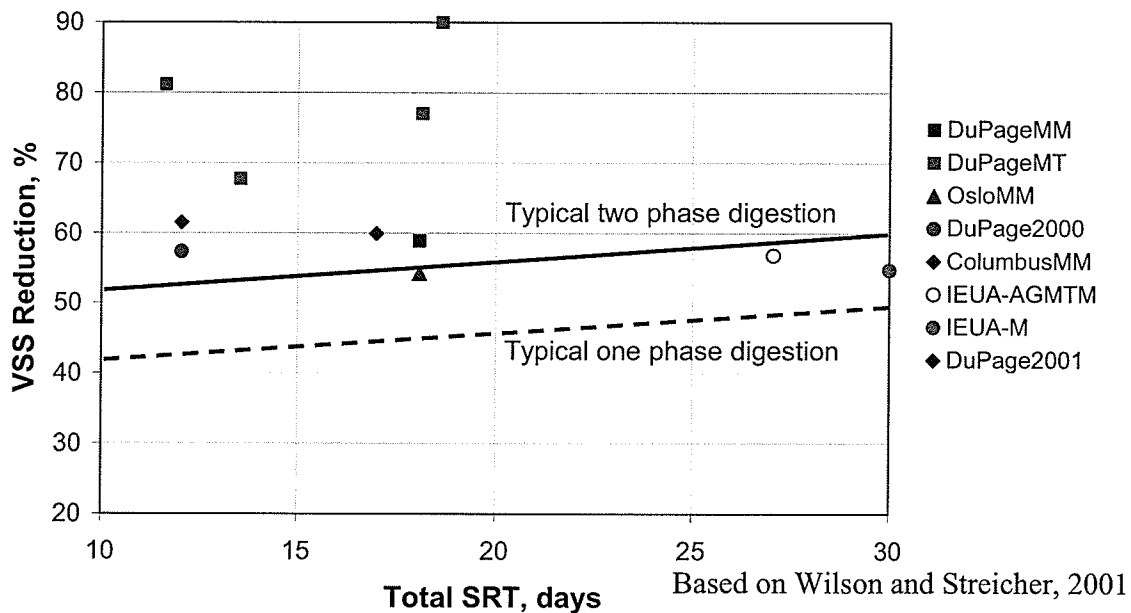


Figure 1- 9. Comparison of VS destruction between plants using two-phase and one-phase AD processes.

## 2.6 Impact of acid-gas digestion on energy recovery

Several improvements to conventional single phase mesophilic anaerobic digestion (MAD), which include: temperature-phased digestion, acid-gas digestion, or recently three-phase and multiple-phase digestion (Ghosh, 1998, Reusser and Zelinka, 2004, Schafer et al., 2003, Wilson et al., 2002), have been researched and implemented in the past two decades. While most of the solutions claim higher biogas production and add stability to the process, there is little information on how these improvements influence overall energy balance for the digestion systems. For instance, using often implemented two-phase, thermophilic-mesophilic anaerobic digestion requires more energy than conventional mesophilic digestion at the same retention time (Shimp et al., 2003). An energy balance for conventional MAD and two-phase, thermophilic acid followed by mesophilic gas digestion is presented in Figure 1-10.

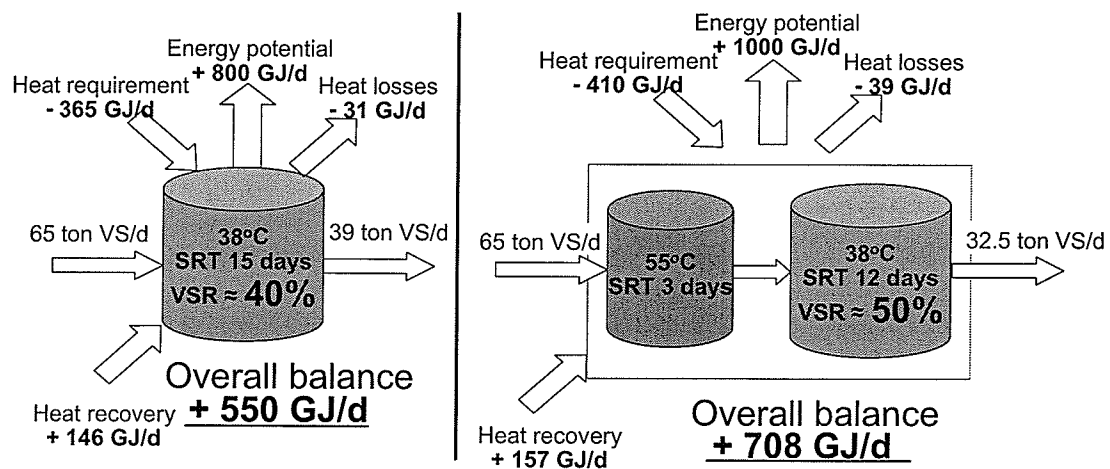
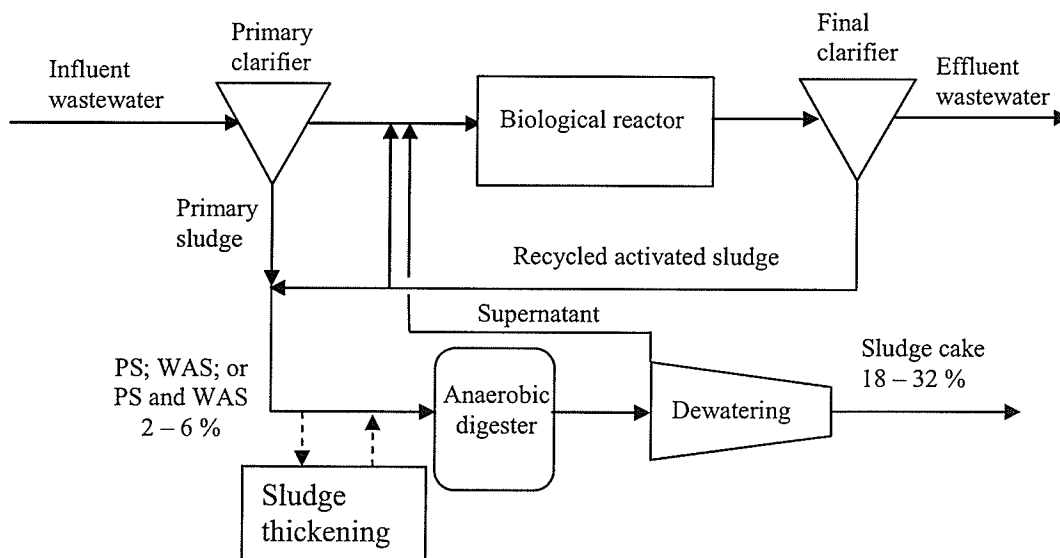


Figure 1- 10. Energy balance for single mesophilic and two - phase, thermophilic - mesophilic anaerobic digester (assumptions as in Figure 1-7).

## 2.7 AD at increased solids content in sludge

Anaerobic digestion is a commonly applied process to stabilize both primary (PS) and/or waste activated sludge (WAS). Typically, the process is applied under sludge solids concentration that varies between 2 – 6% of solids (Quasim, 1999). This represents the sludge concentration coming from primary or secondary sedimentation tanks. A typical wastewater treatment system that employs AD as a process to stabilize sludge is presented in Figure 1-11. Sludge thickening prior to digestion offers the following benefits:

- sludge flow volume reduction - increasing total solids (TS) content of sludge from 3% TS to approximately 6% TS brings approximately 50% reduction in sludge volume (Figure 1-12)
- reduction in digesters' volumes and/or
- increase in retention time at same digestion volume
- reduction in heat requirement (Figure 1-13) of the process (Bailey and Daigger, 1998, Shimp et al., 2003)



**Figure 1- 11. A wastewater treatment system layout with sludge stabilization through anaerobic digestion.**



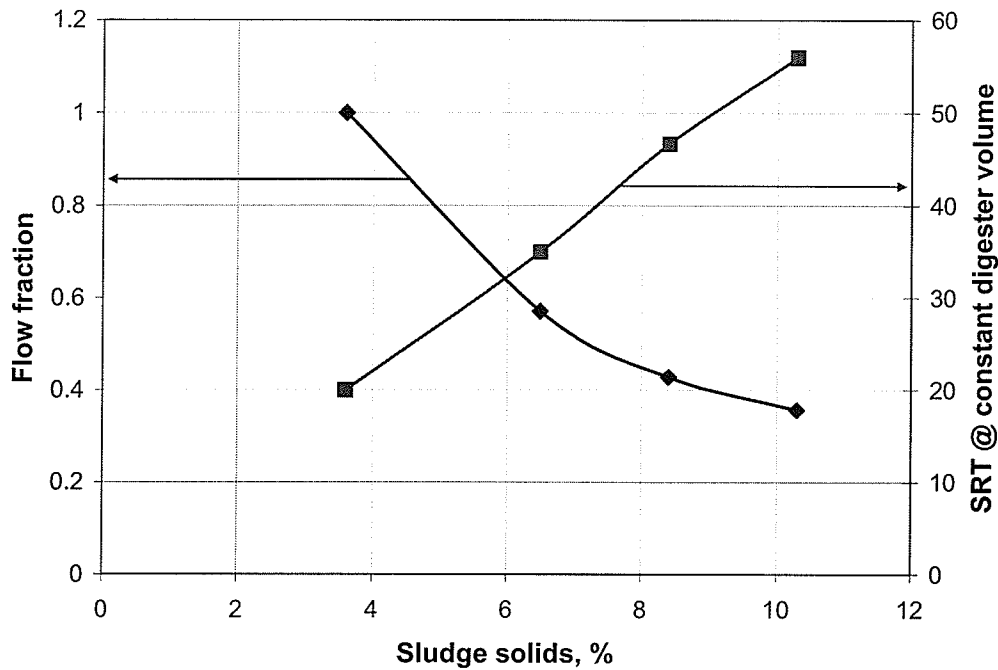


Figure 1- 12. Effect of increased sludge thickness of sludge flow into digester and increase in SRT of digestion at constant digesters volume – based on Biowin simulation (EnviroSim\_Associates\_LTD., 2004).

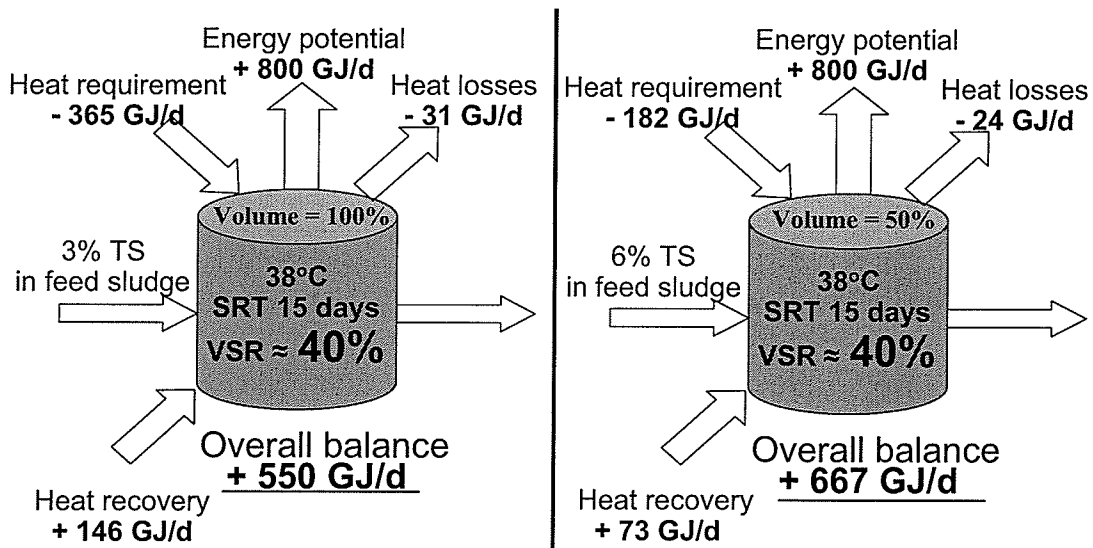


Figure 1- 13. Comparison of energy balance for system that receives sludge with 3% TS (full sludge flow) and system that receives sludge with 6% TS (half of the sludge flow). It was assumed that both systems exhibit equal VS reduction and methane production (other assumptions as in Figure 1-7).

## 2.8 Environmental factors and their influence on acid-phase digestion

Among many parameters that may influence acid-phase digester performance the most important from an operational and research point of view are the concentration of TS in sludge, loading rate, retention time, temperature and pH.

### 2.8.1 TS content and solids loading rate

The impact of higher TS concentrations on hydrolysis-acidification in the AD process has not been broadly explored so far. An interesting example of a study that included variable TS sludge contents (dry animal feed with adjusted moisture content was selected as digester feed) was performed during the development of an innovative-plug flow reactor to determine the extent of the acid-phase part of the reactor as a function of increased loading (Liu and Ghosh, 1997). Phase separation was first observed when the loading increased to 2.05 kg VS/ m<sup>3</sup> day at an HRT of 13.1 days and TS content of 3%. Figures 1-14 presents results when TS content of sludge was increased to 4, 6, 8, and 10%. Increasing the TS from 4% to 10% resulted in an increase in VFA concentration from 3000 mg/l to 10000 mg/l respectively.

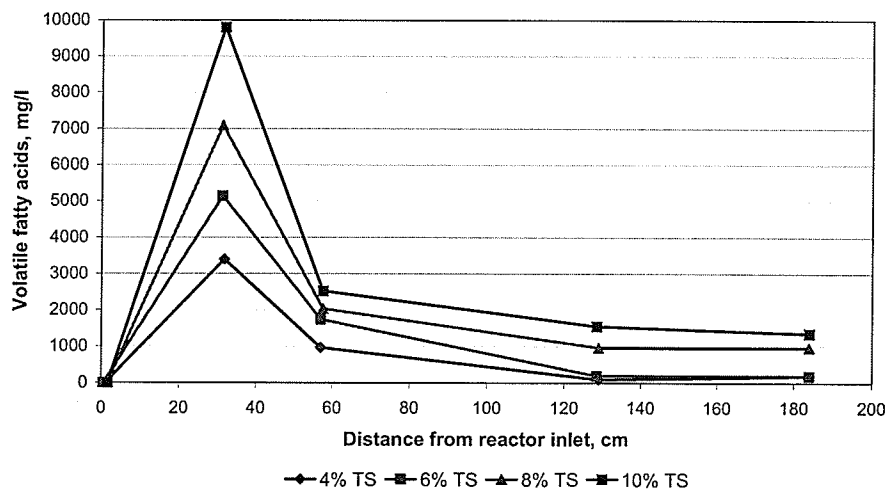
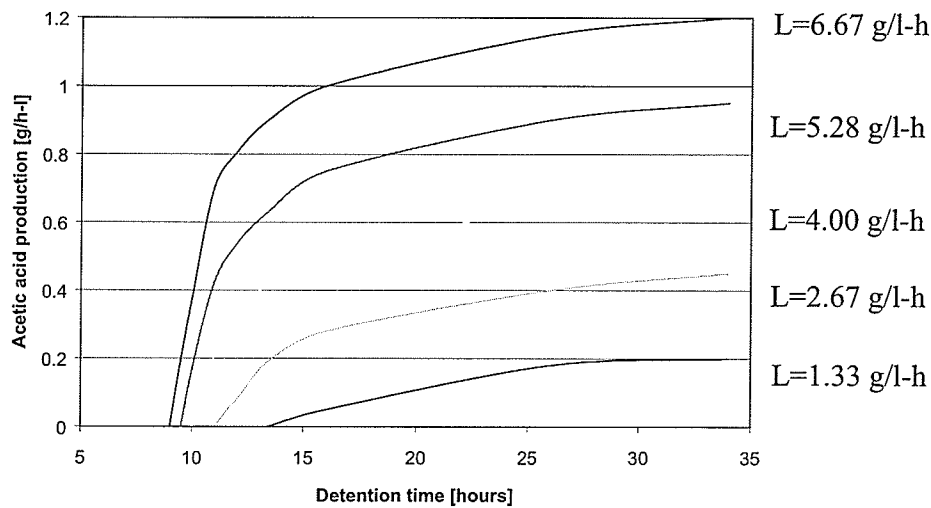


Figure 1- 14. Longitudinal profiles of VFA generated with sludge at different TS content in a plug-flow reactor (based on Liu and Ghosh, 1997).

Theoretically, the rate of acid formation increases with the organic loading. In one of the first works at optimizing acidogenic reactors, it was found that acid-phase digestion may be conducted satisfactory at loading and detention times varying from 1.33 to 6.67 g VS/l hr (activated sludge) and 8.9 to 14.4 hours respectively (Figure 1-15). However, digester operation with loading above 65 kg/m<sup>3</sup> day may not be practical because of problems encountered in pumping and mixing of sludge having consistencies of 9% TS or higher (Ghosh et al., 1975).

A study performed by Chyi and Dague (1994) on the hydrolysis and acidogenesis of crystalline cellulose revealed that soluble COD and VFA increase approximately proportionally to increases in influent cellulose concentration from 4320 mg/l to 16400 mg/l. Moreover, higher concentrations of VFAs did not lessen the overall efficiency of hydrolysis and acidogenesis.

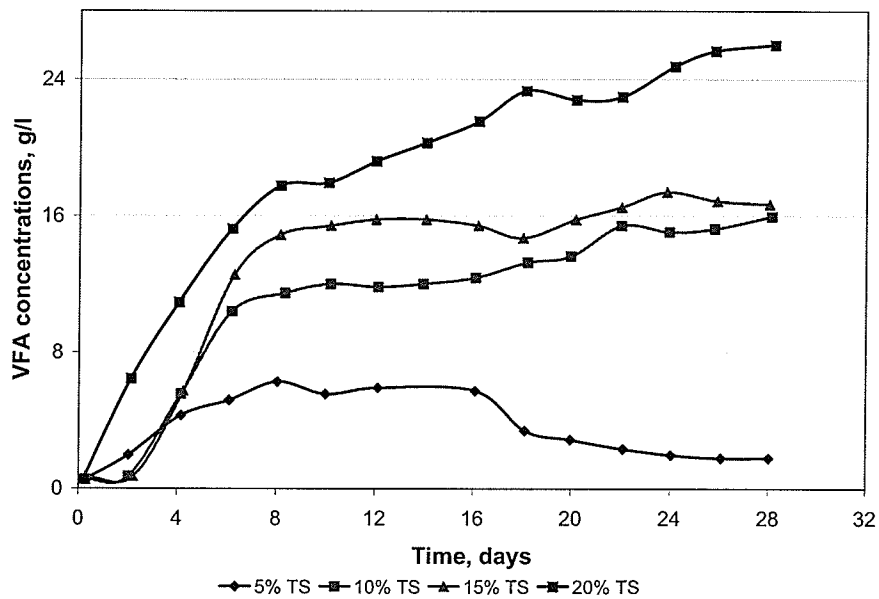


**Figure 1- 15. Curves for volatile acid production rate from acid-phase digester (adapted from Ghosh et al., 1975).**

Recent studies (Vandenburgh and Ellis, 1999) on the effect of solids concentration from 4.4 to 7.9 % on the performance of TPAD showed that the VFA concentration in mesophilic

conditions were only slightly changed by an increase in TS content from around 200 mg/l to 500 mg/l as acetic acid. However, in thermophilic conditions, VFA concentration increased rapidly with an increase in TS content from initially 600 mg/l to approximately 3000 mg/l as acetic acid.

During a study of anaerobic digestion of OFMSW a concentration of TS ranging from 5 to 20% on the production of VFA was examined (D'Addario et al., 1993). An increase in TS content resulted in increase in VFA generation. AD of OFMSW at 5% TS resulted in production of approximately 6 g/l VFA while VFA level at the TS content of 20% exceeded 24 g/l what is shown in Figure 1-16.



**Figure 1- 16. Concentration of VFA vs. TS content in OFMSW (adapted from D'Addario et al., 1993).**

Higher concentrations of TS content imply a proportional increase of the digestible organic matter which results in elevated VFA concentration. The resulting high content of non-ionized acids inhibits methanogenesis but also contributes to the inhibition of acidogenesis.

This can be seen in Figure 1-16 which shows the plateaus are reached at longer retention times. Other factors such as toxic compounds fed with substrate (dissolved phenolic and humic acids lignin related, heavy metals) or produced in the process (partially reduced organic molecules, ammonia, etc.) may also retard acidogenic phase (D'Addario et al., 1993). When AD of swine manure was performed at TS content varying from 60.3 g VS/l to 108 g VS/l it was found that VFA concentration were increasing with an increase in VS in feed from 128 mg VFA /l at 68.3 gVS/l to 6532 mg VFA/l at 108 g VS/l with an exception at the TS concentration of 60.3% when VFA level established at 370 mg/l (Fischer et al., 1984). In another experiment when sludge TS ranged from 2.6 % to 0.43 % it was found that VFA yield resulted in 0.22 mg VFA/mg VS fed at 0.43% TS and only 0.12 mg VFA/mg VS fed at 2.6% TS (Skalsky and Daigger, 1995). It is somehow in contradiction with previously presented findings. As it was explained by authors, more dilute reactor had higher VFA concentration due to better mixing which enhanced hydrolysis and substrate uptake by biomass.

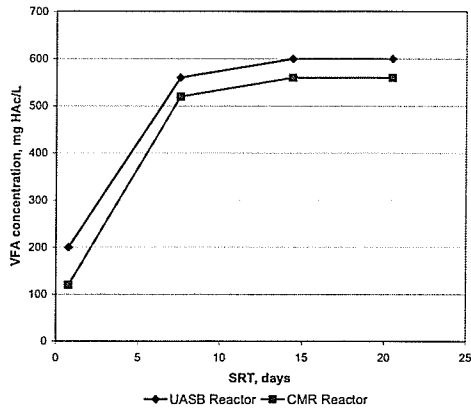
It seems that the enhancement of the hydrolysis-acidogenesis step could be proportional to TS content of sludge at concentrations above 6% TS, while at lower TS ranges, other factor such as mixing may influence overall efficiency of the process. Nevertheless, the question of TS influence on hydrolysis-acidification step remains to be explored.

### **2.8.2 Residence time**

An important operational variable that can be easily manipulated is solids and hydraulic residence time (HRT). HRT can act as a selection parameter for the acidogenic phase if it encourages the growth of acid formers only and suppresses the growth of methane producers. "The ultimate goal of acidogenic digestion can be said to operate at an HRT as high as

possible to minimize reactor volume and associated capital costs, concurrently to maintain a reasonably long SRT in order to promote growth and proliferation of the acid-generation organisms, increase process stability, and minimize sludge production, without inducing growth of methane-forming bacteria” (Kim and Somiya, 2001). In most studies, however, SRT and HRT are equal because of the use of batch reactors or conventional flow systems without solids recycle.

The role of SRT in hydrolysis-acidogenesis of primary sludge was investigated by Elefsiniotis and Oldham (1993). VFA were the main soluble compounds generated during acidogenic digestion. The net VFA production increased slightly with increasing SRT from 10 to 20 days, but it decreased sharply at a 5-day SRT as shown in Figure 1-17. This could indicate that short SRT may challenge the acidogenic population in the reactors. Also, soluble COD concentration as a function of SRT showed great similarity with the VFA production, with the VFA production dropping sharply at an SRT of 5 days and approaching plateau at longer SRTs. However, the COD specific solubilization rates (expressed as mg COD/mg VSS x day) appeared to be independent of SRT as presented in Table 1-2. The reason for this could be that at short SRTs the biochemical pathways followed for VFA production from soluble biopolymers are much more influenced than those involved in hydrolysis. “If the same microbial community is responsible for the conversion of particulate organic matter to VFA, it can be concluded that SRTs below a certain value pose a limit on acidogenic activity; therefore, intermediate soluble products accumulate” (Elefsiniotis and Oldham, 1993).

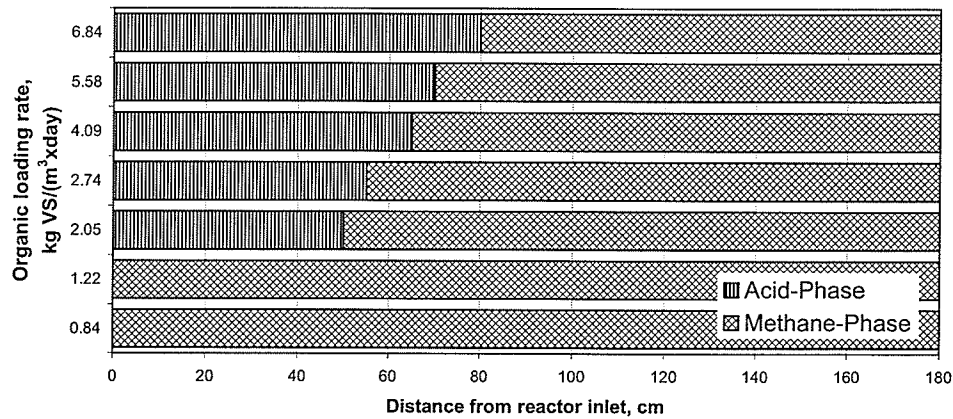


**Figure 1-17. Specific VFA production and COD solubilization (from Elefsiniotis and Oldham, 1993).**

**Table 1-2. Specific rates of VFA production and COD solubilization (from Elefsiniotis and Oldham, 1993).**

| Run | SRT (days) | VFA production rate (mg VFA/mg VSS x day) |       | COD solubilization rate (mg COD/mg VSS x day) |       |
|-----|------------|---|-------|---|-------|
|     |            | CMR                                       | UASB  | CMR   | UASB  |
| 1   | 5          | 0.053                                     | 0.056 | 0.184   | 0.192 |
| 2   | 10         | 0.101                                     | 0.103 | 0.187   | 0.198 |
| 3   | 15         | 0.125                                     | 0.110 | 0.200   | 0.193 |
| 4   | 20         | 0.119                                     | 0.109 | 0.184   | 0.181 |

One of the earliest works on acidogenesis of primary sludge (Ghosh et al., 1975) showed that successful acid-phase digestion may be conducted at detention times between 10 to 24 hours. It seems that at higher solids concentration range acidogenesis of sludge may still be achieved in relatively short time in the range of a few days. Figure 1-18 shows zones of dominant acidogenic and methanogenic activities in plug-flow anaerobic reactor. A more than triple load increase on the digester resulted in only doubling extent of acidogenic zone showing that hydrolysis and acidogenesis is not linearly proportional to the loading applied. If one would assume that higher concentration of TS in sludge would pose higher loading on the digester, the extent of acidogenesis should therefore not increase linearly with the increase of TS content in sludge.

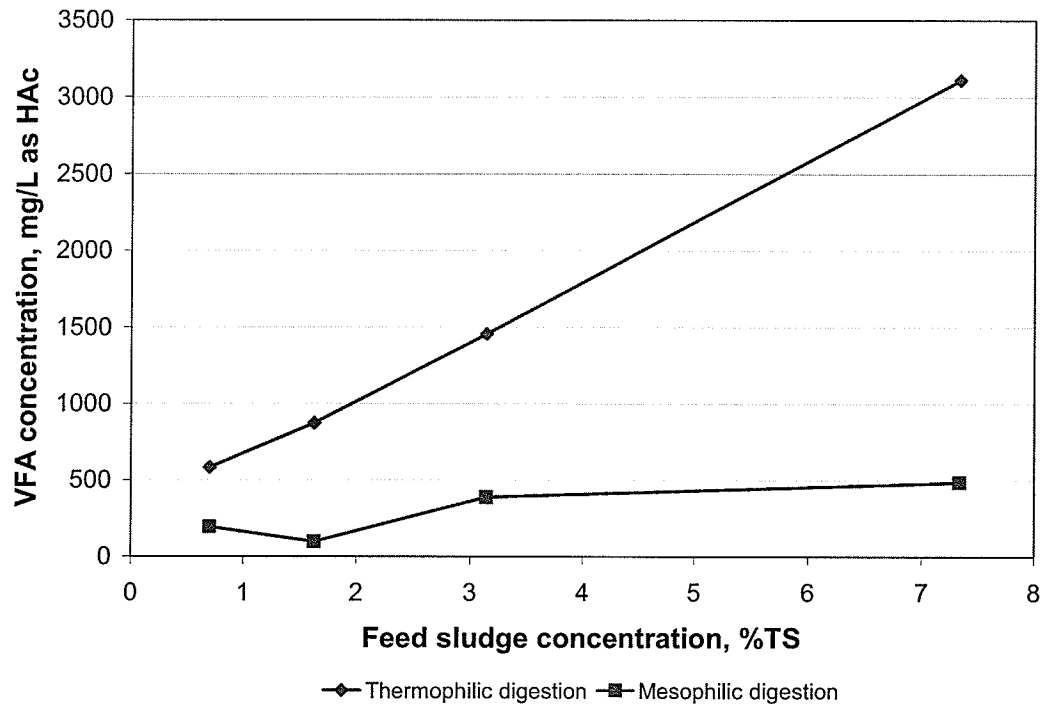


**Figure 1- 18. Zones of dominant acidogenic and methanogenic activities in mesophilic plug-flow reactor (based on Liu and Ghosh, 1997).**

### 2.8.3 Temperature

AD are found to have two different temperatures optima: in the mesophilic range, approximately 35°C and in the thermophilic range, about 55°C. A recent study of the effects of process-control parameters on hydrolysis and acidogenesis (H&A) showed that temperature caused an increase in H&A of proteins and carbohydrates but had no effect on H&A of lipids (Ghosh et al., 1999). In general temperature was found to play a minor role and distinct temperature influence could not be established. However, in most cases thermophilic digestion generated higher H&A of carbohydrates, proteins and lipids at constant pH and HRT. The effect of increased temperature and feed sludge concentration was investigated in another study (Vandenburg and Ellis, 1999). The results are shown in Figure 1-19. AD at thermophilic temperatures resulted in much higher concentrations of VFA at all TS concentration and was especially pronounced at higher ranges of TS concentration.





**Figure 1- 19. VFA concentration versus feed sludge concentration (based on Vandenburg and Ellis, 1999).**

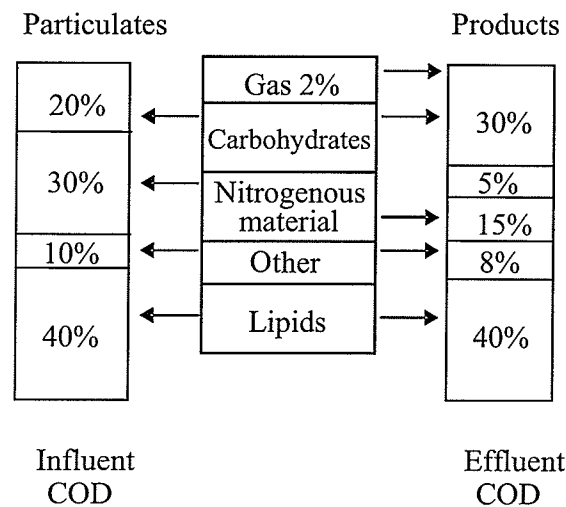
#### 2.8.4 pH

pH was found to have a stronger effect on hydrolysis and VFA formation of sludge than any other process-control parameter. pH 7 resulted in much higher conversion of proteins, carbohydrates and lipids than pH of 5 (Ghosh et al., 1999). The optimum pH for VFA formation of primary-activated sludge at the mesophilic temperatures was found to be between 5.5 and 6.2, while at thermophilic temperatures it was 6. Another study conducted by Zoetemeyer et al., (1982b) concluded that the most stable pH for acidogenic reactor was between 5.7 and 6.0 and stable operation of the acidogenesis of carbohydrates in a single as well as two stage anaerobic process may be hardly possible in the pH range of 6.0 – 8.0. Eastman and Ferguson (1981) found that pH 5.15 was optimal for VFA formation of primary sludge at temperature of 35<sup>0</sup>C (mostly due to inhibition of methanogens). Moreover, acid phase digester was highly resistant to variations in feed strength, converting about 60 to 90%

of the degradable particulates to VFA and biomass within 1 to 3 days. However, an extensive report on acid-phase digestion concluded that the “natural” pH of mesophilic and thermophilic acid-digesters stabilize at around 6.6 (Ghosh et al., 1987). This could suggest that optimal operation may not be reached without pH control.

### 2.8.5 Gas production during acid-phase

There is a certain amount of gas production occurring during acid-phase digestion. This amount may vary significantly depending on the conditions of acid-phase digester. For example, Eastman and Ferguson (1981) reported that only 2% of the influent COD is converted into gaseous products during acid-phase digestion as presented in Figure 1-20.



**Figure 1- 20. General composition of acid phase influent and effluent for domestic primary sludge (Eastman *et al.*, 1981).**

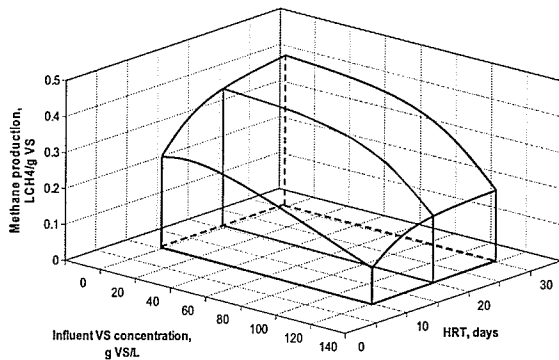
A study performed by Ghosh et al., (1999) concluded that around 5 to 16% of VS can be converted into gas at an HRT of 2 days in mesophilic temperatures. Thermophilic temperatures resulted in decreased gas yield ranging from 1.6 to 7.0% of VS at the same HRT. High-rate two-phase mesophilic digestion of soft-drink bottling wastes resulted in nearly 10% of total gas generated from acid-phase digester (Ghosh, 1981). This is in

agreement with Perot and Amar (1989), and Walden and Andryszak (2003) who reported 6 to 12% of gas generated during acid-phase. The gas composition in the acid-phase digester contained 65% CO<sub>2</sub> and 35% methane. Ghosh et al., (1987) reported no evidence of hydrogen accumulation in the head space in the acid-phase digester except during mesophilic operation at an HRT of 0.9 days. This observation indicated that H<sub>2</sub> utilization by the homoacetogenic bacteria exceeded the rate of H<sub>2</sub> production during oxidation of the sludge hydrolysates. It was also concluded that the critical HRT of the syntrophic methane formers was less than 1 day. However, Zoetemeyer et al., (1982b) reported very high H<sub>2</sub> content in the acid-phase gas from the acid-phase digester fed with glucose. Hydrogen content was as high as 75%. Similar results were reported by Majizat et al., (1997). This suggests that efficient H<sub>2</sub> production could be possible if physical separation of acid and CH<sub>4</sub> formers is provided such as occurs in acid-phase digestion. In a single phase digestion H<sub>2</sub> gas cannot be recovered because it serves as electron-sink for the methanogenic bacteria. Sludge as a organic rich matter is recognized as a potential source of hydrogen production through biomass conversion. However, at this point economical feasibility of H<sub>2</sub> production has not yet been established (Noike and Mizuno, 2000).

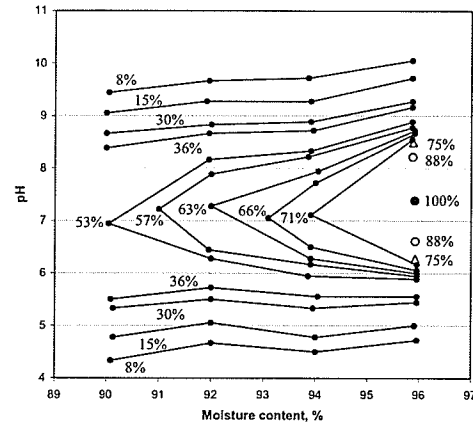
## **2.9 Methane-phase digestion of concentrated wastes**

The influence of moisture content of wastes on methane production has been described by many authors (Fischer et al., 1984, Harikishan and Sung, 2001, Lay et al., 1997a, Lay et al., 1998, Lay et al., 1997b). Figures 1-21 and 1-22 show the relationship between methane production with respect to moisture content, pH and HRT. One general conclusion that can be drawn from these relationships is that methane production per gram VS added decreases with an increase in VS content of wastes and increases with an increase in HRT. pH

influences biogas generation at all range of TS content. Additionally, large quantities of  $\text{NH}_3\text{-N}$  and organic acids may retard or completely inhibit methanogenesis.

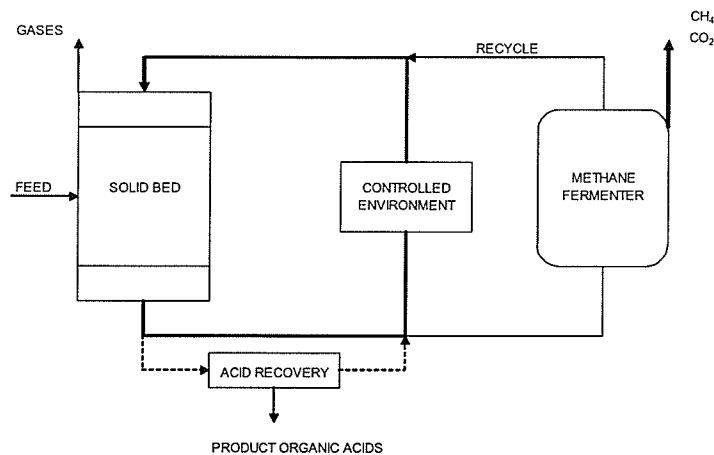


**Figure 1- 21. Theoretical relationship between methane production and HRT versus influent VS concentration (based on Fischer et al., 1984).**



**Figure 1- 22. pH-moisture relationship and equivalent percentage of maximum methanogenic activity (based on Lay et al., 1997a).**

Similar issues were encountered during research on the two-phase digestion of semi-solid feeds with novel upflow bioreactors (Ghosh et al., 1987). The schematic drawing of the system is presented in Figure 1-23. To overcome the difficulties in biogasification of high-solid wastes, unless they are diluted to form slurries, the leachate from the bed, containing mostly liquefaction products was diverted to a separate methane-phase digester. The recycle from the methane phase digester to a leach-bed was applied to conserve the nutrients indigenous to the solids substrate and thus to eliminate or reduce the need for external nutrient addition. This system operated without mechanical mixing. Other advantages over the conventional



**Figure 1- 23. Leach-bed two-phase anaerobic digestion (based on Ghosh et al., 1987).**

AD system included: greater methane production and greater net energy production, much smaller digester volume, higher loading rates, a minimum of feed processing and pre-treatment. The process showed that successful AD of high solid wastes can be achieved if the methane phase is conducted at a different (lower) TS content than the acid production phase.

## 2.10 AD and pathogen inactivation

Wastewater contains the feces and the urine from both humans and animals that carries many disease causing organisms called pathogens. If sludge is to be land applied (which is the case in Winnipeg, Manitoba), the reduction of pathogen numbers during AD process is one of the major concerns. In the United States, the use and disposal of treated sewage sludge (biosolids), including domestic septage, are regulated under 40 CFR Part 503 (E.P.A., 1999). These regulations are to protect public health and the environment through requirements designed to reduce the potential for contact with pathogens in sewage sludge applied to the land or placed on a surface disposal site. These requirements are divided into:

requirements that are designed to control and reduce pathogens in treated sewage sludge (biosolids) and to reduce the ability of the biosolids to attract vectors (insects and other living organisms that can transport biosolids pathogens away from the land application or surface disposal site).

EPA regulations divide biosolids into two classes: A and B. Table 1-3 shows the requirements to meet Class A and B conditions.

**Table 1- 3. Class A and B pathogen requirements according to EPA standards.**

| Class A  | Class B   |
|--|---|
| Pathogens are reduced to below detectable level. Pathogenic microorganisms densities that must be met at all times:<br>Fecal coliform < 1000 MPN/g TS<br>Salmonella < 3 MPN/4g TS<br>Enteric Viruses < 1 PFU/ 4g TS<br>Viable Helminth Ova < 1/4g TS | Pathogens are reduced to levels that are unlikely to pose a threat to public health and environment under the specific use conditions. Pathogenic microorganisms densities that must be met:<br>Fecal coliform < 2,000,000 MPN/g TS |
| No site restrictions.  | Site restrictions with regard to crop harvesting, animal grazing and public access for certain period of time.  |

It should be mentioned here that EPA standards are recognized throughout the world currently there are no formal regulations on federal level as to pathogen content of sludge in Canada. Land application of sewage sludge is regulated by provincial governments, which release permits or licenses to wastewater treatment plants. These permits or licenses usually take into account many factors such as biosolids composition, season of application, site used for application, nutrient application rates, crops planted, impact on ground water, etc. (Wakelin, 2004) In terms of biosolids quality, specifically pathogen content, provincial guidelines are often based on or refer to EPA guidelines (Alberta Environment, 2001, Ontario Ministry of Environment, 2000, Province of Manitoba, 1987, Saskatchewan Environment, 2004). Therefore, it is assumed that AD technology that meets EPA standards for Class A biosolids fulfills a mandate of sufficient pathogen inactivation.

The level of pathogen inactivation, based on typical pathogen levels in unstabilized sewage sludge and limits for Class A sludge, expressed in log reduction are presented in Table 1-4 (E.P.A., 1999, Smith and Farrell, 2001).

According to EPA, sludge can be classified as a Class A product if AD process meets time-temperature regime - Figure 1-24. For most commonly occurred sewage sludges with less than 7% solids, time temperature regime is defined by equation [1-1].

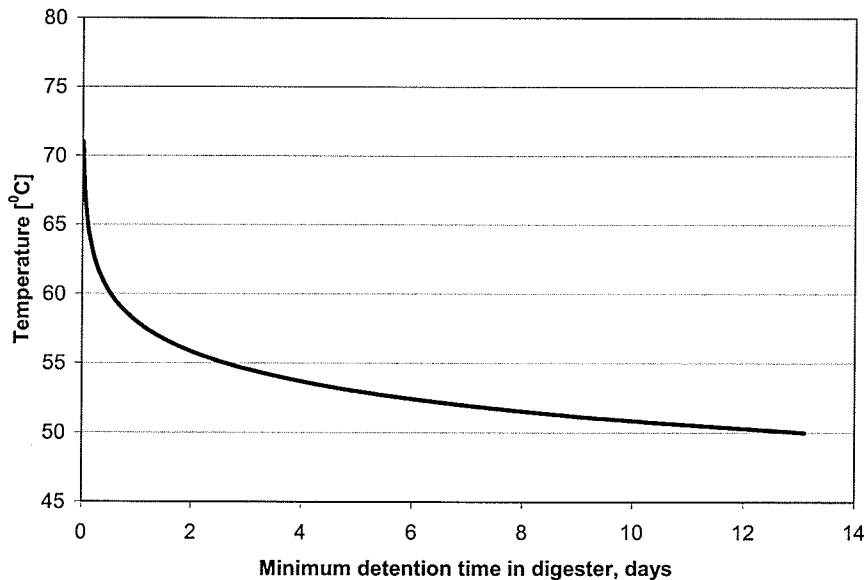
**Table 1- 4. Level of pathogen inactivation for meeting Class A biosolids standards**

| Pathogen       | Typical pathogen level in unstabilized sludge No./100 milliliters* | EPA limit for Class A biosolids | Log reduction |
|----------------|--|---------------------------------|---------------|
| Fecal coliform | 1,000,000,000  | 1000/ g TS                      | 5.4           |
| Salmonella     | 8,000  | 3 MPN/ 4 g TS                   | 3.9           |
| Virus          | 2,500 – 70,000   | 1 PFU/4 g TS                    | 3.4 – 4.85    |
| Helminth Worms | 200 – 1,000  | 1 viable ova/4 g TS             | 2.3 – 3.0     |

\* - 100 ml of sludge corresponds to 4 gram of TS (assumption: sludge is a 50:50 mixture of PS and WAS at 3% solids, 1.33 specific gravity).

**[Equation 1- 1]** 
$$D = \frac{131,000,000}{10^{0.14 \times t}}$$

where D – days, t – temperature and  $D_{\text{minimum}} = 0.0139$  (i.e. 20 minutes).



**Figure 1- 24. Time temperature relationship for meeting Class A requirement.**

These regulations imply that temperature of the treatment cannot be less than 50°C. In other words Class A biosolids quality is in practice restricted to thermophilic processes. However, many WWTP operate their digesters below thermophilic temperatures in which case the only way to comply with Class A standard is to follow Alternative 3, which is to demonstrate adequate pathogen reductions through comprehensive monitoring of bacteria, enteric viruses and viable helminth ova (E.P.A., 1999).

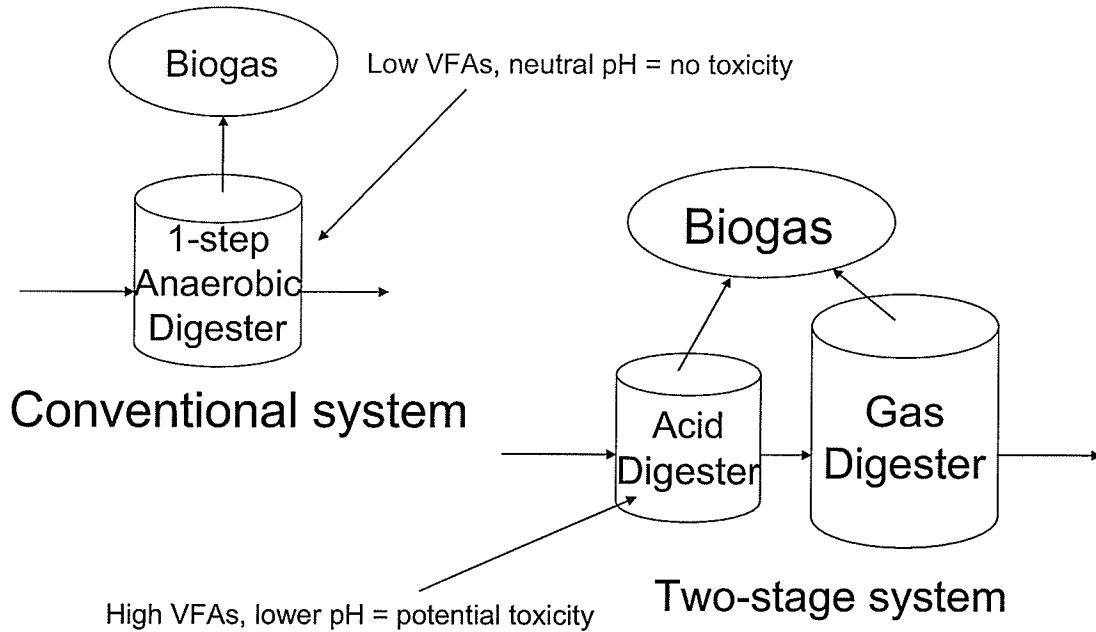
### **2.11 Organic acid toxicity as an alternative method of pathogen inactivation**

Recent advances in digestion technologies, including acid-gas digestion, opened a possibility to provide pathogen destruction without the required heat/energy input. Organic acids, intermediate products of anaerobic decomposition of wastewater sludge, accumulate in acid digesters in high concentrations of up to 13,000 mg/l (Ghosh, 2003, Wilson and Streicher, 2001). Pathogen inactivation caused by organic acids during sludge digestion have been reported since the early 1960's (McCarty and McKinney, 1961), and the latest publications show that there is growing interest in applying acid digestion (Figure 1-25) for pathogen destruction (Matthews and Asadi, 2003, Mayhew et al., 2002, Reimers et al., 1999, Salsali et al., 2004, Wilson et al., 2002).

The antibacterial properties of weak acids have been recognized for quite some time (Bergeim, 1940). Since ancient times people have been using fermentative methods to preserve foods (Russell, 1992), which, in a sense, use the same mechanism that was later discovered to cause inactivation of *E.coli* in the digestive tract of animals (Wolin, 1969).

Lipid permeable, weak acids such as acetic or benzoic acids are still frequently used in food preservation (Roe et al., 1998).

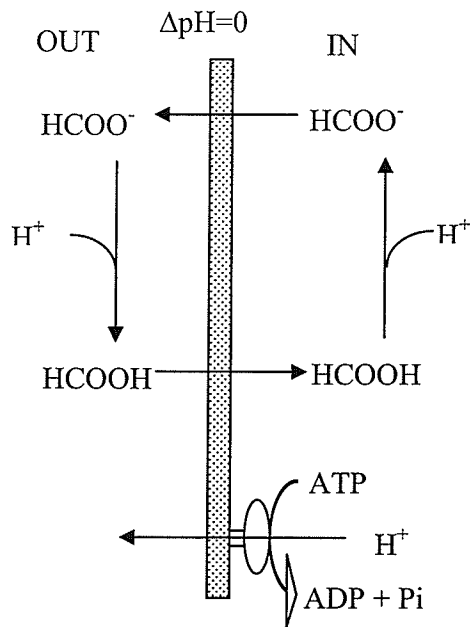




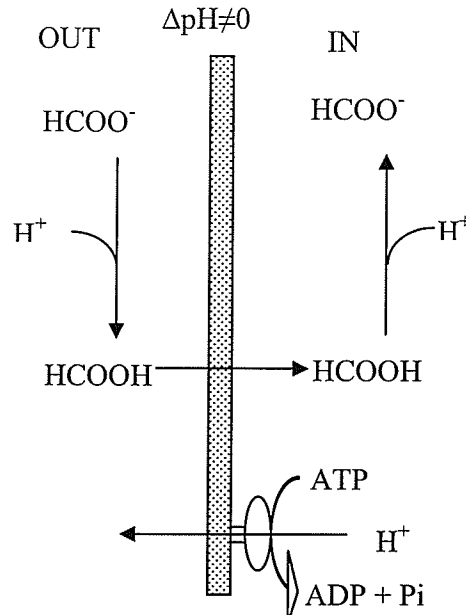
**Figure 1- 25. Main concept of toxicity from organic acids in acid digestion.**

Although the effect of antimicrobial properties of weak acids had been observed, it wasn't until the introduction of the chemiosmotic theory in the 1960s which provided the first insight into the mechanisms and the specificity of this action (Russell, 1992). It was demonstrated that organic weak acids (and bases) could dissipate the pH and electrical gradients across the cell membrane acting as uncouplers.

Uncouplers are synthetic compounds (dinitrophenol, CCCP, FCCP, TCS, SF6847), which are highly lipophilic, and can pass across the membrane in both the dissociated and undissociated form. The undissociated (protonated) species travel across the cell membrane and release a proton inside as a response to pH gradient. The now dissociated species are driven outside by the electrical gradient. On the outside they are protonated and the cycle continues until the proton motive force is dissipated as shown in Figure 1-26.



**Figure 1- 26. A schematic representation of uncouplers action and the dissipation of protonmotive force (Russell, 1992).**

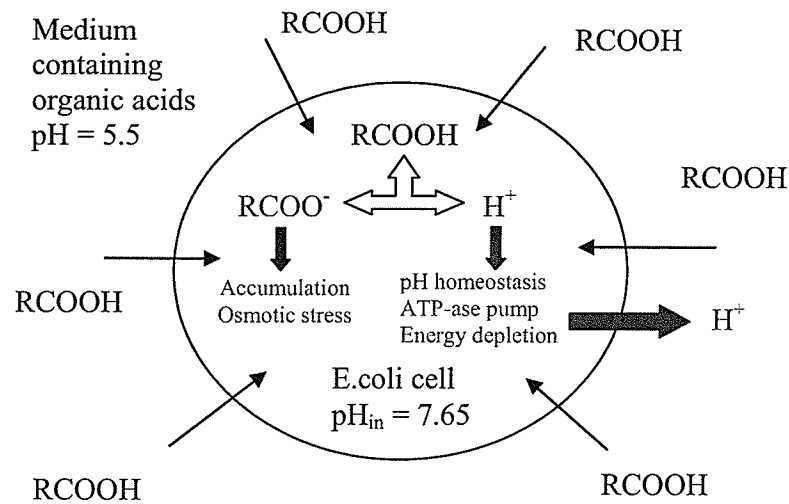


**Figure 1- 27. The flux of fermentation acids into bacteria and their dissociation in the more alkaline interior (Russell, 1992).**

Weak fatty acids act in similar fashion as uncouplers. In aqueous solutions, weak acids exist in pH-dependent equilibria between unionized acid molecules such as acetic acid and charged anions such as acetate. The proportion of undissociated acid increases as the pH declines (Lambert and Stratford, 1999). In their undissociated form organic acids are lipophilic (they are able to permeate freely across the cell membrane) but once they dissociate, their anions are lipophobic (they cannot pass across the membrane). Those lipophobic anions accumulate inside the cell when the intracellular pH is alkaline (Figure 1-27). Bacteria face two problems associated with this situation (Figure 1-28). Organic acids acidify cytoplasm and to overcome this, bacteria must employ a specific mechanism known as H<sup>+</sup>-ATPase pump. This mechanism of course consumes energy (Gauthier, 2002).

Moreover, undissociated organic acids act as protonophores and increase the inward leak of

$H^+$  so that  $H^+$  efflux is not rapid enough to alkalinize the cytoplasm (Russell and Diez-Gonzalez, 1998). Another problem is accumulation of the anionic part of the acid inside the cell. This accumulation becomes toxic to bacteria and leads to osmotic problems (Gauthier, 2002, Roe et al., 1998). The amount of diffusion of the undissociated VFA depends on the pH outside the cell and the pK value of the acid.



**Figure 1- 28. Weak acids toxicity (pH inside the cell as in *E.coli*).**

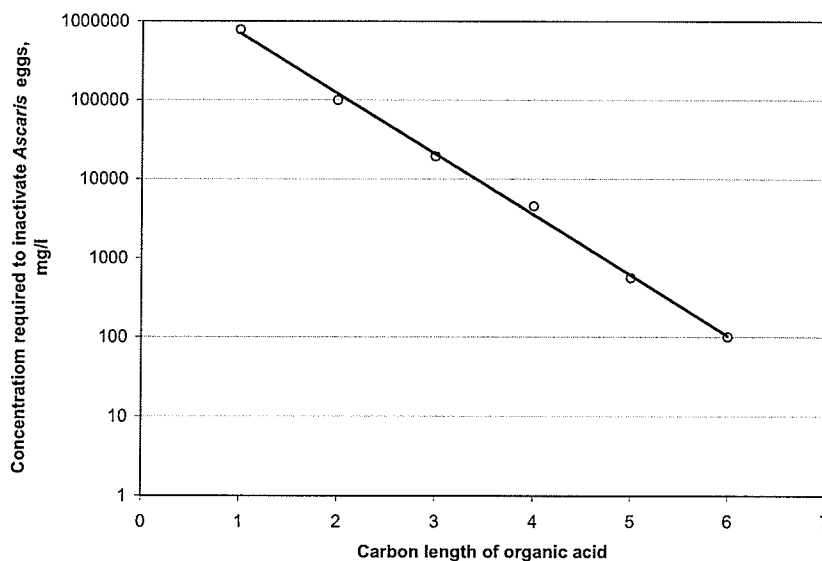
## 2.12 Factors affecting organic acids toxicity

There are several factors that have to be considered for successful pathogen inactivation through organic acids toxicity. These include concentration and carbon length of organic acids, acid solubility, sludge pH, pK value of the acids, and duration of the treatment and temperature.

### 2.12.1 Carbon length of organic acid

Increasing the strength of the wastes through sludge thickening is probably the simplest way to achieve an increase in concentration of volatile fatty acids (VFA) produced in a digester.

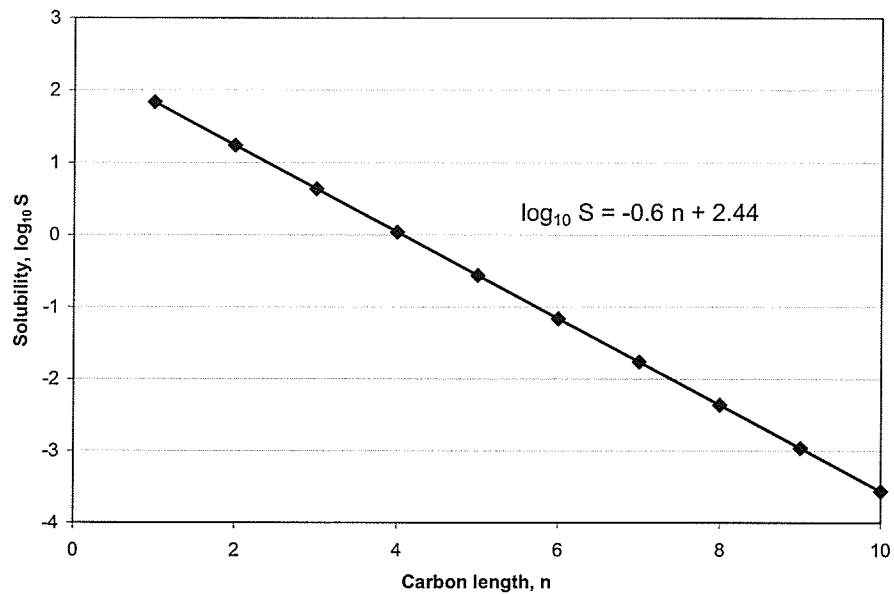
Galbraith and Miller (1973), Galbraith et al., (1971), and Morishita (1971) showed that organic acids toxicity increases with increasing carbon length (i.e., a lower concentration of caproic acid would be needed to achieve pathogen inactivation than acetic acid at the same conditions as shown in Figure 1-29). However, conditions in the digester would more likely involve synergistic effects of the mixture of organic acids on pathogen inactivation.



**Figure 1- 29. Relationship between carbon length of organic acid and the concentration required for inactivation of *Ascaris* eggs (based on experimental data from Takeyama, 1951 cited by Morishita, 1971).**

### 2.12.2 Solubility of organic acids

Solubility is another important factor that needs to be considered besides the carbon length of organic acids. Carbon length of organic acids has a direct impact on solubility of acids (Pryde, 1979) as shown in Figure 1-30. Acetic acids (C2) is more than 100 times more soluble than caproic acid (C6). It is essential for toxicity that acids are soluble in order to enter bacteria cells. Therefore, poor solubility of acids with higher carbon lengths may counteract their higher toxicity as compared to acids with lower carbon length.



**Figure 1- 30. The effect of carbon length on solubility of organic acids in un-ionized form (based on Pryde, 1979).**

### 2.12.3 pH of the environment and pK of organic acids

The pH of the sludge is another important characteristic because it affects the distribution between the un-ionized and ionized forms of organic acids. Because the equilibrium between unionized and ionized molecules of organic acid depends on pH and their pK values, it is possible to determine the proportions of the species (neutral molecule, anion, cation) that are present in the solution according to the equation [1-2] (Perrin et al., 1981). Based on equation [1-2] and dissociation values from Kortöm et al., (1961) a plot of fraction of undissociated acetic, propionic and n-butyric acid in relation to pH of the medium was prepared as shown in Figure 1-31.

[Equation 1- 2]  $\frac{\text{base}}{\text{acid}} = 10^{\text{pH}-\text{pK}_a}$

where:

base – fraction of ionized organic acid

acid – fraction of un-ionized organic acid (u-VFA)

pK – negative logarithm of acid ionization constant

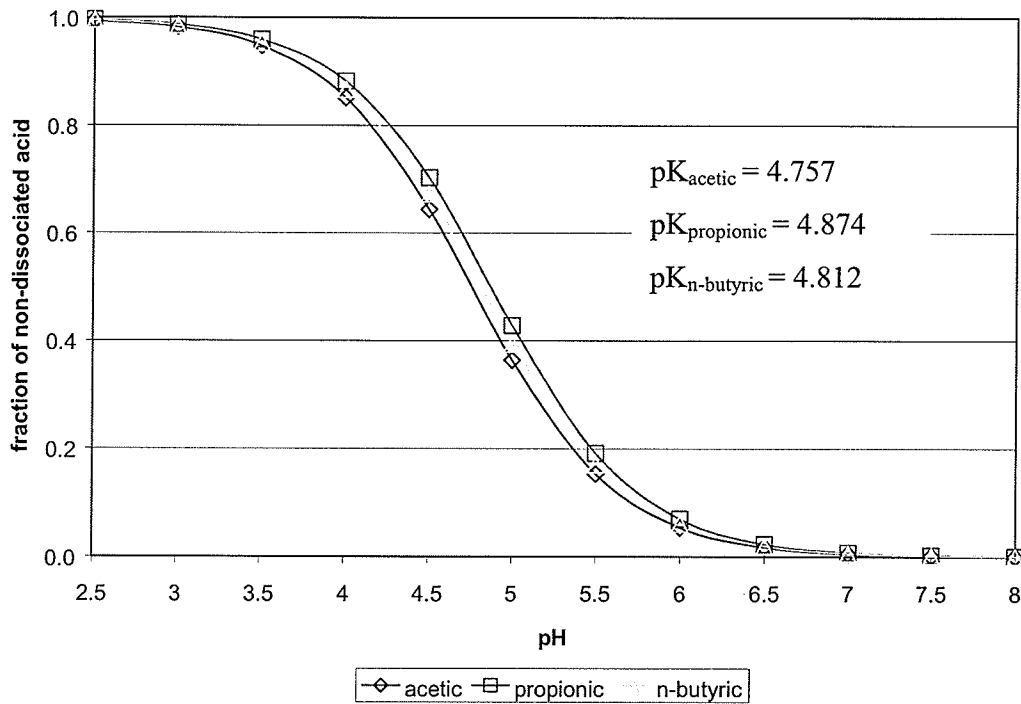


Figure 1- 31. A distribution of acetic, propionic and n-butyric acid in relation to pH of the medium.

#### 2.12.4 Temperature

Finally, temperature of the treatment is a factor to be considered. Though temperature has some certain influence on pK value of organic acid, the impact of temperature is negligible as shown in Table 1-6. Temperature may play a role, though, in the diffusion of organic acids as shown – Figure 1-32. It was found that increase in temperature resulted in increase in rate of diffusion of organic acids. An increase from 4°C to 24°C increased diffusion rate of acetic and propionic acids (through chitosan-based antimicrobial packaging) more than twofold

(Quattara et al., 2000). Diffusion coefficient itself usually follows an Arrhenius relationship (Tiwari et al., 2005) and increases with an increase in temperature as in equation [1-3].

**[Equation 1- 3]**  $D = D_0 \exp \frac{-\Delta H}{RT}$

where:

D – diffusion coefficient, and  $D_0$ ,  $\Delta H$ , and R - constants

### **3 Goals and scope of the project**

On the basis of objectives from section 1.2. referring to issues such as: thermophilic vs. mesophilic sludge treatment and its impact on energy recovery, toxicity of organic acids and its application in anaerobic digestion, and the effect of environmental factors on sludge hydrolysis, project goals were developed and divided into following areas:

#### **3.1 Assessment of anaerobic digestion improvement through thermal sludge treatment**

This part of the project aimed at the assessment of benefits of thermophilic, and thermophilic-mesophilic anaerobic digestion systems over conventional mesophilic digestion. Specifically measurements included biogas production, VS destruction, pathogen inactivation, and effluent quality such as dewaterability and concentration of volatile fatty acids (VFAs) which contribute to odour problems.

#### **3.2 Establishment of hydrolysis kinetics for thermophilic acid digester**

The first-order rate reaction model was used to establish kinetics of wastewater sludge under thermophilic conditions. Reaction kinetics under thermophilic and mesophilic conditions previously published data by Eastman and Ferguson (1981) were compared.

#### **3.3 Optimization of retention time in acid digester**

Acidification index was used as an optimization parameter of retention time in the first digester of a two-stage system. Acidification index, defined as ratio between acidification rate and hydrolysis rate, required calculation of acidification rate. Acidification model included measurements of acidifying biomass.



### **3.4 Evaluation of various temperature and solids concentration on kinetics of anaerobic digestion**

Hydrolysis constants were established for three different digestion temperatures: thermophilic at 55°C, mesophilic at 38°C, and low-mesophilic at 24°C; and three different solids concentration in feed sludge: unadjusted concentration, concentration of solids adjusted to 6% TS, and to 8% TS.

### **3.5 Evaluation of hydrogen (H<sub>2</sub>) generation and recovery from acid anaerobic digestion**

The quantity and quality of hydrogen in biogas from acid digestion was evaluated. Three operating conditions were tested: thermophilic at 55°C, mesophilic at 38°C, and low-mesophilic at 24°C. Four different solids concentration in feed sludge were used: 3.7% TS, 6.0% TS, 8.1% TS, and 9.8% TS.

### **3.6 Evaluation of potential of pathogen inactivation in batch non-thermophilic acid digesters**

This part of the research evaluated the potential of non-thermophilic acid digesters on inactivation of fecal coliforms. Two temperatures were evaluated: mesophilic at 38°C and low-mesophilic at 24°C.

### **3.7 Evaluation of potential of pathogen inactivation in semi-continuously fed non-thermophilic acid digesters**

Continuation of previous research (section 3.6), this part evaluated the potential of semi-continuously fed non-thermophilic acid digesters on inactivation of fecal coliforms. This type

of digester operation is closer to practical situation at the WWTP which in most cases are operated on fully continuous basis.

### **3.8 Comparison of conventional mesophilic digestion with proposed new systems that offer potential for increased energy recovery and Class A product**

During this part of the research three systems were compared. System (1) a conventional mesophilic digester, system (2) staged mesophilic acid-gas digestion at increased solids concentration, and system (3) staged low-mesophilic acid digestion followed by mesophilic gas digestion at increased solids concentration. It was anticipated that thickened sludge would result in higher VFA concentration that should produce higher toxicity compared to non-thickened sludge. Temperature below mesophilic together with thickened sludge was tested to achieve additional energy recovery.

Following this section, each chapter is composed of published papers or part of papers (edited to fit thesis format) with an opening section that explains how the work presented relates to the goals of the project. There is no formal “Materials and Methods” section because each chapter contains appropriate sections.

## 4 Thermal treatment of sludge

This section is composed of two journal publications supplemented with extracts from a conference publication. Key ideas presented in this section refer to the following issues:

- The impact of thermophilic digestion and temperature staged digestion on pathogen inactivation compared to conventional MAD. This part is based on journal publication: Puchajda, B., Oleszkiewicz, J. A. and Bowman, D. D. (2004) Pathogen inactivation in single and two-phase anaerobic digestion. *Journal of Residual Science and Technology*, 1, 183-189.
- The comparison between single thermophilic digestion, temperature staged digestion and conventional MAD on VS destruction, biogas production and composition and capillary suction time. This part is based on extracts from conference publication: Puchajda, B., Oleszkiewicz, J. A., Fogarthy, J. and Bowman, D. D. (2003) Anaerobic Digestion of Municipal Sludge: Single- and Two-Phase System: Hydrolysis, Acidification and Pathogen Destruction, Water Environment Federation, WEFTEC 2003, Los Angeles, CA.
- Optimization of the retention time in the first, thermophilic reactor of the temperature staged digestion through establishment of process kinetic parameters such as hydrolysis and acidification rate. This section is based on journal publication: Puchajda, B. and Oleszkiewicz, J. A. (2005) Single and Two-stage anaerobic digestion: hydrolysis, acidification and sludge stabilization. *Journal of Environmental Engineering and Science*, accepted for publication.

Raw data for all results presented in section 4.0 can be found in Appendix A.

## 4.1 Pathogen inactivation

### PATHOGEN INACTIVATION IN SINGLE AND TWO-PHASE ANAEROBIC DIGESTION

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#### ABSTRACT

The goal of this study was to examine the ability of the two-phase anaerobic digestion system (thermophilic-mesophilic) to reduce pathogen densities in sewage sludge. Comparisons with single-stage digesters operated at thermophilic and mesophilic temperatures were made.

Pathogens tested included fecal coliforms, *Ascaris suum* eggs, and *Clostridium perfringens*.

The single thermophilic reactor and two-phase system achieved Class A biosolids standards with respect to fecal coliforms and *Ascaris* eggs destruction at all SRTs tested. The single mesophilic reactor often failed to produce Class B biosolids when fecal coliforms were used as an indicator organism. Very low destruction of *Ascaris* eggs was observed in mesophilic conditions. None of the systems achieved 1-log reduction of *Clostridium perfringens*. Based on results from this experiment, the minimum (batch) time to achieve Class A biosolids standards was 1 day at 55°C.

#### INTRODUCTION & OBJECTIVES

Pathogens are ubiquitous in sewage sludge and their destruction is one of the main goals during sewage sludge treatment. That issue becomes especially important if sludge is to be land applied. In response to public health concerns the use and disposal of sewage sludge was regulated under EPA 40 CFR Part 503 standards (E.P.A., 1999). Class A sludge is achieved when levels of pathogen are essentially reduced below detection limit, while Class B sludge

may still contain limited levels of pathogens. Many wastewater plants face problems with land applications of Class B sludge and meeting Class A standards is usually cost-prohibitive. The most attractive method for many plants utilizing anaerobic digestion is to apply thermophilic temperatures or switch to two-phase digestion with one stage in the thermophilic range. Even though thermophilic anaerobic digestion is not yet recognized as a process to further reduce pathogens there is a number of studies (Gabb et al., 2001, Han et al., 1997, Huyard et al., 2000, Huyard et al., 1999, Lee et al., 1989) and full scale data (Holbrook and Henderson, 2001, Shao et al., 2002) indicating that these processes are capable of achieving a Class A product.

There are several factors that may directly or indirectly affect pathogen destruction. The combined effect of temperature and duration of the process are the key factors. According to (E.P.A., 1999) once all sludge particles meet the time-temperature requirement regime, the sludge is considered to be Class A. It is therefore important to avoid or minimize the effect of short-circuiting in the digester. One way to accomplish this is to use two or more vessels in series. The City of Vancouver (Annacis Island plant) managed to eliminate pathogen short-circuiting using 4 thermophilic vessels staged in series (Schafer and Farrell, 2000). Even two-phase digestion operated entirely in mesophilic conditions yielded lower pathogen densities than a single mesophilic digestion (Lee et al., 1989, Matthews and Asaadi, 2003). Higher pathogen destruction in the two-phase system was attributed to harsher conditions caused by higher organic acid concentrations in the acid-phase digester than in the conventional digesters or the methane phase digester. The sequence of feeding and drawing sludge is also of great importance. Draw and fill operated digester had over one log higher pathogen reduction than fill and draw operated digester Farrel (1988). Another factor that

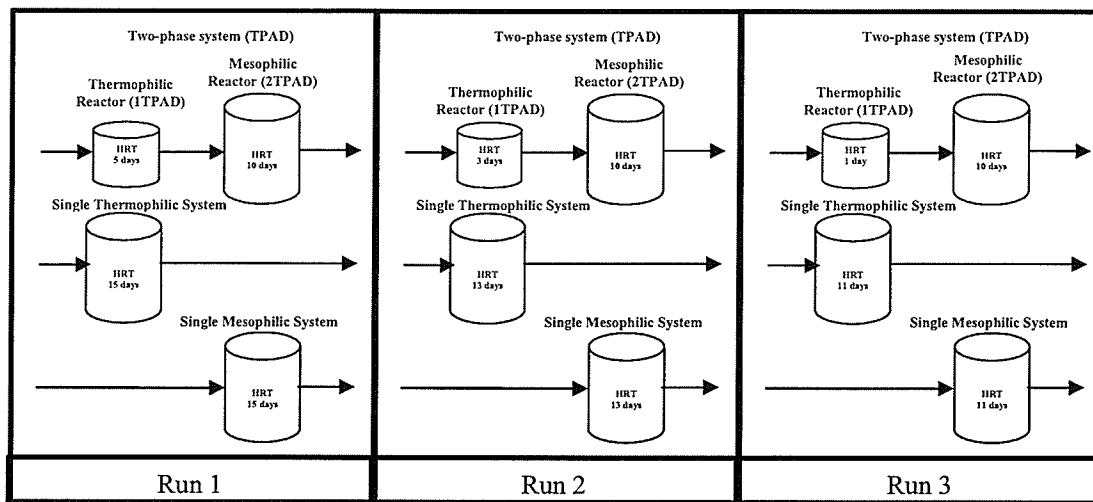
influences pathogen reduction is the frequency of feeding. Lower daily feeding frequency brings the digester closer to the batch reactor mode and thus enhances pathogen reduction capability of the system (Huyard et al., 2000).

The goal of this study was to examine the ability of the two-phase system (thermophilic-mesophilic) to reduce pathogen densities. Comparisons with single-stage digesters operated at thermophilic and mesophilic temperatures were made. Pathogens tested included fecal coliforms, *Ascaris suum* eggs, and *Clostridium perfringens*.

## METHODOLOGY

### Experimental Approach

Three lab scale anaerobic digestion systems were assembled. The main system – two-phase anaerobic digestion (TPAD) – consisted of two reactors: a thermophilic-acid reactor (1TPAD) followed by a mesophilic-gas reactor (2TPAD). Two other systems included a single phase thermophilic digester (Single Thermo) and a single phase mesophilic digester (Single Meso). The thermophilic temperature was set at 55°C and the mesophilic temperature was set at 36°C. In Run 1 of the experiment, SRT was 15 days for the Single Thermo and Single Meso systems, and 5 + 10 days for 1TPAD and 2TPAD, respectively. Run 2 of the experiment consisted of the SRT of 13 days or 3 + 10 days for the TPAD and in Run 3 SRT was set at 11 days and 1 + 10 days in case of the TPAD system (Figure 4-1). Each run was followed by an acclimation period equal to three SRTs, and each testing period lasted for a minimum of three SRTs. Each reactor contained 2 litres of biomass, with the exception of Run 3 of the experiment where the acid reactor (1TPAD) of the two-phase system contained 1 litre of biomass.



**Figure 4 - 1. Reactors' configuration during three runs of the experiment.**

#### Sludge Source and Reactor Operation

Reactors were fed with municipal sludge obtained from the North End Water Pollution Control Center (NEWPCC) in Winnipeg, Manitoba. NEWPCC collects the sludge from two other wastewater treatment plants located in the South End and the West End in Winnipeg. The vast majority of the sludge however comes from the North End Plant which co-thickens primary sludge (PS) with waste activated sludge (WAS) in a primary clarifier. All wastewater treatment plants in Winnipeg are operated to remove only BOD without nitrification or phosphorous removal. Sludge was delivered to the laboratory once a week and kept in refrigerator at 4°C. Prior to feeding, the sludge was sieved through a No.5 sieve (opening size 4 mm). Digesters were emptied and fed manually once a day. Single phase digesters were intermittently mixed (2 – 3 times a day for couple of minutes) in Run 1 and 2 but in Run 3 continuous mixing was maintained to prevent foaming. The thermophilic reactor of the two-phase system was mixed continuously in all runs of the experiment and the mesophilic reactor of the two-phase system was mixed intermittently due to very little foaming in all runs. The pH and alkalinity of the digesters were not controlled.

## Experimental Procedures

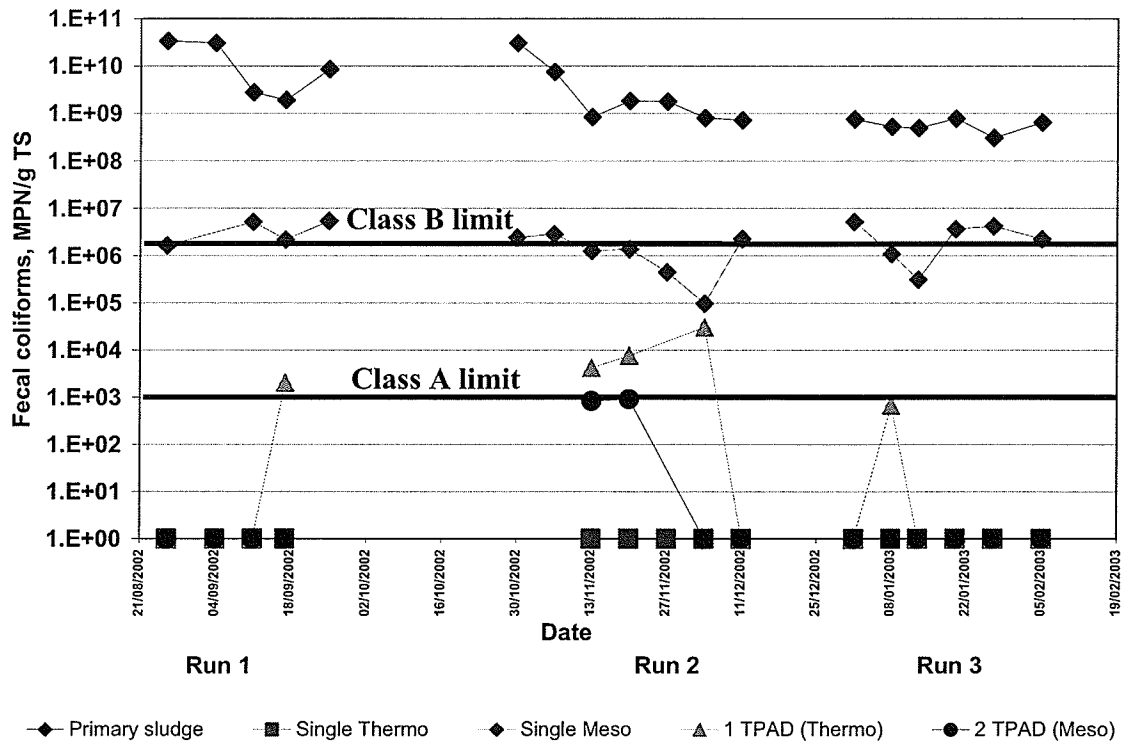
Total solids (TS) were measured twice a week according to SM 2540 B (APHA et al., 1998). Total coliforms were measured once per week according to SM 9221 B (APHA et al., 1998) and then positive samples were tested for fecal coliform presence according to SM 9221 E (APHA et al., 1998). The three-tube method was used and most probable number (MPN) was obtained according to Finstein (1972). *Clostridium perfringens* spores were measured once per week according to the method developed by Bujoczek (2001). *Ascaris* eggs were cultured and counted according to Bowman et al. (2002). Coliform organisms and *Clostridia* were present in the feed sludge, however *Ascaris* eggs were not detected or their number was so low that in order to assess the efficiency of digestion on their inactivation it was necessary to spike digesters with swine *Ascaris suum* prior to testing. In addition to spiking sludge directly with the free floating eggs, helminth ova inactivation was evaluated using sentinel chambers placed inside the digester. Sentinel chambers provided a free exchange of liquid through a semi-permeable membrane on the sides of the sentinels, as eggs were contained inside without possibility of escape. Sentinels were prepared in Ithaca, N.Y. and the procedure consisted of mixing *Ascaris suum* eggs with the same sludge that was fed into digesters, placing this mixture inside sentinels and shipping on dry ice. Free floating eggs viability was analyzed at the University of Manitoba, and viability tests of those enclosed in sentinels were performed at Cornell University. Percent viability corresponds to average value of two or three samples containing 100 eggs each.

## RESULTS & DISCUSSION

As provided by the US.EPA guidelines under 40 CFR Part 503 (E.P.A., 1999) biosolids can be classified as Class A if during process of their stabilization time-temperature requirements



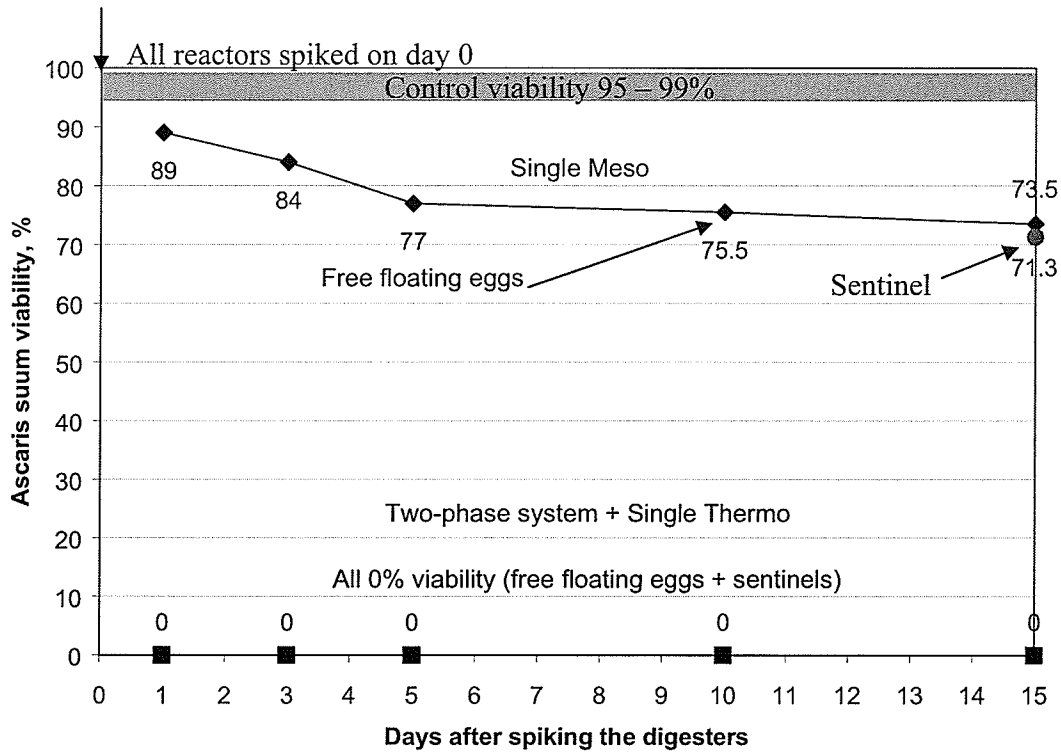
were met or if pathogen densities are reduced to a certain level. For instance, fecal coliforms must be reduced to less than 1 000 MPN per gram total solids (dry weight basis). There must be less than 1 viable helminth ova per 4 grams total solids (dry weight basis). Enteric viruses and *Salmonella sp.* are also regulated but they were not measured during this experiment. However, meeting only one requirement is enough to prove compliance with Class A biosolids. Additionally, densities of *Clostridium perfringens* were monitored. Though this organism is not regulated under 40 CFR Part 503 (E.P.A., 1999) it has been proven to be an emerging indicator organism for the assessment of microbial removal in wastewater and sludge treatment (Hirata et al., 1991). This is a thermophilic organism thus it is resistant to temperature. *Clostridium* species reside in the soil and in the intestinal tracks of animals and man. They include both pathogenic and non-pathogenic species. Specifically, *Clostridium perfringens* species cause wound infections, gangrenous mastitis and enterotoxemias. Thermophilic digestion as well as two-phase digestion met Class A pathogen requirement with respect to fecal coliforms densities as shown in Figure 4-2. Very often complete destruction of fecal coliforms was achieved during the first-stage of the two-phase system with the exception of three instances during Run 2 and one instance during Run 1. The single mesophilic reactor operated at 36°C performed poorly with respect to fecal coliform destruction and many times did not even achieve Class B requirement. Interestingly, mesophilic digesters in NEWPCC in Winnipeg, operated at 38°C and an SRT of approximately 20 days fulfill the requirements of Class B in terms of achieving coliform densities below  $2 \times 10^6$  (Smyrski, 2003).



**Figure 4 - 2. Fecal coliform densities during three consecutive experimental runs.**

Procedure of spiking digesters with free floating eggs included single injection of the suspension containing approximately 200 000 *Ascaris suum* eggs in 10 ml. Eggs were retrieved later while digesters were emptied and incubated for 28 days. In addition, sentinel chambers were placed inside digesters and were taken out after 1, 3, 5, 10 and 15 days of digestion. These procedures allowed tracing the fate of *Ascaris suum* eggs as if in batch system even though the digesters were operated in continuous mode.

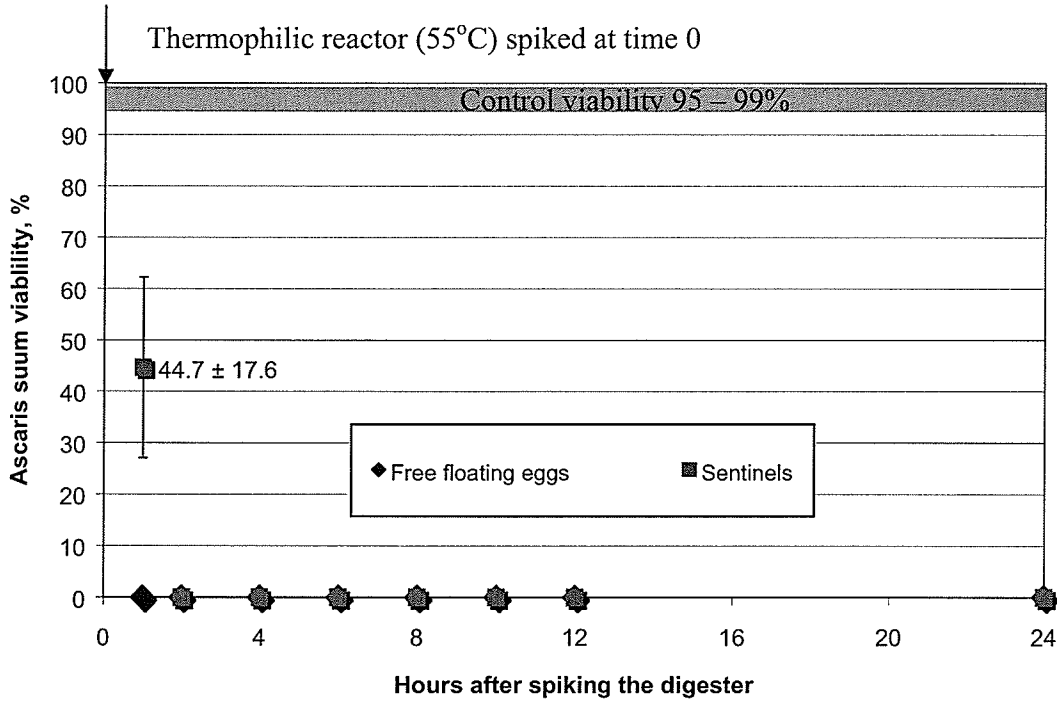
Complete inactivation of *Ascaris* eggs, free floating and enclosed in sentinels, was achieved in the single thermophilic reactor as well as in the two-phase system as shown in Figure 4-3.



**Figure 4 - 3. *Ascaris suum* viability during Run 1 – SRT ~ 15 days, free floating eggs and sentinels.**

Undoubtedly, thermophilic conditions in the first stage of the two-phase system were responsible for inactivation of *Ascaris* eggs. It is assumed that 0% viability would correspond to Class A requirement as in study done by Gabb et al., (2001). Therefore, the two-phase system as well as the single thermophilic digester would comply with Class A biosolids requirements. In the single mesophilic digester very poor destruction of *Ascaris* eggs was observed, and the final viability after 15-day treatment resulted in 73.5% free floating eggs viability compared to 71.3% viability of eggs enclosed in sentinels. To resolve how quickly *Ascaris* eggs were destroyed during thermophilic conditions a 24-hour batch test was performed. After 2 hours at 55°C all *Ascaris* eggs, free floating and enclosed in sentinels, were dead (Figure 4-4) which corresponds exactly with findings of Paulsrud et al., (2003). The differences in viability after 1 hour between the free floating eggs and those contained in

sentinels was attributed to limitations in heat transfer into the sentinels which were kept refrigerated before the test.



**Figure 4 - 4. 24-hours *Ascaris suum* viability test – thermophilic conditions 55°C, free floating eggs and sentinels.**

Very low reductions of *Clostridium perfringens* densities were observed in all treatments as shown in Figure 4-5. None of the treatments achieved 1-log reduction. In the case of the single mesophilic digester, densities of *Clostridium perfringens* spores after treatment were often higher than in primary sludge. Densities in the mesophilic digester of the two-phase system were also often higher than in thermophilic digester. This could suggest that mesophilic treatment provided good conditions for survival and proliferation of *Clostridium sp.* causing the apparent regrowth. The survival of *Clostridium perfringens* suggests that the microorganism can only be used to indicate inactivation by means other than temperature alone.

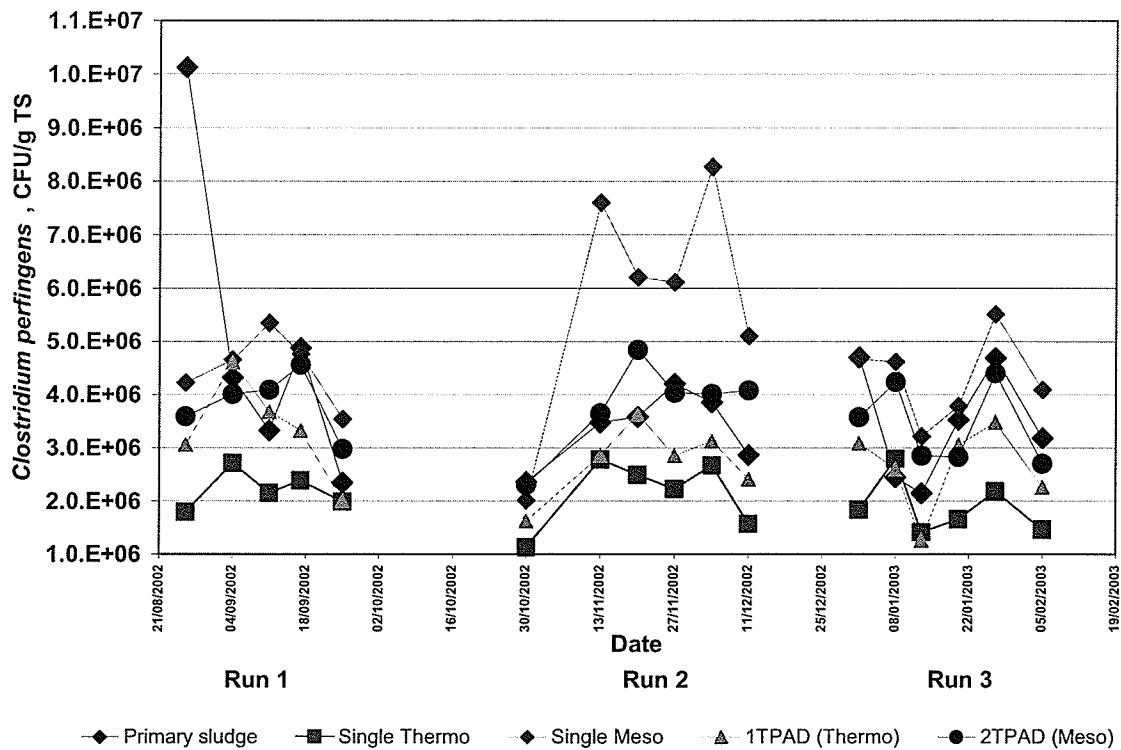


Figure 4 - 5. *Clostridium perfringens* densities during three consecutive experimental runs.

## CONCLUSIONS

The following conclusions were drawn based on the results of this experiment:

- The single thermophilic reactor and two-phase system achieved Class A biosolids standards with respect to fecal coliforms and *Ascaris* eggs destruction at all SRTs tested that included 15, 13 and 11 days for single-phase systems and 5+10, 3+10 and 1+10 for two-phase systems, acid and gas reactor, respectively.
- A 24-hour batch test for *Ascaris* viability at 55°C showed inactivation of free floating *Ascaris* eggs after 1 hour of treatment, and eggs enclosed in sentinels were inactivated within 2 hours.

- The single mesophilic reactor often failed to produce Class B biosolids using fecal coliforms as an indicator organism. Very low inactivation of *Ascaris* eggs was observed in that reactor.
- None of the systems achieved 1-log reduction of *Clostridium perfringens*. Mesophilic conditions led to the apparent regrowth of *Clostridium perfringens*.
- Based on results from this experiment, the minimum time to achieve Class A biosolids standards was 1 day at 55°C operated in batch mode. If the sludge handling facility was to be upgraded in order to achieve Class A biosolids, a two-phase anaerobic digestion with the first digester operated at thermophilic temperatures in a batch mode and retention time of 24 hours offers an effective solution with less energy required than the single-thermophilic digester.

#### ACKNOWLEDGEMENTS

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## 4.2 Biogas production and VS destruction

This chapter supplements information presented in section 4.1. It refers to the same experimental work but for the purpose of this document only excerpts of the paper that refer to biogas production and VS destruction are presented.

### OBJECTIVES

The objectives of this part of the research was to provide comparison between three different options of wastewater sludge digestion and their impact on VS destruction, biogas production, biogas quality and dewaterability of the final effluents by measurements of capillary suction time (CST).

### METHODOLOGY

For details see section 4.1. Description of additional experimental procedures follows.

A volume of biogas was collected and measured daily using liquid displacement method in calibrated air-tight vessels. The liquid was saturated sodium chloride with 5% sulphuric acid and methyl orange to prevent gas from dissolving. Biogas composition – methane ( $\text{CH}_4$ ) and carbon dioxide ( $\text{CO}_2$ ) were measured twice a week using GOW MAC Model 550 gas chromatograph with helium as a gas carrier and Porapak Q column, 80/100, ¼ inch. Hydrogen ( $\text{H}_2$ ) was measured twice a week (only for the first reactor of the two-phase system) using the same gas chromatograph with nitrogen as a gas carrier and Molsieve 5X column, 60/80 Mesh, 1/8 inch. Total (TS) and volatile solids (VS) were measured twice a week according to SM 2540 B and SM 2540 E (APHA et al. 1998). Triton Electronics 304B Capillary Suction Timer was used to measure capillary suction time (CST) of sludge, twice per week to determine dewaterability characteristics of final effluents.

A computer program JMP Start Statistics (Sall et al., 2001) was used for statistical calculations. Statistical analysis included comparing many means using one-way analysis of variance. Specifically pairs of means were compared using Student's t test. To avoid committing Type I error (error of declaring significant difference that is actually zero) results were further confirmed by the Tukey-Kramer Honestly Significant Difference (HSD) test.

## RESULTS AND DISCUSSION

### VS destruction

The US. EPA (E.P.A., 1999) defines the vector attraction reduction (VAR) as a way to determine the degree of stabilization of sludge. The sludge can be described as "stabilized" if the mass of volatile solids (VS) in sewage sludge is reduced by at least 38%. It must be noted that VAR (or VS mass reduction) does not demonstrate the achievement of Class A biosolids, and VAR must be met after or concurrent with pathogen reduction to prevent growth of pathogenic bacteria. The results of VS destruction for all reactors are presented in Table 4-1.

**Table 4 - 1. VS destruction achieved by one stage and two-stage anaerobic digestion.**

| Reactor                   |       | 1 TPAD<br>Thermo         | 2 TPAD<br>Meso | 1+2 TPAD*   | Single MAD  | Single TAD  |
|---------------------------|-------|--------------------------|----------------|-------------|-------------|-------------|
| VS <sub>destruction</sub> |       | Mean ± Std. Deviation, % |                |             |             |             |
|                           | Run 1 | 22.1 ± 14.9              | 31.4 ± 18.9    | 44.9 ± 12.3 | 46.6 ± 11.6 | 41.7 ± 13.6 |
|                           | Run 2 | 16.1 ± 8.8               | 45.5 ± 5.8     | 53.6 ± 3.9  | 53.1 ± 4.0  | 51.5 ± 4.2  |
|                           | Run 3 | 14.5 ± 5.2               | 47.7 ± 11.3    | 51.8 ± 10.2 | 52.2 ± 9.0  | 48.9 ± 8.3  |

\* VS destruction of the two-phase system was calculated for each phase separately, i.e. for the first stage, VS destruction corresponds to the destruction of VS coming from PS, and VS destruction of the second stage corresponds to the additional destruction of VS coming from the first phase reactor 1TPAD. Note: the sum of VS destruction of each phase does not equal the overall VS destruction of the system.

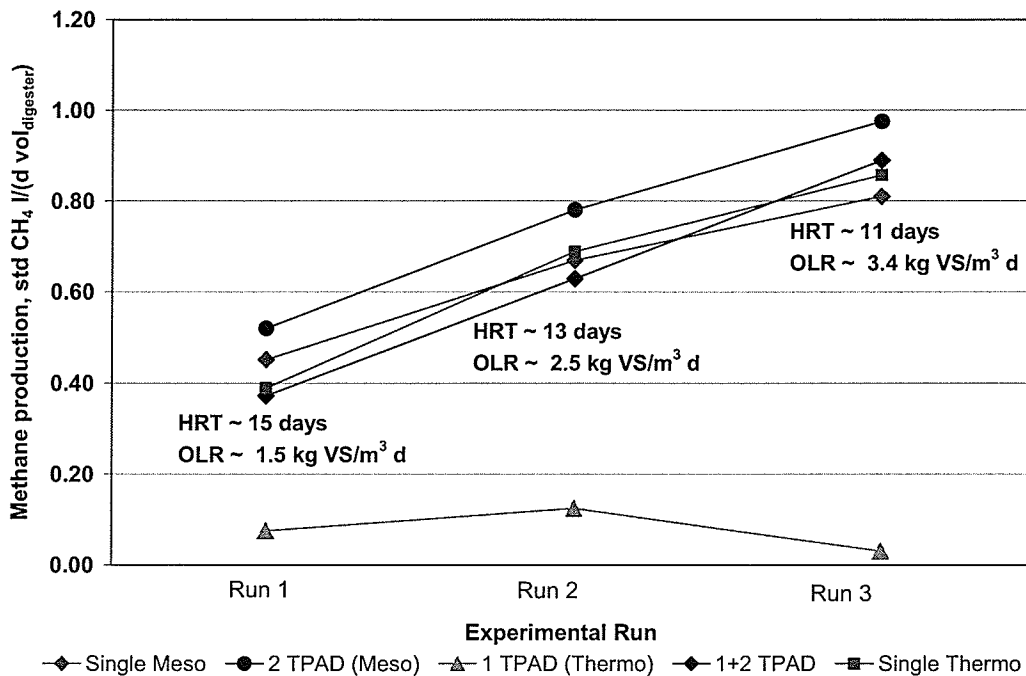
The two-phase system as well as the single stage reactors met the minimum 38% VS destruction requirement for all phases of the experiment. The difference in VS destruction was not statistically different at each run as compared by the Tukey-Kramer HSD test (alpha



= 0.05). Therefore, no improvement of VS destruction by the two-phase system compared to the single stage systems was observed. Furthermore, no significant difference in VS destruction was observed between single digesters.

#### Biogas production

The methane gas production efficiency of all systems was compared. The comparison included overall methane production per day per volume of digestion system and methane production per gram VS added per day as presented in Figures 4-6 and 4-7.



**Figure 4 - 6. Methane production from two-phase system and single phase digesters per digester volume per day.**

As expected the overall methane production increased with an increase in loading rate, but no considerable changes in methane production per gram VS added were observed (Figure 4-7).

Also, no significant difference in biogas production between systems was observed.

However, out of all reactors tested, the second reactor of the two-phase system achieved the

highest biogas production per digester volume for all phases of the experiment (confirmed by statistical analysis). It outperformed other reactors by roughly 15 to 20%. The overall efficiency of the methane production in the two-phase system per system volume was offset by very low biogas production per digester volume in the first reactor of the two-phase system. These facts have to be considered while designing and operating a two-phase anaerobic digestion system. Longer retention times in the hydrolytic-acidogenic reactor resulted in higher contribution of this reactor to the overall methane production of the system – see Figure 4-8. The optimization of the two-phase system should aim at shortening of the SRT in the first reactor while promoting longer retention time in the methane producing reactor.

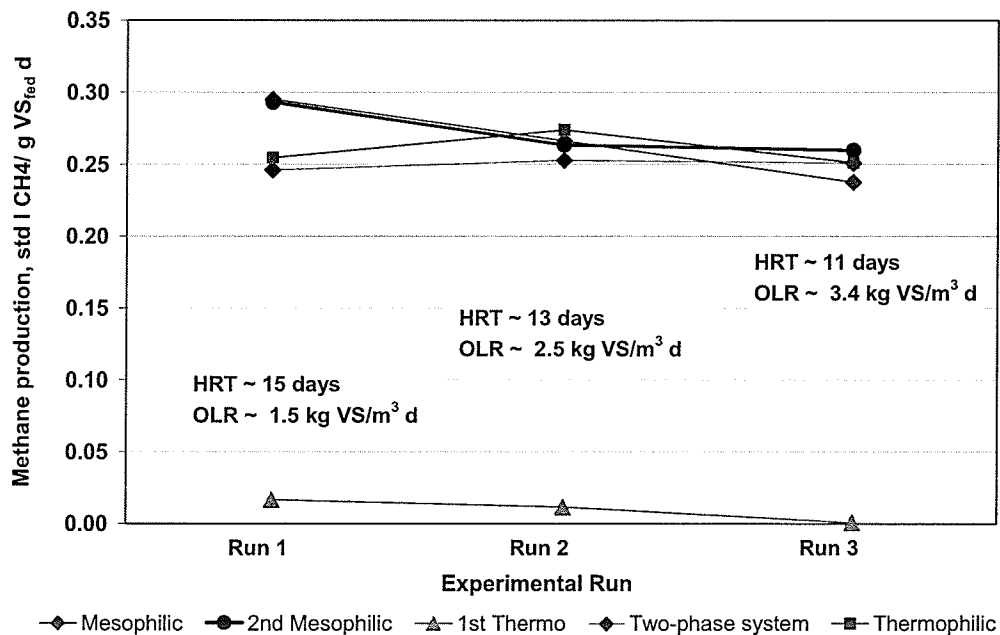
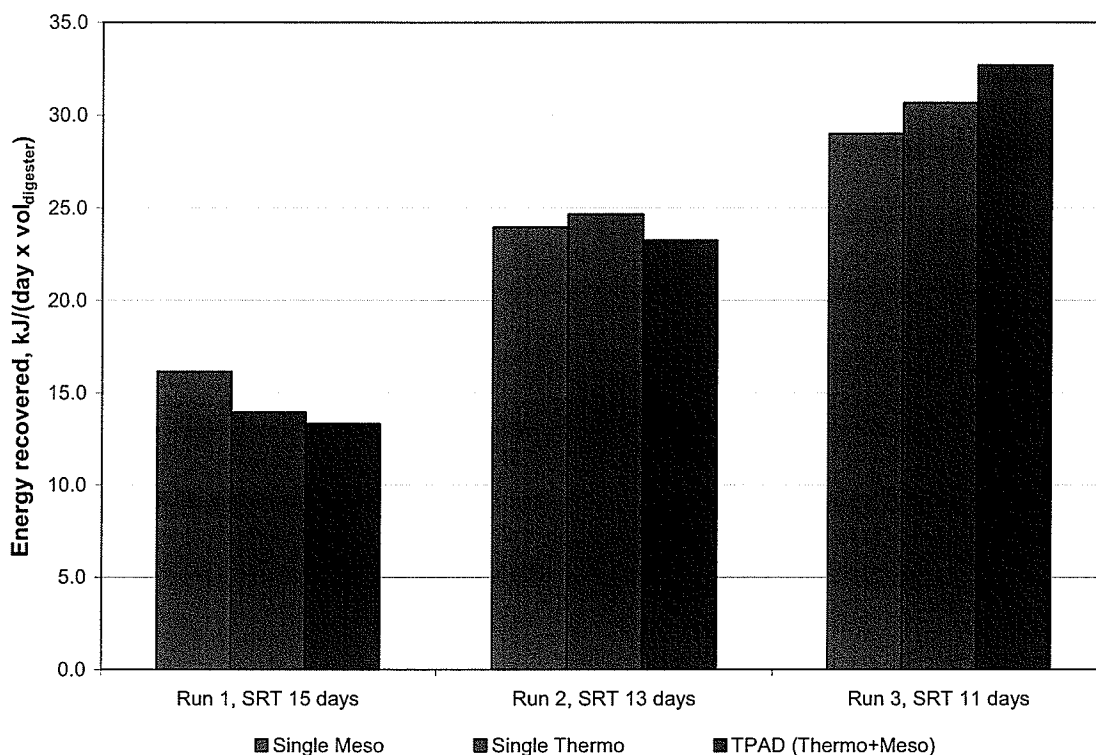


Figure 4 - 7. Methane production from two-phase system and single-phase digesters per gram VS fed per day.



**Figure 4 - 8. Energy recovery from single mesophilic, single thermophilic and two-stage system at each operating SRT.**

An analysis of biogas for methane and hydrogen content is presented in Table 4-2. These two components, if recovered, can be utilized as an energy source. For instance, methane gas is commonly used to heat digesters to mesophilic or thermophilic temperature. Although, economical feasibility of hydrogen production from organic wastes has not yet been established (Noike and Mizuno, 2000) it could be an excellent source of energy for fuel cells.

**Table 4 - 2. Composition of biogas from two-phase system and single phase digesters.**

| Reactor |                 | 1TPAD (Termo)           | 2 TPAD (Meso)  | Single Meso    | Single Thermo  |
|---------|-----------------|-------------------------|----------------|----------------|----------------|
|         |                 | Mean $\pm$ Std. Dev., % |                |                |                |
| Run 1   | CH <sub>4</sub> | 42.8 $\pm$ 2.4          | 64.8 $\pm$ 2.4 | 63.8 $\pm$ 2.3 | 64.5 $\pm$ 2.2 |
| Run 2   | CH <sub>4</sub> | 39.6 $\pm$ 3.2          | 64.5 $\pm$ 2.4 | 64.8 $\pm$ 2.5 | 65.4 $\pm$ 2.1 |
| Run 3   | CH <sub>4</sub> | 7.1 $\pm$ 8.5           | 64.3 $\pm$ 2.3 | 62.5 $\pm$ 2.1 | 64.4 $\pm$ 2.5 |
|         | H <sub>2</sub>  | 12.2 $\pm$ 8.7          | -              | -              | -              |

The single phase digesters and the second digester of the two-phase system consistently produced biogas containing more than 60% of methane. However, methane content in biogas of the 1st reactor of the two-phase system was much lower, approximately 40%. During Run 3 of the experiment, when retention time in 1TPAD was 1 day, methane generation dropped to less than 10 % and at the same time hydrogen recovery occurred.

#### DEWATERABILITY

NEWPCC in Winnipeg uses centrifuges to dewater digested sludge and sludge cake is later land applied. Hence, dewaterability of sludge after anaerobic digestion is an important parameter that affects operation of centrifuges and determines the amount of biosolids that have to be disposed. Improvement in dewaterability could result in considerable savings on polymer and hauling costs. A correlation between CST and dewaterability has been reported by Huisman and van Kesteren (1998), Unno et al. (1983), and Vesilind (1988). Results from CST test are presented in Table 4-3.

**Table 4 - 3. CST after two-phase and single-phase anaerobic digestion.**

|                           | PS       | 1 TPAD<br>(Thermo) | 2 TPAD<br>(Meso) | Single<br>TAD | Single<br>MAD |
|---------------------------|----------|--------------------|------------------|---------------|---------------|
| Mean CST ± Std. Dev., sec |          |                    |                  |               |               |
| Run 1                     | 287 ± 71 | 794 ± 116          | 480 ± 99         | 527 ± 50      | 430 ± 81      |
| Run 2                     | 392 ± 61 | 1128 ± 164         | 506 ± 28         | 693 ± 41      | 461 ± 45      |
| Run 3                     | 450 ± 66 | 1224 ± 199         | 656 ± 55         | 910 ± 40      | 876 ± 79      |

CST values increased with a decrease in SRT for all systems. It should be noted, however, that the average CST of the PS also increased and was probably caused by seasonal changes in PS quality. Very high values of CST measured for 1TPAD were subsequently reduced by 2TPAD. Statistical analysis showed no significant difference between CST values for the Run 1. During the Run 2 only the results from single thermophilic reactor differed

significantly from the other reactors. During the Run 3 no significant difference between the CST of single digesters was observed and 2TPAD reactor produced best results. The two-stage anaerobic digestion system was the least sensitive to changes in loading rates and retention times with respect to CST.

### 4.3 Hydrolysis and fermentation

#### **THERMOPHILIC ANAEROBIC ACID-DIGESTION OF BIOSOLIDS: HYDROLYSIS, FERMENTATION AND OPTIMIZATION OF RETENTION TIME OF ACID-DIGESTION**

B. Puchajda and J. Oleszkiewicz

#### ABSTRACT

The objective of the study was the optimization of the retention time in a thermophilic acid digestion system. To achieve this optimization, fermentation index defined as the ratio between fermentation rate and hydrolysis rate was used. First-order reaction model developed by Eastman and Ferguson (1981) was used to describe hydrolysis of particular matter in sludge. The first-order hydrolysis rate constant was found to be  $2.36 \text{ d}^{-1}$ , and initial concentration of degradable particulate COD was found to be  $10.4 \text{ g/l}$ . A model of acid-digestion was proposed that included acidogenic biomass growth and decay. The total acidogenic population was estimated and kinetic parameters of acidogenic population were: specific growth rate  $0.78 \text{ d}^{-1}$ , net specific growth rate  $0.25 \text{ d}^{-1}$ , decay coefficient  $0.52 \text{ d}^{-1}$ , and observed yield  $0.05 \text{ mg biomass / mg COD consumed}$ . Based on fermentation index, the optimum retention time in thermophilic acid digester was 3 to 3.5 days.

#### INTRODUCTION

Stabilization efficiency of the anaerobic digestion of biosolids depends mainly on the efficiency of the hydrolysis and fermentation processes. These processes are carried out by “primary producers” in the digester ecosystem and products – volatile fatty acids – are further utilized by other groups of microorganisms (Ghosh et al., 1999). In the past 25 – 30 years a lot of attention has been paid to the hydrolytic and acidogenic bacteria which led to

the development of a two-stage anaerobic digestion technology. The anaerobic digestion is carried out in two digesters and solids retention time (SRT) serves as a selector between acidogens and methanogens. Providing a short retention time, mostly in the range of 2 to 5 days (Wilson et al., 2002) in the first digester suppresses methanogenic activity and allows for the proliferation of fast-growing acidogenic bacteria. For instance, the generation time of methane formers was about ten times longer than that of acid formers (Ghosh and Pohland, 1974). Following the hydrolytic digester, a methane digester with longer retention times, allowing methane community to develop, is utilized. Typically retention times in the second digester vary between 10 – 15 days or more (Wilson, 2001). Therefore, in terms of time requirement, methanogenesis can be viewed also as a limiting step of anaerobic digestion (Ghosh and Pohland, 1974). Two-phase digestion separates two-limiting steps: hydrolysis and methanogenesis. This enhances the process by improving reaction kinetics and stability. In the first reactor, hydrolysis is enhanced by more intimate contact between intracellular enzymes and complex substrates. The improvement of hydrolysis is due to the fact that the smaller reactor volume contains primarily hydrolyzing and acidifying biomass which creates a high concentration of enzymes. In the second reactor, acetogenic and methanogenic bacteria proliferate at the same time also allowing a syntrophic relationship (in order to convert solvents and higher organic acids to acetate, hydrogen and carbon dioxide) to take place (Fox and Pohland, 1994).

The most common way to enhance hydrolysis and fermentation is to increase the temperature of digestion to thermophilic range. For instance, Moen et al., (2001) found soluble COD concentrations in the thermophilic reactor at 55<sup>0</sup>C much higher compared to mesophilic reactor at 35<sup>0</sup>C over all SRTs. Others found maximum specific growth rates of thermophilic

and mesophilic acetogens to be  $0.7 \text{ day}^{-1}$  and  $0.4 \text{ day}^{-1}$ , respectively (Ghosh et al., 2001). In another study, the activities of acidogenic bacteria under thermophilic conditions were about 1.8 times higher than those under mesophilic conditions (Kiyohara et al., 2000). Since thermophilic digestion requires a higher energy input to reach the higher temperatures, a two-phase anaerobic digestion offers a potential to offset the costs of thermophilic digestion with benefits of increased hydrolysis and fermentation of sludge and additionally higher pathogen destruction.

A configuration of two-phase digestion when the thermophilic reactor is followed by a mesophilic reactor seems to have somewhat of an advantage over a mesophilic-thermophilic configuration. The latter was reported to produce higher effluent soluble COD, while offering the same COD removal rates as in case of thermophilic-mesophilic configuration (Duran and Speece, 1997).

However, two-phase digestion itself has not been optimized thus far, as there are conflicting views with regards to the required retention time in the first digester as shown in Table 4-4. Reported retention times varied between 2 to 17 days. Similarly, there is lack of reliable source of kinetic parameters that describe behaviour of acid-digester biomass such as yield, growth rate and decay coefficient (Table 4-5).



**Table 4 - 4. Retention times in thermophilic acid digesters in existing plants, pilot plants and lab experiments (Schafer et al., 2002, Wilson and Streicher, 2001).**

| Plant   | Digester size       | Type of sludge | SRT, days | Temperature, °C | Reactor Feeding    |
|---|---------------------|----------------|-----------|-----------------|--------------------|
| Western Lake Superior Sanitary District Duluth, MN                      | 3785 m <sup>3</sup> | WAS            | 5         | 55              | Continuous         |
| Cologne, Germany Stammheim Plant  | 7000 m <sup>3</sup> | WAS            | 7         | 55              | Continuous         |
| Neenah-Menasha  | 2465 m <sup>3</sup> | PS + WAS       | 8         | 55              | Nearly Continuous  |
| Sturgeon Bay, Wisconsin   | 2840 m <sup>3</sup> | PS + WAS       | 17        | 55              | Intermittent       |
| King Co., WA Renton (Pilot Scale)                                       | 2 m <sup>3</sup>    | PS + WAS       | 8         | 55              | Nearly Continuous  |
| City of Los Angeles Hyperion, Waste Water Treatment Plant (Pilot Scale) | 29 m <sup>3</sup>   | PS + WAS       | 10        | 55              | Intermittent       |
| Madison, WI Pilot (Lab Scale)   | 0.75 to 2.5 litre   | PS + WAS       | 5         | 55              | Intermittent       |
| Ville d'Aix, France   | 900 m <sup>3</sup>  | WAS            | 3         | 55              | ?                  |
| City of Los Angeles Hyperion, WWTP (Pilot Scale)                        | 29 m <sup>3</sup>   | PS + WAS       | 2         | 55              | Intermittent       |
| Indianapolis, IN Belmont Waste Water Treatment Plant (IDI Pilot Scale)  | 1 m <sup>3</sup>    | PS + WAS       | 2         | 55 & 60         | 2 or 4 times a day |

PS – Primary Sludge, WAS – Waste Activated Sludge

**Table 4 - 5. Biomass parameters including yield, yield observed, growth rate, and decay coefficient found in previous studies.**

| Biomass parameter                         |                         |                         | Temperature | Substrate                              | Reference                            |
|---|-------------------------|-------------------------|-------------|--|--------------------------------------|
| Yield                                     | Maximum growth rate     | Decay coefficient       |             |  |                                      |
| <i>mg biomass / mg substrate consumed</i> | <i>day<sup>-1</sup></i> | <i>day<sup>-1</sup></i> | <i>°C</i>   |  |                                      |
| 0.17                                      | 30.00                   | 6.00                    | 36          | glucose                                | Ghosh and Pohland, 1974              |
| 0.40                                      | 3.84                    | 1.70                    | 36          | primary sludge                         | Ghosh et al., 1975                   |
| 0.48                                      | -                       | 0.43                    | 35          | primary sludge                         | Eastman and Ferguson, 1981           |
| 0.15                                      | 2.00                    | -                       | 35          | not defined                            | Pavlostathis and Giraldo-Gomez, 1991 |
| 0.15                                      | 0.37                    | 0.10                    | 35          | primary, secondary and tertiary sludge | Siegrist et al., 1993                |
| 0.05                                      | 0.40                    | 0.03                    | 38          | constants used for numerical model     | Azeitero et al., 2001                |
| 0.15                                      | 4.00 and 16.00          | 0.80 and 3.20           | 35 and 55   | IWA numerical model for primary sludge | Batstone et al., 2002                |
| 0.03 to 0.08                              | 0.3 to 0.6              |                         | 20 to 55    | various data                           | Noykova et al., 2002                 |

## OBJECTIVES

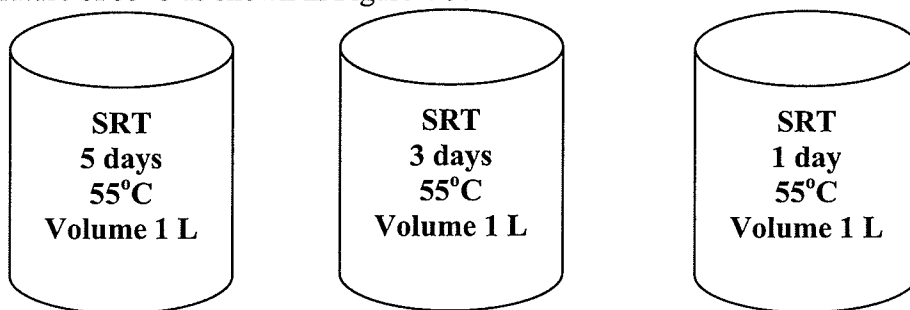
The objective of the study was to optimize the retention time in a thermophilic acid digestion system. The primary objective of this experiment was to assess the impact of different retention times in the thermophilic reactor with respect to hydrolysis and fermentation of sludge. The secondary objective was to enumerate acidogenic population found in acid-

digesters and to establish the following kinetic coefficients for this population as specific and maximum growth rate, observed yield and decay coefficient.

## METHODOLOGY

### Experimental approach

Three lab scale acid-digestion systems were assembled and operated at thermophilic temperature of 55°C as shown in Figure 4-9.



**Figure 4 - 9. Schematic representation of lab scale acid-digesters.**

### Sludge source and reactor operation

See section 4.1.

### Experimental procedures

Chemical oxygen demand (COD) was determined twice a week according to SM 5220 D (APHA et al., 1998). Soluble chemical oxygen demand (SCOD) was determined as COD after filtration through 0.45  $\mu\text{m}$  pore size media. Volatile fatty acids (VFA) including acetic, propionic, i-butyric, n-butyric, i-valeric, n-valeric, i-caproic and n-caproic (C2 to C6) were measured twice a week using gas chromatograph Antek 3000 with flame ionization detector. Volatile fatty acids were stored in the freezer and analyzed every two weeks. A separate experiment was performed to establish initial, viable anaerobic hydrolytic-acidogenic bacteria (AHAB) population. Samples from raw sludge were taken every day for a period of

3 weeks and most probable number (MPN) procedure was performed. Dilutions were made using buffered dilution water. Difco™ Anaerobe Broth MIC (enzymatic digest of casein 10 g/l; peptone 10 g/l; yeast extract 5 g/l; dextrose 1 g/l; sodium chloride 5 g/l; L-arginine 1 g/l; sodium pyruvate 1 g/l; hemin 5 mg/l; and vitamin K<sub>1</sub> 0.5 mg/l) was used as a medium for AHAB growth with the addition of 0.01 g/l of bromcresol purple to detect changes in pH. Tubes were incubated anaerobically in a temperature of 36°C for 24 hours. Tubes that turned yellow (indication that pH was 5.2 or lower) were counted as positive and purple tubes (final pH higher than 5.2) were counted as negative. Most probable number (MPN) was established based on three-tubes per dilution according to Finstein (1972).

## RESULTS AND DISCUSSION

### First-order hydrolysis model

There are commonly two approaches used to establish the extent and the rate of hydrolysis. The first approach calculates hydrolysis based on the mass balance of solids (Ghosh et al., 1999, Ghosh et al., 1987) and in the second approach hydrolysis is calculated based on the COD mass balance (Miron et al., 2000, Schmit and Ellis, 1998). Similarly, the fermentation process can be considered in both ways. This paper will focus on COD calculations.

Hydrolysis of a particulate matter at constant temperature and pH was proposed (Eastman and Ferguson, 1981, Pavlostathis and Giraldo-Gomez, 1991) to follow a first-order reaction with respect to the concentration of degradable particulate organic matter. The derivation of particulate COD mass balance proposed by Eastman and Ferguson (1981) for a completely mixed, continuous-flow reactor at steady state is presented in equations [4-1] and [4-2].

**Equation 4 - 1** Inflow of Particulates – Outflow of Particulates – Rate of Hydrolysis = 0

**Equation 4 - 2**  $Q \times (F_0 - F) - V \times k_h \times F = 0$

Equation [4-3] was derived after transforming equation [4-2]

**Equation 4 - 3**  $F = \frac{F_0}{1 + k_h \text{SRT}}$

Where:

Q – flow, L.day

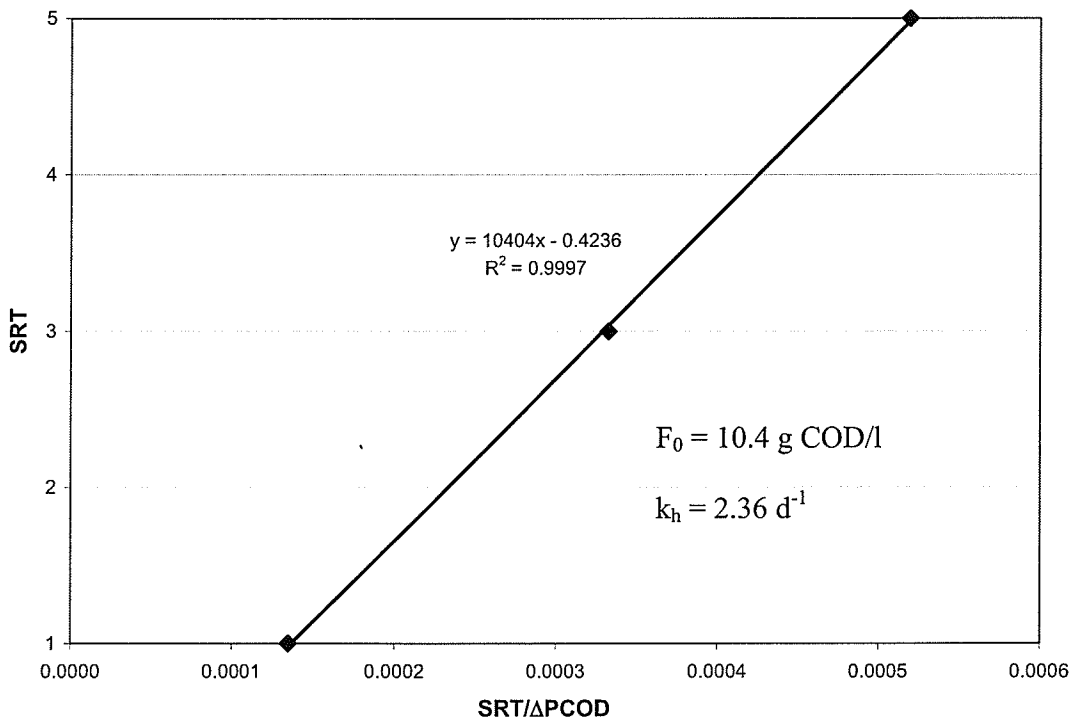
V – reactor volume, L

$\frac{dF}{dt}$  - hydrolysis rate, mg COD/L-day

F – concentration of degradable particulate COD, mgCOD/L

F<sub>0</sub> – initial concentration of degradable particulate COD, mgCOD/L

k<sub>h</sub> - first-order hydrolysis rate constant, d<sup>-1</sup>



**Figure 4 - 10. Graphic solution of hydrolysis rate constant (k<sub>h</sub>) and initial concentration of degradable particulate matter (F<sub>0</sub>).**

Based on the acid-phase model, the slope as indicated in Figure 4-10 corresponds to F<sub>0</sub> and the Y-intercept to -1/k<sub>h</sub>. The first-order hydrolysis rate constant found by Eastman and

Ferguson (1981) at 35°C was 3.0 d<sup>-1</sup>. In this study, the hydrolysis rate constant at 55°C was 2.36 d<sup>-1</sup>. The influent concentration of particulate COD found in this study was 10.4 g/l and previous study indicated it was 7.3 g/l. It should be noted that pH in this research varied between 6.3 and 7.1, and pH values varied between 5.13 and 5.21 in the study by Eastman and Ferguson (1981). It was also found that an increase in pH from 5.15 to 6.6 resulted in an increase in degradation at constant temperature (Eastman and Ferguson 1981). Therefore, most probably the combined effect of increased temperature and higher pH resulted in the increased hydrolysis rate observed in this study as shown in Figure 4-11. A comparison between results from this study and results previously published by Eastman and Ferguson (1981) are presented in Table 4-6. Calculated hydrolysis rates at different SRT are presented in Figure 4-10.

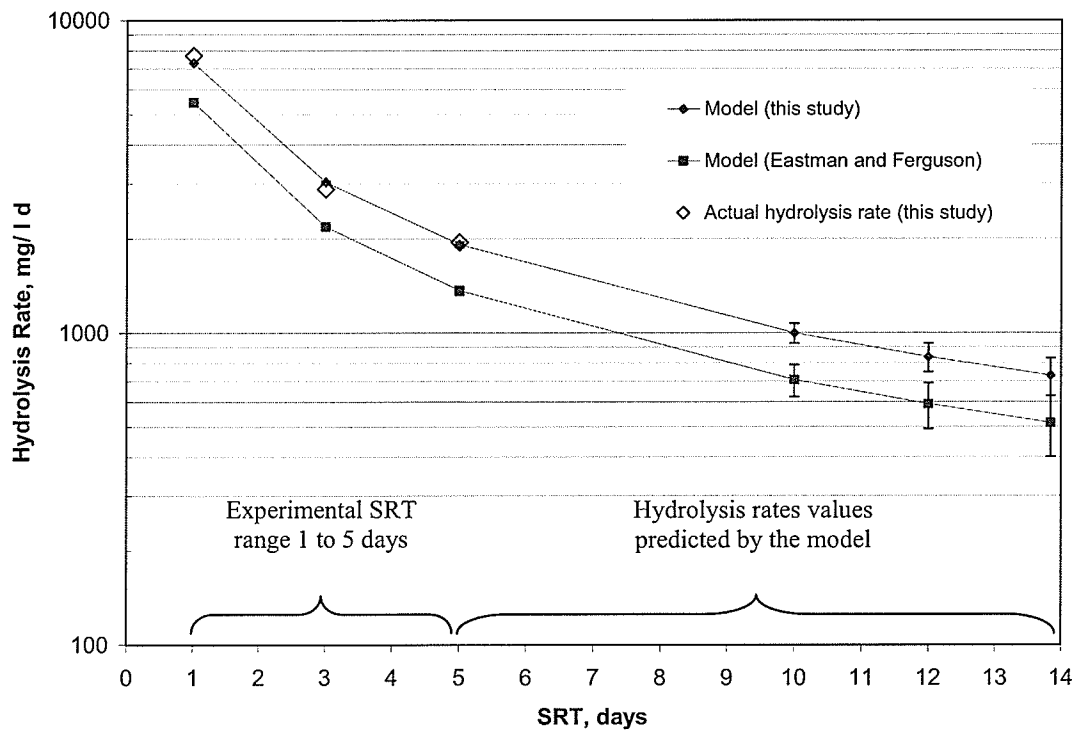
**Table 4 - 6. Comparison of results from this study and values found by Eastman and Ferguson.**

| Comparison   | This study                 | Eastman and Ferguson 1981*  |
|--|----------------------------|-----------------------------|
| Type of sludge used  | PS + WAS                   | PS                          |
| Temperature of digestion   | 55°C                       | 35°C                        |
| First-rate hydrolysis constant                                     | 2.36 ± 0.34d <sup>-1</sup> | 3.00 ± 0.38 d <sup>-1</sup> |
| Initial concentration of degradable particulate COD in feed sludge | 10.40 ± 0.17 g/l           | 7.30 ± 0.15 g/l             |

\* - errors were calculated by authors based on results presented in the paper

Hydrolysis rates in Figure 4-10 were calculated based on the proposed model according to equations [4-1] and [4-2]. Calculations included hydrolysis rates within experimental range of SRT (1, 3 and 5 days SRT) followed by hydrolysis rates at higher SRT based on the model prediction. Error bars in Figure 3 were calculated based on standards error established for

slope and Y-intercept for which analysis was performed by computer program JMP Start Statistics version 4 (Sall et al., 2001). Accuracy of the coefficients used in the model indicated that difference in calculated hydrolysis rates between this study and based on results provided Eastman and Ferguson (1981) becomes statistically insignificant when SRT reaches approximately 14 days (Figure 4-11). It is very likely, that expected benefits of higher VS destruction and biogas recovery, which are attributed to thermophilic anaerobic digestion over conventional mesophilic digestion, may be expected only at short retention times (particularly using results presented in this study when operating SRT is less than 14 days).



**Figure 4 - 11. Comparison between hydrolysis rates obtained from this study with findings of Eastman and Ferguson (1981).**

Fermentation rate

The proposed model to evaluate the fermentation rate of wastewater sludge is presented in Figure 4-12. The main components of this model are soluble products (namely volatile organic acids) and acidogenic biomass concentration. The proposed model includes methanogenic activity in the acid-phase reactor. However, recent findings suggest that methanogenic community in the acid phase digester account for less than 1% of the total microbiological population (Solera et al., 2001). It is therefore proposed to neglect methanogenic biomass growth and decay processes, and include loss of COD associated with biogas production to the COD of acid phase products (Figure 4-12). This will result in simplification of the model. It is also assumed that biomass decay will result in cell lysis and increase in soluble products (equations [4-4] and [4-5]) as in the model proposed by Eastman and Ferguson (1981).

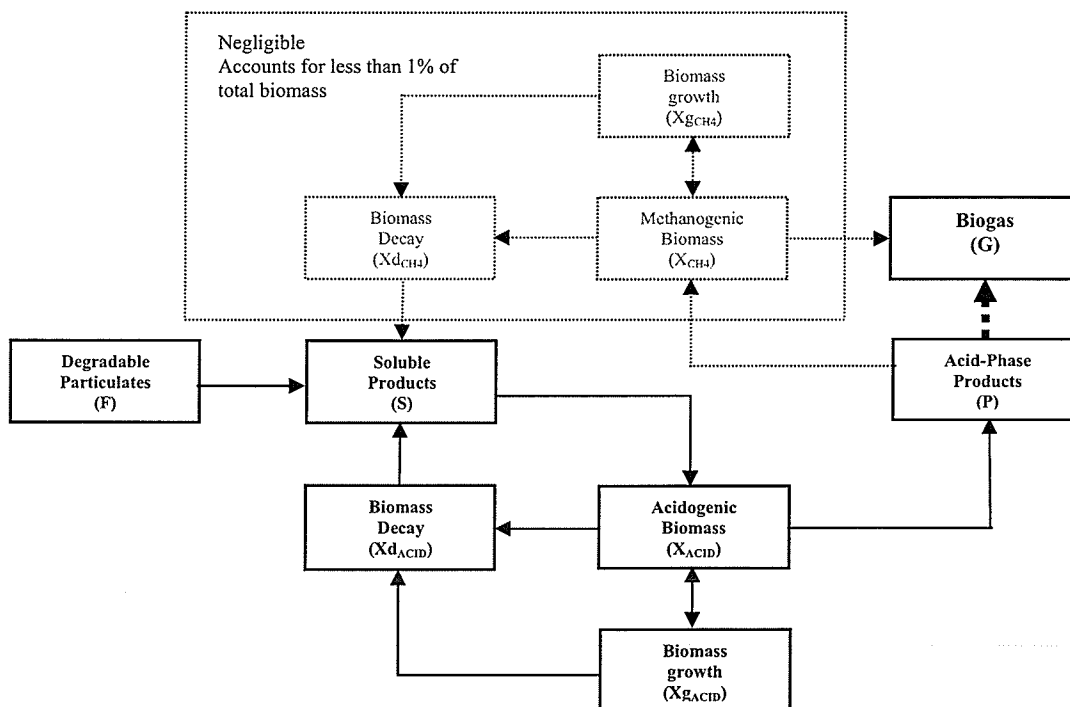


Figure 4 - 12. The proposed model of the acid-phase digester.



**Equation 4 - 4** [Acid-Phase Products in effluent] - [Acid-Phase Products in inflow] - [Biomass Decay] + [Biomass growth] + [Biogas] - [Fermentation rate] = 0

or

**Equation 4 - 5** Fermentation rate =  $Q (VFA_{eff} - VFA_{inf} - Xd_{ACID} + Xg_{ACID} + COD_{inf} - COD_{eff})$

The important components of the proposed model are those associated with biomass growth and decay. In order to establish those components an estimation of acidogenic biomass had to be performed. Calculation of total acidogenic biomass (anaerobic hydrolytic-acidogenic bacteria – AHAB) in the acid-phase reactor was possible after the initial, cultivable population ( $X_0$ ) was established with the use of MPN procedure (Table 4-7). These findings are within the range previously published by other researchers as presented in Table 4-8.

**Table 4 - 7. Initial viable anaerobic hydrolytic-acidogenic bacteria (AHAB) –  $X_0$ .**

| Initial AHAB, MPN/ml   | Conversion Factor, g cell per ml | Reference          | Initial AHAB, mg/l |
|------------------------|----------------------------------|--------------------|--------------------|
| 1.08 x 10 <sup>8</sup> | 2.50 x 10 <sup>12</sup>          | White et al., 2002 | 43.15              |
|                        | 0.35 x 10 <sup>12</sup>          | Norland, 1993      | 31.07              |
| Average                |                                  |                    | <b>37.11</b>       |

**Table 4 - 8. Bacterial populations found in different aquatic environments.**

| Environment                                     | Bacterial Population, cells per gram | Reference              |
|---|--------------------------------------|------------------------|
| Anaerobic acidogenic digesters operated at 55°C | 2.70 to 25.10 x 10 <sup>8</sup>      | Solera et al., 2001    |
| Subsurface sediments                            | 2.5 x 10 <sup>12</sup>               | White et al., 2002     |
| Deep subsurface sediments                       | 10 <sup>4</sup> to 10 <sup>5</sup>   | Ogram et al., 1995     |
| Marine sediments and neotropical mud flats      | 10 <sup>9</sup>                      | White et al., 2002     |
| Cold deep sea sediments                         | 10 <sup>9</sup>                      | Li et al., 1999        |
| Different soil types                            | 10 <sup>6</sup> to 10 <sup>9</sup>   | Atlas and Bartha, 1993 |
| Hydrothermally influenced sediments             | 10 <sup>5</sup>                      | Summit et al., 2000    |

Calculation of acidogenic biomass concentration included solving equations 5 to 10 simultaneously. The only parameter assumed for calculations was the biomass yield of 0.15

(Table 4-9). Equations [4-5] and [4-6] were used for a reactor operated at SRT of 1 day. It could be done knowing that all biomass developed in this reactor came from substrate utilization and from initial viable biomass (Table 4-6).

$$\text{Equation 4 - 6 } X_{\text{ACID}} = X_0 + Y_{\text{obs}} \times (\text{COD}_{\text{inf}} - \text{COD}_{\text{eff}}), \text{ g biomass / l}$$

$$\text{Equation 4 - 7 } \frac{dX_{\text{ACID}}}{dt} = \frac{X_{\text{ACID}} - X_0}{dt} = \mu \times X_0, \text{ g biomass / l}$$

Slightly different approach was used to calculate active acidogenic biomass in acid-phase reactors when the operating SRT was 3 and 5 days. During these operating conditions when some amount of active biomass is added to reactor viable biomass, the sludge age and biomass concentration can be calculated solving simultaneously equations [4-7] and [4-8] according to Rittmann (1996).

$$\text{Equation 4 - 8 } \text{SRT}_X = \frac{X_{\text{ACID}} \times V}{Q \times X_{\text{ACID}} - Q \times X_0} = \frac{\text{SRT}}{1 - X_0/X_{\text{ACID}}}, \text{ days}$$

$$\text{Equation 4 - 9 } X_{\text{ACID}} = \frac{\text{SRT}_X}{\text{SRT}} \frac{Y(\text{COD}_{\text{inf}} - \text{COD}_{\text{eff}})}{1 + k_d \times \text{SRT}_X}, \text{ g COD biomass}$$

The biomass growth only was calculated using true yield instead of observed yield in equation [4-6] or maximum specific growth rate value instead of specific growth rate in equation [4-7] or assuming that decay coefficient equal zero ( $k_d = 0$ ) in equation [4-9]. The difference between actual values for biomass concentration and values calculated for growth only was assumed to be caused by decay of biomass. Observed yield and maximum specific growth rate values were obtained from calculations according to equations [4-10] and [4-11], respectively. Saturation constant,  $K_s$ , was assumed to be 160 mg/l (Noykova et al., 2002).

Kinetic parameters for acidogenic biomass obtained from this study are provided in Table 4-9.

**Table 4 - 9. Kinetic parameters of acidogenic biomass.**

| Parameter                    | Symbol      | Unit                       | Value |
|------------------------------|-------------|----------------------------|-------|
| Net specific growth rate     | $\mu$       | $d^{-1}$                   | 0.25  |
| Maximum specific growth rate | $\mu_{max}$ | $d^{-1}$                   | 0.78  |
| Decay coefficient            | $k_d$       | $d^{-1}$                   | 0.52  |
| Observed yield               | $Y_{obs}$   | <i>mg biomass / mg COD</i> | 0.05  |
| True yield (assumed)         | $Y$         | <i>mg biomass / mg COD</i> | 0.15  |

**Equation 4 - 10**  $Y_{obs} = Y \times \left( 1 - \frac{k_d}{\mu_{max}} \frac{K_s + COD_{eff}}{COD_{eff}} \right)$ , mg biomass / mg COD utilized

**Equation 4 - 11**  $\mu = \mu_{max} \frac{COD_{eff}}{K_s + COD_{eff}} - k_d$ ,  $d^{-1}$  - net specific growth rate

Table 4-10 presents results of the calculations of fermentation rates including acidogenic biomass components. Established biomass concentrations were converted to COD values where 1 mg biomass equals to 1.42 mg COD.

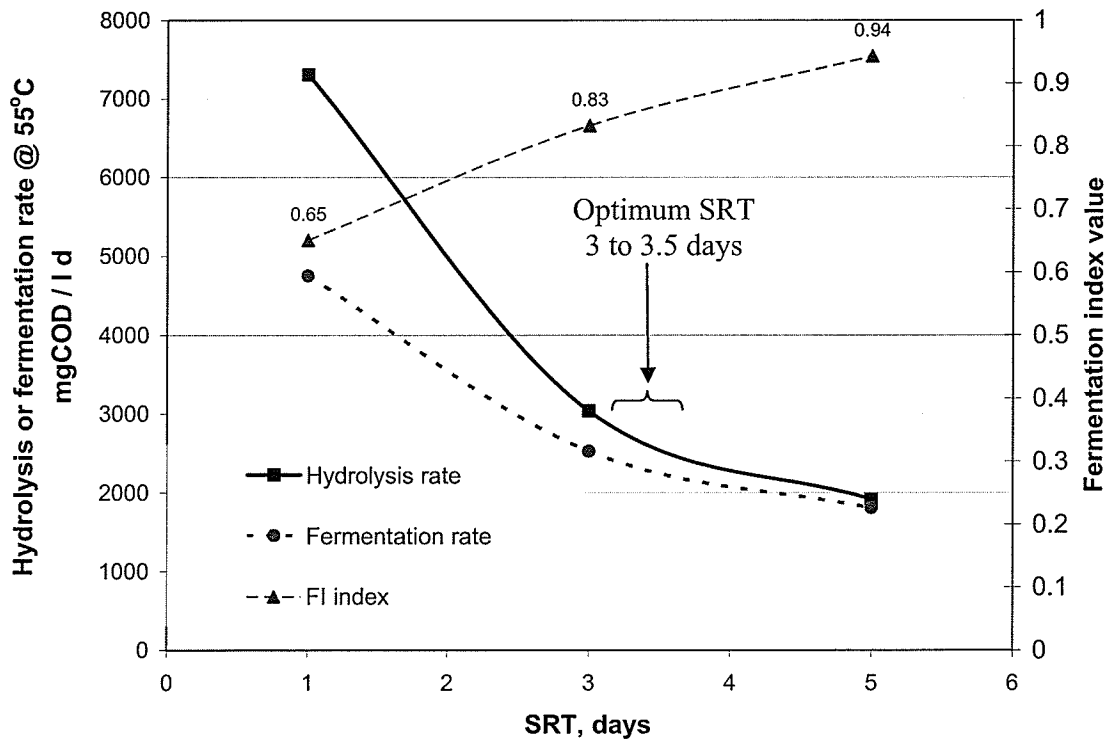
**Table 4 - 10. Fermentation rates established for thermophilic acid-digesters.**

| Calculated fermentation rates at different SRT |                  |             |             |             |
|--|------------------|-------------|-------------|-------------|
|  | Unit             | SRT 1 d     | SRT 3 d     | SRT 5 d     |
| Flow   | <i>l/d</i>       | 1.00        | 0.33        | 0.20        |
| VFA <sub>effl</sub>                            | <i>mgCOD/l</i>   | 6300        | 7512        | 5046        |
| VFA <sub>inf</sub>                             | <i>mgCOD/l</i>   | 2168        | 2332        | 1392        |
| X <sub>dACID</sub>                             | <i>mgCOD/l</i>   | 129         | 440         | 1004        |
| X <sub>gACID</sub>                             | <i>mgCOD/l</i>   | 298         | 649         | 1309        |
| COD <sub>inf</sub>                             | <i>mgCOD/l</i>   | 38193       | 39873       | 38632       |
| COD <sub>effl</sub>                            | <i>mgCOD/l</i>   | 37742       | 37596       | 33547       |
| <b>Fermentation rate</b>                       | <i>mgCOD/l d</i> | <b>4752</b> | <b>2530</b> | <b>1809</b> |

By definition, anaerobic decomposition of sludge in acid-digester is not at equilibrium, with hydrolysis and acidogenesis (fermentation) dominating over extremely inhibited

methanogenesis. As shown in Figure 4-13, hydrolysis and acidogenesis rates change over time. At longer SRTs those rates decrease, and so does the ratio between fermentation rate and hydrolysis rate. Ideally, at long retention times when anaerobic decomposition is balanced, hydrolysis and fermentation rates would be equal. To evaluate the degree of fermentation the empirical fermentation index (*FI*) was introduced (Ghosh et al., 1999) calculated according to equation [4-11].

**Equation 4 - 12**  $FI = \text{Fermentation rate} / \text{Hydrolysis rate}$



**Figure 4 - 13. Graphic representation of hydrolysis and fermentation rates and fermentation index (FI) values at 55°C.**

It is proposed that the fermentation index could be used as an optimization parameter for the retention time in acid digester. The *FI* value increases as more time is provided for digestion but that increase becomes slower at longer retention times (Figure 4-12). An arbitrary limit

on a minimum increase in *FI* value could be imposed that would limit retention time. If one assumes that an increase in retention time is reasonable if *FI* value increases by at least 0.05 (in the absence of known engineering cut off value 5% or 0.05 is commonly used) per day then in case of this study an operation of acid digester at SRT of 3 to 3.5 days would be considered as optimized with respect to both hydrolysis and VFA formation. From a practical view point, optimization of retention time in acid digestion is important when one considers possible savings of energy (especially if acid digestion is operated at thermophilic temperatures) and digester's volumes.

## CONCLUSIONS

The following conclusions were drawn based on the results of this experiment:

- First-order reaction model developed by Eastman and Ferguson (1981) was used to describe hydrolysis of the particular matter in sludge. The first-order hydrolysis rate constant during thermophilic (55°C) anaerobic digestion of wastewater sludge was found to be  $2.36 \text{ d}^{-1}$ , and initial concentration of degradable particulate COD was found to be 10.4 g/l.
- Hydrolysis rates established for this study on blended sludge were higher than those established by Eastman and Ferguson (1981) on raw primary sludge. The difference between hydrolysis rates diminishes at longer retention times showing potential benefits of thermophilic digestion over conventional mesophilic digestion at relatively short retention times (below 14 days SRT in case of this study).
- A model of acid-digestion was proposed that includes acidogenic biomass growth and decay. The total acidogenic population was estimated and kinetics parameters of acidogenic population were: maximum specific growth rate  $0.78 \text{ d}^{-1}$ , net specific growth

rate  $0.25 \text{ d}^{-1}$ , decay coefficient  $0.52 \text{ d}^{-1}$ , and observed yield  $0.05 \text{ mg biomass / mg COD}$  consumed.

- It was proposed that fermentation index - *FI* - could be used as optimization parameter for the retention time in acid-phase digester with respect to hydrolysis and fermentation. This study found that retention time in acid-digester was optimized when operating SRT was 3 to 3.5 days.

#### ACKNOWLEDGMENTS

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## **5 Impact of environmental factors on acid digestion**

This section consists of previously unpublished material. The focus of this part of the research was twofold: (1) it is a continuation of work on hydrolysis presented in section 4.3. except that more environmental conditions and their effect on hydrolysis rates were studied; and (2) as indicated in section 4.2. acid digestion can lead to production of hydrogen and experimental work was designed to find the amount of hydrogen produced, optimum conditions for hydrogen production, and ultimately practical and economical feasibility of hydrogen recovery.

Raw data for all results presented in section 5.0 can be found in Appendix B.

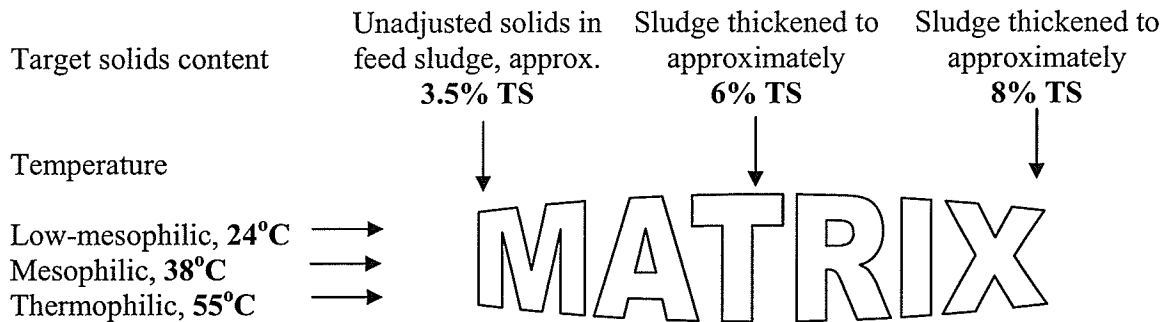
## 5.1 Hydrolysis rates

### INTRODUCTION AND OBJECTIVES

This section presents results of the experimental work on the effect of different environmental factors on the hydrolysis rate of wastewater sludge. It is a continuation of work presented in section 4.3. but this study also aims at finding whether the hydrolysis rate can be correlated with efficiency of anaerobic digestion and/or biogas production. It is commonly accepted that hydrolysis rates increase with increase in temperature, which should cause increase in sludge stabilization. This was not supported by findings from section 4.2., when either VS destruction or biogas production from a single thermophilic digester, a single mesophilic digester and a temperature staged thermophilic-mesophilic digestion system did not show any significant differences. In light of the widely accepted anaerobic digestion model (see section 2.2 and 2.3) hydrolysis is considered to be the limiting step of anaerobic digestion. Therefore the knowledge of hydrolysis rate constant could also be used to predict the impact of environmental factors such as temperature and solids content in feed sludge on optimum conditions for anaerobic digestion. This information could be used as a design parameter, for instance defining optimum solids content in sludge if sludge was to be thickened prior to digestion, or as a calibration parameter in computer model of anaerobic digestion. Hydrolysis is the single most important and sensitive parameter (Batstone et al., 2002, Christ et al., 2000, El-Fadel et al., 1996, El-Fadel et al., 1997, Pavlostathis and Giraldo-Gomez, 1991, Sanders et al., 2002) while modelling anaerobic digestion of any type of wastes, yet a comprehensive study on the effect of environmental factors (such as temperature and solids concentration in feed sludge) on hydrolysis rates of wastewater sludge could not be found. Therefore this experimental work was designed as a matrix (Figure 5-1)



with solids content in sludge and process temperature and their combined effect on hydrolysis rate of wastewater sludge.



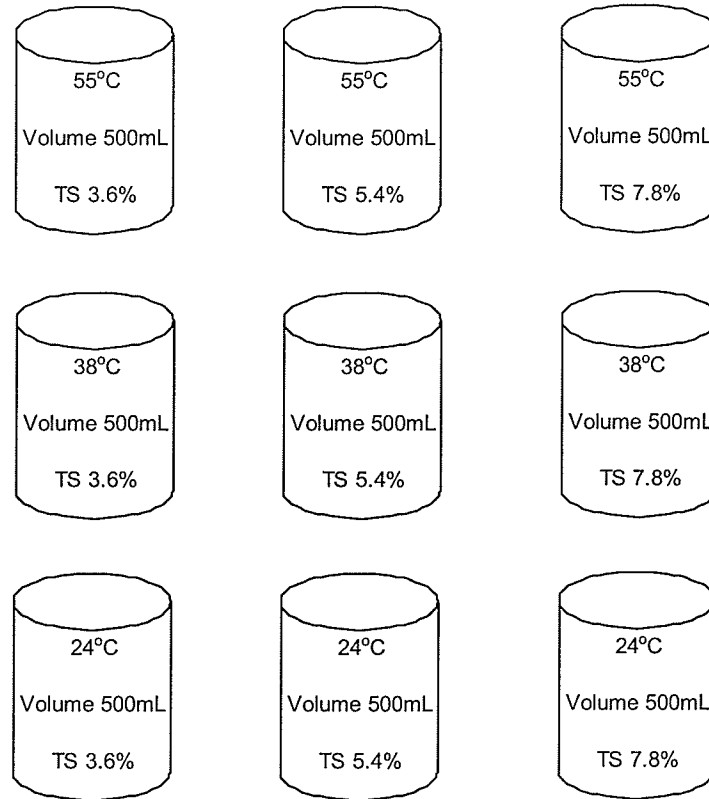
**Figure 5 - 1. Matrix for research of simultaneous effects of different solids content in feed sludge and temperature of the process on hydrolysis rate of wastewater sludge.**

#### METHODOLOGY

Experimental work included running nine digesters at constant SRT for six weeks (as presented in Figure 5-2), acclimation period and another experimental run at constant SRT. The main variables were temperature and solids concentration (target solids concentrations are presented in Figure 5-1). Three operating SRTs were chosen: 5 days, 3.2 days and 2.2 days. Digesters were mixed manually 3 times per day. pH was not adjusted. Temperature of the waterbaths was controlled through Isotemp 2100 circulators.

Source of sludge as in section 4.0.

Total solids (TS) were measured according to Standard Method (SM) 2540 B (APHA et al., 1998). Total chemical oxygen demand (TCOD) was measured according to SM 5220 D (APHA et al., 1998). Soluble chemical oxygen demand (SCOD) was determined as COD after filtration through 0.45 µm pore size filter.



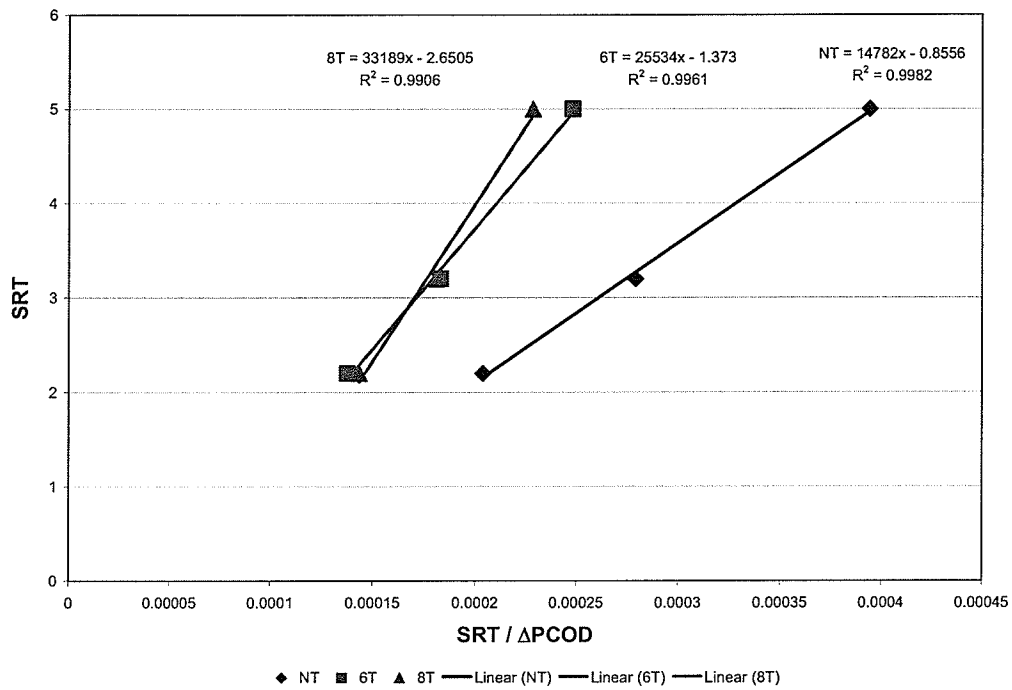
**Figure 5 - 2. Experimental layout – nine sets of acid digesters each operated at different temperature and solids content in feed sludge.**

## RESULTS AND DISCUSSION

Hydrolysis constants and rates were established according to the model described in section 4.3 (equations 4-1 and 4-2). Hydrolysis was established based on COD mass balance and the COD results from experimental runs are presented in Table B-1 (Appendix B).

During the time of experimentation, primary sludge exhibited a certain degree of variability such as differences in average COD strength from one experimental phase to another. To eliminate the effect of change in parameters of primary sludge the results were recalculated or normalized according to following assumptions: (1) average particulate COD of primary sludge over the course of all phases were established at each solids level (Table B-2); (2) percentage of particulate COD remaining was established for each acid digester at each

condition (Table B-3); and results were recalculated using the same strength of feed sludge in terms of particulate COD value and remaining particulate COD based to the fraction established in Table B-3 and results are provided in Table B-4 (Appendix B). On the basis of results presented in Table B-4 the following graphs were created (Figures 5-3, 5-4 and 5-5) and first-order hydrolysis constants and initial concentration of degradable particulate COD are presented in Table 5-5.



**Figure 5 - 3. Graphic solution of hydrolysis rate constants and initial concentrations of degradable particulate COD for acid digesters operated at thermophilic temperature.**

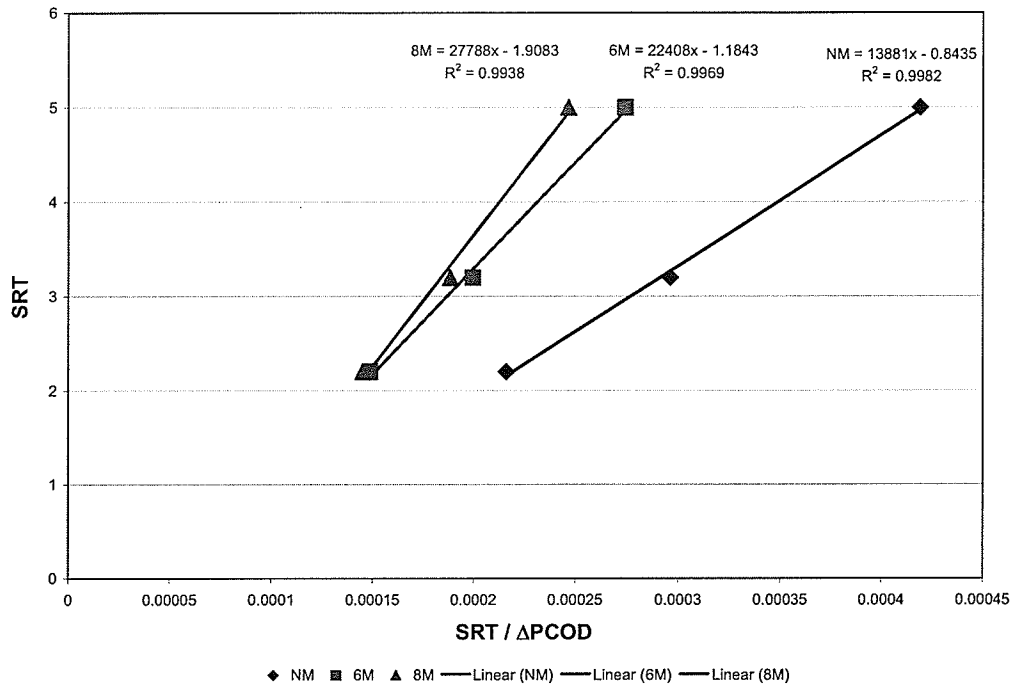


Figure 5 - 4. Graphic solution of hydrolysis rate constants and initial concentrations of degradable particulate COD for acid digesters operated at mesophilic temperature.

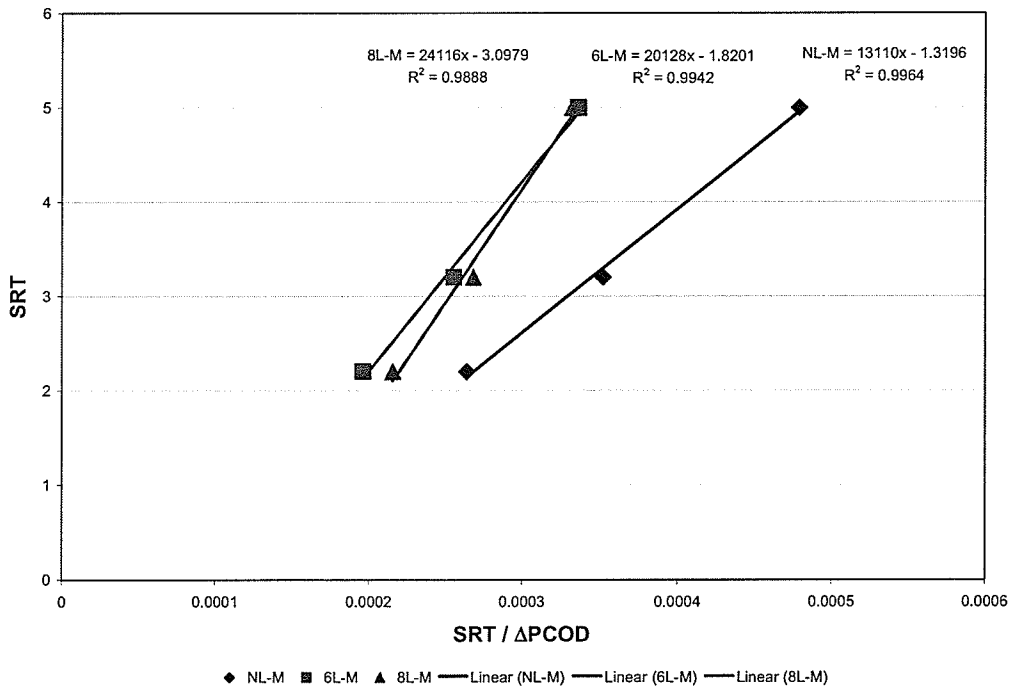
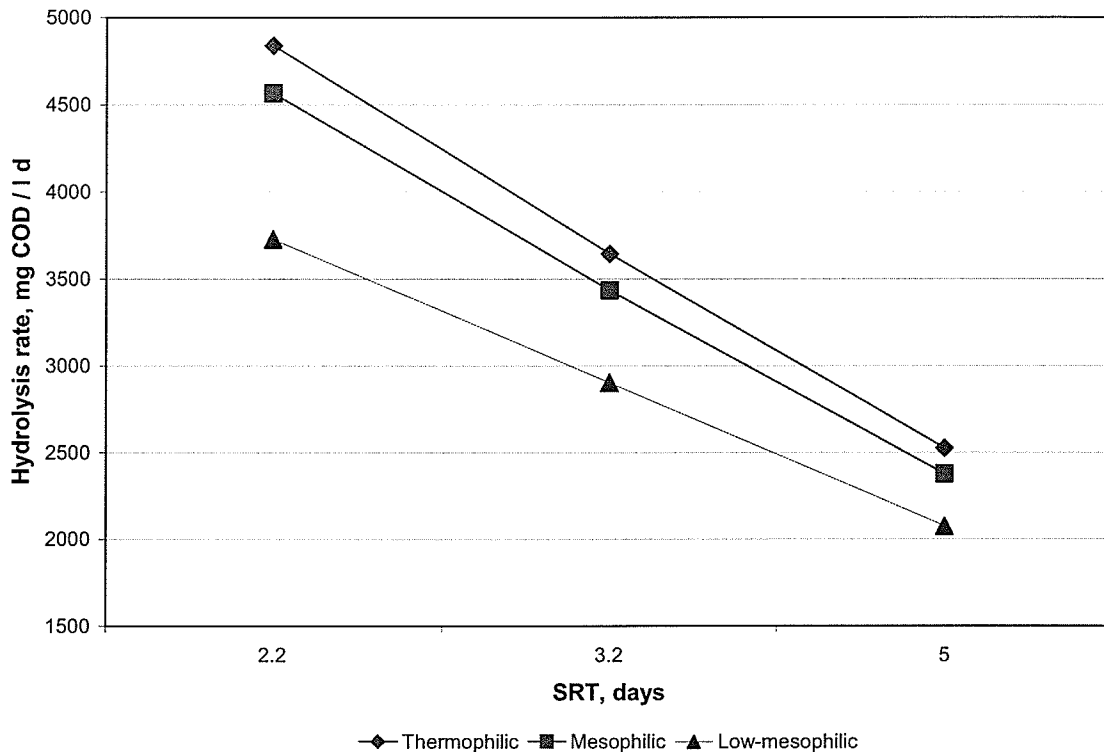


Figure 5 - 5. Graphic solution of hydrolysis rate constants and initial concentrations of degradable particulate COD for acid digesters operated at low-mesophilic temperature.

**Table 5 - 1. First-order hydrolysis constants and concentrations of degradable particulate COD for all treatments tested.**

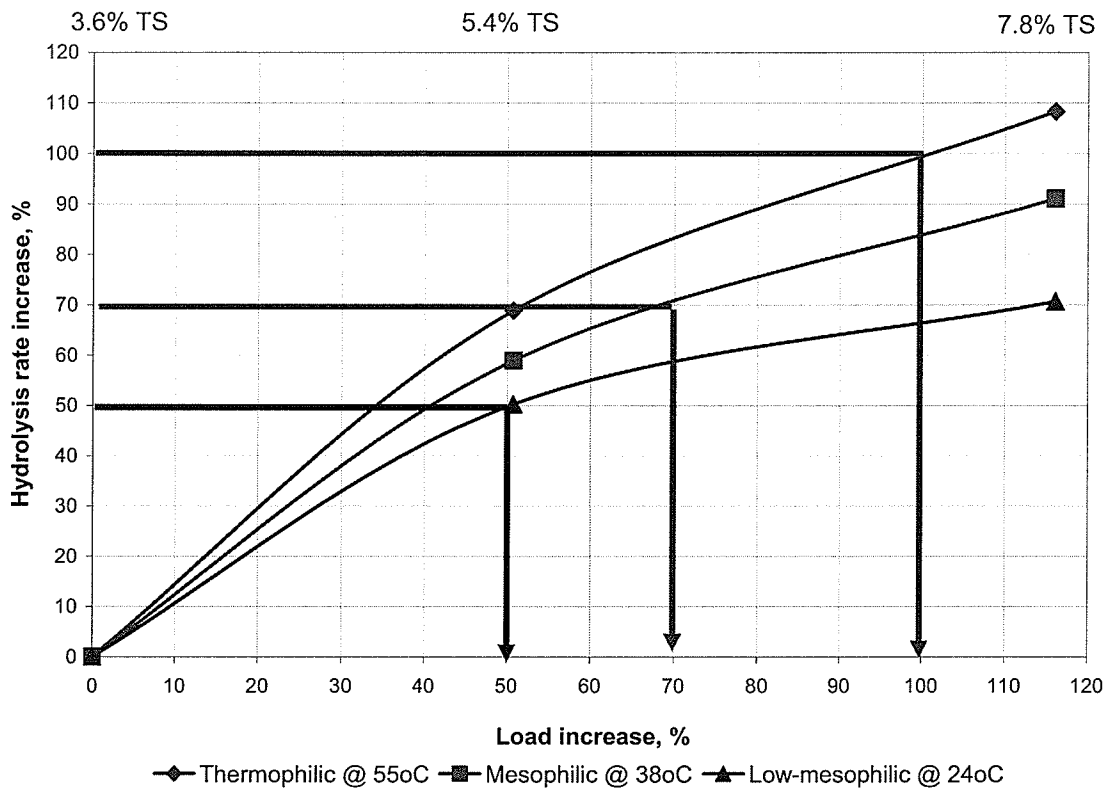
| Treatment   | Unit     | Thermophilic @ 55°C |                 |                 | Mesophilic @ 38°C |                 |                 | Low-mesophilic @ 24°C |                 |                 |
|---|----------|---------------------|-----------------|-----------------|-------------------|-----------------|-----------------|-----------------------|-----------------|-----------------|
| Sludge thickness                                    | % TS     | 3.6                 | 5.4             | 7.8             | 3.6               | 5.4             | 7.8             | 3.6                   | 5.4             | 7.8             |
| First-order hydrolysis rate constant                | $d^{-1}$ | 1.17<br>± 22%       | 0.73<br>± 23%   | 0.38<br>± 23%   | 1.19<br>± 21%     | 0.84<br>± 22%   | 0.52<br>± 23%   | 0.76<br>± 23%         | 0.55<br>± 23%   | 0.32<br>± 23%   |
| Initial concentration of degradable particulate COD | mg/l     | 14782<br>± 633      | 25534<br>± 1590 | 33189<br>± 3230 | 13881<br>± 588    | 22408<br>± 1247 | 27788<br>± 2191 | 13110<br>± 792        | 20128<br>± 1536 | 24116<br>± 2569 |

As seen from Table 5-1 increase in temperature creates an increase in concentration of degradable particulate COD. Also, increase in solids content in feed sludge creates higher concentrations of biodegradable particulate COD, but at the same time it lowers first-order hydrolysis rate constants at all temperatures. Hydrolysis rate is a function of the initial concentration of biodegradable particulate COD and first-order hydrolysis constant (see equations 4-1 and 4-2) and it is also a function of SRT. Hydrolysis rates, presented in Figure 5-6, at unadjusted solids content in feed sludge (approx. 3.6% TS), were found to be the function of temperature with highest rates at thermophilic temperature, followed by mesophilic and low-mesophilic (differences not statistically significant between thermophilic and mesophilic conditions). However, the average difference in hydrolysis rates between thermophilic and mesophilic temperatures were less than 6%. A more noticeable difference in hydrolysis rates was seen at low-mesophilic temperature which was almost 20% lower than thermophilic hydrolysis rate.



**Figure 5 - 6. Wastewater sludge hydrolysis rates at unadjusted solids content in feed sludge (approx. 3.6% TS) as a function of temperature and SRT.**

The knowledge of hydrolysis rates at higher solids content in sludge may be useful in defining the optimum sludge solids content at each operating temperature. According to the anaerobic digestion model, hydrolysis of particulate matter in wastewater sludge affects methane generation, which means lower hydrolysis rate results in lower biogas production. A change in solids content in feed sludge has been shown to affect biogas production (see Figures 1-21 and 1-22). Generally, increasing solids content in sludge results in lower amount of biogas produced per gram VS added. Optimum solids sludge content can be defined as solids content that does not inhibit hydrolysis rates at any given operating temperature. Hydrolysis rates as functions of increasing loading resulted from increased solids content in feed sludge is presented in Figure 5-7.



**Figure 5 - 7. Hydrolysis rates as function of solids content in feed sludge (at SRT of 22 days).**

Increase in solids content in feed sludge (loading on digester), from initial concentration of 3.6% TS, that would be accompanied by proportional increase in hydrolysis rate (required for equal treatment in terms of VS destruction and biogas production) was found to be approximately 50% or up to 5.4% TS for low-mesophilic conditions, 70% or up to 6.1% TS for mesophilic conditions, and 100% or up to 7.2% TS for thermophilic conditions (at SRT of 22 days which is totals SRT of systems presented in section 7.0).

## CONCLUSIONS

The kinetic parameters of sewage sludge hydrolysis were established for three different digestion temperatures: thermophilic, mesophilic and low-mesophilic and three different solids concentration levels in feed sewage sludge: 3.6% TS (unadjusted solids content), 5.4%

TS, and 7.8% TS. It was found that these environmental variables had an impact on the hydrolysis rate constant and concentration of biodegradable particulate COD. The increase in solids content caused increase in initial concentration of biodegradable COD which was the highest at thermophilic conditions, followed by mesophilic conditions and low-mesophilic conditions at all solids levels in feed sludge. The first-order hydrolysis rate constant was found to be highest in mesophilic conditions, followed by thermophilic and low-mesophilic operating temperatures. Hydrolysis rate which is a function of a concentration of biodegradable particulate COD and first-order hydrolysis rate constant was found to be the highest for thermophilic conditions, followed by mesophilic conditions, and the lowest for low-mesophilic conditions. However, the differences between hydrolysis rates obtained in thermophilic and mesophilic conditions were less than 6%, while hydrolysis rates obtained in low-mesophilic conditions were 20% lower than thermophilic hydrolysis rates.

The knowledge of hydrolysis rates allowed to define optimum solids content for anaerobic digestion. The optimum solids content was defined as a solids content that does not inhibit hydrolysis rates at any given operating temperature. Increase in solids content in feed sludge from the initial concentration of 3.6% TS, that would be accompanied by proportional increase in hydrolysis rate (required for equal treatment in terms of VS destruction and biogas production) was found to be approximately 50% or up to 5.4% TS for low-mesophilic conditions, 70% or up to 6.1% TS for mesophilic conditions, and 100% or up to 7.2% TS for thermophilic conditions.



## 5.2 Hydrogen recovery

### INTRODUCTION

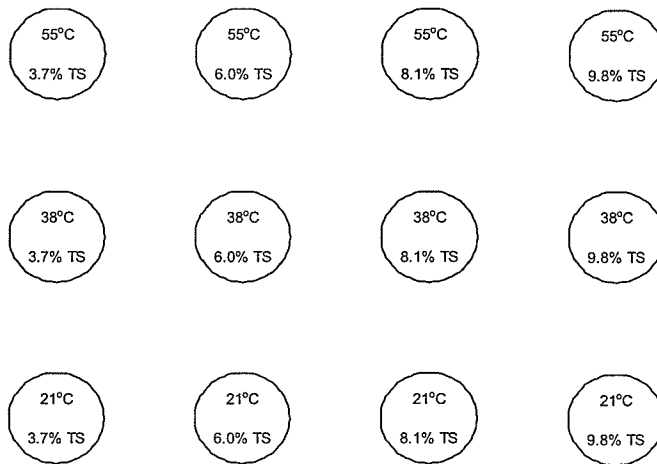
Acid digestion creates an opportunity to recover hydrogen from the biogas produced at this stage. A detail description of hydrogen generation in anaerobic digestion can be found in section 2.2, but it is expected that total biogas (including H<sub>2</sub>, CH<sub>4</sub>, and CO<sub>2</sub>) production in an acid phase digestion accounts for approximately 6% of total biogas produced during anaerobic digestion (Ghosh, 2003, Walden and Andryszak, 2003). However, some models suggest that up to 26% of initial COD could be potentially converted to and recovered as hydrogen gas (Batstone et al., 2002; Siegrist et al. 2002). Therefore, this part of the research investigates the quality of hydrogen in biogas recovered from acid digesters and optimum operating conditions for hydrogen recovery in terms of SRT and temperature.

### METHODOLOGY

Lab-scale digesters (1.5 to 2.0 litres in volume) were assembled and operated in a batch mode for 5 days as shown in Figure 5-8. Reactors were fed daily with a municipal sludge obtained from the North End Water Pollution Control Center in Winnipeg, Manitoba. The main variables were temperature and concentration of total solids (TS) in the sludge fed to digesters (similar to matrix presented in Figure 5-1). Three operating temperatures were chosen: thermophilic @ 55°C, mesophilic @ 38°C (operating temperature at NEWPCC), and low-mesophilic @ 21°C. Before feeding, the sludge was sieved through sieve No. 5 (opening size 4 mm) and a portion of the sludge was thickened using a Himac (Tokyo, Japan) centrifuge. Solids concentration in feed sludge included: 3.7% TS (unadjusted solids content), 6.0% TS, 8.1% TS and 9.8% TS. To obtain accurate total solids concentration, sludge was later re-suspended with collected supernatant. Before feeding digesters, the

sludge was kept refrigerated at 4 °C for up to one week. The pH of raw sludge and digesters was not adjusted. The temperature of the waterbaths was controlled through Isotemp2100 circulators (Fisher Scientific, Ottawa, Ontario).

Biogas was collected and measured daily using liquid displacement method in calibrated air-tight vessels. The liquid was saturated sodium chloride with 5% sulphuric acid and methyl orange to prevent gas from dissolving. Biogas composition – methane (CH<sub>4</sub>) and carbon dioxide (CO<sub>2</sub>) were measured using GOW MAC Model 550 gas chromatograph with helium as a gas carrier. Hydrogen (H<sub>2</sub>) was measured using the same gas chromatograph with nitrogen as a carrier gas.



**Figure 5 - 8. Layout of the experimental setup. All reactors operated for 5 days in batch mode.**

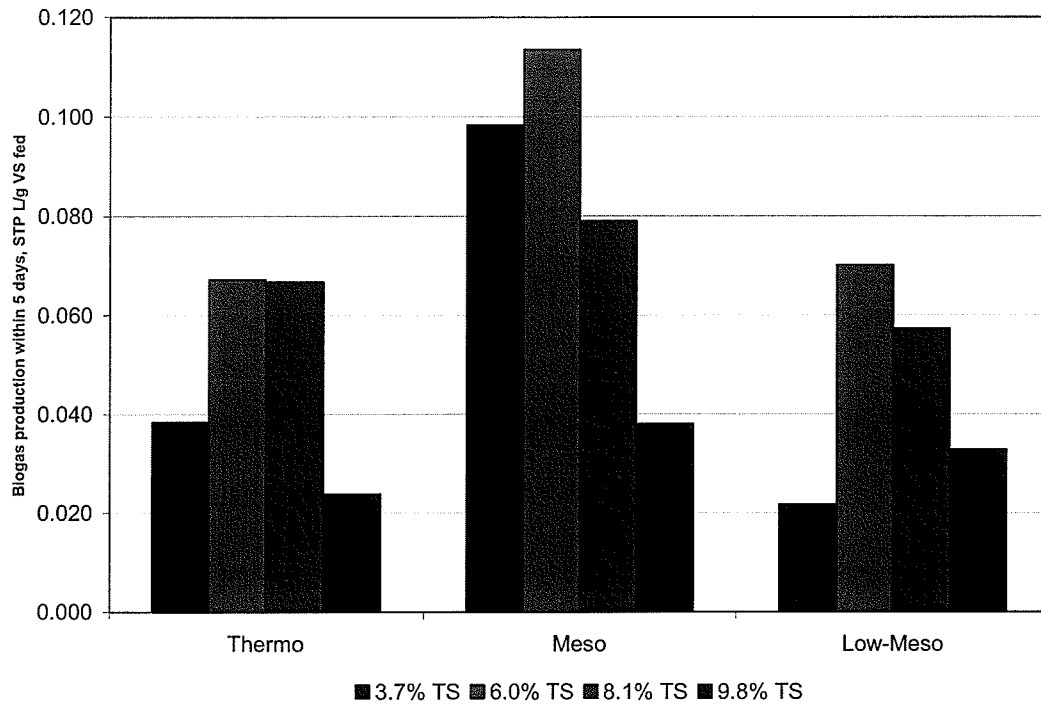
Energy from biogas was calculated according to the equation [5- 1].

**Equation 5 - 1** Energy from digester biogas = Volume of Biogas (CH<sub>4</sub> + CO<sub>2</sub> + H<sub>2</sub>) x  
 (%CH<sub>4</sub> x 34,000 kJ/L + %H<sub>2</sub> x 10,200 kJ/L) / Volume of digester

## RESULTS AND DISCUSSION

Experiments resulted in the following findings:

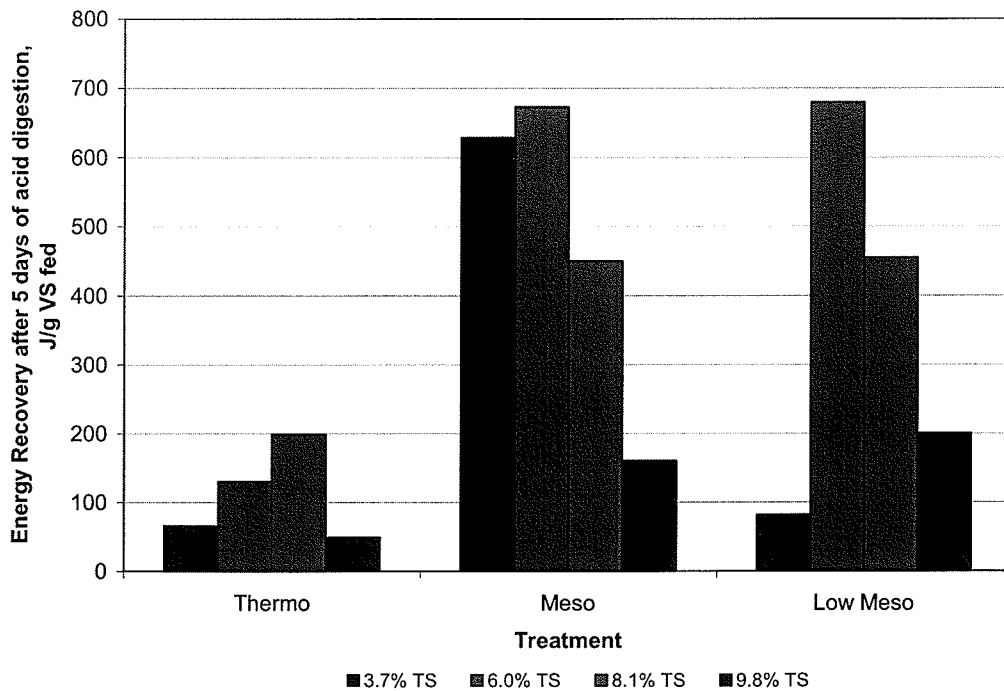
- feasible solids content in sludge was found to be up to 6% TS. Thickening the sludge to 8% TS did not lead to any increase in biogas production (in mesophilic and low-mesophilic temperatures) and sludge at 10% TS showed less biogas than 6% TS and 8% TS (Figure 5-9). These findings are in agreement with findings from the hydrolysis study and suggest that there is limitation most probably due to diminished hydrolysis rates.
- Hydrogen production occurred only in thermophilic treatment (55°C) mostly within first day of the treatment as shown in Table 5-6.
- The overall amount of recoverable energy from biogas (in the form of methane or hydrogen or both) is presented in Figure 5-10.
- Estimated hydrogen recovery as COD presented a very small fraction of initial COD in feed sludge as presented in Table 5-3. Highest hydrogen recovery, measured as a fraction of COD associated with hydrogen gas of total COD associated with digester feed, showed that at approximately 8.1% TS in feed sludge it would be possible to recover 0.22 % COD in the form of hydrogen gas.



**Figure 5 - 9. Biogas production per gram VS fed in form of methane, carbon dioxide and hydrogen after 5 days of batch treatment at three different temperatures and four solids content in feed sludge.**

**Table 5 - 2. Gas composition from batch acid digestion.**

| Gas composition - Thermophilic acid digesters   |      |      |         |      |      |         |      |      |         |      |      |
|---|------|------|---------|------|------|---------|------|------|---------|------|------|
| 3.7% TS   |      |      | 6.0% TS |      |      | 8.1% TS |      |      | 9.8% TS |      |      |
| CH4   | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   |
| 0.0   | 84.1 | 15.9 | 0.0     | 85.3 | 14.7 | 0.0     | 80.8 | 19.2 | 0.0     | 81.4 | 18.6 |
| 0.0   | 88.9 | 11.1 | 0.0     | 84.4 | 15.6 | 0.0     | 84.0 | 16.0 | 0.0     | 80.9 | 19.1 |
| 0.0   | 93.4 | 6.6  | 0.0     | 87.1 | 12.9 | 0.0     | 85.6 | 14.4 | 0.0     | 83.7 | 16.3 |
| 3.9   | 94.1 | 2.0  | 6.4     | 88.6 | 5.0  | 8.0     | 84.4 | 7.6  | 3.4     | 86.5 | 10.1 |
| 7.4   | 91.9 | 0.8  | 8.6     | 88.3 | 3.1  | 15.6    | 81.2 | 3.2  | 11.3    | 85.0 | 3.7  |
| Gas composition - Mesophilic acid digesters     |      |      |         |      |      |         |      |      |         |      |      |
| 3.7% TS   |      |      | 6.0% TS |      |      | 8.1% TS |      |      | 9.8% TS |      |      |
| CH4   | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   |
| 34.8  | 65.2 | 0.0  | 30.8    | 69.2 | 0.0  | 31.2    | 68.8 | 0.0  | 15.5    | 84.5 | 0.0  |
| 39.2  | 60.8 | 0.0  | 38.8    | 61.2 | 0.0  | 33.4    | 66.6 | 0.0  | 33.1    | 66.9 | 0.0  |
| 37.8  | 62.2 | 0.0  | 34.3    | 65.7 | 0.0  | 31.8    | 68.2 | 0.0  | 35.4    | 64.6 | 0.0  |
| 40.1  | 59.9 | 0.0  | 37.0    | 63.0 | 0.0  | 33.3    | 66.7 | 0.0  | 16.4    | 83.6 | 0.0  |
| 40.8  | 59.2 | 0.0  | 42.9    | 57.1 | 0.0  | 35.9    | 64.1 | 0.0  | 15.9    | 84.1 | 0.0  |
| Gas composition - Low-mesophilic acid digesters |      |      |         |      |      |         |      |      |         |      |      |
| 3.7% TS   |      |      | 6.0% TS |      |      | 8.1% TS |      |      | 9.8% TS |      |      |
| CH4   | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   |
| 6.6   | 93.4 | 0.0  | 27.5    | 72.5 | 0.0  | 16.5    | 83.5 | 0.0  | 5.3     | 94.7 | 0.0  |
| 8.6   | 91.4 | 0.0  | 31.2    | 68.8 | 0.0  | 25.7    | 74.3 | 0.0  | 21.2    | 78.8 | 0.0  |
| 11.7  | 88.3 | 0.0  | 28.0    | 72.0 | 0.0  | 24.5    | 75.5 | 0.0  | 21.4    | 78.6 | 0.0  |
| 12.7  | 87.3 | 0.0  | 27.1    | 72.9 | 0.0  | 24.8    | 75.2 | 0.0  | 22.4    | 77.6 | 0.0  |
| 13.8  | 86.2 | 0.0  | 26.2    | 73.8 | 0.0  | 24.5    | 75.5 | 0.0  | 21.5    | 78.5 | 0.0  |



**Figure 5 - 10. Energy recovery in form of methane and hydrogen after 5 days of batch treatment at three different temperatures and four solids content in feed sludge.**

**Table 5 - 3. Hydrogen recovery in form of COD as a percentage of COD in feed.**

| Sludge type                             | Units         | 3.7% TS | 6.0% TS | 8.1% TS | 9.8% TS |
|---|---------------|---------|---------|---------|---------|
| COD                                     | <i>mg/L</i>   | 30272   | 57438   | 69299   | 91136   |
| Volume                                  | <i>L</i>      | 1.8     | 1.8     | 1.8     | 1.6     |
| Total COD In                            | <i>mg COD</i> | 54489   | 103389  | 124738  | 145818  |
| Total amount of hydrogen recovered      | <i>L</i>      | 0.093   | 0.248   | 0.374   | 0.208   |
| Total COD recovered in form of hydrogen | <i>mg COD</i> | 67      | 179     | 269     | 150     |
| COD recovery in form of hydrogen        | <i>%</i>      | 0.12    | 0.17    | 0.22    | 0.10    |

## CONCLUSIONS

Hydrogen production occurred only in thermophilic acid digestion. In all other cases the conditions still lead to active methanogenesis removing the H<sub>2</sub> and a portion of the CO<sub>2</sub>. It decreased steadily within first five days of acid digestion and after three days of thermophilic acid digestion the production of methane began (as shown in Table 5-2). The amount of energy in the form of biogas (both hydrogen and methane component) from thermophilic acid digesters was the lowest compared to mesophilic and low-mesophilic conditions. It was calculated that highest hydrogen recovery amounted to 0.22% on the COD basis. Lowest energy recovery from thermophilic treatment was a result of the overall lowest biogas production from thermophilic reactors compared with mesophilic and low-mesophilic reactors and low concentration of hydrogen in biogas. It can be concluded that due to low concentration of hydrogen in the biogas and overall low energy recovery from thermophilic acid digestion, this type of treatment is not an attractive option for energy recovery through hydrogen production.

## **6 Acid digestion and pathogen inactivation**

This section is composed of three journal publications and one unpublished section on inactivation of pathogens during acid digestion process:

- Initial study on pathogen inactivation through un-ionized VFA action. This part is based on the journal publication: Puchajda, B., Oleszkiewicz, J. A., Sparling, R. and Reimers, R. S. (2005) Low Temperature Inactivation of Fecal Coliforms in Sludge Digestion. *Water Environment Research*, accepted for publication.
- Continuation of the study, paper presents results from batch and two-stage acid digestion systems. This part is based on the journal publication: Puchajda, B. and Oleszkiewicz, J. A. (2005) Extended acid-digestion for fecal coliforms inactivation. *Water Environment Research*, accepted for publication.
- This paper contains results from three-stage acid digestion experiments. This part is based on the journal publication: Puchajda, B. and Oleszkiewicz, J. A. (2005) Extended acid-gas digestion for Class A. *Water Research*, submitted for publication.
- Unpublished section on experimental results from study on comparison between pressurized and not pressurized batch acid digesters operated at mesophilic and low-mesophilic conditions.

Raw data for all results presented in section 6.0 can be found in Appendix C.

## 6.1 Low-temperature inactivation of fecal coliforms

### LOW TEMPERATURE INACTIVATION OF FECAL COLIFORMS IN SLUDGE DIGESTION

Bartek Puchajda, Jan Oleszkiewicz, Richard Sparling and Robert Reimers

#### ABSTRACT

The goal of this research was to demonstrate the ability to achieve Class A pathogen standards in non thermophilic acid digesters. It was proposed that the key mechanism responsible for fecal coliform inactivation was the presence of un-ionized volatile fatty acids. Lab-scale acid digesters were assembled and operated in a batch mode for 5 days at mesophilic (38 °C) and low-mesophilic (21 °C) temperatures and at different solids concentrations. The key issue recognized for successful pathogen inactivation was pH, which is the main factor driving the shift in organic acids toward the un-ionized form. Compared to conventional mesophilic acid digestion, low-mesophilic acid digestion was effective in fecal coliforms inactivation because the process maintained lower pH throughout the duration of the experiment, offered continuous release of organic acids, and showed higher concentrations of organic acids in un-ionized form, including acetate, propionate, butyrate, and valerate.

#### INTRODUCTION

According to current U.S. Environmental Protection Agency regulations, sludge is considered as Class A if, according to Alternative 1, time-temperature requirements are met (which, in practice, limits Class A to thermophilic processes) and, according to Alternative 2, sludge is treated in high pH and high temperature processes (which, in practice, means a pH level higher than 12 and a temperature above 52 °C) (E.P.A., 1999). None of these conditions



exist in acid digesters operated at non thermophilic temperatures. When these processes cannot fulfill Alternatives 1 or 2, Class A compliance can be achieved according to Alternative 3 through comprehensive monitoring of bacteria, enteric viruses, and viable helminth ova to demonstrate adequate reduction of pathogens.

Pathogen inactivation caused by organic acids during anaerobic digestion has been reported since the early 1960s (Jarrel et al., 1987, Koster and Cramer, 1987, McCarty and McKinney, 1961). Some researchers reported that, in addition to organic acids, the presence of alcohols, aldehydes, ketones, and so on contributes to pathogen inactivation (McCarty and McKinney, 1961, Reimers et al., 1999, Wilson et al., 2002). All of these compounds are intermediate products of sludge fermentation. However, organic acids are present in high concentrations in the acid digester of the acid–gas digestion system. An acid digester operated with a short solids retention time (SRT) that is usually between 2 and 5 days (Wilson and Streicher, 2001) only allows for hydrolysis and acidification (fermentation) of sludge. Under these conditions when organic acids are not used by methanogens, they tend to accumulate in this digester (commonly called an acid digester) at concentrations above 6,000 mg/L (Wilson and Streicher, 2001) and up to more than 13,000 mg/L (Ghosh, 2003). Configuration of the process, where an acid digester is followed by a gas digester, has become an increasingly used solution after it was shown that acid digestion can improve overall performance of anaerobic digestion in terms of volatile solids destruction, biogas yield, and stability of the whole process.

For additional information see section 2.11 and 2.12.

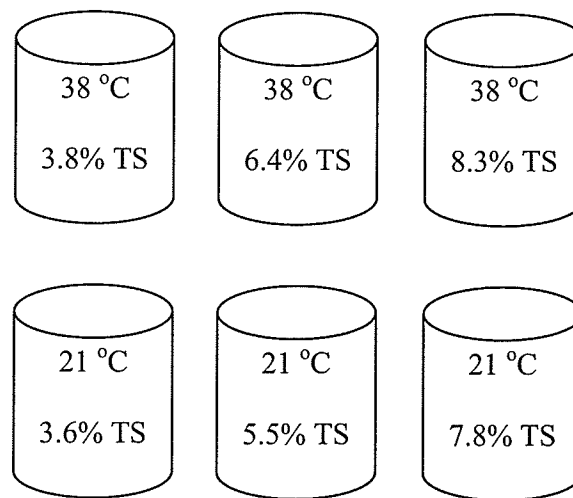
## OBJECTIVES

The objective of this experiment was to assess the ability of acid-phase digestion at achieving fecal coliform inactivation through the presence of organic acids in un-ionized form at temperatures lower than thermophilic. The following two operating temperatures were chosen: mesophilic (38 °C) and low mesophilic (21 °C). The experimental objectives also included the effect of sludge concentration on the release of organic acids as well as its effect on sludge pH and the composition of VFA.

## METHODOLOGY

Acid-phase digesters were operated in a batch mode for 5 days. Source of sludge as in section 4.0. Digester volumes were 1.6 to 1.8 L, and approximately 50 ml of sludge was removed every day for analyses. Digesters were mixed manually 3 times per day; the pH was not adjusted. The temperature of the waterbaths was controlled through Isotemp 2100 circulators (Fisher Scientific, Ottawa, Ontario).

The main variables were temperature of the process and concentration of total solids in sludge fed to digesters, as shown in Figure 6-1.



**Figure 6 - 1. Experimental setup: acid-phase digesters with varying operating temperatures and solids concentration.**

Total solids were measured every day according to *Standard Method* 2540 B (APHA et al., 1998). Volatile fatty acids including acetic, propionic, i-butyric, n-butyric, i-valeric, n-valeric, i-caproic and n-caproic, enanthic, n-caprylic, pelargonic, and n-capric (C2 to C10) were measured every day using a gas chromatograph Hewlett-Packard (Palo Alto, California) 5890A with flame ionization detector (FID). The FID detector consists of a small hydrogen-air diffusion flame. When a sample containing organic compounds (including organic acids) is introduced into the flame from the column, electrically charged species (ions) are formed that are later collected by applying voltage across the flame (Grob, 1985). Before analysis, VFA samples were stored in the freezer. The un-ionized VFA fraction, which existed in the digester, was calculated based on the equation [1-2] by Perrin et al., (1981):

$$\text{Equation 1 - 2 } \frac{\text{base}}{\text{acid}} = 10^{\text{pH}-\text{pK}}$$

where: base = fraction of ionized organic acid; acid = fraction of un-ionized organic acid (u-VFA); and pK = negative logarithm of acid ionization constant.

The pK values adopted from Kortöm et al., (1961) for calculation of u-VFA concentrations are presented in Table 6-1.

pH was measured every day using pH electrode Accumet (Fisher Scientific, Ottawa, Ontario) AP 50. Total coliforms were measured on day 0, 1, 2, 3, and 5 according to *Standard Method* 9221 B (APHA et al., 1998) and then positive samples were tested for fecal coliform presence according to *Standard Method* 9221 E (APHA et al., 1998). The three-tube method was used and the most probable number was obtained according to Finstein (1972).

**Table 6 - 1. pK values of organic acids.**

| <i>Acid</i>                | <i>Formula</i>                                       | <i>Number of Carbon atoms</i> | <i>pK</i> |
|----------------------------|--|-------------------------------|-----------|
| Acetic acid                | CH <sub>3</sub> COOH                                 | 2                             | 4.757     |
| Propionic acid (propanoic) | CH <sub>3</sub> CH <sub>2</sub> COOH                 | 3                             | 4.874     |
| n-Butyric acid (butanoic)  | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>2</sub> COOH | 4                             | 4.812     |
| i-Butyric acid (butanoic)  | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>2</sub> COOH | 4                             | 4.840     |
| n-Valeric acid (pentanoic) | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>3</sub> COOH | 5                             | 4.835     |
| i-Valeric acid (pentanoic) | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>3</sub> COOH | 5                             | 4.767     |
| n-Caproic acid (hexanoic)  | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>4</sub> COOH | 6                             | 4.849     |
| i-Caproic acid (hexanoic)  | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>4</sub> COOH | 6                             | 4.853     |
| Enanthic acid (heptanoic)  | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>5</sub> COOH | 7                             | 4.893     |
| Caprylic acid (octanoic)   | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>6</sub> COOH | 8                             | 4.894     |
| Pelargonic acid (nonanoic) | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>7</sub> COOH | 9                             | 4.955     |
| Capric acid (decanoic)     | CH <sub>3</sub> (CH <sub>2</sub> ) <sub>8</sub> COOH | 10                            | 4.845     |

## RESULTS AND DISCUSSION

Fecal coliform profiles during acid-digestion treatment are presented in Figures 6-2 and 6-3. Fecal coliform destruction at mesophilic and low-mesophilic conditions cannot be explained by temperature action. It is hypothesized that inactivation was caused by the un-ionized fraction of the organic acids present in these digesters. There are three important issues associated with pathogen inactivation through un-ionized VFA: (1) the concentration of VFA shown in Table 6-2, (2) pH shown in Table 6-3, and (3) the duration of these conditions in the digester. The first two factors contribute to the concentration of u-VFA. Profiles comparing u-VFA concentrations in mesophilic and low-mesophilic conditions are shown in Figures 6-4, 6-5, and 6-6.

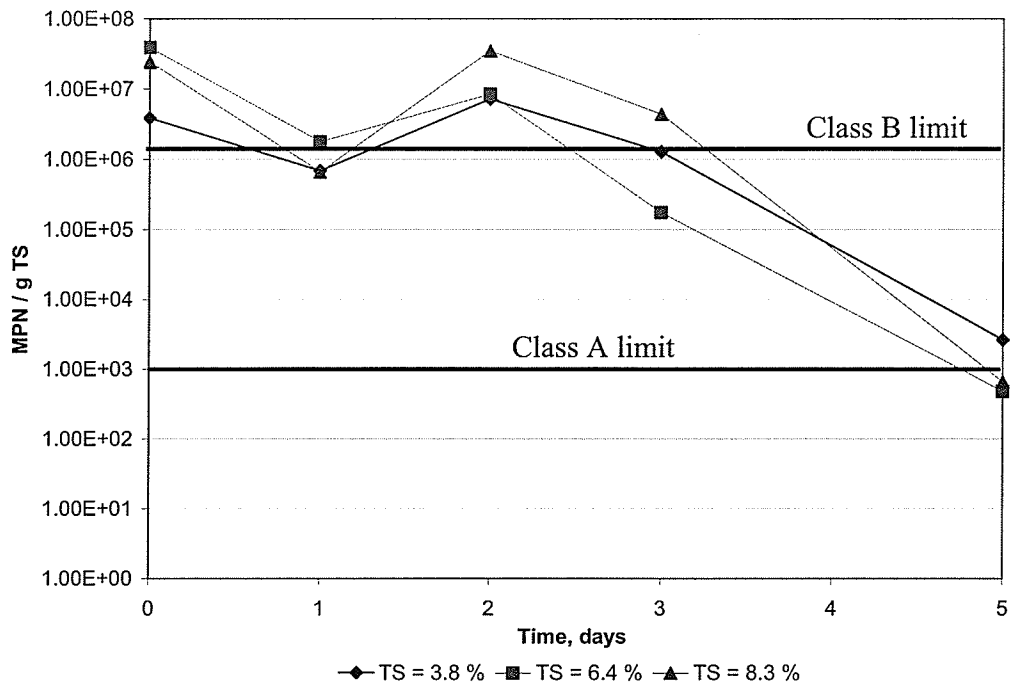


Figure 6 - 2. Fecal coliform densities in the outflow from the acid-phase reactor operated at mesophilic temperature (38 °C).

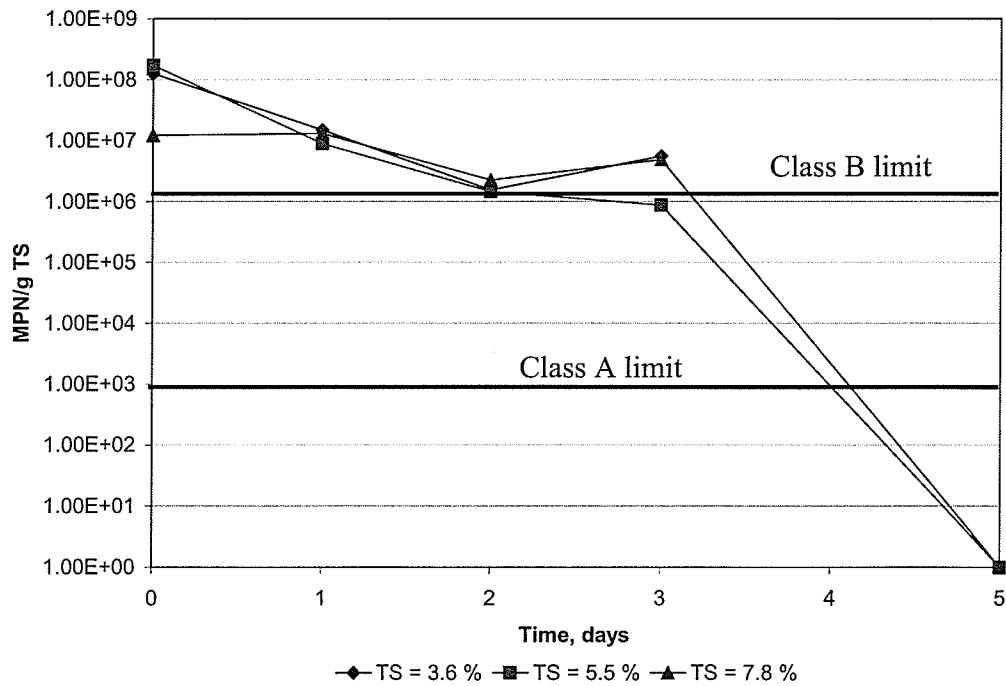


Figure 6 - 3. Fecal coliform densities in the outflow from the acid-phase reactor operated at low-mesophilic temperature (21 °C).

**Table 6 - 2. Total VFA concentration in mesophilic and low-mesophilic acid digesters.**

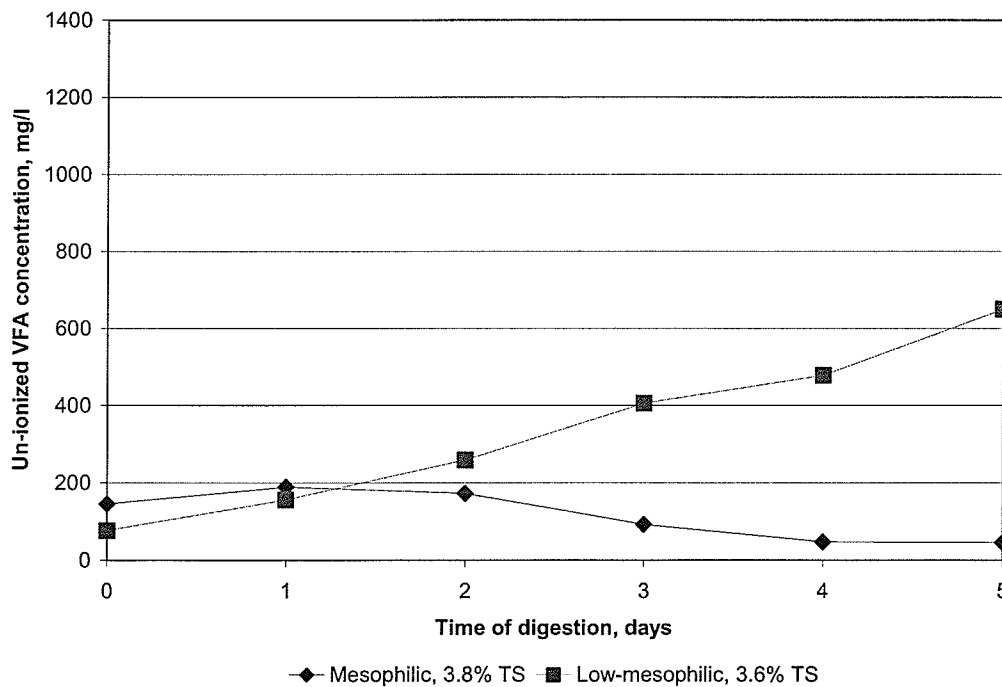
| Days | Total VFA                 |            |            |                               |            |            |
|------|---------------------------|------------|------------|-------------------------------|------------|------------|
|      | Mesophilic acid digesters |            |            | Low-mesophilic acid digesters |            |            |
|      | TS = 3.8 %                | TS = 6.4 % | TS = 8.3 % | TS = 3.6 %                    | TS = 5.5 % | TS = 7.8 % |
|      | mg/l                      | mg/l       | mg/l       | mg/l                          | mg/l       | mg/l       |
| 0    | 2840                      | 2833       | 2963       | 1111                          | 1649       | 1528       |
| 1    | 3269                      | 3699       | 3966       | 1710                          | 2499       | 3114       |
| 2    | 3297                      | 4625       | 7118       | 2047                          | 3544       | 4059       |
| 3    | 2997                      | 5235       | 7182       | 2916                          | 3511       | 4705       |
| 4    | 2873                      | 5327       | 6081       | 3115                          | 3904       | 6194       |
| 5    | 3030                      | 4654       | 6050       | 3564                          | 4777       | 7020       |

TS = total solids

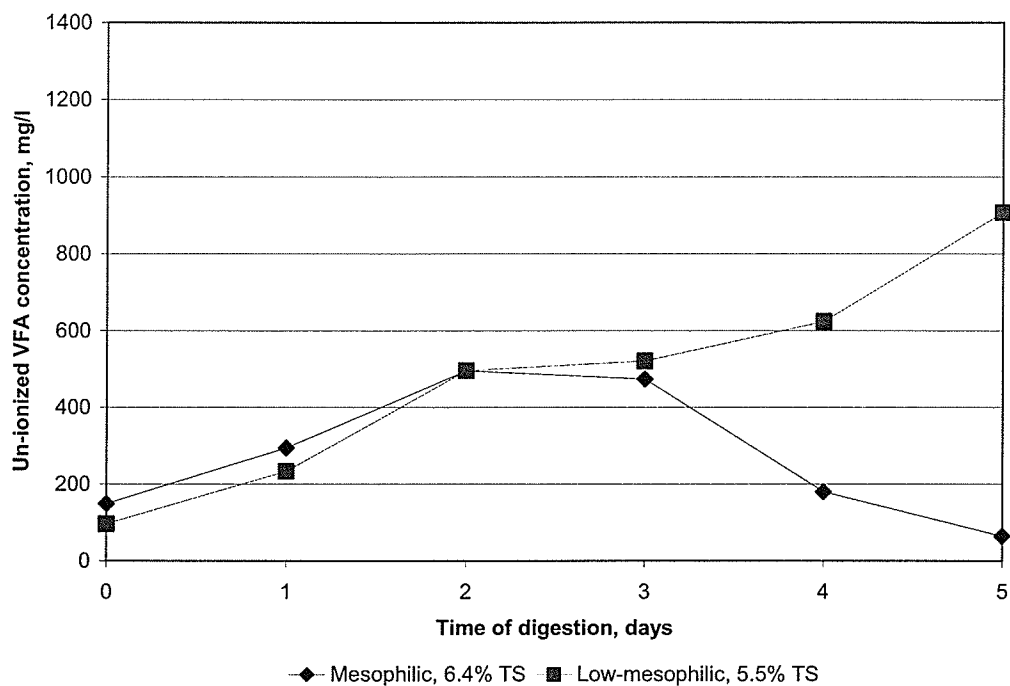
**Table 6 - 3. pH in acid digesters.**

| Day | pH                        |            |            |                               |            |            |
|-----|---------------------------|------------|------------|-------------------------------|------------|------------|
|     | Mesophilic acid digesters |            |            | Low-mesophilic acid digesters |            |            |
|     | TS = 3.8 %                | TS = 6.4 % | TS = 8.3 % | TS = 3.6 %                    | TS = 5.5 % | TS = 7.8 % |
| 0   | 5.92                      | 5.91       | 5.97       | 5.93                          | 6.01       | 6.03       |
| 1   | 5.87                      | 5.71       | 5.97       | 5.80                          | 5.79       | 5.84       |
| 2   | 5.91                      | 5.55       | 5.66       | 5.64                          | 5.59       | 5.61       |
| 3   | 6.14                      | 5.65       | 5.68       | 5.59                          | 5.56       | 5.56       |
| 4   | 6.40                      | 6.10       | 6.05       | 5.54                          | 5.52       | 5.48       |
| 5   | 6.44                      | 6.48       | 6.40       | 5.45                          | 5.43       | 5.42       |

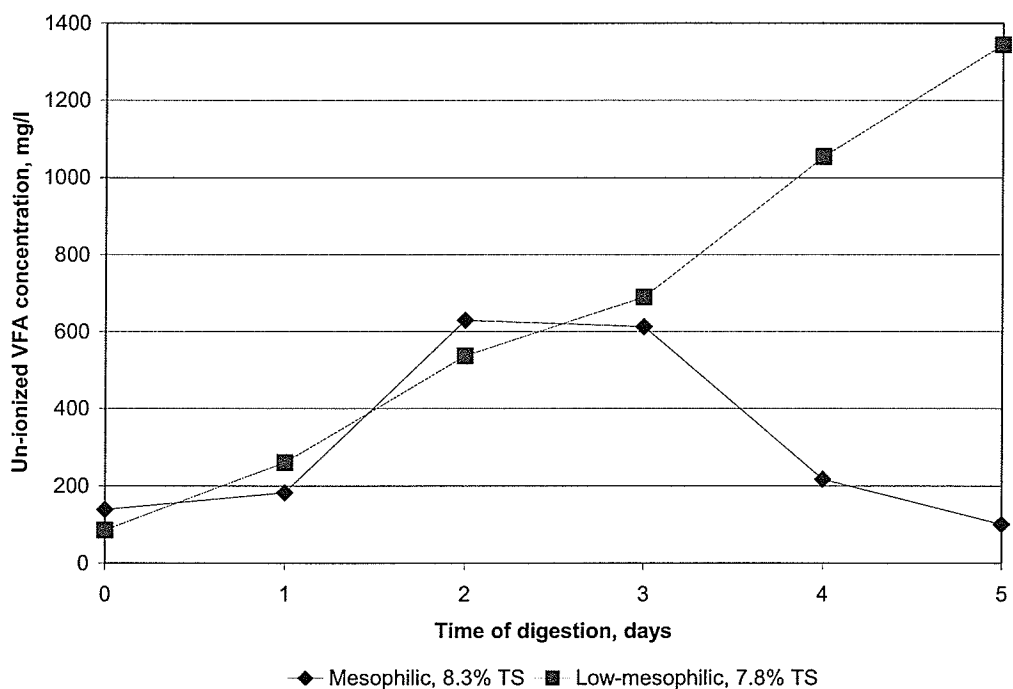
TS = total solids



**Figure 6 - 4. Un-ionized VFA concentration in acid digesters with approximately 3.7% total solids in the feed sludge.**



**Figure 6 - 5. Un-ionized VFA concentration in acid digesters with approximately 6% total solids in the feed sludge.**



**Figure 6 - 6. Un-ionized VFA concentration in acid digesters with approximately 8% total solids in the feed sludge.**

The key factors that contributed to inactivation of fecal coliforms were VFA concentration, pH, which contributed to the formation of un-ionized VFA, and the duration of treatment. Digesters operated at low-mesophilic temperature showed continuous increase in VFA concentration throughout the duration of the experiment. In the case of digesters operated under mesophilic conditions, the VFA concentration resembled the pattern seen for u-VFA release, with a peak concentration between day 2 and day 4 (Table 6-2). At the same time, mesophilic digesters exhibited much higher methane production (data not shown). Because most methanogenic bacteria are mesophiles (Gerardi, 2003), it is reasonable to assume that at 38<sup>0</sup>C the longer retention times were sufficient to allow reestablishment of an active methanogenic populations leading to a faster and higher rate of methane production which resulted in VFA use and led to the increase in pH in mesophilic conditions. Thus, the inhibition caused by u-VFA was considerably shortened and mitigated. Gas production correlates well with a drop in concentration of acetate in mesophilic conditions, whereas, in low-mesophilic conditions, concentration of acetate continuously increased (Figures 6-7 and 6-8).

Methanogens are also affected by pH, the optimum value of which is between 6.5 and 8.0 (van Lier et al., 2001). Ghosh et al., (1999) found that the optimum pH for acidification (fermentation) of sludge was approximately 5.7. Similarly, Zoetemeyer et al., (1982b) found that the most stable acid digestion occurred at pH 5.7 to 6.0. These facts could explain that at pH lower than 6.0 organic acids accumulate inside digester and sufficient concentration and contact time may cause fecal coliforms inactivation.

Profiles of u-VFA reveal important differences that contributed to inactivation of fecal coliforms in low-mesophilic conditions. A continuous increase of u-VFA was calculated for

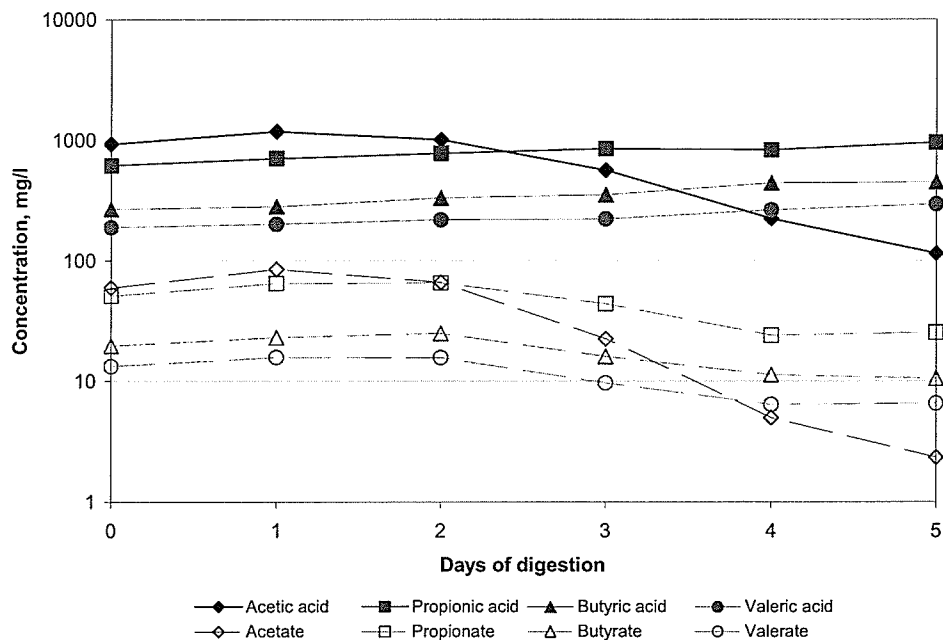


digesters operated at 21 °C, whereas digesters operated at 38 °C after the initial increase showed a decline in u-VFA concentration. In mesophilic conditions, the pH after feeding reactors dropped somewhat during the first 2 to 3 days and later increased to values above 6.0. At the same time, pH in low-mesophilic reactors stayed at approximately 5.5 (Table 6-3). This created a situation in which mesophilic conditions provided harmful conditions for fecal coliforms for only a few days, whereas, in low-mesophilic conditions, fecal coliforms were constantly under stress from increasing concentrations of u-VFAs throughout the 5 days. It is essential to maintain pH in a digester below 6.0 (around 5.5) otherwise the fraction of un-ionized VFA is so low that effective pathogen destruction may be impossible to achieve even at high VFA concentrations.

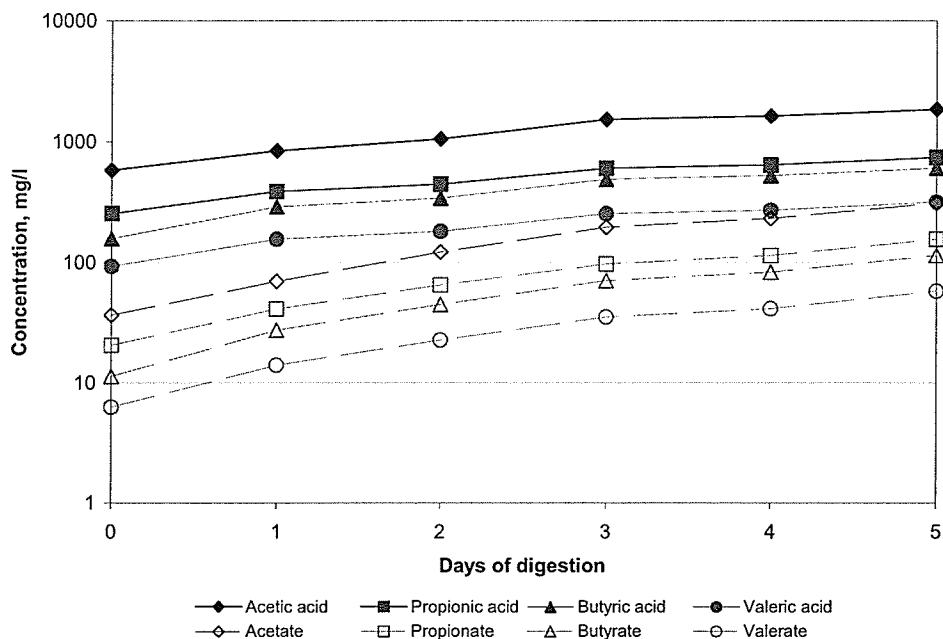
Increased solids content in sludge due to thickening resulted in higher VFA concentrations in all cases (Table 6-2). However, increased VFA concentrations exhibited little effect on sludge pH (Table 6-3). Therefore, the temperature of acid digestion in relation to faster startup and higher activity of the methanogenic community is a decisive factor in maintaining low pH.

Another aspect of un-ionized VFAs toxicity is the composition of VFAs. Theoretically, the longer the carbon length the more harmful effect is brought by weak acid. Investigation of VFAs composition included organic acids with carbon length C2 to C10. Acids higher than C6 were not detected. It is most likely attributed to their low solubility. The solubility of organic acids is proportional to carbon length and acetic acid (C2) is approximately 250 times more soluble than caproic acid (C6) at 25 °C (Pryde, 1979). An important aspect of organic acid composition in relation to temperature of acid digestion was described by Zoetemeyer et al., (1982a). The predominant products of acidification at mesophilic

temperatures are acetic and propionic acid, whereas, at low-mesophilic temperatures, production of butyric acid is favourable. In terms of inhibitory effect, butyric acid has higher carbon length (C4) than acetic (C2) and propionic (C3) acids and, therefore, may be more effective in pathogen inactivation during acid digestion. The distribution of the four major acids, acetic, propionic, butyric (sum of i-butyric and n-butyric), and valeric (sum of i-valeric and n-valeric), showed that acid digesters operated at low-mesophilic temperatures resulted in much higher concentrations of un-ionized fractions of those acids than digester operation at mesophilic temperature (the dashed lines in Figures 6-7 and 6-8). Trends shown in Figures 6-7 and 6-8 for the lowest sludge concentration were also true at the higher solids concentration in sludge (Table C-4 and C-5 in the Appendix C). Therefore, this feature of the acid digester operated at low-mesophilic temperature must contribute to improved fecal coliform inactivation.



**Figure 6 - 7. Concentrations of four major organic acids and their un-ionized fractions in a mesophilic digester with 3.8% total solids in the feed sludge. These acids represent 99.2% of total VFA concentration.**



**Figure 6 - 8. Concentrations of four major organic acids and their un-ionized fractions in a low-mesophilic digester with 3.6% total solids in the feed sludge. These acids represent 98.8% of total VFA concentration.**

## CONCLUSIONS

This experiment demonstrated the ability of low-mesophilic acid digestion on the inactivation of fecal coliforms through the presence of organic acids in un-ionized form. The key issue recognized for successful fecal coliforms inactivation was pH, which is the main factor driving the shift in organic acids toward un-ionized form. Low-mesophilic digestion (21 °C) exhibited a drop in pH without later increase, as in the case of mesophilic (38 °C) digesters. In addition, a continuous increase in concentration of VFA was observed in low-mesophilic conditions, whereas mesophilic conditions achieved a peak VFA concentration within 2 to 4 days. Both of these factors (i.e., low pH and continuous increase in VFA concentration) contributed to attaining Class A levels of fecal coliform in all reactors operated at low-mesophilic conditions (21 °C) after 5 days of treatment operated in a batch mode. The pH in these digesters after 5 days was approximately 5.5, with almost 20% of

VFA in their un-ionized form. Increased solids content in feed sludge resulted in higher VFA concentration, but did not affect the sludge pH. Analysis of the composition of VFA showed that organic acids higher than C6 were not detected because of their low solubility. Low-mesophilic digestion exhibited a higher concentration of butyrate (only slightly lower than propionate), which may have contributed to higher pathogen inactivation because higher carbon length may be more toxic to bacteria. Low-mesophilic acid digestion was more effective at fecal coliform inactivation compared to conventional mesophilic acid digestion because the process:

- maintained lower pH throughout the duration of the experiment;
- offered continuous release of organic acids;
- did not exhibit a decrease in acetate, which was probably caused by methane production (data not shown); and
- showed higher concentrations of organic acids in un-ionized form including acetate, propionate, butyrate, and valerate.

#### ACKNOWLEDGMENTS

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## 6.2 Extended acid digestion for fecal coliforms inactivation

### EXTENDED ACID-DIGESTION FOR FECAL COLIFORMS INACTIVATION

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#### ABSTRACT

The objective of this research was to establish the correlation between inactivation of fecal coliforms caused by organic acids in their un-ionized form in batch acid-digesters and semi-continuously fed acid-digesters at both mesophilic (38°C) and low-mesophilic (24°C) temperatures. Batch acid digesters achieved Class A level of fecal coliforms within 6 to 7 days of digestion at both temperatures. Semi-continuously fed, staged acid digestion systems achieved Class A standard on average only at mesophilic temperature at SRT of 11 days. Systems operated at low-mesophilic temperature did not achieve Class A. It was attributed to lower diffusion rates most probably caused by lower vapour pressure of organic acids at lower temperatures.

#### INTRODUCTION

Inactivation of pathogens found in sewage sludge becomes an increasingly important task for wastewater treatment plants (WWTP). Anaerobic digestion, one of the most commonly applied methods for sewage sludge stabilization, may achieve high level of pathogen destruction which depends mainly on the temperature of the process. Sewage sludge is classified as a Class A product if, according to Alternative 1, treatment complies with "time-temperature" relationship defined by EPA regulations (E.P.A., 1999). These regulations imply that the temperature of the treatment cannot be less than 50°C. In other words Class A

biosolids quality is in practice restricted to thermophilic processes. However, many WWTP operate their digesters below thermophilic temperatures, in which case the only way to comply with Class A standard is to follow Alternative 3, which is to demonstrate adequate pathogen reductions through comprehensive monitoring of bacteria, enteric viruses and viable helminth ova (E.P.A., 1999). This paper will focus only on one indicator, which is fecal coliform.

Recent advances in digestion technologies, including acid-gas digestion, opened a possibility to provide pathogen destruction without necessary heat/energy input. Organic acids, intermediate products of anaerobic decomposition of wastewater sludge, accumulate in acid digesters in high concentrations up to 13,000 mg/l (Ghosh, 2003, Wilson and Streicher, 2001). Pathogen inactivation caused by organic acids during sludge digestion have been reported since early 1960's (McCarty and McKinney, 1961), and latest publications show that there is growing interest in applying acid digestion for pathogen destruction (Matthews and Asaadi, 2003, Mayhew et al., 2002, Puchajda and Oleszkiewicz, 2004b, Reimers et al., 1999, Salsali et al., 2004, Wilson et al., 2002).

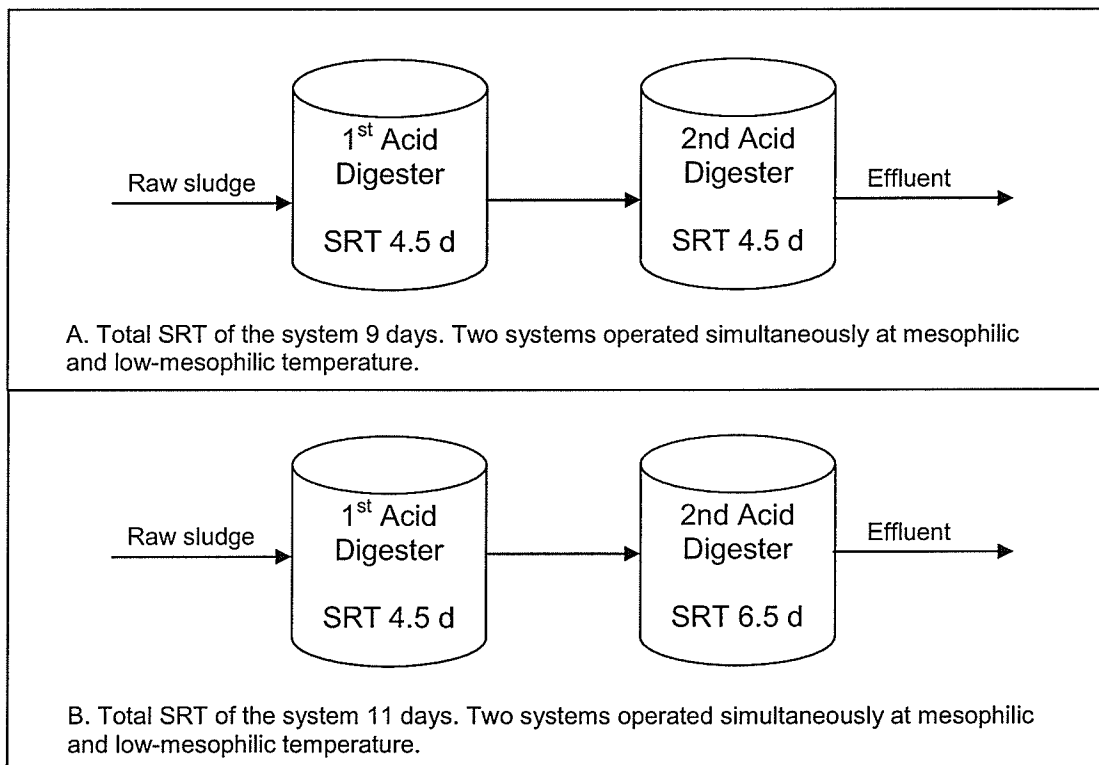
For further information see section 2.11, 2.12, and introduction to section 6.1.

## OBJECTIVES

Previous experimental work (Puchajda et al., 2005) showed potential of low-temperature inactivation of fecal coliforms during batch acid-digestion below Class A limit. The objectives of this research were to establish the correlation between inactivation of fecal coliforms in batch digesters and semi-continuously fed digesters at both mesophilic (38°C) and low-mesophilic temperatures (24°C).

## METHODOLOGY

Acid digesters were operated in batch and semi-continuously fed mode. Batch acid digesters were fed at the beginning of the experiment (total volume of approximately 2.5 l) and a portion of sludge (approximately 50 ml) was sampled on daily basis. Results presented in this paper were averages from 3 runs that were performed in a batch mode. Semi-continuous digesters' operation refers to essentially sequencing batch operation where a portion of completely mixed liquid is removed daily and replaced with raw sludge (Figure 6-9). Results from semi-continuously operated digesters are considered an approximation of continuous systems that are used in full scale operations.



**Figure 6 - 9. Layout of semi-continuously fed, staged acid digestion systems (A – system with total SRT of 9 days; and B – system with total SRT of 11 days).**

Source of sludge as in section 4.0. Additionally, it was estimated that PS:WAS mixture corresponded to approximately 50:50 ratio. At the time of experimentation sludge contained approximately 4.2 % total solids of which 66.3% were volatile solids (Zaleski, 2005). Total solids (TS) were measured according to SM 2540 B (APHA et al., 1998). Volatile fatty acids (VFA) including acetic, propionic, i-butyric, n-butyric, i-valeric, n-valeric, i-caproic and n-caproic (C2 to C6) were measured using gas chromatograph Hewlett-Packard 5890A with flame ionization detector (FID). The FID detector consists of a small hydrogen-air diffusion flame and when a sample containing organic compounds (including organic acids) is introduced into the flame from the column electrically charged species (ions) are formed that are later collected by applying voltage across the flame (Grob, 1985). Before analysis volatile fatty acid samples were stored in the freezer. The unionized VFA fraction, which existed in the digester, was calculated based on equation [1-2] (Perrin et al., 1981).

$$\text{Equation 1 - 2 } \frac{\text{base}}{\text{acid}} = 10^{\text{pH}-\text{pK}}$$

where: base – fraction of ionized organic acid , acid – fraction of un-ionized organic acid (u-VFA), pK – negative logarithm of acid ionization constant

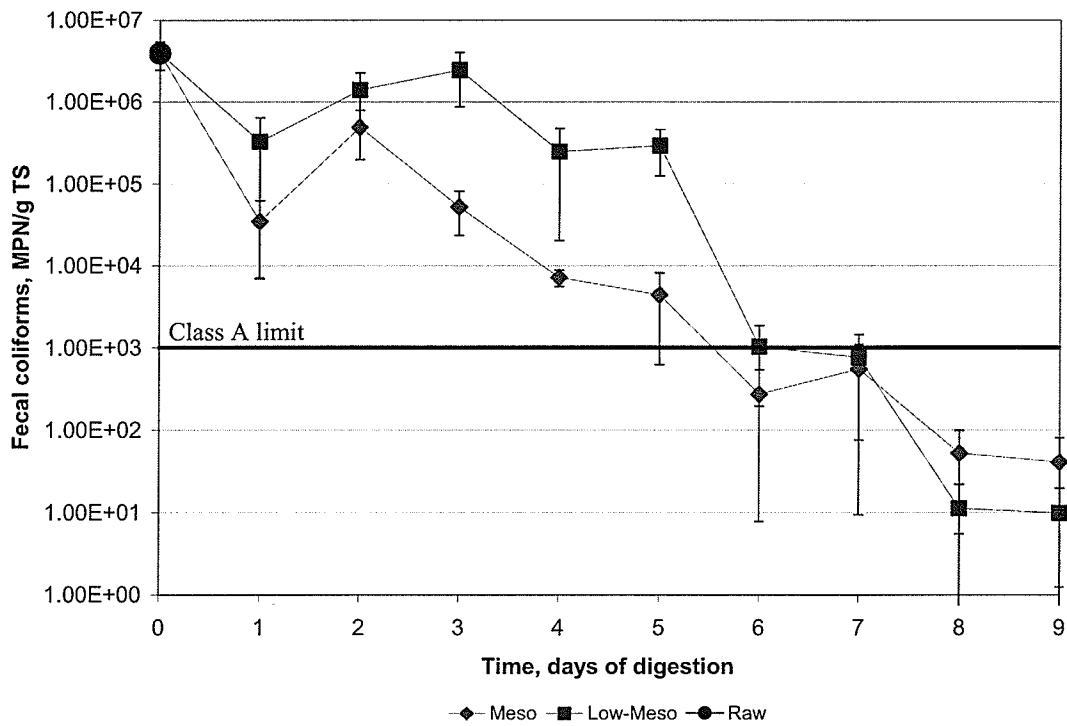
pK values adopted from Kortöm et al., (1961) for calculation of u-VFA concentrations are presented in Table 6-1.

pH was measured using pH electrode Accumet AP 50. Total coliforms were measured according to SM 9221 B (APHA et al., 1998) and then positive samples were tested for fecal coliform presence according to SM 9221 E (APHA et al., 1998). The three-tube method was used and most probable number (MPN) was obtained according to Finstein (1972).



## RESULTS & DISCUSSION

Results from batch digesters' operation showed that Class A limit using fecal coliforms was achieved in both mesophilic and low-mesophilic conditions (Figure 6-10). Experiments were terminated on day 9, which is after Class A limit was achieved consecutively for 3 days.



**Figure 6 - 10. Fecal coliform profiles in the influent and effluent sludge from batch digesters (average from three runs).**

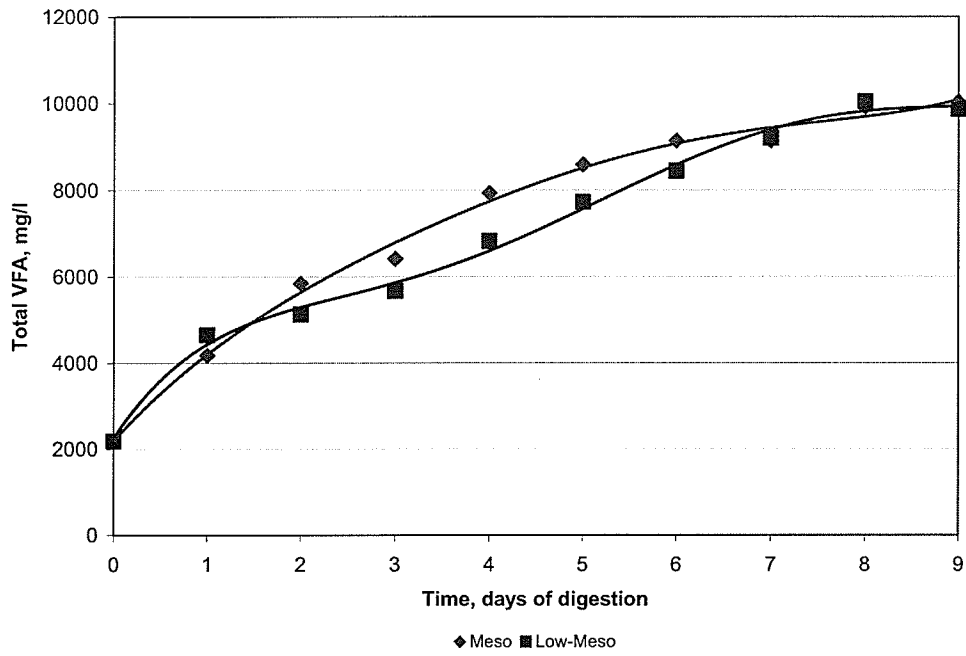


Figure 6 - 11. Total VFA profiles in batch digesters (average from three runs).

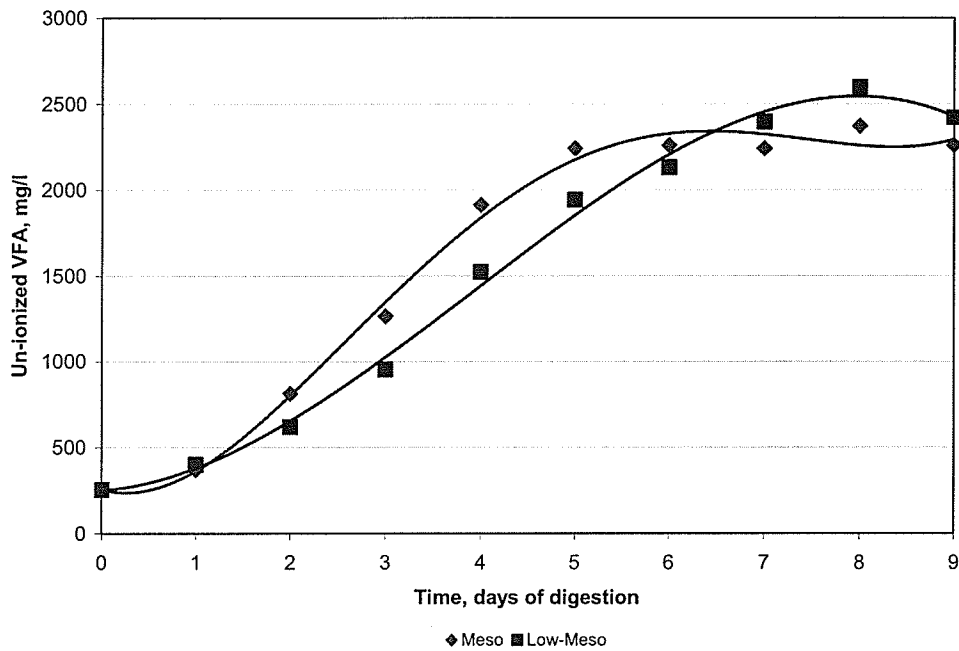
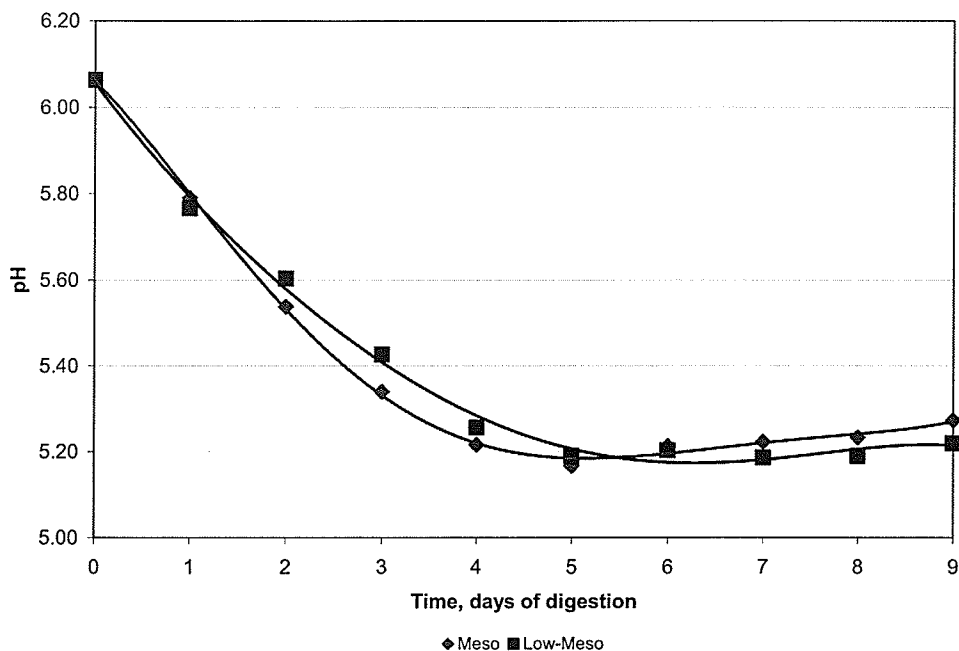


Figure 6 - 12. Un-ionized VFA profiles in batch digesters (average from three runs).



**Figure 6 - 13. pH profiles in batch digesters (average from three runs).**

Fecal coliform profiles correlate well with profiles of un-ionized VFA concentration in digesters (Figure 6-12). Similar U-VFA profiles were attributed to very similar VFA release (Figure 6-11) in both conditions (mesophilic and low-mesophilic) and very similar pH values in digesters (Figure 6-13). In conclusion, this part of the research showed that, increasing concentration of VFA in un-ionized form correlated well with the declining numbers of fecal coliforms in batch digestion. Temperature seemingly did not play a role in fecal coliforms inactivation in batch systems. To achieve Class A level a minimum of 6 to 7 days in batch conditions was required at both temperatures. After this time concentrations of U-VFA of 2300 to 2500 mg/l was observed which corresponded to approximately 25% of total VFA. Following, semi-continuously fed acid digesters were tested (layout of the systems is presented in Figure 6-9 A).

The semi-continuously fed digester system consisted of two-digesters (staged acid digestion) because: it can help prevent short-circuiting (escaping sludge particles to effluent before designed retention time has passed) which is very likely to occur at this retention time; and a two-stage system showed potential of maintaining lower pH values as compared to single acid digester operated at the same retention time which is important due to ionization constant of organic acids. Retention time in the first acid digester was set at 4.5 days which should produce a complete pH drop in this digester based on results from batch tests (Figure 6-13).

In the first experiment, when total SRT of the system was 9 days (4.5 days + 4.5 days in the first and second acid digester, respectively) the effluent concentration of fecal coliforms was much higher than results indicated after batch tests and Class A was not achieved at either temperature (Figure 6-14). When comparing concentrations of U-VFA in semi-continuously fed acid digestion systems to batch systems, it can be seen that much lower concentration of U-VFA were observed in the semi-continuous systems (Table 6-4). This could account for higher concentrations of fecal coliforms found in the effluent of semi-continuously fed systems. Also, a considerable difference was noticed between the mesophilic and low-mesophilic conditions when fecal coliforms concentrations were compared. The difference between low-mesophilic and mesophilic staged acid digestion systems cannot be attributed to almost identical concentrations of U-VFA. Moreover, batch results indicated that temperature did not have any significant effect on inactivation of fecal coliforms. Additional analysis of speciation of organic acids in digesters was performed to detect if different VFA makeup could contribute to higher fecal coliforms inactivation in mesophilic conditions. However, no significant changes were found in composition of VFA as shown in Table 6-5.

Mathews and Asaadi (2003) in similar studies concluded that acid digestion at 24°C had very little effect on fecal coliforms reduction while digestion at 35°C showed much better results. However, no explanation was provided to account for the differences in fecal coliforms inactivation at those temperatures. Even though microbial responses to varying environmental conditions including the presence of organic acids and temperature (which both can be inhibitory at certain level) are very complex there may be a simpler justification to this phenomenon. It was found that increased temperature resulted in increased rate of diffusion of organic acids. An increase from 4°C to 24°C increased diffusion rate (through chitosan-based antimicrobial packaging) of acetic and propionic acids more than twofold (Quattara et al., 2000). The diffusion coefficient itself usually follows an Arrhenius relationship (Tiwari et al., 2005) and increases with an increase in temperature as in equation [6-3].

**Equation 6 - 1**  $D = D_0 \exp \frac{\Delta H}{RT}$

where:

D – diffusion coefficient, and  $D_0$  – maximum diffusion coefficient,  $\Delta H$  – change in enthalpy (constant), and R – universal gas constant, T - temperature

Due to the fact that Class A was not achieved in either system, the retention time in the second acid digester was extended to 6.5 days, which resulted in overall systems SRT of 11 days (Figure 6-9 B). It was assumed that this should result in longer contact time between unionized VFA and fecal coliforms, which should lower their concentrations. Concentrations of U-VFA in acid digesters during this experiment are shown in Table 6-6. U-VFA concentrations increased slightly from previous results (Table 6-4) and extension of SRT in second acid digester resulted in Class A standard on average in the effluent from mesophilic staged acid digestion was achieved (Figure 6-14) with four out of sixteen samples not

complying with Class A standard. It should be noted that for Class A analytical results must not be averaged according to EPA regulations. Therefore, due to non-compliance of four samples Class A was not achieved by the system.

Virtually no improvement was observed under low-mesophilic conditions. As in the case of the previous operational regime, speciation of individual organic acids showed no significant differences in VFA composition (Table 6-7) that could contribute to enhanced pathogen inactivation.

**Table 6 - 4. U-VFA in raw sludge and effluent from acid-digestion system operated at total SRT of 9 days (4.5 + 4.5 days).**

|   | Unit | Raw sludge  | Mesophilic digestion system |             | Low-mesophilic digestion system |             |
|---|------|-------------|-----------------------------|-------------|---------------------------------|-------------|
|   |      |             | 1st reactor                 | 2nd reactor | 1st reactor                     | 2nd reactor |
| Average VFA concentration                           | mg/l | 1601 ± 535  | 6841 ± 975                  | 8294 ± 840  | 6565 ± 1933                     | 7601 ± 1085 |
| Average pH in digester                              | -    | 6.14 ± 0.14 | 5.33 ± 0.07                 | 5.27 ± 0.11 | 5.39 ± 0.14                     | 5.21 ± 0.09 |
| Average un-ionized VFA concentration                | mg/l | 64±21       | 1443 ± 206                  | 1948 ± 197  | 1342 ± 393                      | 1980 ± 283  |
| Average un-ionized VFA concentration for the system | mg/l |             | 1695 ± 202                  |             | 1661 ± 338                      |             |

**Table 6 - 5. Contribution of individual acids to total VFA concentration in raw sludge and effluent from acid-digestion system operated at total SRT of 9 days (4.5 + 4.5 days).**

| Type of sludge   | Acid  |           |           |           |           |           |           |           | Total |
|------------------|---|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-------|
|                  | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic |       |
|                  | <i>Contribution of individual acids to total concentration, %</i> |           |           |           |           |           |           |           |       |
| Raw              | 29.6  | 29.4      | 8.7       | 15.5      | 8.9       | 4.6       | 2.3       | 1.0       | 100.0 |
| Low-mesophilic 1 | 52.8  | 24.1      | 3.6       | 10.7      | 4.3       | 3.6       | 0.5       | 0.4       | 100.0 |
| Low-mesophilic 2 | 54.6  | 23.4      | 3.6       | 9.8       | 4.3       | 3.6       | 0.5       | 0.4       | 100.0 |
| Mesophilic 1     | 50.7  | 24.8      | 4.2       | 9.9       | 5.3       | 4.1       | 0.5       | 0.5       | 100.0 |
| Mesophilic 2     | 51.2  | 24.5      | 4.1       | 9.2       | 5.6       | 4.4       | 0.5       | 0.4       | 100.0 |

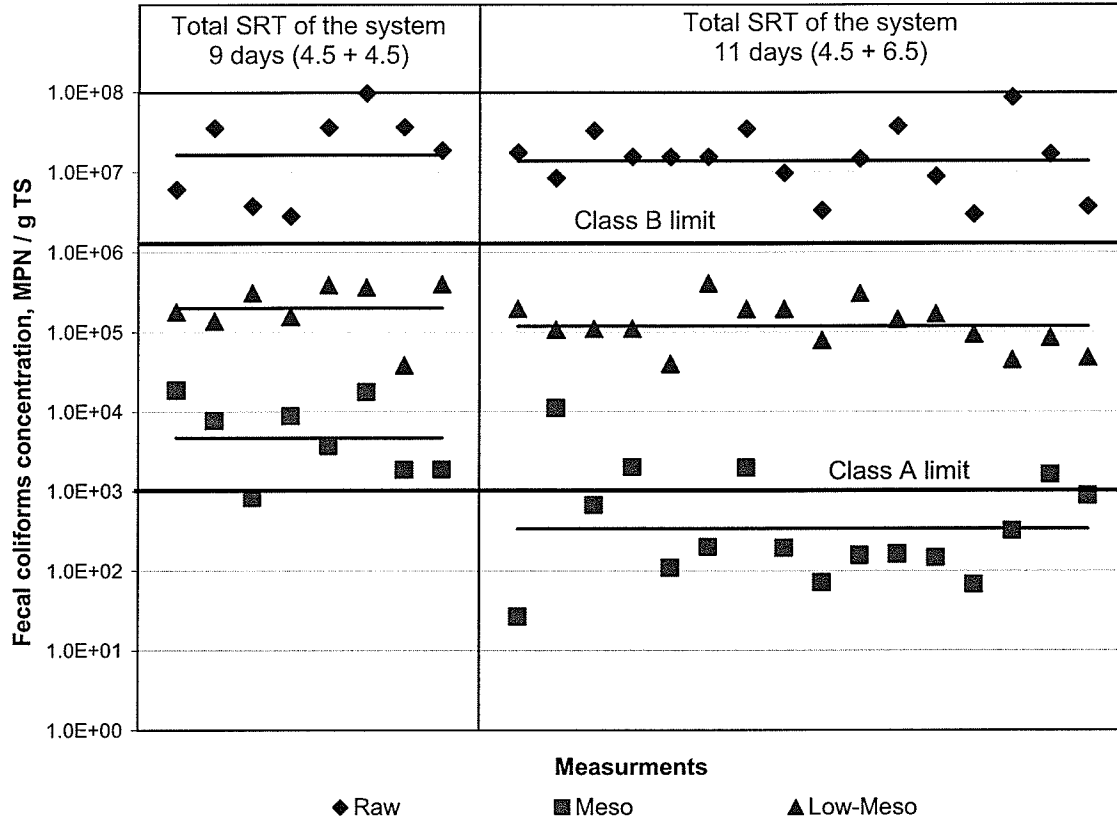


Figure 6 - 14. Fecal coliform concentrations in the raw wastewater and effluent from semi-continuously fed, staged acid digesters operated at total SRT of 9 and 11 days.

Table 6 - 6. U-VFA in raw sludge and effluent from acid-digestion system operated at total SRT of 11 days (4.5 + 6.5 days).

|   | Unit | Raw sludge  | Mesophilic digestion system |             | Low-mesophilic digestion system |             |
|---|------|-------------|-----------------------------|-------------|---------------------------------|-------------|
|   |      |             | 1st reactor                 | 2nd reactor | 1st reactor                     | 2nd reactor |
| Average VFA concentration                           | mg/l | 2219 ± 618  | 7781 ± 1540                 | 8127 ± 1540 | 6531 ± 672                      | 9058 ± 1018 |
| Average pH in digester                              | -    | 6.13 ± 0.05 | 5.27 ± 0.10                 | 5.31 ± 0.13 | 5.30 ± 0.10                     | 5.27 ± 0.12 |
| Average un-ionized VFA concentration                | mg/l | 54 ± 28     | 1827 ± 362                  | 1777 ± 317  | 1543 ± 157                      | 2127 ± 239  |
| Average un-ionized VFA concentration for the system | mg/l |             | 1798 ± 335                  |             | 1888 ± 205                      |             |

Table 6 - 7. Contribution of individual acids to total VFA concentration in raw sludge and effluent from acid-digestion system operated at total SRT of 11 days (4.5 + 6.5 days).

| Type of sludge   | Contribution of individual acids to total concentration, % |           |           |           |           |           |           |           |       |
|------------------|--|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-------|
|                  | Acetic   | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | Total |
| Raw              | 63.8   | 19.7      | 1.5       | 10.3      | 2.2       | 1.7       | 0.7       | 0.2       | 100.0 |
| Low-mesophilic 1 | 63.5   | 18.4      | 1.7       | 9.9       | 2.2       | 3.5       | 0.4       | 0.3       | 100.0 |
| Low-mesophilic 2 | 68.1   | 16.3      | 1.6       | 8.3       | 2.2       | 3.0       | 0.3       | 0.3       | 100.0 |
| Mesophilic 1     | 60.5   | 21.8      | 2.3       | 8.2       | 2.8       | 3.7       | 0.4       | 0.3       | 100.0 |
| Mesophilic 2     | 61.1   | 20.8      | 2.1       | 8.2       | 3.1       | 4.0       | 0.3       | 0.3       | 100.0 |

## CONCLUSIONS

Fecal coliforms could be successfully reduced below Class A level in batch acid digestion within 6 to 7 days in mesophilic and low-mesophilic conditions. This reduction was attributed to inhibitory effects of organic acids in their un-ionized form. The key factor that enables the inhibition is pH of the environment which affects distribution of ionized versus un-ionized form of organic acid. Semi-continuously fed, staged acid digestion system operated in mesophilic conditions and total SRT of 11 days achieve Class A standard on average while systems operated in low-mesophilic conditions showed very little reduction of fecal coliforms and did not achieve Class A level. Extended retention time of semi-continuously fed digestion systems compared with batch systems needed to achieve Class A level of fecal coliforms was probably caused by short-circuiting that may occur at this SRT. Concluding, a successful destruction of fecal coliforms using inhibitory effect of un-ionized organic acids depends on pH of the sludge, retention time, temperature and hydraulic properties of digestion system.

## ACKNOWLEDGEMENTS

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### 6.3 Extended acid-gas digestion for Class A biosolids

#### EXTENDED ACID-GAS DIGESTION FOR CLASS A BIOSOLIDS

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#### ABSTRACT

Two multi-phase digestion systems: meso acid/meso gas and low-meso acid/meso gas and conventional mesophilic anaerobic digester were tested and compared with respect to their ability to inactivate pathogens including fecal coliforms and *Ascaris suum* ova. Meso acid/meso gas system achieved Class A limit with respect to fecal coliforms, while two other systems did not. Enhanced fecal coliforms inactivation in multi-phase digestion systems compared to conventional mesophilic digestion was attributed to toxicity from VFA in un-ionized form in acid digesters. Effluent from meso acid/meso gas system showed the lowest viability of *Ascaris suum* ova from all the systems tested. Free floating eggs showed 2.8% viability while eggs contained in sentinels showed 0% viability. Two other systems showed poor inactivation of *Ascaris suum* ova showing their viability in the range of 60 to 80% in the final effluent.

#### INTRODUCTION

Conventional mesophilic anaerobic digestion (MAD) stabilizes sludge organics and leads to production of biogas, but it performs very poorly in terms of pathogen inactivation. Usually it can reduce fecal coliforms concentration in the range of 0.5 to 2.0 log (Barber, 2005, Watanabe et al., 1997). Pathogen content in digested sludge has become one of the most important parameter of digested sludge, especially if it is to be land applied. US. EPA agency

classifies biosolids depending on pathogen density as Class A and Class B product (E.P.A., 1979). Class A biosolids having lower density of pathogenic microorganisms can be applied to land without restrictions, which apply to Class B product. Also, public perception of biosolids plays an important role and safety of the final product became not only a matter of science but a necessary ingredient of communal acceptance (Matthews, 2005).

Several technologies have been tested and implemented to improve pathogen inactivation achieved by conventional MAD. Commonly applied methods for improved pathogen inactivation include: thermophilic digestion, temperature-staged digestion with at least one stage at thermophilic temperature, or pre-pasteurization (Gabb et al., 2001, Ghosh, 1998, Huyard et al., 2000, Lee et al., 1989, Roediger, 2000, Wilson et al., 2002). All these systems require additional energy to elevate sludge temperature to thermophilic temperature range. This results in decreased energy recovery from anaerobic digestion facilities. An example of energy balance for two systems utilizing conventional MAD and thermophilic anaerobic digestion was presented in Figure 1-7. Assuming that thermophilic digestion causes additional 10% volatile solids (VS) destruction (which is not always the case) accompanied by biogas production, overall balance results in approximately 1% more energy available for recovery from thermophilic system. Moreover, effluents from thermophilic digesters have worse dewaterability characteristics and are more odorous than effluents from mesophilic digesters. That is why some researchers (Ahring et al., 2001, De León and Jenkins, 2002, Watanabe et al., 1997) suggest that the main reason for thermophilic digestion is the need for improved pathogen inactivation.

Interestingly, some research results show that improved fecal coliforms inactivation can be achieved using staged-mesophilic digestion or mesophilic acid digestion followed by mesophilic gas digestion (De León and Jenkins, 2002, Lee et al., 1989). Previous experimental work (section 6.1 and 6.2.) showed that fecal coliforms inactivation was correlated with toxicity from un-ionized form of VFA and Class A biosolids standard with respect to fecal coliforms concentration may be achieved in mesophilic and low-mesophilic batch digestion and that two-stage mesophilic acid digestion may achieve on average Class A limit with respect to fecal coliforms concentrations.

The mechanism of toxicity from un-ionized VFA was presented in section 2.11 and 2.12.

There are several factors that can improve pathogen inactivation through un-ionized VFA toxicity. An increase in concentration of VFA in digester can be achieved using sludge thickening. Additionally, due to thickening, a considerable sludge flow reduction can be achieved and energy savings due to decreased heat requirements as shown in Figure 1-13.

Short-circuiting of acid digester (the escape of sludge particles before the designed retention time has passed) may pose a problem in achieving maximum pathogen inactivation. A successful acid digestion system would have to comprise of several digesters in series.

Recent example of applying enzymic hydrolysis digestion in Bromborough, which consist of six acid digesters in series (Matthews, 2005) allowed a four-log reduction of fecal coliforms (Shea, 2005).

## OBJECTIVES

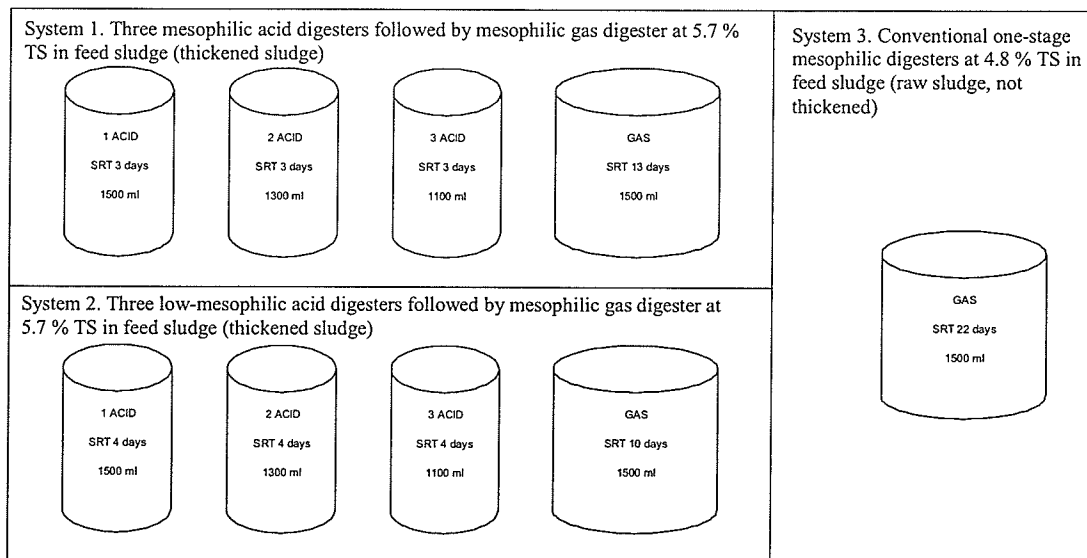
The objective of this experiment was to evaluate and compare the ability of two multi-phase digestion systems and the conventional mesophilic anaerobic digester, to inactivate pathogens such as fecal coliforms and *Ascaris suum* ova found in sewage sludge.

Multiphase-digestion systems consist of three acid digesters in series, either at mesophilic or low-mesophilic temperature, followed by gas digestion at mesophilic temperature for both systems (Figure 6-15). A previous study showed that enhanced fecal coliforms removal may be achieved due to high concentrations of organic acids in un-ionized form and Class A could be potentially achieved in a flow-through system having more than one acid digester in series to avoid short-circuiting (section 6.1 and 6.2.). The impact of high concentrations of VFA in un-ionized form on *Ascaris suum* viability was not known. The issue of potential regrowth of fecal coliforms in gas digester that follows acid digestion needed to be addressed.

## METHODOLOGY

Three lab-scale anaerobic digestion systems (Figure 6-15) were assembled:

- 1 – three mesophilic acid digesters followed by mesophilic gas digester (also referred to as meso acid/meso gas system)
- 2 – three low-mesophilic acid digesters followed by mesophilic gas digester (also referred to as low-meso acid/meso gas system)
- 3 – control, one phase mesophilic anaerobic digester (also referred to as conventional mesophilic anaerobic digester – MAD)



**Figure 6 - 15. Experimental setup. Three anaerobic digestion systems: (1) 3-stage mesophilic (38°C) acid followed by mesophilic gas digester; (2) 3-stage low-mesophilic (24°C) acid digesters followed by mesophilic (38°C) gas digester; (3) conventional one-stage mesophilic (38°C) digester.**

All methods and sludge source as in previous sections.

Additionally, because *Ascaris* eggs were not detected in raw sludge, in order to assess the efficiency of digestion on their inactivation it was necessary to spike digesters with swine *Ascaris suum* prior to testing. Besides spiking sludge directly with the free floating eggs, helminth ova inactivation was evaluated also using sentinel chambers placed inside the digester. Sentinel chambers provided a free exchange of liquid through a semi-permeable membrane on the sides of the sentinels, but eggs were contained inside without possibility of escape. Sentinels were prepared in Ithaca, N.Y. and procedure consisted of mixing *Ascaris suum* eggs with the same sludge that was fed into digesters, placing this mixture inside sentinels and shipping on dry ice. *Ascaris suum* eggs culture and viability counts were performed in Cornell University according to Bowman et al., (2002).

## RESULTS & DISCUSSION

Fecal coliform concentration was measured at the effluent from acid digesters, gas digesters as well as control and raw sludge as shown in Figures 6-16 and 6-17. VFA concentrations, pH and calculated concentration of VFA in un-ionized form are provided in Tables 6-8, 6-9 and 6-10.

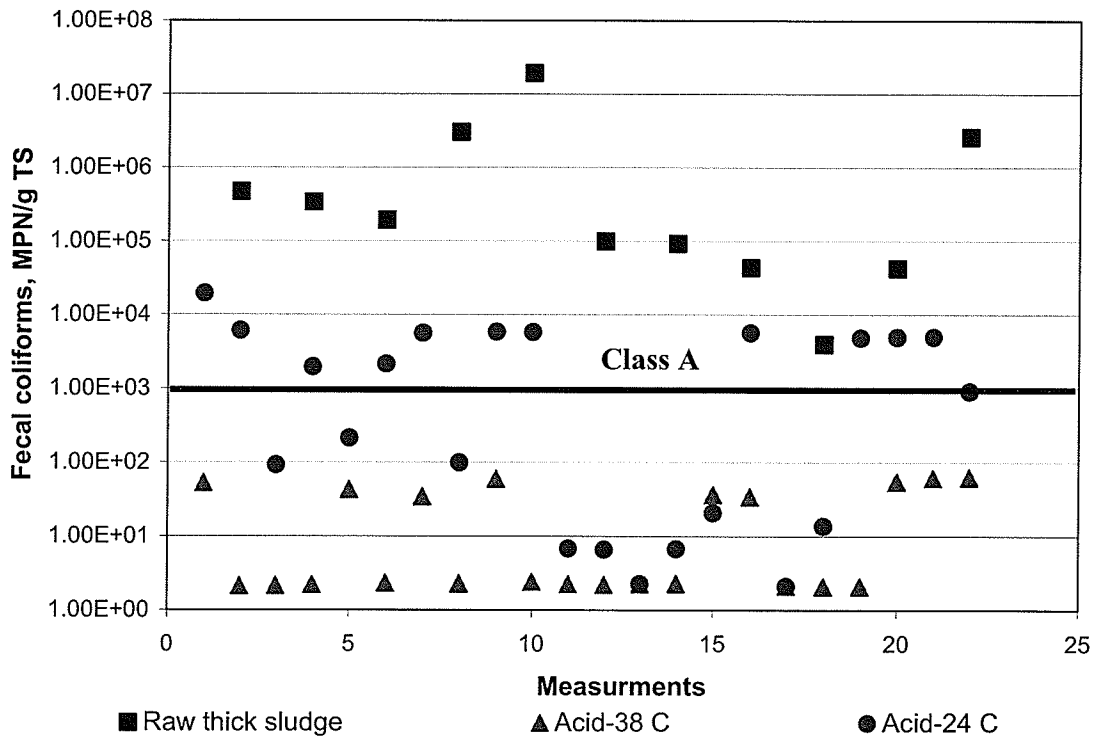


Figure 6 - 16. Fecal coliform concentrations in the raw-thickened wastewater sludge and effluent from three-phase acid digester operated in mesophilic (38 °C) and low-mesophilic temperatures (24 °C).

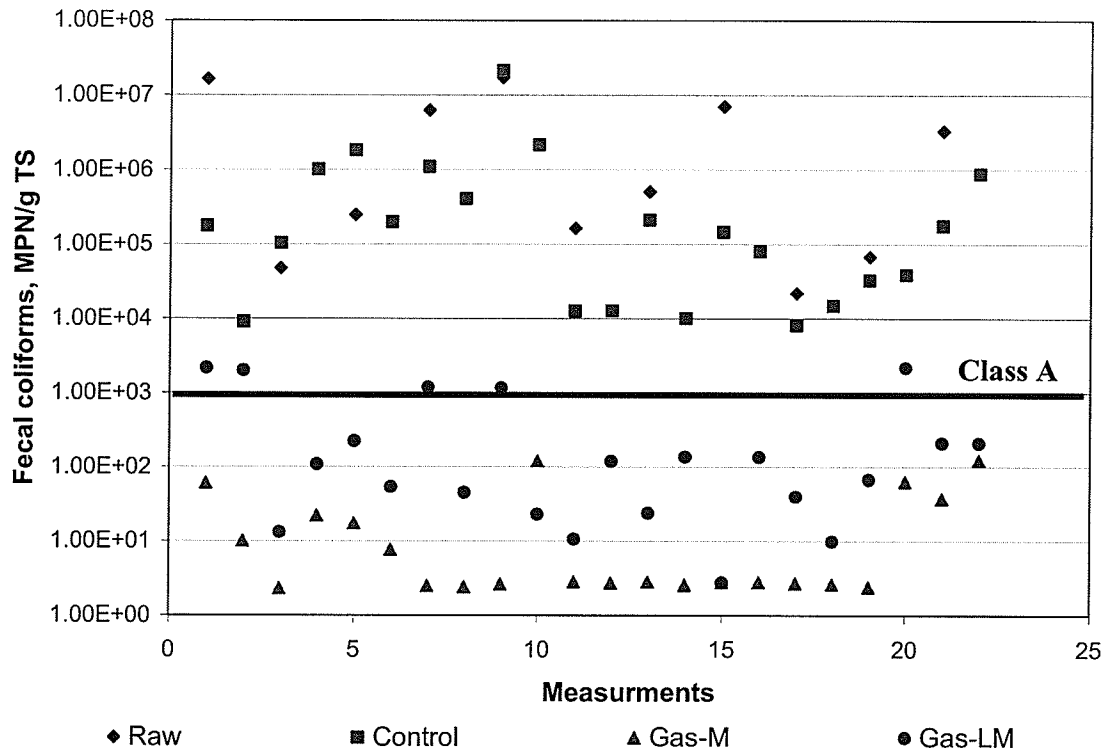


Figure 6 - 17. Fecal coliform concentrations in the raw sludge, raw-thickened sludge and effluent from meso gas/meso gas and low-meso acid/meso gas digestion system and control digester.

Table 6 - 8. VFA concentration, pH and un-ionized VFA concentration in raw and raw-thickened sludge.

|                                      | Unit | Raw sludge    | Raw-thick sludge |
|--------------------------------------|------|---------------|------------------|
| Average VFA concentration            | mg/l | 2296 ± 1399   | 2645 ± 1341      |
| Average pH in digester               | -    | 6.20 ± 0.26   | 6.36 ± 0.22      |
| Average un-ionized VFA concentration | mg/l | 103.23 ± 60.9 | 65 ± 29          |

**Table 6 - 9. VFA concentration, pH and un-ionized VFA concentration in mesophilic and low-mesophilic acid digesters.**

|                                      |      | Mesophilic acid digesters     |              |             |
|--------------------------------------|------|-------------------------------|--------------|-------------|
|                                      | Unit | 1                             | 2            | 3           |
| Average VFA concentration            | mg/l | 9696 ± 2961                   | 10619 ± 3881 | 8101 ± 3535 |
| Average pH in digester               | -    | 5.58 ± 0.13                   | 5.39 ± 0.11  | 5.45 ± 0.33 |
| Average un-ionized VFA concentration | mg/l | 1282 ± 342                    | 2058 ± 839   | 1250 ± 468  |
|                                      |      | Low-Mesophilic acid digesters |              |             |
|                                      | Unit | 1                             | 2            | 3           |
| Average VFA concentration            | mg/l | 8503 ± 3182                   | 9163 ± 3017  | 9580 ± 2966 |
| Average pH in digester               | -    | 5.61 ± 0.18                   | 5.38 ± 0.20  | 5.30 ± 0.22 |
| Average un-ionized VFA concentration | mg/l | 990 ± 281                     | 1730 ± 487   | 2125 ± 588  |

**Table 6 - 10. VFA concentration, pH and un-ionized VFA concentration in control digester, and gas digesters of meso acid/meso gas system and low-meso acid/meso gas system.**

|                                      | Unit | Control MAD | Gas Meso/Meso | Gas Low-meso/Meso |
|--------------------------------------|------|-------------|---------------|-------------------|
| Average VFA concentration            | mg/l | 408 ± 328   | 2878 ± 1987   | 3623 ± 2668       |
| Average pH in digester               | -    | 7.55 ± 0.21 | 7.38 ± 0.14   | 7.23 ± 0.24       |
| Average un-ionized VFA concentration | mg/l | 0.6 ± 0.3   | 5.3 ± 3.3     | 8.8 ± 6.9         |

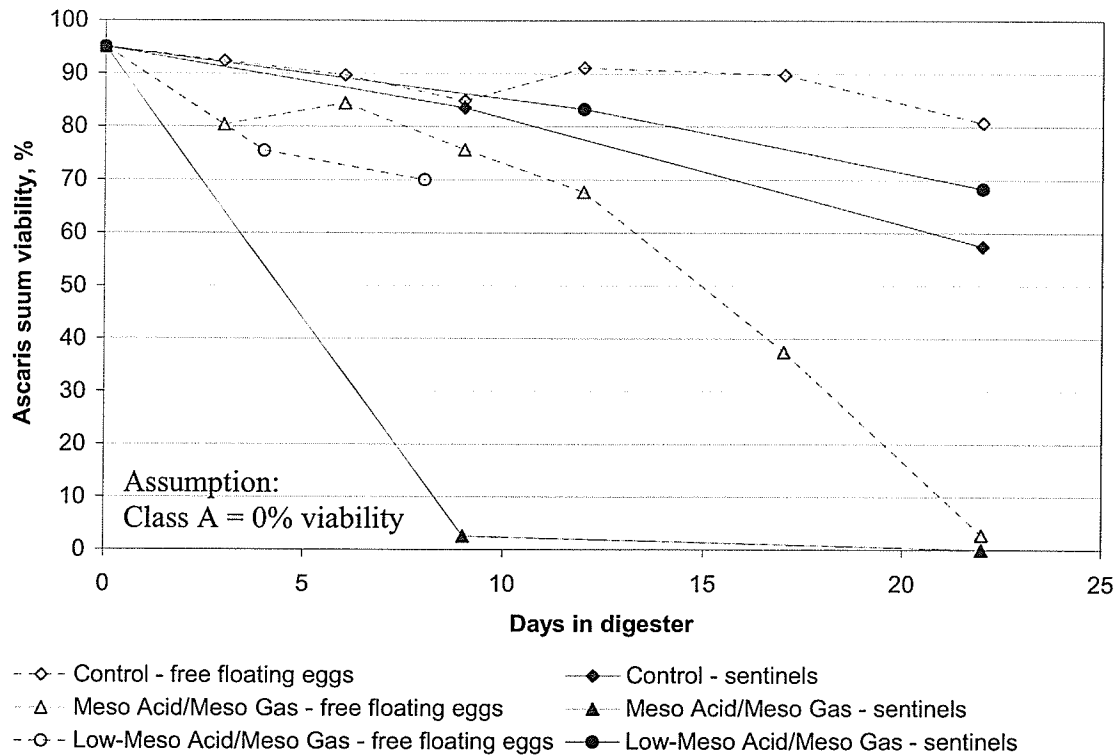
The average fecal coliforms concentration in raw, thickened sludge was  $2.54 \times 10^5$  MPN/g TS. Mesophilic acid digestion effluent showed an average concentration of fecal coliforms of  $7.8 \times 10^0$  MPN/g TS while low-mesophilic acid digestion average fecal coliform concentration was  $3.4 \times 10^2$  MPN/g TS. Even though both acid digestion systems on average achieved Class A standards, it was only the mesophilic acid digesters that could comply 100% with Class A limit what is required by US. EPA regulation. It should be noted that total SRT for mesophilic acid digestion was 9 days while SRT in low-mesophilic acid digestion was 12 days. Acid digestion under mesophilic conditions was limited by the start of methanogenic activity in the last acid digestion reactor at longer retention times, which at times lowered concentration of VFA in this reactor which causing increase in pH and decrease in



concentration of VFA in un-ionized form. Greater inactivation of fecal coliforms in mesophilic conditions compared to low-mesophilic conditions was attributed to increased diffusion rates at higher temperature (Puchajda and Oleszkiewicz, 2005).

The effluent from meso acid/meso gas system consistently complied with Class A limits achieving average concentration of fecal coliforms of  $7.5 \times 10^0$  MPN/g TS. The effluent from low-meso acid/meso gas system showed average concentration of fecal coliforms of  $1.1 \times 10^2$  MPN/g TS but Class A limit was exceeded on 5 occasions out of 22 measurements therefore 100% compliance with the Class A limit was not achieved. As shown in Table 6-10 gas digesters in both systems had very little (virtually non-existent) concentration of VFA in un-ionized form, but effluent from gas digesters showed comparable concentrations of fecal coliforms as effluents from acid digesters for both systems. It means that additional fecal coliforms inactivation in gas digesters did not occur, but more importantly their regrowth was not observed. At the same time, conventional MAD digesters (or control digester), which lacked VFA present in un-ionized form, achieved average concentration of fecal coliforms in the effluent of  $1.39 \times 10^5$  MPN/g TS while raw sludge has average concentration of fecal coliforms of  $7.77 \times 10^5$  MPN/g TS.

Results from *Ascaris suum* viability tests are presented in Figure 6-11.



**Table 6 - 11. *Ascaris suum* viability results from free floating eggs and sentinels.**

Control MAD system and low-meso acid/meso gas system showed high viability of ascaris ova in the effluent, in the range of 60 to 80% viability, after 22 days of digestion. Note: free floating eggs could not be found in quantities allowing statistical analysis in the effluent from low-meso acid/meso gas system after day 8 of digestion which was caused most probably due to washout. The only system that could provide higher level of inactivation of ascaris ova was meso acid/meso gas system with 0% viability of ova in sentinels (3-log reduction) and 2.8% viability of free floating ova (1.6-log reduction). The difference in viability profiles between free floating eggs and sentinels could be explained by the short-circuiting that is commonly occurring hydraulic phenomena in digesters. Ova contained in sentinels were placed and kept in digesters for desired duration while free floating eggs were moving with

liquid. As expected, most ova contained in sentinels were inactivated during first nine days when sentinel was placed in acid digester. However, free floating eggs showed still high viability at the effluent from acid digestion. Interestingly, the viability was decreasing in the gas digester even though un-ionized VFA toxicity was not present due to neutral pH in these digesters as shown in Table 6-13. This could suggest that accumulation of anion may occur and loss of viability could be attributed to osmotic stress. Overall, effluent from meso acid/meso gas system showed much lower concentration of pathogens than effluents from other systems.

## CONCLUSIONS

Two multi-phase digestion systems: meso acid/meso gas and low-meso acid/gas were tested and compared to conventional mesophilic anaerobic digester with respect to their ability to inactivate fecal coliforms and *Ascaris suum* ova. The effluent from meso acid/meso gas system consistently met Class A standards achieving on average fecal coliforms concentration of  $7.5 \times 10^0$  MPN/g TS. The effluent from low-meso acid/meso gas system achieved on average with fecal coliform concentration of  $1.1 \times 10^2$  MPN/g TS but Class A limit was exceeded on five occasions. This would disqualify this system from meeting Class A product. Fecal coliforms inactivation was attributed to high concentrations of VFA in un-ionized form in acid digesters. Their regrowth was not observed in gas digestion where un-ionized VFA was virtually non-existent due to neutral pH. Conventional MAD digester (control digester) operated at mesophilic temperature and SRT of 22 days (same SRT as for multi-phase digestion systems) achieved very poor reduction of fecal coliforms with effluent average concentration of  $1.4 \times 10^5$  MPN/g TS. Effluent from meso acid/meso gas system showed the lowest viability of *Ascaris suum* ova from all the systems tested. Free floating

eggs showed 2.8% viability while eggs contained in sentinels showed 0% viability. Two other systems showed poor inactivation of *Ascaris suum* ova showing their viability in the range of 60 to 80% in the final effluent.

#### ACKNOWLEDGEMENTS

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## 6.4 Pressure tests with acid digestion

### INTRODUCTION

As indicated in sections 6.1, 6.2 and 6.3. temperature may have strong influence on diffusion coefficient of organic acids and therefore impact inactivation of pathogens. This factor, besides concentrations of un-ionized VFA themselves, may enhance pathogen inactivation. According to Henry's law, at any given temperature there exists an equilibrium between concentration of any species in aqueous phase and partial pressure of those species in the gas phase (Nazaroff and Alvarez-Cohen, 2001) as shown in equation [6-3].

**Equation 6 - 2**       $c = k_H \times P_p$

where:  $c$  – concentration of species in aqueous phase,  $k_H$  – Henry's constant, and  $P_p$  – partial pressure of species in gas phase.

Therefore, if pressure in the gas phase increases, so does the concentration of any species in aqueous phase. This leads to the assumption that if the pressure in the headspace of the acid digester is increased, then the concentrations of organic (volatile) acids in the sludge should increase. One way to increase the pressure in the digester's headspace is to not allow for the digester's gas to escape. Gas production that results from sludge digestion, should build up positive pressure in an air-tight sealed digester.

It is hypothesized that an increase in partial pressure of organic acids caused by increase in headspace may lead to increase in diffusion of organic acids and therefore, it may lead to increased toxicity from VFA. The effect of increased partial pressure on diffusion is showed in following equations [6-3] to [6-6].

General equation for diffusion (Berg, 1983).

$$\text{Equation 6 - 3 } J = -D \frac{\Delta C}{x} = -D \frac{C_2 - C_1}{x}$$

where:

J - diffusion flux, mol/m<sup>2</sup>-sec

D - diffusion coefficient, m<sup>2</sup>/sec

ΔC - concentration gradient, mol/L

x - distance, m

According to Henry's law

$$\text{Equation 6 - 4 } C_2 = \frac{P_{p2}}{k_{H2}} \quad \text{and} \quad \text{Equation 6 - 5 } C_1 = \frac{P_{p1}}{k_{H1}}$$

Assuming equal temperature, Henry's constants stay the same, then  $k_{H1} = k_{H2}$

Then,

$$\text{Equation 6 - 6 } J = -D \frac{\frac{P_{p2}}{K_{H1}} - \frac{P_{p1}}{K_{H1}}}{x} = \frac{-D}{K_{H1}} \frac{P_{p2} - P_{p1}}{x} = \frac{-D}{K_{H1}} \frac{\Delta P_p}{x}$$

where:

ΔPp = difference in partial pressure, atm

It should be noted that Henry's constants for organic acids decrease with an increase in temperature what is presented in Table 6-12 based on data provided by Sander, 1999.

Overall, Henry's constants for organic acids are very high, which means that portion that exists in gas phase (partial pressure of the compound in gas phase) is very small compared to the amount of substance in solution. For comparison, Henry's constant for oxygen is  $1.3 \times 10^{-3}$  mol/L/atm and for carbon dioxide it is  $3.4 \times 10^{-2}$  mol/L/atm (both values at 25 Celsius).

**Table 6 - 12. Henry's constants for organic acids at 25 Celsius and 38 Celsius (based on data provided by Sanders, 1999).**

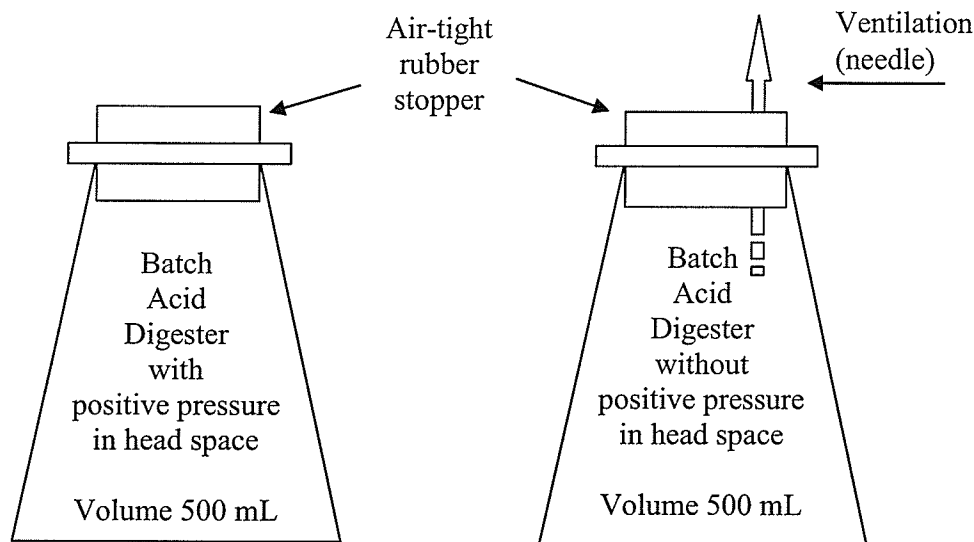
| Acid       | Henry's constant |            | Henry's constant                                |            |
|------------|------------------|------------|---|------------|
|            | <i>mol/L/atm</i> |            | <i>dimensionless<br/>mol gas / mol solution</i> |            |
|            | 25 Celsius       | 38 Celsius | 25 Celsius                                      | 38 Celsius |
| Acetic     | 5.7E+03          | 2.4E+03    | 7.2E-06   | 1.7E-05    |
| Proptionic | 4.1E+03          | 1.7E+03    | 1.0E-05   | 2.4E-05    |
| Butyric    | 2.8E+03          | 1.2E+03    | 1.5E-05   | 3.5E-05    |
| Valeric    | 2.2E+03          | 9.2E+02    | 1.9E-05   | 4.4E-05    |
| Caproic    | 1.3E+03          | 5.4E+02    | 3.1E-05   | 7.5E-05    |

## OBJECTIVES

The objective of this part of experimental work was to assess if pressure generated by production of biogas may have an impact on inactivation of fecal coliforms and environmental conditions in digester that contribute to pathogen inactivation such as: concentration of VFAs and pH

## METHODOLOGY

Batch acid digesters were operated (in duplicates) at mesophilic (38°C) and low-mesophilic (24°C) temperatures. At any temperature there were two sets of digesters: (1) digesters that did not allow for biogas venting which resulted in positive pressure in head space; and (2) digesters that allow for biogas venting and did not exhibit positive pressure in head space as shown in Figure 6-18.



**Figure 6 - 18. Schematic representation of two types of batch acid digesters: pressurized and vented, used in experiment.**

All methods and sludge source as in previous sections.

Additionally, pressure was measured everyday in room temperature using Ergonomical Digital Tire Gauge with LCD readout display with range 5 to 99 psi  $\pm 0.5$  psi (Canadian Tire, Toronto, Ottawa).

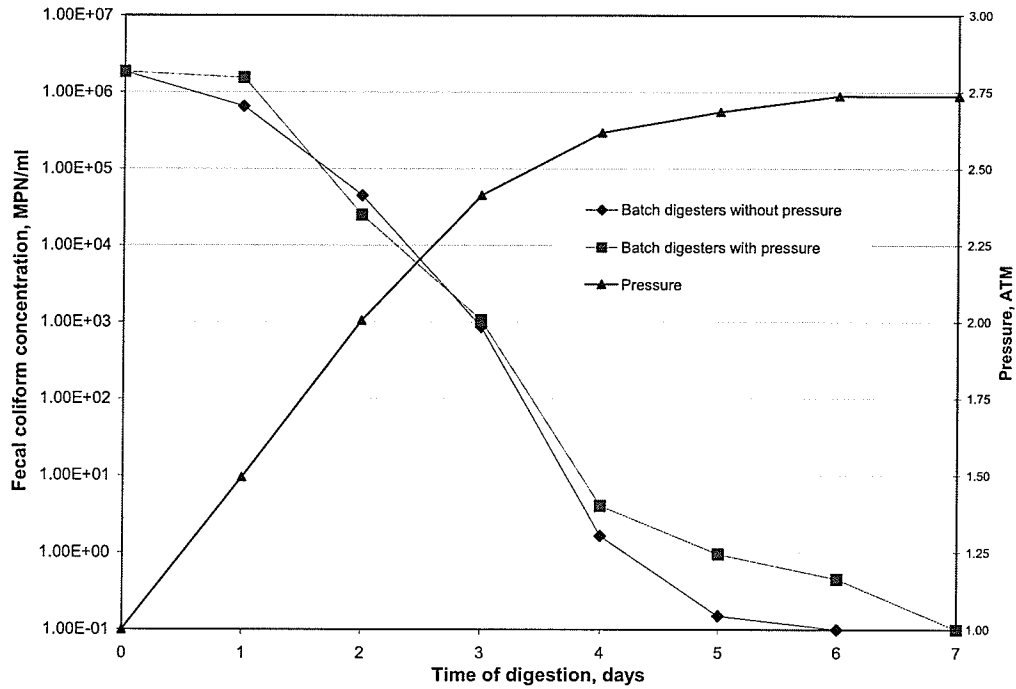
## RESULTS AND DISCUSSION

Fecal coliforms profiles (averages from duplicate digesters) from pressurized and vented batch acid digesters at mesophilic and low-mesophilic temperatures are presented in Figures 6-19 and 6-20 and concentrations of VFA, and un-ionized VFA concentrations in those digesters are presented in Figures 6-21 and 6-22.

Because measurements of VFA concentrations were performed after opening of pressurized digester, an estimation of concentration before opening was performed. This estimation was based on the following assumptions: an increase in VFA concentration would be proportional to increase in partial pressure, which is proportional to overall increase in pressure. For



instance, at 2 atm pressure in headspace, partial pressure of VFA would double (compared to vented digester with approximately 1 atm pressure in headspace), which would result in doubling the concentration of VFA in solution.



**Figure 6 - 19. Fecal coliform profiles in batch acid digesters operated with and without pressure build-up operated at mesophilic temperature.**

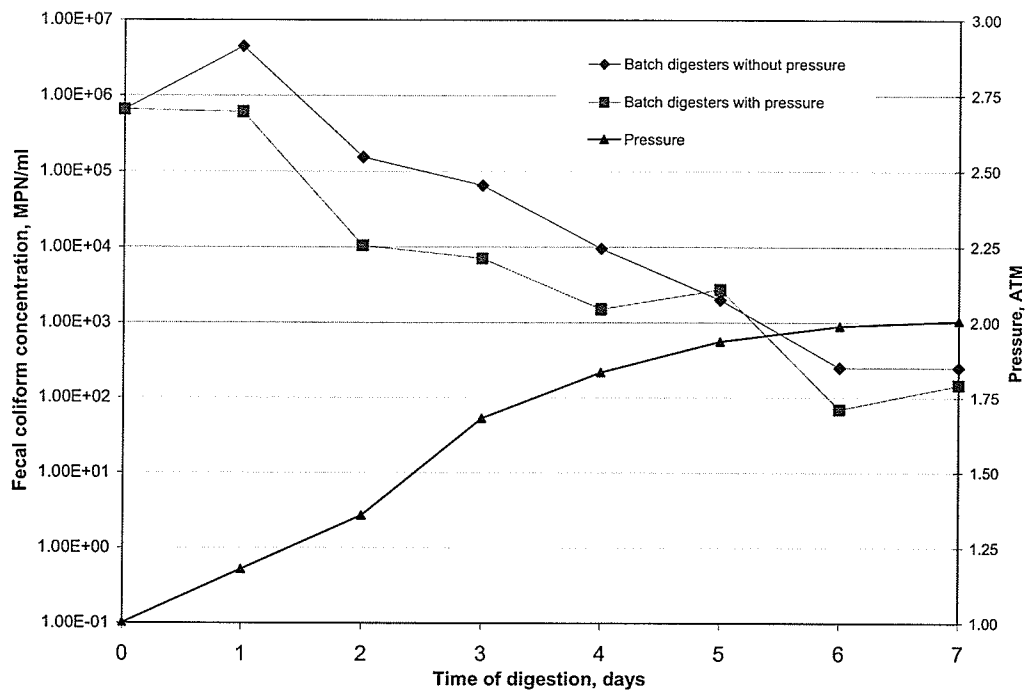


Figure 6 - 20. Fecal coliform profiles in batch acid digesters operated with and without pressure build-up operated at low-mesophilic temperature.

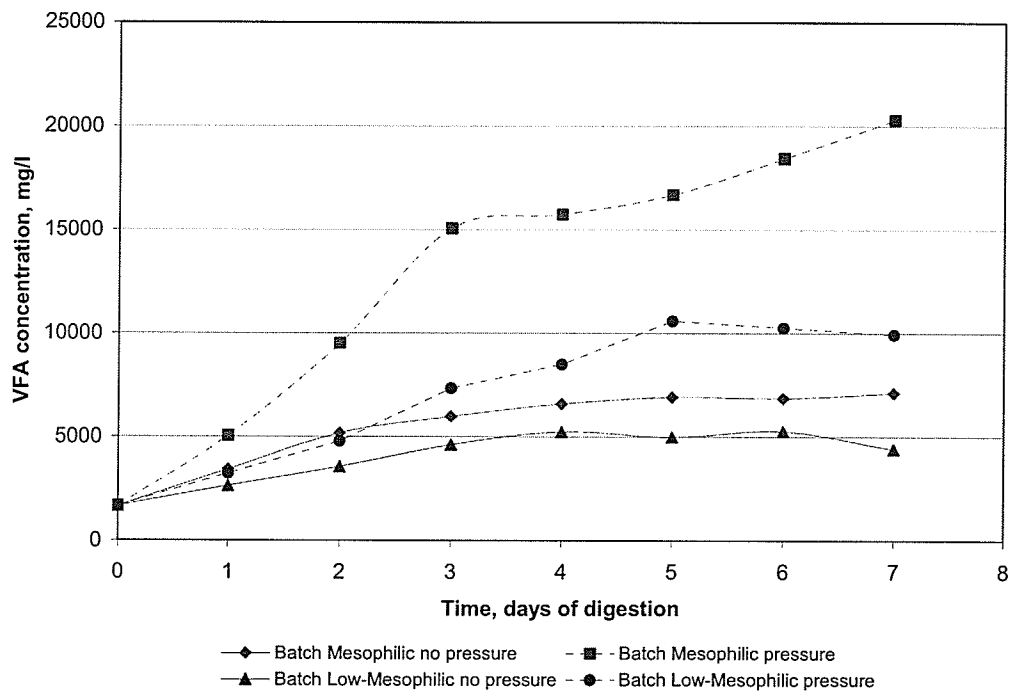
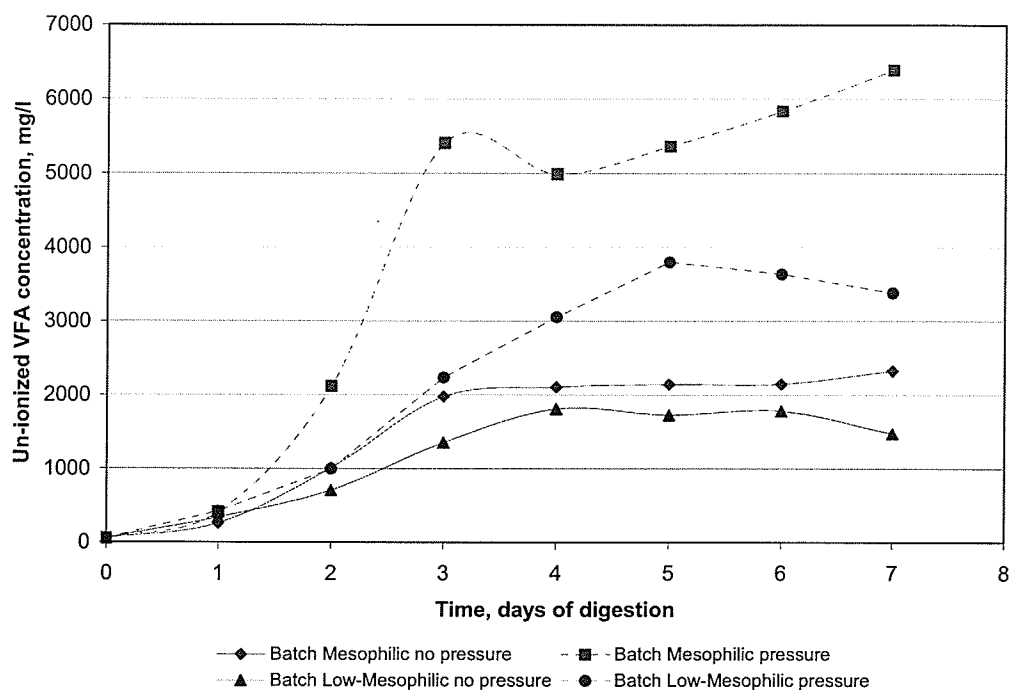


Figure 6 - 21. Total VFA concentrations in pressurized and vented acid digesters at mesophilic and low-mesophilic operating temperatures.



**Figure 6 - 22. Un-ionized VFA concentrations in pressurized and vented acid digesters at mesophilic and low-mesophilic operating temperatures.**

Batch mesophilic digesters at mesophilic temperatures (both pressurized and vented) lowered fecal coliforms densities to below detectable levels, while batch low-mesophilic digesters (both pressurized and vented) achieved final concentrations of fecal coliforms after 7 days of digestion at the level slightly higher than  $10^2$  MPN/ml. Batch pressurized digesters operated at mesophilic temperatures achieved almost 2.75 ATM pressure after 7 days of digestion while, pressurized digesters operated at low-mesophilic temperatures achieved lower pressure of 2 ATM after the same digestion time. It was most likely caused by higher gas production that occurs at mesophilic temperature (the actual amount of biogas production was not measured but it is congruent with findings in section. 5.2 and 6.3). There was no

considerable difference in fecal coliform profiles between pressurized and vented digesters at any temperature tested.

This is an important observation, because an increase in headspace pressure showed considerable effect on increase in total and un-ionized VFA concentration, yet fecal coliforms inactivation pattern was not observed. It is possible that increase in concentration of un-ionized VFA (caused for instance by increase in pressure in headspace or increased in solids concentration) may not contribute to enhanced inactivation of fecal coliforms. It could also partially explain findings from section 6.1. when three different levels of solids in sludge generated different levels of VFA concentration yet overall fecal coliform profiles were very similar in all conditions (see Figures 6-2 and 6-3).

These findings suggest that there may be other factors that may affect fecal coliforms inactivation besides temperature and presence of un-ionized VFA concentration. For instance, different intermediate digestion products, such as lactate or ethanol (which is produced at low pH as alternative to acetate), may influence fecal coliform inactivation.

## **7 Increased energy recovery and enhanced pathogen removal through anaerobic digestion of thickened wastewater sludge**

This section consists of a paper submitted for journal publication on increased energy recovery and inactivation of pathogens during novel acid-gas digestion process:

- Study presented in the paper is extension of section 6.3, except the main focus of this paper is energy recovery from novel anaerobic digestion processes compared to conventional MAD process: Puchajda, B. and Oleszkiewicz, J. A. (2005) Increased Energy Recovery from Anaerobic Digestion of Thickened Wastewater Sludge. Journal of Residual Science and Technology, submitted for publication.

Raw data for all results presented in section 7.0 can be found in Appendix D.

## 7.1 Energy recovery from novel extended acid-gas anaerobic digestion systems

### INCREASED ENERGY RECOVERY FROM ANAEROBIC DIGESTION OF THICKENED WASTEWATER SLUDGE

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#### ABSTRACT

This paper presents results from experimental work on multiple-phase anaerobic digestion of thickened wastewater sludge and its effect on overall energy balance compared to conventional mesophilic anaerobic digestion. Three labs-scale anaerobic digestion systems were assembled: (1) three-phase mesophilic acid digestion followed by mesophilic gas digestion; (2) three phase low-mesophilic acid digestion followed by mesophilic gas digestion and (3) conventional mesophilic anaerobic digestion. System (1) and (2) were operated on thickened wastewater sludge (5.7 % TS) while system (3) was fed raw, un-thickened sludge (4.8% TS). Calculations of energy balance included the following scenarios: all systems operated at equal SRT of 22 days, conventional mesophilic digestion operated at lower solids concentration in feed sludge (3.9% TS compared to 4.8% TS), and systems operated at equal volumes, which resulted in extension of the retention time in gas digesters of multiple-phase digestion systems. The latter scenario resulted in highest amount of energy available for recovery (for meso acid/meso gas system) from any configurations evaluated.

#### INTRODUCTION

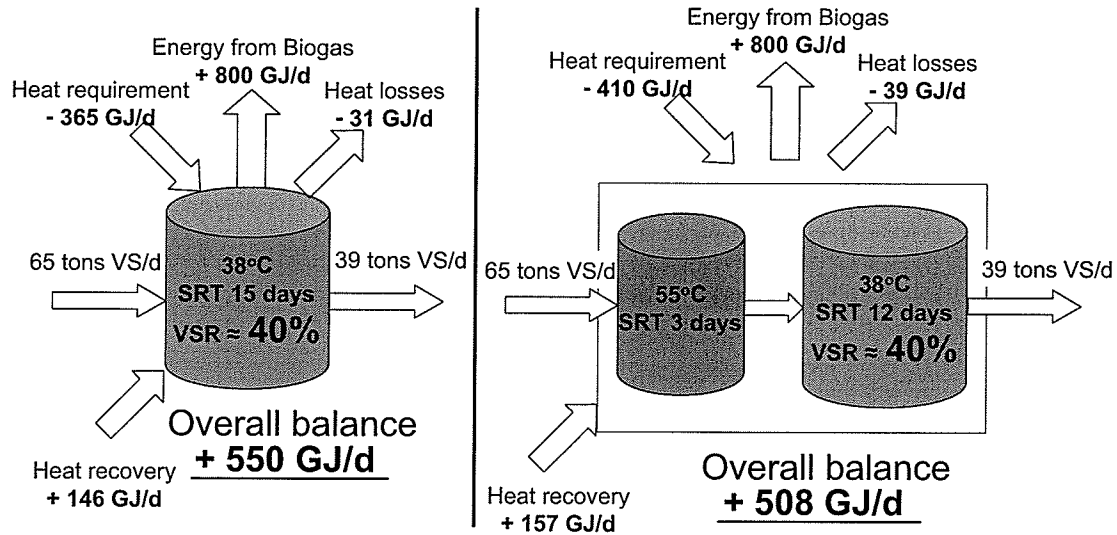
The two most important challenges for anaerobic digestion of wastewater sludge is to minimize the costs of the process and to improve biosolids production in order to comply

with Class A standards (E.P.A., 1999). Several improvements to conventional single stage mesophilic anaerobic digestion (MAD) have been researched and implemented in the past two decades, which include: temperature-phased digestion, acid-gas digestion, or recently three-phase and multiple-phase digestion (Ghosh, 1998, Reusser and Zelinka, 2004, Schafer et al., 2003, Wilson et al., 2002). While most of the solutions claim a higher biogas production and add stability to the process, there is little information on how these improvements influence the overall energy balance for the digestion systems. For instance, using two-phase, thermophilic-mesophilic anaerobic digestion will result in a Class A biosolids product (if the system complies with the temperature-time relationship as defined by EPA) but overall it requires more energy than conventional mesophilic digestion at the same retention time (Shimp et al., 2003). An example of energy balance for the single thermophilic and single mesophilic digester is provided in Figure 1-7.

The energy balance includes the following components: energy associated with methane content of biogas and heat requirement, and energy losses and recovery for the digester. It is also assumed that thermophilic digestion results in 50% volatile solids (VS) destruction and mesophilic in 40% VS destruction. Even though, it was assumed that thermophilic digestion destroyed more VS and produced more biogas, the overall energy balance for thermophilic digestion is only slightly better (approximately 1% more) than mesophilic digestion.

To offset costs associated with heat requirements, two-phase, thermophilic-mesophilic digestion is an often used solution. It is a desirable solution from an energy view point only if additional biogas production can offset additional heat requirements. This means, that, because of the additional heat requirement, the additional VS destruction achieved by two-phase, thermophilic-mesophilic system is required when compared to conventional MAD

(Figure 1-10). Previous work on two-phase, thermophilic-mesophilic digestion system (Puchajda et al., 2003) showed no statistical difference in biogas production between conventional MAD which would result in a lower overall energy balance for the two-phase system compared to a conventional one stage MAD (Figure 7-1).



**Figure 7 - 1. Energy balance for conventional mesophilic digester (MAD) and two-phase, thermophilic-mesophilic digestion system. Assumptions as in Figure 1-7 except no increase in biogas production from two-phase system compared to conventional MAD system.**

Another solution frequently applied is sludge thickening prior to digestion which reduces sludge volume and overall digesters' volumes as well as the heat requirement of the process (Bailey and Daigger, 1998, Shimp et al., 2003). Increasing total solids (TS) content of sludge from 3% TS to approximately 6% TS brings approximately a 50% reduction in sludge volume. Alternatively, using the same digester's volume, sludge thickening and volume reduction may result in an increase in solids retention time (SRT) of the digestion and therefore provide additional VS destruction and biogas production – Figure 1 - 12.



Assuming that digestion of thickened wastewater sludge results in the same VS destruction at the same retention time and the same amount of biogas produced, it may provide an additional 21% of recoverable energy (thickening costs not included) - Figure 1 - 13. Additional energy savings could be achieved if successful pathogen reduction could be obtained without thermophilic process or pre pasteurization which is often practiced in Europe (Shimp et al., 2003). Besides, many existing digestion systems cannot be upgraded to work at thermophilic temperatures due to structural limitations of the digesters, which is the case in Winnipeg North End pollution Control Centre (Szoke, 2003). Previous work (Puchajda and Oleszkiewicz, 2005, Puchajda et al., 2005) showed the potential of multiple-phase acid digestion, at mesophilic (38°C) and low-mesophilic (24°C) temperatures to lower fecal coliform concentrations below the Class A limit. Retention times needed to achieve fecal coliforms removal below Class A limit were approximately 9 days at mesophilic temperature and 12 days at low mesophilic temperature (both three-stage acid digestion systems).

## OBJECTIVES

The objective of this experimental work was to compare two multiple-phase anaerobic digestion systems at increased solids concentrations in feed sludge to a conventional one-stage mesophilic anaerobic digestion (MAD). Parameters used for comparison included: VS destruction, gas production, methane content in biogas, effluent quality such as SCOD and VFA content. The second objective of this work was to establish and compare energy balance for the systems. Energy components included in the energy balance were: energy component of biogas, heat requirement, recovery and heat losses, energy required for sludge thickening, and recoverable energy in the form of heat and/or electricity generation.

## METHODOLOGY

As in section 6.3.

Additionally, total solids (TS) and volatile solids (VS) were measured three times per week according to Standard Method (SM) 2540 B and SM 2540 E, respectively (APHA et al., 1998). Total chemical oxygen demand (TCOD) was measured three times per week according to SM 5220 D (APHA et al., 1998). Soluble chemical oxygen demand (SCOD) was determined as COD after filtration through 0.45  $\mu\text{m}$  pore size media. Volatile fatty acids including acetic, propionic, i-butyric, n-butyric, i-valeric, n-valeric, i-caproic and n-caproic (C2 to C6) were measured three times per week using a gas chromatograph Hewlett-Packard (Palo Alto, California) 5890A with flame ionization detector (FID). Before analysis, VFA samples were stored in the freezer. A volume of biogas was collected and measured daily using the liquid displacement method in calibrated air-tight vessels. The liquid was saturated sodium chloride with 5% sulphuric acid and methyl orange to prevent gas from dissolving. Biogas composition – methane ( $\text{CH}_4$ ) and carbon dioxide ( $\text{CO}_2$ ) was measured twice a week using gas chromatograph GOW MAC Model 550 (Bridgewater, N.J.) with helium as a gas carrier. pH was measured twice per week using pH electrode Accumet AP 50 (Fisher Scientific, Ottawa, Ontario).

## RESULTS AND DISCUSSION

Main parameters of all systems obtained from bench-scale experiments are presented in Table 7-1. Multiple-phase anaerobic digestion systems 1 and 2 (as in Figure 6-15) fed with thickened wastewater sludge achieved 43.5 and 39.9% VS destruction, respectively, compared to 57.8% VS destruction achieved by control MAD system fed with raw sludge with unadjusted solids content. Even though VS destruction (in %) of multiple-phase

digestion systems was much lower than VS destruction achieved by conventional mesophilic digester, all systems achieved a minimum of 38% of VS destruction required by EPA (E.P.A., 1999) for Class A limit. VS destruction on the mass basis resulted in 0.96 g VS/d destroyed by conventional MAD system, and 0.92 g VS/d and 0.85 g VS/d destroyed by multiple phase meso acid/meso gas and low-meso acid/meso gas digestion systems, respectively. At the same time, multiple-phase digestion systems exhibited much higher SCOD and VFA content in the effluent which: (1) showed potential for additional gas production at longer retention times; and (2) would result in increased odour and worse dewaterability characteristics of digested sludge. Gas digesters in multiple-phase digestion systems showed much higher specific gas production - 0.84 STD L/ g VS<sub>destroyed</sub> /Volume (both systems), compared to conventional MAD - 0.46 STD L/ g VS<sub>destroyed</sub> /Volume. Also, the quality of biogas from gas digesters of multiple-phase digestion systems showed higher methane content than from the conventional MAD digester which was statistically different as compared by Student's t-test (alpha 0.05) and Tukey-Kramer HSD test (Sall et al., 2001). Energy calculations for all digestion systems were based on equations [7-1], [7-2], and [7-3] and are graphically presented in Figure 7-2. An additional component included for multiple-phase digestion systems was energy loss associated with thickening. For the purpose of the calculation it was assumed that a gravity thickener would be used. The construction, operation and financing costs of gravity thickener presented in Table 7-2 were converted into energy equivalent, based on the assumption that energy price is \$0.06 CAD per kWh (which was the price of energy in Winnipeg in year 2005).

**Table 7 - 1. Main parameters obtained from bench scale studies used for comparison of conventional mesophilic digestion systems to three-staged acid digestion(at mesophilic temperature) followed by mesophilic gas reactor and three-staged acid digestion(at low-mesophilic temperature) followed by mesophilic gas reactor.**

| Parameter              | Unit                                       | Raw Sludge | Thickened Raw Sludge | Control | Mesophilic Acid / Mesophilic Gas System |        |        |       | Low-Mesophilic Acid / Mesophilic Gas System |        |        |       |
|------------------------|--|------------|----------------------|---------|---|--------|--------|-------|---|--------|--------|-------|
|                        |  |            |                      |         | 1 Acid                                  | 2 Acid | 3 Acid | Gas   | 1 Acid                                      | 2 Acid | 3 Acid | Gas   |
| Digester volume        | L  |            |                      | 1.5     | 1.5                                     | 1.3    | 1.1    | 1.5   | 1.5   | 1.3    | 1.1    | 1.5   |
| Total Solids           | g TS/L                                     | 47.94      | 57.02                | 25.14   | -                                       | -      | 44.33  | 38.56 | -   | -      | 46.35  | 39.41 |
| Volatile Solids        | g VS/L                                     | 24.43      | 31.17                | 10.31   | -                                       | -      | 23.04  | 17.62 | -   | -      | 24.44  | 18.72 |
| Volatile Solids added  | g VS <sub>added</sub> /day                 |            |                      | 1.67    | 2.13                                    |        |        |       | 2.13  |        |        |       |
| VS destruction         | %  |            |                      | 57.77   |   |        | 26.06  | 43.47 |   |        | 21.58  | 39.94 |
| VS destroyed           | g/d  |            |                      | 0.96    |   |        |        | 0.92  |   |        |        | 0.85  |
| Gas Production         | STD L/day                                  |            |                      | 0.66    | 0.40                                    | 0.13   | 0.11   | 1.16  | 0.20  | 0.12   | 0.04   | 1.07  |
| Gas production         | STD L/gVS <sub>destr.</sub> /Unit Volume   |            |                      | 0.46    |   |        |        | 0.84  |   |        |        | 0.84  |
| Gas Production         | STD L/day/Unit Volume                      |            |                      | 0.44    | 0.27                                    | 0.10   | 0.10   | 0.78  | 0.13  | 0.09   | 0.03   | 0.71  |
| Average gas production | STD L/gVS <sub>added</sub>                 |            |                      | 0.26    | 0.25                                    |        |        |       | 0.17  |        |        |       |
| Average gas production | STD L/gVS <sub>destr.</sub> /System Volume |            |                      | 0.46    | 0.49                                    |        |        |       | 0.44  |        |        |       |
| Average gas production | STD L/day/System Volume                    |            |                      | 0.44    | 0.45                                    |        |        |       | 0.37  |        |        |       |
| Gas Composition        | % CH <sub>4</sub>                          |            |                      | 63.72   | 29.07                                   | 31.78  | 46.56  | 67.10 | 31.16                                       | 30.14  | 30.80  | 66.48 |
|                        | % CO <sub>2</sub>                          |            |                      | 36.28   | 70.93                                   | 68.22  | 53.44  | 32.90 | 68.84                                       | 69.86  | 69.20  | 33.52 |
| Total COD              | mg COD/L                                   | 44926      | 59441                | 21349   | -                                       | -      | 55835  | 40904 | -   | -      | 56806  | 44833 |
| Soluble COD            | mg COD/L                                   | 3756       | 4554                 | 1211    | -                                       | -      | 14579  | 6781  | -   | -      | 14402  | 8260  |
| TotalCOD destruction   | %  |            |                      | 52      |   |        | 6      | 31    |   |        | 4      | 25    |
| VFA                    | mg VFA/L                                   | 2696       | 2645                 | 409     | 9695                                    | 10620  | 8101   | 2878  | 8503  | 9163   | 9580   | 3623  |
| VFA                    | mg COD/L                                   | 3726       | 3729                 | 560     | 13337                                   | 14964  | 11250  | 3909  | 11633                                       | 12845  | 13499  | 5180  |
| pH                     | -  | 6.20       | 6.36                 | 7.55    | 5.58                                    | 5.39   | 5.45   | 7.38  | 5.61  | 5.38   | 5.30   | 7.23  |

**Equation 7 - 1**            Energy Demand = Heat Requirement - Heat recovery from Heat Exchangers + Heat Losses + Costs of Thickening

**Equation 7 - 2**            Recoverable Energy = 80% Energy from Biogas

**Equation 7 - 3**            Net Energy = Recoverable Energy – Energy Demand

Energy from the biogas can be recovered through a gas engine and converted into hot water and electricity. Efficiencies of energy recovery are as follows (Burrowes, 2005): energy of hot water equals 50% of energy from biogas, electricity generation equals 30% of energy from biogas, and total recoverable energy equals 80% of energy from biogas.

In case electricity generation is not feasible due to high heat demand, all biogas can be fired into the hot water boiler and energy can be recovered in the form of hot water. Efficiency of energy recovery through a hot water boiler is assumed to be 80% (Burrowes, 2005).

**Table 7 - 2. Costs of thickening and corresponding energy loss.**

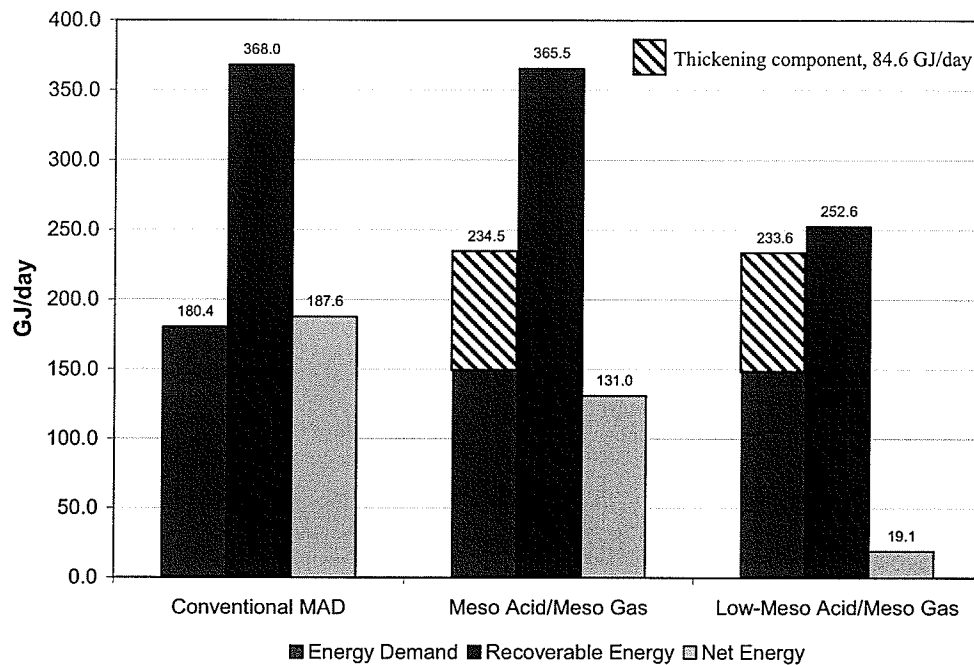
| Description   | Costs       | Source         |
|---|-------------|----------------|
| Construction + Engineering + Administrations (in year 2000)           | \$6.9M US   | E.P.A., 2003   |
| Inflation adjustment to year 2005 (x 1.122)                           | \$7.75M US  | Bruce, 1996    |
| Capital recovery factor (8% in 20 years = 0.10185)                    | \$8.54M US  | Vesilind, 1997 |
| Operation and maintenance in 20 years @ 490 hr/year and \$20 per hour | \$0.19M US  | E.P.A., 2003   |
| Total costs in 20 years (2005 - 2025)                                 | \$8.73M US  |                |
| Conversion to CAD @ 1.18  | \$10.3M CAD | XE.COM, 2004   |
| Conversion to GJ/d @ \$0.06CAD* per kWh                               | 84.6 GJ/d   |                |

\* - price of energy in Winnipeg, Manitoba

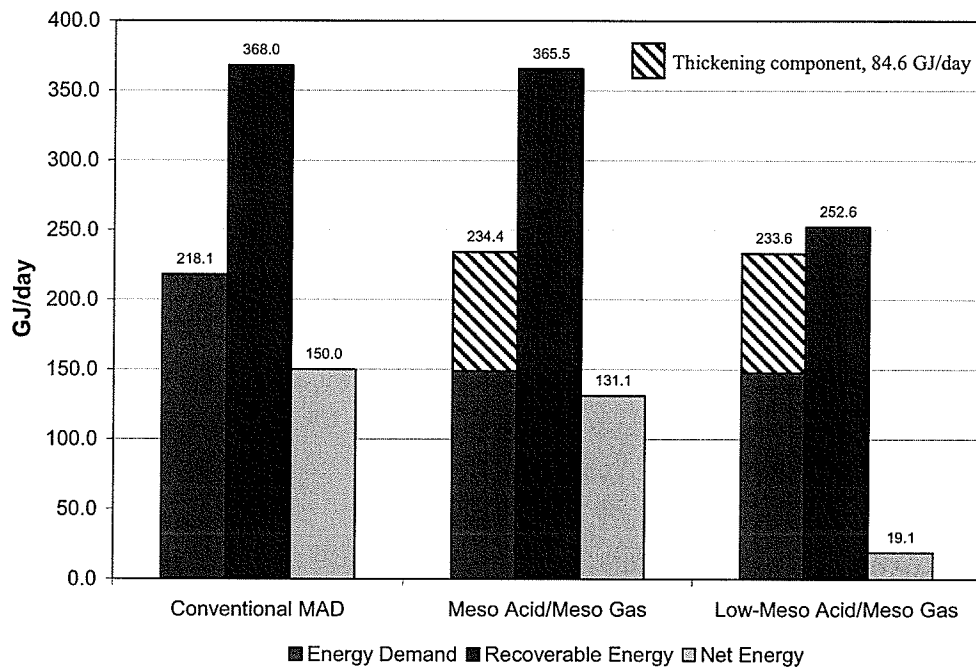
It is evident looking at Figure 7-2 that, after balancing energy components of all systems, the conventional one staged mesophilic digestion ranked the best of all. The main factors that contributed to this result were: (1) the fact that at the time of the experimentation, incoming

sludge had higher than usual TS content (4.8 %TS compared to 3.9% TS); and (2) the energy balance for multi staged digestion systems has an additional component related to the costs of thickening. Net energy balance for the conventional MAD system showed that this system would offer approximately 30% more energy than a meso acid/meso gas system and 90% more energy than a low-meso acid/meso gas system. However, if it was assumed that incoming sludge had only 3.9% TS and was thickened to 5.7% TS (scenario #2 in Table 7-4) the overall balance of energy for digestion systems showed that the configuration of meso acid/meso gas provided only 13% less net energy than conventional MAD digestion system as shown in Figure 7-3. The above calculations refer to the scenario in which all digestion systems would be operated at the same SRT. Therefore, volumes of system that digest thickened wastewater sludge would be smaller. This would result in a smaller plant footprint and lower investment costs. These factors were not included in the calculations.

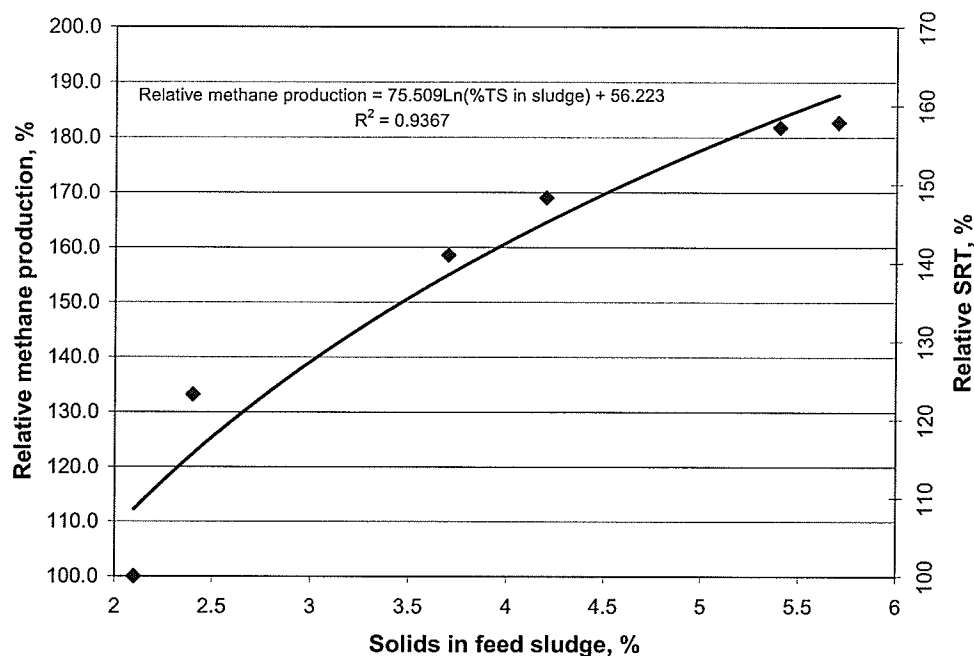
Another scenario worth presenting is the situation where digestion systems that receive thickened and un-thickened sludge are operated at the same solids retention times (scenario #3 in Table 7-4). This results in an extension of SRT in the digestion systems that receive thickened sludge. Extended SRT in turn results in additional VS destruction and additional methane production. The increase in methane production was calculated based on numerical modeling results (Figure 7-4) and is provided in Table 7-3.



**Figure 7 - 2. Results of energy calculations - control MAD at 4.8% TS in feed sludge and multi-staged digestion systems at 5.7% TS in feed sludge. All systems at loading rate of 100 tons TS/day and equal SRT.**



**Figure 7 - 3. Results of energy calculations - control MAD at 3.9% TS in feed sludge and multi-staged digestion systems at 5.7% TS in feed sludge. All systems at loading rate of 100 tons TS/day and equal SRT.**



**Figure 7 - 4. Calculated increase in methane production that results from volume reduction due to sludge thickening and extension in SRT at constant digesters' volume - based on Biowin simulation (EnviroSim\_Associates\_LTD., 2004).**

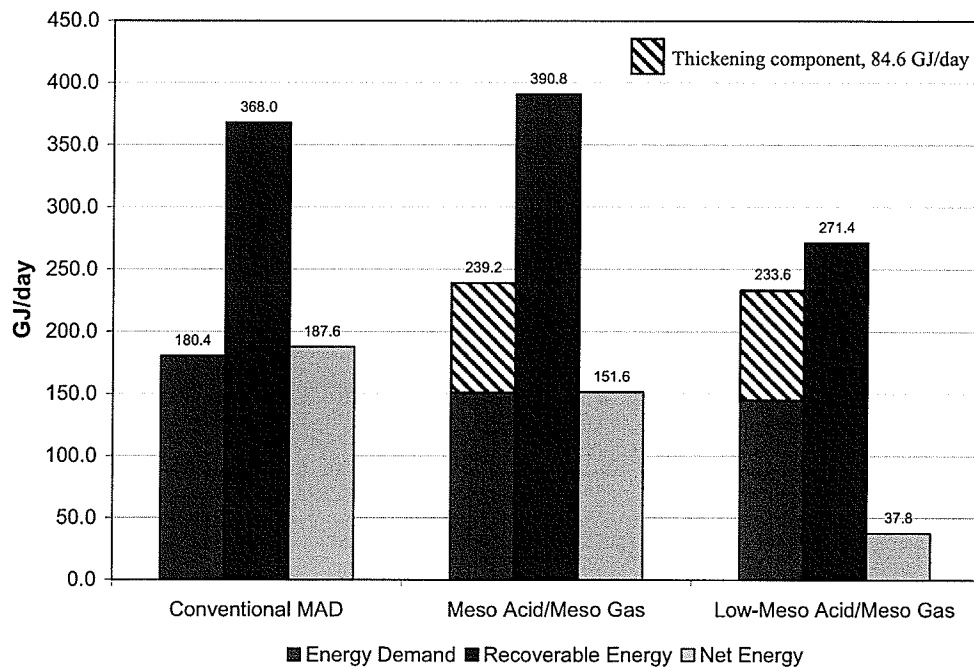
**Table 7 - 3. Effect of sludge thickening on SRT and methane production.**

| Increase in solids content |                  | Increase in SRT |                  | Increase in methane production after thickening |
|----------------------------|------------------|-----------------|------------------|---|
| Initial TS                 | After thickening | Initial SRT     | After thickening |   |
| %                          | %                | days            | days             | %   |
| 4.8                        | 5.7              | 22              | 26.1             | 6.9   |
| 3.9                        | 5.7              | 22              | 32.2             | 15.3  |

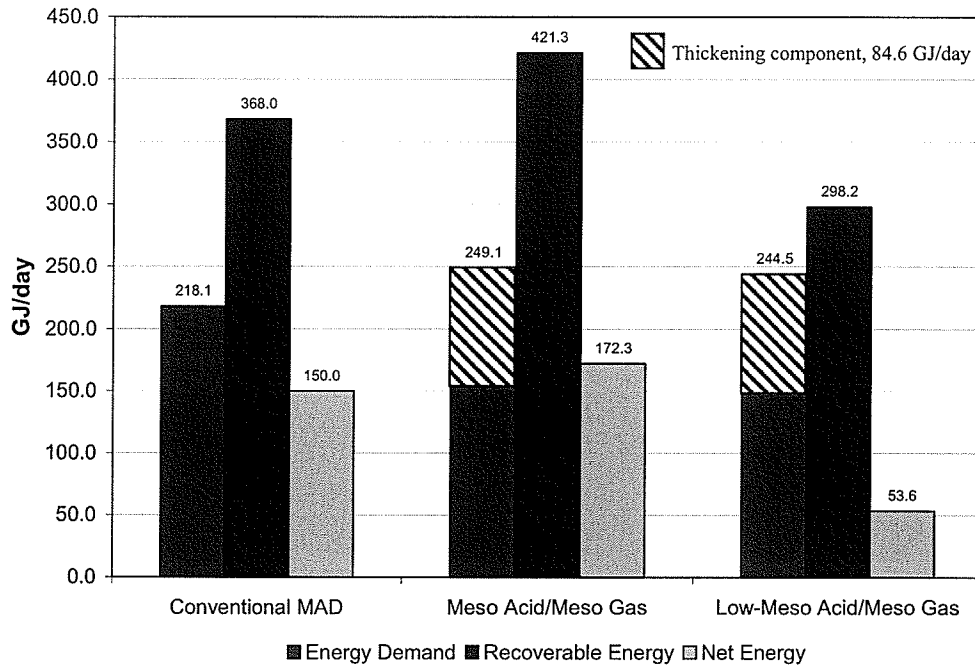
Energy balances calculated for the digestion systems of equal volumes showed that this solution is the most beneficial for the maximization of energy recovery. Thickening the sludge from 4.8% TS to 5.7% TS would result in an improvement of the net energy balance for multiple-digestion systems compared to conventional MAD due to the fact that additional benefits from extra methane production would exceed additional energy spending due to the larger digestion volumes used (Figure 7-5). Sludge thickening from 3.9% TS to 5.7% TS



(scenario #4 in Table 7-4) would result in approximately 13% more net energy generated by meso acid/meso gas system compared to conventional MAD system (costs of thickening included) as shown in Figure 7-6. Additionally, an enhanced pathogen inactivation would be achieved. In all cases, net energy balance for low-mesophilic acid/mesophilic gas system showed the least amount of net energy available at any operational configuration evaluated and from energy view point is at least desirable to implement. The only advantage this system exhibited over conventional MAD was enhanced fecal coliforms reduction (results described in section 6.3).



**Figure 7 - 5. Results of energy calculations - control MAD at 4.8% TS in feed sludge and multi-staged digestion systems at 5.7% TS in feed sludge. All systems at loading rate of 100 tons TS/day and equal volumes.**



**Figure 7 - 6. Results of energy calculations - control MAD at 3.9% TS in feed sludge and multi-staged digestion systems at 5.7% TS in feed sludge. All systems at loading rate of 100 tons TS/day and equal volumes.**

**Table 7 - 4. Summary of conditions in all scenarios presented.**

|                  | Unit                | Scenario # 1     |                            | Scenario # 2     |                            | Scenario # 3     |                            | Scenario # 4     |                            |
|------------------|---------------------|------------------|----------------------------|------------------|----------------------------|------------------|----------------------------|------------------|----------------------------|
|                  |                     | Control Digester | Advanced Digestion Systems | Control Digester | Advanced Digestion Systems | Control Digester | Advanced Digestion Systems | Control Digester | Advanced Digestion Systems |
| SRT              | days                | 22.0             | 22.0                       | 22.0             | 22.0                       | 22.0             | 26.1                       | 22.0             | 32.2                       |
| Digestion volume | m <sup>3</sup>      | 45833            | 38596                      | 56410            | 38596                      | 45833            | 45833                      | 45833            | 45833                      |
| Load             | tons TS/day         | 100              | 100                        | 100              | 100                        | 100              | 100                        | 100              | 100                        |
| Sludge thickness | % TS                | 4.8              | 5.7                        | 3.9              | 5.7                        | 4.8              | 5.7                        | 3.9              | 5.7                        |
| Sludge volume    | m <sup>3</sup> /day | 2083             | 1754                       | 2564             | 1754                       | 2083             | 1754                       | 2564             | 1754                       |

VS/TS = 0.65 for all scenarios

Advanced digestion systems = multiple-phase digestion systems fed with thickened wastewater sludge

## CONCLUSIONS

On the basis of experimental results and energy balance computations, the following conclusions were drawn:

- At total systems SRT of 22 days, conventional MAD achieved 57.8% VS destruction at 4.8% TS in feed sludge compared to 43.5% VS destruction achieved by meso

acid/meso gas systems at 5.7 %TS in feed sludge, and 39.9% achieved by low-meso acid/meso gas system also at 5.7% TS in feed sludge. Nevertheless, all of the systems tested achieved a minimum of 38% of VS destruction to comply with Class A requirement as defined by (E.P.A., 1999).

- VS destruction on the mass basis resulted in 0.96 g VS/d destroyed by conventional MAD system, and 0.92 g VS/d and 0.85 g VS/d destroyed by multiple phase meso acid/meso gas and low-meso acid/meso gas digestion systems, respectively.
- Quality of biogas from gas digesters of multiple-phase digestion systems showed higher methane content (up to 72% methane in biogas) than from conventional MAD digester which was statistically different.
- Using biogas production obtained during experimental work at total systems SRT of 22 days and assuming load of 100 kg TS/d, conventional MAD would show 187.6 GJ/d net available energy, which is energy available for recovery at 4.8% TS. Multiple-phase digestion systems at higher solids content in feed sludge (5.7% TS) resulted in 131.0 GJ/d and 19.1 GJ/d of recoverable energy for meso acid/meso gas systems and low-meso acid/meso gas system, respectively.
- The main factors that contributed to those results were the following:
  - at the time of the experimentation incoming sludge had higher than usual TS content (4.7% TS compared to 3.6% TS for NEWPCC).
  - additional energy is required for multiple-phase systems to thicken the sludge (calculated to be 84.6 GJ/d) and
  - compared to 22 days SRT in conventional MAD, retention times of gas digesters in multiple-phase systems were 13 days (meso acid/meso gas

system) and 10 days (low-meso acid/meso gas system). Even though specific gas production in those digesters was  $0.84 \text{ STP L/ g VS}_{\text{destroyed}}/\text{Volume}$  (for both systems) compared to  $0.46 \text{ STP L/ g VS}_{\text{destroyed}}/\text{Volume}$  for conventional MAD, the overall methane production from multiple-phase systems was not higher than from conventional MAD.

- Extended retention time of gas digestion in multiple-phase digestion systems would be required because at 22 days total systems SRT effluent showed high SCOD and high VFA content which would contribute to odor problems and poor dewaterability. Besides, higher VS destruction is expected at longer retention times, which would contribute to greater biogas production.
- Benefits of additional methane production at extended SRT would exceed additional energy spending due to larger volumes of multiple-digestion systems at increased solids in feed sludge (based on simulated scenarios).
- Energy calculations showed that multiple-phase digestion system (meso acid/meso gas) at increased TS content in feed sludge would be attractive from energy recovery view point if sludge would be thickened from 3.9% TS to 5.7% TS and equal digestions volumes would be used. This would allow for approximately 10 days longer SRT, 15% higher biogas production and overall 13% higher net energy recovery from meso acid/meso gas system as compared to conventional MAD (at loading of 100 kg TS/d).
- Three-stage, low-mesophilic acid digestion followed by mesophilic gas digester showed least amount of energy available for recovery and from energy view point was least desirable.

## ACKNOWLEDGMENTS

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## **8 Research overview**

Due to the current expansion of wastewater treatment plants in Winnipeg, there is an ongoing discussion, among many other topics, about sewage sludge management options. Under current treatment, sewage sludge is anaerobically digested at mesophilic temperatures, dewatered and applied to land. The quality of the final product corresponds to Class B biosolids. This study was originally initiated to determine whether thermal sludge treatment may be beneficial from an energy view point or can it be justified only for inactivation of pathogen found in sewage. In the course of this study, research was performed using wastewater sludge obtained from the North End Wastewater Pollution Control Centre, the same sludge that is anaerobically digested in Winnipeg.

Sewage sludge exhibits a wide range of chemical, biological, and physical characteristics that are dependent on local conditions and treatments applied. Therefore it should be noted, that the results and conclusions from this study are specific to local conditions in Winnipeg. However, methodology, the proposed technological changes, and results obtained could be used as guidance in other situations.

### **8.1 Summary and conclusions**

#### **8.1.1 Thermophilic vs. mesophilic anaerobic digestion ( based on section 4.2)**

The research has shown that no improvement, in terms of higher VS destruction and biogas production, as well as biogas quality, was observed between conventional mesophilic digestion of sewage sludge (essentially the same treatment that is currently applied in Winnipeg) and thermophilic digestion and two-stage, thermophilic-mesophilic digestion. Additionally, thermophilic treatments resulted in higher soluble COD and VFA in the

effluent as compared to conventional MAD treatment, which would contribute to odour problems and worse dewaterability properties indicated by much higher capillary suction times.

### **8.1.2 Thermophilic vs. mesophilic hydrolysis (based on section 4.3 and 5.1)**

The impact of temperature on sludge hydrolysis was investigated. Hydrolysis is considered a bottleneck of the digestion process that may be considerably improved by application of higher temperature. It was found out that the hydrolysis rate of wastewater sludge digested in thermophilic conditions was on average only 6% higher than the hydrolysis rate of wastewater sludge digested under mesophilic conditions (difference statistically not significant). Thermophilic temperature had very little impact on the improvement of hydrolysis rates compared to mesophilic temperatures. The fact that the thermophilic digesters contained higher soluble COD and VFA in the effluent (that could be potentially converted into biogas) resulted in no overall improvement of biogas production and VS destruction compared to mesophilic digestion. Therefore, thermophilic digestion was not found beneficial from the energy view point of the process.

However, thermophilic processes resulted in a high level of pathogen inactivation including fecal coliforms and *Ascaris suum* eggs, which would allow for the final product to meet Class A standards, while mesophilic digestion provided much lower levels of pathogen inactivation and was able to produce Class B biosolids. Therefore, an alternative, non-thermophilic method of pathogen inactivation was investigated.

### **8.1.3 Pathogen inactivation through non-thermophilic acid digestion (based on chapter 6)**

The mechanism responsible for inactivation of pathogens under non-thermophilic conditions was associated with high concentrations of un-ionized organic acids. The un-ionized form of organic acid exists in environment with low pH. The toxicity from un-ionized VFA is based on the cytoplasm acidification from protons released after acid ionization inside the bacteria cell and anion accumulation inside the cell which may lead to osmotic stress. It was possible to create these conditions in acid digesters at mesophilic (38°C) and low-mesophilic (24°C) conditions.

Batch acid digesters were able to achieve Class A biosolids standards with respect to fecal coliforms concentrations in both operating conditions: mesophilic and low-mesophilic. Semi-continuously fed acid digesters (essentially sequencing batch reactors) were most effective when operated in series (possibly plug-flow conditions would be the best). Three acid digesters operated in series under mesophilic conditions were able to consistently achieve Class A level for fecal coliforms, while three acid digesters operated in series in low-mesophilic conditions were able to achieve, on average, compliance with Class A limit, however not on a consistent basis (which is essential).

The pattern of fecal coliforms inactivation observed in section 6.1 differs substantially from later observations. Class A was achieved later in low-mesophilic conditions only in batch mode at a longer retention time of approximately 7 to 9 days (section 6.2.) as opposed to 5 days (presented in section 6.1.). Mesophilic conditions achieved Class A in subsequent experiments in batch mode (section 6.2.) and in semi-continuous operation of staged acid digestion (section 6.3.). The discrepancy of results does not undermine theory presented. The



main reasons for successful fecal coliforms inactivation were attributed to low pH and the presence of VFA in un-ionized form. The critical component of toxic environment - low pH - did not last long enough in mesophilic conditions in section 6.1 to provide adequate fecal coliforms inactivation. In subsequent experiments, when low pH was present in mesophilic conditions for the duration of acid digestion, excellent fecal coliforms inactivation was observed. At the same time, low-mesophilic conditions never showed the potential to achieve Class A within 5 days in batch conditions (indicated in section 6.1.) nor low-mesophilic conditions could achieve Class A during semi-continuous staged acid digestion. Later observations at low-mesophilic conditions in batch mode (section 6.2.) and semi-continuous operated staged acid digestion systems (section 6.2. and 6.3.) were consistent, therefore findings presented in section 6.1. were not confirmed later.

#### **8.1.4 Impact of sludge thickening on hydrolysis rates (based on section 5.1)**

In order to increase concentrations of organic acids, sludge was thickened prior to digestion. On the other hand, increasing solids content in sludge results in lower amount of biogas produced per gram VS added. To estimate the effect of environmental conditions on digestion, including sludge thickness and operating temperature, hydrolysis tests were performed. Hydrolysis rates of raw, un-thickened sludge were found to be a function of temperature, with highest hydrolysis rate under thermophilic conditions (55°C) and lowest under low-mesophilic conditions (24°C). (It should be noted again that the thermophilic hydrolysis rate being 6% higher than the mesophilic rate did not result in increased VS destruction and/or additional biogas production). Optimum sludge solids content was defined as solids content that does not inhibit hydrolysis rates at any given operating temperature, which should result in uninhibited VS destruction and biogas production. Increase in solids

content in raw, un-thickened sludge, from initial concentration of 3.6% TS accompanied by proportional increase in hydrolysis rate was found to be approximately 50% or up to 5.4% TS for low-mesophilic conditions, 70% or up to 6.1% TS for mesophilic conditions, and 100% or up to 7.2% TS for thermophilic conditions.

#### **8.1.5 Impact of sludge thickening on anaerobic digestion (based on chapter 7)**

Thickening the sludge results in volume reduction which can be used for extension of SRT at constant digestion volume or smaller digestion volume required. Extension of SRT brings additional VS destruction and biogas production, while volume reduction brings operational savings (less heat required to maintain process temperature) and/or space and construction savings. Both scenarios were investigated and it was concluded that additional digestion time achieved from sludge thickening would be the most cost effective solution if infrastructure is available (which is the case in Winnipeg). Extension of SRT depends mainly on the degree of sludge thickening; for instance thickening the sludge from 3.9% TS (long term average at NEWPCC) at 22 days SRT, it is possible to achieve 32.2 days SRT at 5.7% TS in sludge. Increasing the solids content by 0.9 %TS (from 4.8% TS to 5.7% TS – laboratory experiment) or 1.7% TS (from 3.9% TS to 5.7% TS – simulated data) would increase digestion SRT by additional 4 to 10 days. It could potentially increase methane production by 6.9% to 15.3%.

#### **8.1.6 Conventional MAD vs. novel systems – comparison (based on section 6.3 and chapter 7)**

The last stage of experimental research involved comparison of conventional mesophilic anaerobic digestion and two multi-phase digestion systems: three-phase mesophilic acid

digestion followed by mesophilic gas digestion (meso acid/meso gas) and three-phase low-mesophilic acid digestion followed by mesophilic gas digestion (low-meso acid/meso gas). Multi-phase digestion systems were operated on thickened wastewater sludge (approximately 5.7% TS) while the conventional digestion system was operated on raw, un-thickened sludge (approximately 4.8% TS). Parameters compared included pathogen inactivation potential using two indicator organisms: fecal coliform and *Ascaris suum* ova, digestion parameters such as VS destruction, gas production and gas quality needed to assess energy balance for the systems, and the effluent quality such as soluble COD and VFA content.

In terms of pathogen inactivation, the meso acid/meso gas system achieved Class A limit with respect to fecal coliforms, low-meso acid/meso gas achieved the Class A limit on average but not consistently, and conventional MAD showed the highest concentrations of fecal coliforms within Class B range. Effluents from the meso acid/meso gas system showed the lowest viability of *Ascaris suum* ova from all the systems tested with complete inactivation of eggs enclosed in sentinels (controlled retention time) and 2.8% viability of free floating eggs (possibly due to short-circuiting). The other two systems showed poor inactivation of *Ascaris suum* ova showing their viability in the range of 60% to 80% in the final effluent.

Calculations of energy balance for three digestions systems included the following scenarios: (1) all systems operated at equal SRT of 22 days (based on laboratory results), (2) conventional mesophilic digestion operated at lower solids content in feed sludge - 3.9% TS (which was long-term average for sludge from North End Pollution Control Centre in Winnipeg) versus 4.8% TS which was average solids content in sludge during the time of experimentation (simulated results), (3) digestion systems operated at equal volumes

(extended SRT for multi-phase digestion systems) and conventional digester operated on raw, un-thickened sludge with 4.8% TS content (simulated results), and (4) digestion systems operated at equal volumes (extended SRT for multi-phase digestion systems) and conventional digester operated on raw, un-thickened sludge with 3.9% TS content (simulated results). All calculations were based on 100 kg TS/day load. It was also assumed that recoverable energy equalled 80% of energy from biogas.

Using biogas production obtained during experimental work at a total systems SRT of 22 days (scenario #1) conventional digester showed 187.6 GJ/d net available energy, which is energy available for recovery at 4.8% TS. Multiple-phase digestion systems at higher solids content in feed sludge (5.7% TS) resulted in 131.0 GJ/d and 19.1 GJ/d of recoverable energy for meso acid/meso gas systems and low-meso acid/meso gas system, respectively. The main factors that contributed to those results were the following: at the time of the experimentation incoming sludge had higher than usual TS content (4.7% TS compared to 3.6% TS for NEWPCC), additional energy is required for multiple-phase systems to thicken the sludge (calculated to be 84.6 GJ/d) and compared to 22 days SRT in conventional MAD, retention times of gas digesters in multiple-phase systems were 13 days (meso acid/meso gas system) and 10 days (low-meso acid/meso gas system). Even though specific gas production in those digesters was  $0.84 \text{ STP L/g VS}_{\text{destroyed}}/\text{Volume}$  (for both systems) compared to  $0.46 \text{ STP L/g VS}_{\text{destroyed}}/\text{Volume}$  for conventional MAD, overall methane production from multiple-phase systems was not higher than from conventional MAD.

Extended retention time of gas digestion in multiple-phase digestion systems would be required because at 22 days total systems SRT effluent showed high SCOD and high VFA content which would contribute to odor problems and poor dewaterability. Besides, higher

VS destruction is expected at longer retention times, which would contribute to greater biogas production.

Energy calculations showed that one of the multiple-phase digestion systems tested -meso acid/meso gas - at increased TS content in feed sludge would be attractive from an energy recovery view point if the sludge would be thickened from 3.9% TS to 5.7% TS at the same digestion volume as conventional digestion system. This would allow for approximately 10 days longer SRT in the multi-phase digestion system, 15% higher biogas production and an overall 13% higher net energy recovery (including costs of thickening) from meso acid/meso gas system as compared to conventional digester. Benefits of additional methane production at extended SRT would exceed additional energy spending due to operating larger volumes of multiple-digestion system at increased solids in feed sludge. Besides, quality of biogas from gas digesters from multiple-phase digestion systems showed higher methane content (up to 72% methane in biogas) than from conventional digesters, which was statistically different.

## 8.2 Engineering significance

The work presented addressed several issues related to anaerobic digestion technology. Probably the most significant contribution to the current state of knowledge in this area was related to the development of alternative methods of non-thermophilic pathogen inactivation through the toxicity from un-ionized form of VFA. Application of knowledge from different fields of science such as microbiology and food science, combined with two-phase, acid-gas anaerobic digestion technology, allowed for enhanced pathogen inactivation in acid, non-thermophilic digestion. Many wastewater treatment facilities are equipped with several digestion vessels, which would allow for practical application of the findings presented in this thesis through proper sludge flow management. Instead of using several anaerobic digesters operated in parallel as conventional one-phase mode, redirecting the flow to achieve in-series digestion operation, would allow for the creation of the acid stage of digestion in some vessels with enhanced pathogen inactivation, and gas digestion in remaining vessels. The advantage of this solution is relative low-cost associated with flow redirection, and no additional construction and/or operational costs, for instance additional heat exchangers. The proposed solution may be especially attractive for plants that need to upgrade their sludge quality when application of thermophilic digestion is not possible due to structural limitations of digestion facility, such as the case of Winnipeg.

Another aspect of this work is related to improvement of energy recovery from anaerobic digestion. This could be achieved through sludge thickening and flow reduction, which would allow for additional retention time resulting in additional VS destruction and biogas production. Additional construction and operational costs associated with sludge thickening could be offset by additional energy recovery. Cost effectiveness of this solution is dependent

on the initial solids content in raw, un-thickened sludge, size of the facility and price of energy. It should be remembered that digestion of thickened wastewater sludge generates higher concentrations of VFAs which is beneficial for pathogen inactivation in acid stage of digestion.

Finally, this work contains one of the most comprehensive study on hydrolysis of wastewater sludge with analysis of many environmental factors including operating temperature and solids content in sludge. Knowledge of hydrolysis rates under different environmental conditions may be useful in defining optimum solids content for digestion at any given operating temperature. The optimum solids content was defined as the solids content that did not decrease hydrolysis rates from increased loading on digester.

### 8.3 Future research

There are several areas that require additional research to complete the study:

- Under US. EPA requirements for Class A biosolids Alternative 3: Sewage Sludge Treated in Other Processes (which is applicable in this case) Class A is obtained through comprehensive monitoring of bacteria, enteric viruses and helminth ova to demonstrate adequate reduction of pathogens. Enteric viruses were not measured in this study and future research should address the issue of inactivation of enteric viruses.
  - As indicated in section 4.1. thermophilic and mesophilic digestion systems achieved very poor inactivation of *Clostridium perfringens*. Possibly, better results could be achieved through un-ionized VFA toxicity.
- It was assumed in this study that sludge thickening can be achieved through a gravity thickener. Additional research should assess the ability of gravity thickeners to achieve the desired solids level in sludge.
- Additional experiments with acid digesters should investigate the presence of different intermediate digestion products, such as lactate or ethanol that may influence inactivation of pathogens in sludge.
- Further optimization of SRT in acid digestion should be studied. There is a possibility of adding more digesters in series and shortening SRT in the acid digestion part of the process.



## 9 References

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**Table A - 2. Results of free floating *Ascaris suum* viability study.**

| Day | Meso           |        |            |          |        |            |             |
|-----|----------------|--------|------------|----------|--------|------------|-------------|
|     | Sample 1       |        |            | Sample 1 |        |            | Average     |
|     | Total          | Viable | Non-viable | Total    | Viable | Non-viable | % viable    |
| 1   | 100            | 88     | 12         | 100      | 90     | 10         | <b>89</b>   |
| 3   | 100            | 81     | 19         | 100      | 87     | 13         | <b>84</b>   |
| 5   | 100            | 74     | 26         | 100      | 80     | 20         | <b>77</b>   |
| 10  | 100            | 77     | 23         | 100      | 74     | 26         | <b>75.5</b> |
| 15  | 100            | 75     | 25         | 100      | 72     | 28         | <b>73.5</b> |
| Day | Thermo         |        |            |          |        |            |             |
|     | Sample 1       |        |            | Sample 1 |        |            | Average     |
|     | Total          | Viable | Non-viable | Total    | Viable | Non-viable | % viable    |
| 1   | 100            | 0      | 100        | 100      | 0      | 100        | <b>0</b>    |
| 3   | 100            | 0      | 100        | 100      | 0      | 100        | <b>0</b>    |
| 5   | 43             | 0      | 43         |          |        |            | <b>0</b>    |
| 10  | 100            | 0      | 100        | 29       | 0      | 29         | <b>0</b>    |
| 15  | 100            | 0      | 100        |          |        |            | <b>0</b>    |
| Day | 1TPAD - Thermo |        |            |          |        |            |             |
|     | Sample 1       |        |            | Sample 1 |        |            | Average     |
|     | Total          | Viable | Non-viable | Total    | Viable | Non-viable | % viable    |
| 1   | 100            | 0      | 100        | 100      | 0      | 100        | <b>0</b>    |
| 3   | 100            | 0      | 100        | 100      | 0      | 100        | <b>0</b>    |
| 5   | 100            | 0      | 100        | 100      | 0      | 100        | <b>0</b>    |
| Day | 2TPAD - Meso   |        |            |          |        |            |             |
|     | Sample 1       |        |            | Sample 1 |        |            | Average     |
|     | Total          | Viable | Non-viable | Total    | Viable | Non-viable | % viable    |
| 1   | 0              | 0      | 0          | 0        | 0      | 0          | <b>-</b>    |
| 3   | 8              | 0      | 8          |          | 0      |            | <b>0</b>    |
| 5   | 100            | 0      | 100        | 16       | 0      | 16         | <b>0</b>    |
| 10  | 100            | 0      | 100        | 100      | 0      | 100        | <b>0</b>    |
| 15  | 1              | 0      | 1          |          | 0      |            | <b>0</b>    |

**Table A - 3. Results of a 24-hour Ascaris suum viability test in thermophilic digester – sentinels.**

| Hours: | Sample #: | # Larvated | # Unlarvated | % Viable: | Average |
|--------|-----------|------------|--------------|-----------|---------|
| 24     | 1         | 0          | 100          | 0         | 0       |
| 24     | 2         | 0          | 100          | 0         |         |
| 24     | 3         | 0          | 100          | 0         |         |
| 12     | 1         | 0          | 100          | 0         | 0       |
| 12     | 2         | 0          | 100          | 0         |         |
| 12     | 3         | 0          | 100          | 0         |         |
| 10     | 1         | 0          | 100          | 0         | 0       |
| 10     | 2         | 0          | 100          | 0         |         |
| 10     | 3         | 0          | 100          | 0         |         |
| 8      | 1         | 0          | 100          | 0         | 0       |
| 8      | 2         | 0          | 100          | 0         |         |
| 8      | 3         | 0          | 100          | 0         |         |
| 6      | 1         | 0          | 100          | 0         | 0       |
| 6      | 2         | 0          | 100          | 0         |         |
| 6      | 3         | 0          | 100          | 0         |         |
| 4      | 1         | 0          | 100          | 0         | 0       |
| 4      | 2         | 0          | 100          | 0         |         |
| 4      | 3         | 0          | 100          | 0         |         |
| 2      | 1         | 0          | 100          | 0         | 0       |
| 2      | 2         | 0          | 100          | 0         |         |
| 2      | 3         | 0          | 100          | 0         |         |
| 1      | 1         | 50         | 50           | 50        | 44.7    |
| 1      | 2         | 65         | 45           | 59        |         |
| 1      | 3         | 25         | 75           | 25        |         |

**Table A - 4. Results of a 24-hour Ascaris suum viability test in thermophilic digester – free floating eggs.**

| Hour | % Viable: |
|------|-----------|
| 1    | 0         |
| 2    | 0         |
| 4    | 0         |
| 6    | 0         |
| 8    | 0         |
| 10   | 0         |
| 12   | 0         |
| 24   | 0         |

Table A - 5. Clostridium perfringens counts for all sludges and all experimental phases.

Table with 48 columns and multiple rows. Columns include Date, Day, PS, Thermo, Meso, and 1TPAD. Rows are organized into four sections for different experimental phases: Phase 1, Phase 2, and Phase 3. Each row contains dilution factors, results in MPN/ml and TS/g, and sample identifiers.



**Table A - 6. Volatile solids – all sludges and all experimental phases.**

|                |              | Volatile Solids - Phase 1 |              |              |              |              |              |              |              |              |  |
|----------------|--------------|---------------------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--|
| Week of        | PS           |                           | M            |              | 1TP          |              | 2TP          |              | T            |              |  |
|                | Total        | Volatile                  | Total        | Volatile     | Total        | Volatile     | Total        | Volatile     | Total        | Volatile     |  |
| 21/08/2002     | 32.13        | 19.72                     | 26.75        | 13.11        | 32.19        | 18.94        | 28.00        | 12.55        |              |              |  |
|                | 31.55        | 19.49                     | 27.20        | 13.32        | 32.80        | 19.35        | 28.40        | 13.32        |              |              |  |
|                | 13.40        | 8.81                      | 27.39        | 13.66        | 29.57        | 17.70        | 29.24        | 14.10        | 28.32        | 13.41        |  |
| 26/08/2002     | 13.26        | 8.69                      | 27.35        | 13.82        | 29.66        | 17.93        | 29.02        | 14.12        | 28.52        | 13.59        |  |
|                | 29.56        | 18.87                     | 24.23        | 11.97        | 24.72        | 14.58        | 28.92        | 14.01        | 25.95        | 11.91        |  |
|                | 29.31        | 18.77                     | 24.15        | 11.90        | 24.52        | 14.41        | 28.68        | 14.02        | 26.36        | 12.28        |  |
| 04/09/2002     | 30.61        | 21.79                     | 20.70        | 12.15        | 23.96        | 14.57        | 23.77        | 11.53        | 23.09        | 10.93        |  |
|                | 31.29        | 22.79                     | 20.39        | 11.94        | 23.90        | 14.52        | 23.92        | 11.70        | 23.10        | 10.91        |  |
|                | 26.47        | 18.93                     | 18.33        | 9.65         | 26.01        | 17.06        | 21.91        | 11.24        | 23.06        | 11.81        |  |
| 11/09/2002     | 27.00        | 19.20                     | 18.28        | 9.75         | 26.04        | 17.24        | 21.46        | 11.16        | 22.50        | 11.27        |  |
|                | 34.81        | 23.53                     | 18.35        | 9.74         | 22.04        | 13.37        | 21.70        | 11.32        | 25.49        | 16.84        |  |
|                | 33.36        | 22.44                     | 18.37        | 9.67         | 22.08        | 11.21        | 21.34        | 11.12        | 26.85        | 17.87        |  |
| 17/09/2002     | 23.46        | 16.07                     | 20.47        | 10.93        | 29.76        | 19.64        | 18.69        | 9.72         | 22.60        | 11.66        |  |
|                | 23.42        | 15.91                     | 20.39        | 10.80        | 29.34        | 19.37        | 18.90        | 10.01        | 22.90        | 11.76        |  |
|                | 60.14        | 31.14                     | 24.50        | 11.39        | 44.36        | 23.66        | 21.97        | 10.99        | 25.79        | 11.91        |  |
| 25/09/2002     | 62.94        | 31.86                     | 24.40        | 11.42        | 43.30        | 23.00        | 22.14        | 11.09        | 26.09        | 12.11        |  |
|                | 52.97        | 29.12                     | 27.60        | 12.93        | 47.94        | 25.54        | 25.30        | 12.62        | 29.38        | 13.85        |  |
|                | 51.81        | 28.67                     | 27.74        | 12.90        | 47.91        | 25.45        | 25.26        | 12.58        | 29.32        | 13.64        |  |
| <b>Average</b> | <b>36.30</b> | <b>22.39</b>              | <b>22.62</b> | <b>11.47</b> | <b>31.30</b> | <b>18.24</b> | <b>23.77</b> | <b>11.81</b> | <b>25.58</b> | <b>12.86</b> |  |
|                |              | Volatile Solids - Phase 2 |              |              |              |              |              |              |              |              |  |
| Week of        | PS           |                           | M            |              | 1TP          |              | 2TP          |              | T            |              |  |
|                | Total        | Volatile                  | Total        | Volatile     | Total        | Volatile     | Total        | Volatile     | Total        | Volatile     |  |
| 30/10/2002     | 30.37        | 25.82                     | 20.02        | 11.02        | 29.44        | 21.03        | 20.30        | 11.34        | 20.67        | 11.08        |  |
|                | 35.73        | 26.97                     | 19.82        | 11.04        | 29.07        | 20.81        | 20.60        | 11.42        | 21.14        | 11.36        |  |
|                | 35.49        | 25.93                     | 19.83        | 11.84        | 31.40        | 22.92        | 20.66        | 12.35        | 19.79        | 11.60        |  |
| 06/11/2002     | 35.83        | 26.11                     | 19.98        | 11.70        | 30.92        | 22.53        | 21.17        | 12.64        | 19.90        | 11.53        |  |
|                | 36.14        | 26.03                     | 20.67        | 12.09        | 27.14        | 18.80        | 20.07        | 11.79        | 20.85        | 11.99        |  |
|                | 36.18        | 26.37                     | 20.69        | 12.07        | 27.44        | 19.00        | 20.47        | 12.07        | 21.30        | 12.22        |  |
| 13/11/2002     | 32.87        | 24.24                     | 20.27        | 11.57        | 34.41        | 23.99        | 19.67        | 11.26        | 20.81        | 11.78        |  |
|                | 32.40        | 23.36                     | 20.66        | 11.71        | 33.28        | 23.15        | 19.30        | 11.07        | 21.15        | 11.98        |  |
|                | 30.53        | 21.92                     | 20.34        | 11.51        | 28.71        | 19.78        | 21.07        | 11.90        | 21.15        | 11.90        |  |
| 18/11/2002     | 30.33        | 21.74                     | 20.46        | 11.54        | 29.13        | 20.10        | 20.69        | 11.58        | 20.95        | 11.68        |  |
|                | 33.69        | 24.75                     | 19.82        | 11.14        | 25.88        | 17.97        | 20.45        | 11.42        | 20.26        | 11.50        |  |
|                | 33.07        | 24.35                     | 20.00        | 11.18        | 28.41        | 19.67        | 20.33        | 11.32        | 20.14        | 11.34        |  |
| 25/11/2002     | 34.54        | 25.33                     | 19.24        | 11.03        | 30.61        | 21.54        | 18.98        | 10.55        | 20.46        | 11.72        |  |
|                | 34.52        | 25.22                     | 20.00        | 11.34        | 29.63        | 20.74        | 19.08        | 10.64        | 20.45        | 11.67        |  |
|                | 30.42        | 22.77                     | 20.49        | 11.59        | 30.40        | 21.07        | 19.95        | 10.95        | 20.95        | 11.71        |  |
| 02/12/2002     | 31.32        | 23.53                     | 21.11        | 11.77        | 31.19        | 21.55        | 20.09        | 11.07        | 20.68        | 11.58        |  |
|                | 29.71        | 22.50                     | 19.91        | 11.60        | 26.84        | 19.38        | 19.32        | 10.99        | 18.50        | 12.50        |  |
|                | 29.25        | 22.13                     | 19.83        | 11.49        | 27.06        | 19.73        | 19.18        | 10.93        | 18.30        | 10.67        |  |
| 09/12/2002     | 41.23        | 27.08                     | 19.03        | 10.85        | 26.54        | 19.08        | 18.73        | 10.54        | 19.93        | 11.68        |  |
|                | 41.83        | 27.48                     | 19.12        | 10.92        | 25.41        | 18.22        | 18.87        | 10.58        | 19.90        | 11.71        |  |
| <b>Average</b> | <b>34.69</b> | <b>24.70</b>              | <b>20.75</b> | <b>11.57</b> | <b>30.81</b> | <b>21.12</b> | <b>20.58</b> | <b>11.45</b> | <b>21.37</b> | <b>11.89</b> |  |
|                |              | Volatile Solids - Phase 3 |              |              |              |              |              |              |              |              |  |
| Week of        | PS           |                           | M            |              | 1TP          |              | 2TP          |              | T            |              |  |
|                | Total        | Volatile                  | Total        | Volatile     | Total        | Volatile     | Total        | Volatile     | Total        | Volatile     |  |
| 01/01/2003     | 39.60        | 25.06                     | 18.34        | 10.38        | 25.80        | 19.74        | 22.33        | 13.41        | 21.55        | 12.08        |  |
|                | 39.63        | 25.05                     | 18.10        | 10.25        | 25.22        | 19.27        | 20.19        | 11.23        | 21.67        | 12.20        |  |
|                | 41.10        | 26.36                     | 21.97        | 12.10        | 37.97        | 23.63        | 13.17        | 5.68         | 25.09        | 13.66        |  |
| 06/01/2003     | 40.69        | 25.95                     | 22.13        | 12.12        | 38.15        | 23.78        | 17.14        | 9.58         | 25.11        | 13.67        |  |
|                | 56.23        | 34.59                     | 22.90        | 12.08        | 44.96        | 28.06        | 24.75        | 13.15        | 24.89        | 13.34        |  |
|                | 58.11        | 35.65                     | 22.74        | 12.01        | 44.76        | 27.93        | 24.78        | 13.17        | 24.65        | 13.25        |  |
| 13/01/2003     | 60.61        | 36.94                     | 28.72        | 14.39        | 55.39        | 32.77        | 29.46        | 14.95        | 30.53        | 15.53        |  |
|                | 60.89        | 37.09                     | 28.65        | 14.35        | 55.01        | 32.40        | 29.41        | 14.83        | 29.84        | 15.29        |  |
|                | 31.83        | 23.99                     | 31.13        | 15.35        | 64.52        | 38.13        | 29.83        | 14.67        | 31.93        | 15.84        |  |
| 20/02/2003     | 31.76        | 23.93                     | 30.88        | 15.18        | 65.58        | 38.86        | 29.86        | 14.56        | 31.32        | 15.61        |  |
|                | 31.32        | 23.63                     | 26.19        | 13.50        | 30.23        | 21.64        | 27.82        | 14.22        | 27.25        | 14.57        |  |
|                | 31.43        | 23.63                     | 25.97        | 13.49        | 30.24        | 21.70        | 26.87        | 13.67        | 26.99        | 14.46        |  |
| 27/02/2003     | 30.76        | 23.68                     | 25.40        | 13.61        | 29.64        | 21.90        | 26.04        | 13.82        | 25.47        | 14.01        |  |
|                | 31.12        | 23.93                     | 25.49        | 13.58        | 28.52        | 20.77        | 26.29        | 13.92        | 25.29        | 13.90        |  |
|                | 29.78        | 22.59                     | 22.40        | 12.03        | 26.40        | 19.68        | 22.55        | 12.39        | 22.05        | 12.36        |  |
| 03/02/2003     | 29.37        | 22.38                     | 22.73        | 12.41        | 26.51        | 19.64        | 22.56        | 12.36        | 21.90        | 12.26        |  |
|                | 34.80        | 25.86                     | 20.50        | 11.22        | 28.49        | 21.04        | 21.69        | 12.19        | 21.26        | 12.21        |  |
|                | 35.42        | 26.43                     | 20.35        | 11.12        | 28.61        | 21.06        | 20.42        | 11.12        | 21.23        | 12.22        |  |
| 10/02/2003     | 33.11        | 24.19                     | 21.23        | 11.76        | 35.38        | 25.30        | 22.14        | 12.61        | 21.27        | 12.19        |  |
|                | 32.73        | 23.62                     | 21.11        | 11.70        | 35.58        | 25.45        | 21.68        | 12.32        | 21.09        | 12.08        |  |
|                | 38.37        | 27.38                     | 20.32        | 11.19        | 31.64        | 22.70        | 21.08        | 12.07        | 20.84        | 12.03        |  |
| <b>Average</b> | <b>38.95</b> | <b>26.79</b>              | <b>23.52</b> | <b>12.50</b> | <b>37.31</b> | <b>24.93</b> | <b>23.69</b> | <b>12.63</b> | <b>24.65</b> | <b>13.40</b> |  |

**Table A - 7. Gas production – experimental Phase 1.**

| Date       | Phase 1                  |                  |                  |                     |                   |
|------------|--------------------------|------------------|------------------|---------------------|-------------------|
|            | Gas production STD L/day |                  |                  |                     |                   |
|            | Meso<br>std L/d          | 2TPAD<br>std L/d | 1TPAD<br>std L/d | 1+2 TPAD<br>std L/d | Thermo<br>std L/d |
| 26/08/2002 | 0.5954                   | 0.9207           | 0.0406           | 0.7513              | 0.4745            |
| 27/08/2002 | 0.5872                   | 0.7850           | 0.0371           | 0.6444              | 0.3617            |
| 28/08/2002 | 0.6340                   | 0.5653           | 0.0450           | 0.4915              | 0.4092            |
| 29/08/2002 | 0.7076                   | 0.5764           | 0.0453           | 0.5002              | 0.4447            |
| 30/08/2002 | 0.5654                   | 0.4919           | 0.0337           | 0.4194              | 0.6135            |
| 31/08/2002 | 0.5304                   | 0.5391           | 0.0464           | 0.4738              | 0.5180            |
| 01/09/2002 | 0.6533                   | 0.5723           | 0.0491           | 0.5029              | 0.4418            |
| 02/09/2002 | 0.6257                   | 0.6612           | 0.0386           | 0.5537              | 0.5925            |
| 03/09/2002 |                          | 0.7050           | 0.0423           | 0.5923              | 0.5693            |
| 04/09/2002 |                          | 0.5782           | 0.0551           | 0.5163              | 0.6461            |
| 05/09/2002 |                          | 0.5107           | 0.0285           | 0.4257              | 0.4741            |
| 06/09/2002 |                          | 0.5827           | 0.0331           | 0.4867              | 0.6556            |
| 07/09/2002 |                          | 0.6529           | 0.0571           | 0.5754              | 0.6267            |
| 08/09/2002 |                          | 0.6397           | 0.0518           | 0.5575              | 0.7746            |
| 09/09/2002 | 0.6728                   | 0.8050           | 0.0403           | 0.6641              | 0.6702            |
| 10/09/2002 | 0.7182                   | 0.7649           | 0.0272           | 0.6145              | 0.5959            |
| 11/09/2002 | 0.7759                   | 0.6466           | 0.0452           | 0.5527              | 0.7020            |
| 12/09/2002 | 0.8064                   | 0.6557           | 0.0329           | 0.5411              | 0.5126            |
| 13/09/2002 | 0.7184                   | 0.6650           | 0.0310           | 0.5453              | 0.5769            |
| 14/09/2002 | 0.7803                   | 0.7356           | 0.0549           | 0.6341              | 0.6443            |
| 15/09/2002 | 0.8647                   | 0.8463           | 0.0521           | 0.7129              | 0.7773            |
| 16/09/2002 | 0.7559                   | 0.8343           | 0.0373           | 0.6817              | 0.6299            |
| 17/09/2002 | 0.6297                   | 0.7915           | 0.0278           | 0.6353              | 0.5110            |
| 18/09/2002 | 0.5768                   | 0.7054           | 0.0211           | 0.5606              | 0.5593            |
| 19/09/2002 | 0.5967                   | 0.6911           | 0.0176           | 0.5447              | 0.2938            |
| 20/09/2002 | 0.7363                   | 0.7597           | 0.0297           | 0.6143              | 0.8622            |
| 21/09/2002 | 0.8604                   | 0.8645           | 0.0399           | 0.7082              | 0.8978            |
| 22/09/2002 | 0.9049                   | 0.8420           |                  |                     | 0.9402            |
| 23/09/2002 | 0.7630                   | 0.8836           | 0.0339           | 0.7136              | 0.8851            |
| 24/09/2002 | 0.9610                   | 0.9742           | 0.0112           | 0.7474              | 0.4668            |
| 25/09/2002 | 0.7082                   | 0.7136           | 0.0392           | 0.5940              | 0.6041            |
| Average    | <b>0.7107</b>            | <b>0.7084</b>    | <b>0.0383</b>    | <b>0.5808</b>       | <b>0.6042</b>     |
| Std. Dev   | 0.1154                   | 0.1258           | 0.0114           | 0.0890              | 0.1582            |

**Table A - 8. Gas production – experimental Phase 2.**

| Phase 2         |                          |               |               |               |               |
|-----------------|--------------------------|---------------|---------------|---------------|---------------|
| Date            | Gas production STD L/day |               |               |               |               |
|                 | Meso                     | 2TPAD         | 1TPAD         | 1+2 TPAD      | Thermo        |
|                 | std L/d                  | std L/d       | std L/d       | std L/d       | std L/d       |
| 30/10/2002      | 1.0646                   | 1.4103        | 0.0126        | 0.9440        | 1.0887        |
| 31/10/2002      | 0.8838                   | 1.4134        | 0.0297        | 0.9830        | 0.8042        |
| 01/11/2002      | 1.0295                   | 1.5016        | 0.0055        | 0.9879        | 1.3074        |
| 02/11/2002      | 0.8934                   | 1.1994        | 0.0398        | 0.8660        | 1.1976        |
| 03/11/2002      | 0.9691                   | 1.2007        | 0.0349        | 0.8562        | 1.0550        |
| 04/11/2002      | 0.9449                   | 1.4356        | 0.0181        | 0.9724        | 0.8321        |
| 05/11/2002      | 0.6469                   | 1.7916        | 0.0342        | 1.2387        | 0.9886        |
| 06/11/2002      | 0.9717                   | 1.5720        | 0.0271        | 1.0806        | 0.9463        |
| 07/11/2002      | 1.0486                   | 1.5704        | 0.0408        | 1.1091        | 1.1431        |
| 08/11/2002      | 1.2365                   | 1.5358        | 0.0273        | 1.0574        | 1.2297        |
| 09/11/2002      | 1.1318                   | 1.5066        | 0.0199        | 1.0223        | 1.0245        |
| 10/11/2002      | 0.9648                   |               | 0.0368        |               | 1.0273        |
| 11/11/2002      | 1.0470                   |               | 0.0336        |               | 1.0742        |
| 12/11/2002      | 1.1164                   |               | 0.0375        |               | 1.1827        |
| 13/11/2002      | 1.1734                   |               | 0.0372        |               | 1.1734        |
| 14/11/2002      | 1.1115                   |               | 0.0377        |               | 1.0650        |
| 15/11/2002      | 1.0869                   |               | 0.0358        |               | 1.0959        |
| 16/11/2002      | 1.0218                   |               | 0.0270        |               | 0.9610        |
| 17/11/2002      | 1.0477                   |               | 0.0370        |               | 0.9554        |
| 18/11/2002      | 1.2022                   |               | 0.0462        |               | 1.1789        |
| 19/11/2002      | 0.9673                   |               | 0.0371        |               | 1.1787        |
| 20/11/2002      | 1.2539                   |               | 0.0350        |               | 1.1732        |
| 21/11/2002      | 1.0765                   |               | 0.0296        |               | 1.0065        |
| 22/11/2002      | 1.0371                   |               | 0.0427        |               | 1.0118        |
| 23/11/2002      | 0.9355                   |               | 0.0345        |               | 0.9492        |
| 24/11/2002      | 0.9928                   |               | 0.0338        |               | 0.9691        |
| 25/11/2002      | 1.0807                   |               | 0.0345        |               | 0.9902        |
| 26/11/2002      | 1.1276                   |               | 0.0303        |               | 1.0183        |
| 27/11/2002      | 0.9758                   |               | 0.0293        |               | 0.9377        |
| 28/11/2002      | 1.1195                   |               | 0.0293        |               | 1.0102        |
| 29/11/2002      | 1.1539                   | 1.5907        | 0.0495        | 1.1412        | 1.0652        |
| 30/11/2002      | 1.0368                   | 1.3003        | 0.0387        | 0.9291        | 1.0451        |
| 01/12/2002      | 1.0617                   | 1.3239        | 0.0384        | 0.9438        | 1.0955        |
| 02/12/2002      | 0.8129                   | 1.2969        | 0.0391        | 0.9276        | 1.1230        |
| 03/12/2002      | 1.2337                   | 1.3441        | 0.0357        | 0.9510        | 1.1118        |
| 04/12/2002      | 1.2556                   | 1.4109        | 0.0383        | 1.0001        | 1.1538        |
| 05/12/2002      | 1.0288                   | 1.5251        | 0.0254        | 1.0463        | 1.0463        |
| 06/12/2002      | 0.9875                   | 1.4462        | 0.0346        | 1.0149        | 1.0597        |
| 07/12/2002      | 0.8822                   | 1.3732        | 0.0366        | 0.9718        | 0.9187        |
| 08/12/2002      | 0.9484                   | 1.3835        | 0.0380        | 0.9817        | 0.9530        |
| 09/12/2002      | 0.8313                   | 1.5869        | 0.0408        | 1.1198        | 1.0286        |
| 10/12/2002      | 1.1054                   | 1.2746        | 0.0434        | 0.9226        |               |
| 11/12/2002      | 0.9240                   | 1.3884        | 0.0298        | 0.9671        |               |
| <b>Average</b>  | <b>1.0331</b>            | <b>1.4326</b> | <b>0.0336</b> | <b>1.0014</b> | <b>1.0531</b> |
| <i>Std. Dev</i> | <i>0.1254</i>            | <i>0.1389</i> | <i>0.0083</i> | <i>0.0891</i> | <i>0.1052</i> |

**Table A - 9. Gas production – experimental Phase 3.**

| Phase 3         |                          |               |               |               |               |
|-----------------|--------------------------|---------------|---------------|---------------|---------------|
| Date            | Gas production STD L/day |               |               |               |               |
|                 | Meso                     | 2TPAD         | 1TPAD         | 1+2 TPAD      | Thermo        |
|                 | std L/d                  | std L/d       | std L/d       | std L/d       | std L/d       |
| 31/12/2002      | 1.0794                   | 1.4300        | 0.0106        | 1.0885        | 1.0857        |
| 01/01/2003      | 1.0577                   | 1.5023        | 0.0125        | 1.1454        | 1.0729        |
| 02/01/2003      | 1.0410                   | 1.6065        | 0.0091        | 1.2185        | 1.1567        |
| 03/01/2003      | 1.0677                   | 1.5356        | 0.0123        | 1.1702        | 1.0318        |
| 04/01/2003      | 1.0789                   | 1.3742        | 0.0126        | 1.0495        | 1.0362        |
| 05/01/2003      | 1.0570                   | 1.5380        | 0.0159        | 1.1774        | 1.2275        |
| 06/01/2003      | 1.0788                   | 1.6332        | 0.0103        | 1.2403        | 1.0876        |
| 07/01/2003      | 1.1361                   | 1.6133        | 0.0109        | 1.2263        | 1.0026        |
| 08/01/2003      | 1.3618                   | 1.6771        | 0.0178        | 1.2845        | 1.4141        |
| 09/01/2003      | 1.5793                   | 1.8698        | 0.0200        | 1.4324        | 1.5793        |
| 10/01/2003      | 1.4858                   | 1.7961        | 0.0171        | 1.3727        | 1.4496        |
| 11/01/2003      | 1.6023                   | 2.0464        |               |               |               |
| 12/01/2003      | 1.8052                   | 2.1652        | 0.0121        | 1.6420        | 1.7626        |
| 13/01/2003      | 1.6202                   | 1.9536        | 0.0112        | 1.4820        | 1.5776        |
| 14/01/2003      | 1.6042                   | 1.8367        | 0.0193        | 1.4065        | 1.4121        |
| 15/01/2003      | 1.4562                   | 2.0600        | 0.0165        | 1.5698        | 1.4412        |
| 16/01/2003      | 1.3180                   | 1.8610        | 0.0165        | 1.4205        | 1.1685        |
| 17/01/2003      | 1.3322                   | 2.1162        | 0.0187        | 1.6152        | 1.2218        |
| 18/01/2003      | 1.3723                   | 2.0702        | 0.0152        | 1.5754        | 1.3974        |
| 19/01/2003      | 1.3049                   | 1.5673        | 0.0145        | 1.1972        | 1.6311        |
| 20/01/2003      | 1.2317                   | 1.7881        | 0.0137        | 1.3617        | 1.2650        |
| 21/01/2003      | 1.2933                   | 1.9335        | 0.0105        | 1.4658        | 1.1107        |
| 22/01/2003      | 1.0241                   | 2.3699        | 0.0117        | 1.7949        | 1.5811        |
| 23/01/2003      | 0.8255                   | 1.1484        | 0.0087        | 0.8744        | 0.9188        |
| 24/01/2003      | 1.1838                   | 1.7352        | 0.0108        | 1.3176        | 1.3845        |
| 25/01/2003      | 1.2391                   | 1.7767        | 0.0080        | 1.3444        | 1.3630        |
| 26/01/2003      | 1.3306                   | 2.0051        | 0.0184        | 1.5315        | 1.5212        |
| 27/01/2003      | 1.2282                   | 1.8311        | 0.0103        | 1.3887        | 1.3219        |
| 28/01/2003      | 1.2420                   | 1.7077        | 0.0139        | 1.3017        | 1.4154        |
| 29/01/2003      | 1.2248                   | 2.0423        | 0.0087        | 1.5447        | 1.0717        |
| 30/01/2003      | 1.2680                   | 2.1296        | 0.0172        | 1.6229        | 1.5072        |
| 31/01/2003      | 1.4142                   | 2.2358        | 0.0153        | 1.6999        | 1.1969        |
| 01/02/2003      | 1.4791                   | 1.2439        |               |               | 1.2377        |
| 02/02/2003      | 1.5064                   | 2.2913        | 0.0108        | 1.7347        | 1.5608        |
| 03/02/2003      | 1.3580                   | 2.1051        |               |               | 1.4236        |
| 04/02/2003      | 1.3707                   | 2.2436        | 0.0150        | 1.7052        | 1.6916        |
| 05/02/2003      | 1.3088                   | 2.1956        | 0.0000        | 1.6467        | 1.5506        |
| 06/02/2003      | 1.2964                   | 1.8388        | 0.0131        | 1.3988        | 1.3299        |
| <b>Average</b>  | <b>1.3023</b>            | <b>1.8498</b> | <b>0.0132</b> | <b>1.3831</b> | <b>1.3246</b> |
| <i>Std. Dev</i> | 0.2033                   | 0.2950        | 0.0042        | 0.2029        | 0.2059        |

**Table A - 10. Gas composition – Phase 1.**

| Phase 1          | Gas composition |                 |             |                 |                 |             |                 |                 |             |                 |                 |             |
|------------------|-----------------|-----------------|-------------|-----------------|-----------------|-------------|-----------------|-----------------|-------------|-----------------|-----------------|-------------|
|                  | Meso            |                 |             | 2TPAD           |                 |             | 1TPAD           |                 |             | Thermo          |                 |             |
|                  | CH <sub>4</sub> | CO <sub>2</sub> | Rest        | CH <sub>4</sub> | CO <sub>2</sub> | Rest        | CH <sub>4</sub> | CO <sub>2</sub> | Rest        | CH <sub>4</sub> | CO <sub>2</sub> | Rest        |
|                  | 66.1            | 29.9            | 4.0         | 63.2            | 33.8            | 3.0         | 41.4            | 55.8            | 2.8         | 62.2            | 34.6            | 3.2         |
|                  | 62.1            | 35.9            | 2.0         | 64.2            | 35.1            | 0.7         | 40.7            | 60.3            | -1.0        | 64.7            | 38.7            | -3.4        |
|                  | 64.3            | 33.2            | 2.5         | 65.4            | 32.5            | 2.1         | 41.7            | 55.9            | 2.4         | 63.8            | 34.7            | 1.5         |
|                  | 60.2            | 39.5            | 0.3         | 66.0            | 32.5            | 1.5         | 46.8            | 51.5            | 1.7         | 62.3            | 34.6            | 3.1         |
|                  | 65.8            | 33.8            | 0.4         | 68.5            | 33.8            | -2.3        | 41.3            | 57.2            | 1.5         | 67.4            | 34.6            | -2.0        |
|                  | 64.1            | 32.4            | 3.5         | 61.6            | 31.7            | 6.7         | 44.8            | 47.9            | 7.3         | 66.6            | 33.1            | 0.3         |
| <b>Average</b>   | <b>63.77</b>    | <b>34.12</b>    | <b>2.1</b>  | <b>64.82</b>    | <b>33.23</b>    | <b>2.0</b>  | <b>42.78</b>    | <b>54.77</b>    | <b>2.5</b>  | <b>64.50</b>    | <b>35.05</b>    | <b>0.4</b>  |
| <b>Std. Dev.</b> | <b>2.26</b>     | <b>3.28</b>     | <b>1.54</b> | <b>2.39</b>     | <b>1.23</b>     | <b>2.95</b> | <b>2.44</b>     | <b>4.40</b>     | <b>2.72</b> | <b>2.17</b>     | <b>1.89</b>     | <b>2.70</b> |

**Table A - 11. Gas composition – Phase 2.**

| Phase 2          | Gas composition |                 |              |                 |                 |             |                 |                 |             |                 |                 |              |
|------------------|-----------------|-----------------|--------------|-----------------|-----------------|-------------|-----------------|-----------------|-------------|-----------------|-----------------|--------------|
|                  | Meso            |                 |              | 2TPAD           |                 |             | 1TPAD           |                 |             | Thermo          |                 |              |
|                  | CH <sub>4</sub> | CO <sub>2</sub> | Rest         | CH <sub>4</sub> | CO <sub>2</sub> | Rest        | CH <sub>4</sub> | CO <sub>2</sub> | Rest        | CH <sub>4</sub> | CO <sub>2</sub> | Rest         |
|                  |                 |                 | 100.0        |                 |                 | 100.0       |                 |                 | 100.0       |                 |                 | 100.0        |
|                  | 66.7            | 35.4            | -2.1         | 66.2            | 33.3            | 0.5         | 43.4            | 55.3            | 1.3         | 62.8            | 36.2            | 1.0          |
|                  | 64.6            | 35.4            | 0.0          | 64.6            | 30.5            | 4.9         | 40.7            | 58.1            | 1.2         | 67.2            | 33.3            | -0.5         |
|                  | 63.3            | 36.1            | 0.6          | 61.0            | 38.8            | 0.2         | 34.2            | 62.9            | 2.9         | 64.5            | 34.2            | 1.3          |
|                  | 69.4            | 34.8            | -4.2         | 68.8            | 33.9            | -2.7        | 42.0            | 60.3            | -2.3        | 67.2            | 36.5            | -3.7         |
|                  | 63.4            | 35.9            | 0.7          | 65.7            | 32.2            | 2.1         | 39.7            | 59.4            | 0.9         | 62.1            | 33.7            | 4.2          |
|                  | 66.8            | 30.0            | 3.2          | 64.0            | 27.4            | 8.6         | 34.8            | 43.5            | 21.7        | 66.5            | 27.6            | 5.9          |
|                  | 62.9            | 36.9            | 0.2          | 63.2            | 34.9            | 1.9         | 40.9            | 60.8            | -1.7        | 67.7            | 38.8            | -6.5         |
|                  | 61.2            | 36.2            | 2.6          | 61.5            | 34.0            | 4.5         | 38.7            | 56.9            | 4.4         | 64.2            | 38.0            | -2.2         |
|                  | 65.2            | 37.8            | -3.0         | 65.5            | 34.6            | -0.1        | 42.4            | 61.0            | -3.4        | 66.8            | 36.4            | -3.2         |
| <b>Average</b>   | <b>64.83</b>    | <b>35.39</b>    | <b>-0.22</b> | <b>64.50</b>    | <b>33.29</b>    | <b>2.21</b> | <b>39.64</b>    | <b>57.58</b>    | <b>2.78</b> | <b>65.44</b>    | <b>34.97</b>    | <b>-0.41</b> |
| <b>Std. Dev.</b> | <b>2.493</b>    | <b>2.21</b>     | <b>2.46</b>  | <b>2.42</b>     | <b>3.15</b>     | <b>3.35</b> | <b>3.24</b>     | <b>5.76</b>     | <b>7.52</b> | <b>2.09</b>     | <b>3.34</b>     | <b>3.95</b>  |

**Table A - 12. Gas composition – Phase 3.**

| Phase 3          | Gas composition |                 |             |                 |                 |             |                 |                 |                |              |                 |                 |             |
|------------------|-----------------|-----------------|-------------|-----------------|-----------------|-------------|-----------------|-----------------|----------------|--------------|-----------------|-----------------|-------------|
|                  | Meso            |                 |             | 2TPAD           |                 |             | 1TPAD           |                 |                |              | Thermo          |                 |             |
|                  | CH <sub>4</sub> | CO <sub>2</sub> | Rest        | CH <sub>4</sub> | CO <sub>2</sub> | Rest        | CH <sub>4</sub> | CO <sub>2</sub> | H <sub>2</sub> | Rest         | CH <sub>4</sub> | CO <sub>2</sub> | Rest        |
|                  | 67.2            | 38.4            | -5.6        | 64.7            | 31.4            | 3.9         | 11              | 65.5            |                | 23.5         | 67.8            | 40.2            | -8.0        |
|                  | 63.9            | 33.5            | 2.6         | 66.9            | 31.8            | 1.3         | 0               | 73.6            |                | 26.4         | 61.6            | 37.7            | 0.7         |
|                  | 61.1            | 35.3            | 3.6         | 65.3            | 30.1            | 4.6         | 0               | 74.8            |                | 25.2         | 61.7            | 37              | 1.3         |
|                  | 61              | 35.6            | 3.4         | 65              | 34.4            | 0.6         | 0               | 69.7            |                | 30.3         | 61.9            | 38              | 0.1         |
|                  | 61.1            | 37.9            | 1.0         | 61.6            | 37.9            | 0.5         | 0               | 70.1            | 23.9           | 6.0          | 64.2            | 35              | 0.8         |
|                  | 62.2            | 36.8            | 1.0         | 61.7            | 33.4            | 4.9         | 0               | 73.5            | 16.1           | 10.4         | 63.4            | 35.1            | 1.5         |
|                  | 65.3            | 38.9            | -4.2        | 67.8            | 34              | -1.8        | 18              | 76.4            | 0              | 5.6          | 64.3            | 25.7            | 10.0        |
|                  | 61.4            | 37.4            | 1.2         | 61.9            | 33              | 5.1         | 5.5             | 67.5            | 18.7           | 8.3          | 67.4            | 38.8            | -6.2        |
|                  | 61.4            | 37.3            | 1.3         | 63.5            | 32.4            | 4.1         | 24.6            | 80.1            | 2              | -6.7         | 66              | 35.9            | -1.9        |
|                  | 62.2            | 35.3            | 2.5         | 62.2            | 32.3            | 5.5         | 12.9            | 71.8            | 14.5           | 0.8          | 62.7            | 33.5            | 3.8         |
|                  | 61              | 36.2            | 2.8         | 66.6            | 35.4            | -2.0        | 5.7             | 78.8            | 10             | 5.5          | 67.8            | 30.6            | 1.6         |
| <b>Average</b>   | <b>62.53</b>    | <b>36.60</b>    | <b>0.87</b> | <b>64.29</b>    | <b>33.28</b>    | <b>2.43</b> | <b>7.06</b>     | <b>72.89</b>    | <b>12.17</b>   | <b>7.87</b>  | <b>64.44</b>    | <b>35.23</b>    | <b>0.34</b> |
| <b>Std. Dev.</b> | <b>2.07</b>     | <b>1.60</b>     | <b>3.02</b> | <b>2.25</b>     | <b>2.13</b>     | <b>2.80</b> | <b>8.55</b>     | <b>4.54</b>     | <b>8.73</b>    | <b>12.08</b> | <b>2.45</b>     | <b>4.12</b>     | <b>4.76</b> |

**Table A - 13. COD results for Primary Sludge and 1<sup>st</sup> Acid Thermophilic Reactor – Phase 1.**

| Date       | Phase 1   |         |         |         |
|------------|-----------|---------|---------|---------|
|            | COD, mg/l |         |         |         |
|            | PS        |         | 1TPAD   |         |
|            | Total     | Soluble | Total   | Soluble |
| 28/08/2002 | 41221.3   | 1616.3  | 40103.6 | 3265.2  |
| 04/09/2002 | 31395.1   | 1632.6  | 26828.3 | 6029.2  |
| 09/09/2002 | 34339.0   | 3067.0  | 34339.0 | 7383.9  |
| 11/09/2002 | 36397.5   | 1354.9  | 32846.4 | 8629.6  |
| 16/09/2002 | 29520.5   | 3214.8  | 40773.8 | 8812.5  |
| 18/09/2002 | 39089.6   | 2919.9  | 26392.8 | 7210.7  |
| 23/09/2002 | 49438.0   | 1913.3  | 40773.8 | 7210.7  |
| 25/09/2002 | 38540.0   | 1913.3  | 40206.3 | 6697.8  |
| Average    | 38631.5   | 2252.0  | 33547.3 | 6621.6  |

**Table A - 14. COD results for Primary Sludge and 1<sup>st</sup> Acid Thermophilic Reactor – Phase 2.**

| Date       | Phase 2   |         |         |         |
|------------|-----------|---------|---------|---------|
|            | COD, mg/l |         |         |         |
|            | PS        |         | 1TPAD   |         |
|            | Total     | Soluble | Total   | Soluble |
| 30/10/2002 | 35875.2   | 3363.5  | 37457.8 | 8996.6  |
| 04/11/2002 | 35875.2   | 3363.5  | 47457.2 | 8448.0  |
| 06/11/2002 | 41347.4   | 2773.7  | 37457.8 | 8996.6  |
| 13/11/2002 | 44927.3   | 3663.4  | 36397.5 | 9937.3  |
| 18/11/2002 | 52201.0   | 3363.5  | 39645.0 | 8996.6  |
| 20/11/2002 | 38540.0   | 2773.7  | 35358.1 | 9368.9  |
| 25/11/2002 | 32846.4   | 4584.3  | 39645.0 | 9368.9  |
| 27/11/2002 | 41927.3   | 19544.8 | 34339.0 | 9746.5  |
| 02/12/2002 | 39645.0   | 14303.8 | 45548.4 | 10714.9 |
| 04/12/2002 | 34339.0   | 2054.7  | 52201.0 | 9937.3  |
| 09/12/2002 | 32846.4   | 3067.0  | 36397.5 | 9746.5  |
| 11/12/2002 | 48109.2   | 2054.7  | 42513.7 | 9557.0  |
| Average    | 39873.3   | 3106.2  | 37595.6 | 9484.6  |

**Table A - 15. COD results for Primary Sludge and 1<sup>st</sup> Acid Thermophilic Reactor – Phase 3.**

| Date       | Phase 3   |         |         |         |
|------------|-----------|---------|---------|---------|
|            | COD, mg/l |         |         |         |
|            | PS        |         | 1TPAD   |         |
|            | Total     | Soluble | Total   | Soluble |
| 30/12/2002 | 33339.2   | 4584.3  | 35875.2 | 12980.4 |
| 01/01/2003 | 35358.1   | 2054.7  | 39645.0 | 10913.0 |
| 06/01/2003 | 42513.7   | 1354.9  | 39089.6 | 10323.2 |
| 08/01/2003 | 45548.4   | 1632.6  | 48109.2 | 8629.6  |
| 13/01/2003 | 48769.4   | 3966.8  | 47457.2 | 10129.6 |
| 15/01/2003 | 32358.2   | 3067.0  | 52201.0 | 10323.2 |
| 20/01/2003 | 34846.1   | 3663.4  | 34846.1 | 8812.5  |
| 22/01/2003 | 36397.5   | 1080.1  | 34846.1 | 9557.0  |
| 27/01/2003 | 33339.2   | 2919.9  | 34339.0 | 8629.6  |
| 29/02/2003 | 35875.2   | 1493.4  | 38540.0 | 9557.0  |
| 02/02/2003 | 42513.7   | 3363.5  | 45548.4 | 8812.5  |
| 04/02/2003 | 37457.8   | 1632.6  | 45548.4 | 9368.9  |
| Average    | 38193.0   | 2726.5  | 37742.2 | 9836.4  |

**Table A - 16. VFA results for Primary Sludge and 1<sup>st</sup> Acid Thermophilic Reactor – Phase 1.**

| Date       | Phase 1   |                  |           |                  |
|------------|-----------|------------------|-----------|------------------|
|            | PS        |                  | 1TPAD     |                  |
|            | VFA, mg/l | VFA as COD, mg/l | VFA, mg/l | VFA as COD, mg/l |
| 26/08/2002 | 1041.2    | 1576.4           | 2589.2    | 4118.5           |
| 28/08/2002 | 504.8     | 854.6            | 2115.6    | 3404.2           |
| 04/09/2002 | 950.4     | 1382.7           | 2743.2    | 4094.3           |
| 09/09/2002 | 887.6     | 1326.5           | 3418      | 4859.8           |
| 11/09/2002 | 827.6     | 1193.0           | 3010      | 4238.7           |
| 16/09/2002 | 1316.4    | 1881.2           | 4280.4    | 6052.4           |
| 18/09/2002 | 1506.8    | 2222.8           | 3638      | 5168.4           |
| 23/09/2002 | 1340.4    | 1902.9           | 5241.2    | 7398.7           |
| 25/09/2002 | 490.8     | 765.1            | 4369.6    | 6082.6           |
| 26/09/2002 | 985.1     | 1456.1           |           |                  |
| 27/09/2002 | 337.0     | 747.3            |           |                  |
| Average    | 926.2     | 1391.7           | 3489.5    | 5046.4           |

**Table A - 17. VFA results for Primary Sludge and 1<sup>st</sup> Acid Thermophilic Reactor – Phase 2.**

| Date       | Phase 2   |                  |           |                  |
|------------|-----------|------------------|-----------|------------------|
|            | PS        |                  | 1TPAD     |                  |
|            | VFA, mg/l | VFA as COD, mg/l | VFA, mg/l | VFA as COD, mg/l |
| 30/10/2002 | 1626.8    | 2344.9           | 5164.0    | 7240.5           |
| 04/11/2002 | 2033.6    | 3005.1           | 4515.6    | 6439.7           |
| 06/11/2002 | 1432.0    | 2061.9           | 4808.4    | 6719.3           |
| 13/11/2002 | 1203.6    | 1744.1           | 4903.2    | 6983.6           |
| 18/11/2002 | 1759.6    | 2621.1           | 5077.2    | 7092.2           |
| 20/11/2002 | 1313.6    | 1920.4           | 5255.6    | 7333.2           |
| 25/11/2002 | 2197.6    | 3248.7           | 5298.0    | 7444.4           |
| 27/11/2002 | 1207.6    | 1716.0           | 5053.2    | 7160.4           |
| 02/12/2002 | 2125.2    | 3077.5           | 6824.8    | 9276.5           |
| 04/12/2002 | 830.8     | 1221.1           | 7200.8    | 9760.8           |
| 09/12/2002 | 2267.6    | 3316.8           | 4952.8    | 6889.0           |
| 11/12/2002 | 1177.6    | 1702.3           | 5637.2    | 7808.8           |
| Average    | 1598.0    | 2331.7           | 5390.9    | 7512.4           |

**Table A - 18. VFA results for Primary Sludge and 1<sup>st</sup> Acid Thermophilic Reactor – Phase 3.**

| Date       | Phase 3   |                  |           |                  |
|------------|-----------|------------------|-----------|------------------|
|            | PS        |                  | 1TPAD     |                  |
|            | VFA, mg/l | VFA as COD, mg/l | VFA, mg/l | VFA as COD, mg/l |
| 30/12/2002 | 1754.0    | 2551.2           | 2529.2    | 3661.9           |
| 01/01/2003 | 959.6     | 1395.8           | 3538.0    | 5128.3           |
| 06/01/2003 | 1714.4    | 2482.8           | 3385.2    | 5002.4           |
| 08/01/2003 | 846.0     | 1233.5           | 3198.4    | 4776.7           |
| 13/01/2003 | 2053.2    | 2965.4           | 4595.6    | 6823.1           |
| 15/01/2003 | 1390.8    | 1991.4           | 4730.4    | 7145.8           |
| 20/01/2003 | 2008.0    | 2966.0           | 4304.4    | 6374.4           |
| 22/01/2003 | 775.2     | 1093.0           | 3963.2    | 5797.9           |
| 27/01/2003 | 1856.4    | 2740.5           | 4730.8    | 7172.7           |
| 29/02/2003 | 1037.2    | 1520.1           | 6222.8    | 10024.0          |
| 02/02/2003 | 2094.8    | 3191.5           | 3709.2    | 5881.3           |
| 04/02/2003 | 1264.4    | 1885.1           | 5106.4    | 7814.6           |
| Average    | 1479.5    | 2168.0           | 4167.8    | 6300.3           |



**Table A - 19. Count of initial number of Anaerobic Hydrolytic Acidogenic Bacteria – AHAB.**

| Date       | AHAB count in Primary Sludge |                  |                  |          |
|------------|------------------------------|------------------|------------------|----------|
|            | Raw                          |                  |                  | Result   |
|            | Dilution                     |                  |                  |          |
|            | 10 <sup>-5</sup>             | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   |
| 17/08/2004 | +                            | +                | +                | 1.40E+07 |
|            | +                            | +                | +                |          |
|            | +                            | +                | +                |          |
| 18/08/2004 | 10 <sup>-5</sup>             | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   |
|            | +                            | +                | +                | 1.40E+07 |
|            | +                            | +                | +                |          |
| 19/08/2004 | 10 <sup>-5</sup>             | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   |
|            | +                            | +                | +                | 1.40E+07 |
|            | +                            | +                | +                |          |
| 24/08/2004 | 10 <sup>-6</sup>             | 10 <sup>-7</sup> | 10 <sup>-8</sup> | MPN/ml   |
|            | +                            | +                | -                | 4.50E+06 |
|            | +                            | -                | -                |          |
| 25/08/2004 | 10 <sup>-7</sup>             | 10 <sup>-8</sup> | 10 <sup>-9</sup> | MPN/ml   |
|            | +                            | +                | -                | 2.50E+08 |
|            | +                            | +                | -                |          |
| 26/08/2004 | 10 <sup>-7</sup>             | 10 <sup>-8</sup> | 10 <sup>-9</sup> | MPN/ml   |
|            | +                            | +                | -                | 2.50E+08 |
|            | +                            | +                | -                |          |
| 27/08/2004 | 10 <sup>-7</sup>             | 10 <sup>-8</sup> | 10 <sup>-9</sup> | MPN/ml   |
|            | +                            | +                | +                | 4.50E+08 |
|            | +                            | +                | -                |          |
| 28/08/2004 | 10 <sup>-7</sup>             | 10 <sup>-8</sup> | 10 <sup>-9</sup> | MPN/ml   |
|            | +                            | +                | -                | 9.50E+07 |
|            | +                            | -                | -                |          |
| 28/08/2004 | 10 <sup>-7</sup>             | 10 <sup>-8</sup> | 10 <sup>-9</sup> | MPN/ml   |
|            | +                            | -                | -                | 2.50E+07 |
|            | +                            | -                | -                |          |
| 29/08/2004 | 10 <sup>-7</sup>             | 10 <sup>-8</sup> | 10 <sup>-9</sup> | MPN/ml   |
|            | +                            | -                | -                | 2.50E+07 |
|            | +                            | -                | -                |          |
| 31/08/2004 | 10 <sup>-7</sup>             | 10 <sup>-8</sup> | 10 <sup>-9</sup> | MPN/ml   |
|            | +                            | -                | -                | 4.50E+07 |
|            | +                            | -                | -                |          |
| Average    |                              |                  |                  | 1.08E+08 |

**Table A - 20. Results of capillary suction time (CST) measurements – all sludges and all experimental phases.**

| Phase 1          |               |              |              |              |              |
|------------------|---------------|--------------|--------------|--------------|--------------|
| Date             | CST, sec      |              |              |              |              |
|                  | 1TPAD         | TP           | 2TPAD        | MD           | PS           |
| 27/08/2002       | 666.6         | 495.9        | 433          | 388.1        | 159.3        |
| 29/08/2002       | 539           | 460          | 414.5        | 381.7        |              |
| 05/09/2002       | 894.7         | 498.7        | 445.4        | 478.4        | 244.8        |
| 10/09/2002       | 927.2         | 496.1        | 437.9        | 386.3        | 377.5        |
| 12/09/2002       | 813.5         | 516          | 418.9        | 323.2        | 323.6        |
| 17/09/2002       | 875.4         | 470.8        | 401.5        | 341.8        | 358.1        |
| 19/09/2002       | 809           | 498.6        | 488.4        | 377.9        | 241.5        |
| <b>Average</b>   | <b>789.3</b>  | <b>490.9</b> | <b>434.2</b> | <b>382.5</b> | <b>284.1</b> |
| <i>Std. Dev.</i> | <i>139.2</i>  | <i>19.0</i>  | <i>28.2</i>  | <i>49.1</i>  | <i>83.3</i>  |
| Phase 2          |               |              |              |              |              |
| Date             | CST, sec      |              |              |              |              |
|                  | 1TPAD         | TP           | 2TPAD        | MD           | PS           |
| 01/11/2002       | -             | 589.4        | 476.2        | 412.3        | 397.8        |
| 05/11/2002       | 945.6         | 659.3        | 511.6        | 429.8        | 397.8        |
| 07/11/2002       | 1163.6        | 697.2        | 446.2        | 412.9        | 347.6        |
| 12/11/2002       | 1101.0        | 737.5        | 516.2        | 448.1        | 406.8        |
| 14/11/2002       | 901.9         | 750.8        | 534.5        | 504.8        | 295.7        |
| 19/11/2002       | 1192.8        | 694.8        | 507.9        | 408.6        | 300.2        |
| 22/11/2002       | 907.6         | 697.6        | 539.2        | 468.5        | 365.1        |
| 26/11/2002       | 1131.1        | 680.2        | 536.1        | 440.7        | 487          |
| 29/11/2002       | 1065.8        | 710.9        | 519.9        | 471.5        | 434.4        |
| 03/12/2002       | 1370.7        | 687.1        | 510.2        | 542.5        | 490.2        |
| 06/12/2002       | 1330.5        | 697.1        | 492.6        | 465.6        | 391.7        |
| 10/12/2002       | 1294.4        | 716.6        | 476.5        | 530.7        | 387.6        |
| <b>Average</b>   | <b>1127.7</b> | <b>693.2</b> | <b>505.6</b> | <b>461.3</b> | <b>391.8</b> |
| <i>Std. Dev.</i> | <i>164.5</i>  | <i>40.8</i>  | <i>28.1</i>  | <i>45.3</i>  | <i>61.2</i>  |
| Phase 3          |               |              |              |              |              |
| Date             | CST, sec      |              |              |              |              |
|                  | 1TPAD         | TP           | 2TPAD        | MD           | PS           |
| 31/12/2002       | 1474.6        | 870          | 564.5        | 808.1        | 464.8        |
| 02/01/2003       | 1449.3        | 898.3        | 619.7        | 848.3        | 404.9        |
| 07/01/2003       | 886.9         | 901.7        | 639.4        | 796.4        | 378          |
| 09/01/2003       | 1320.2        | 915.8        | 748.4        | 812          | 360          |
| 14/01/2003       | 1269.1        | 1015.9       | 703          | 898.7        | 390.3        |
| 16/01/2003       | 1165.6        | 917          | 625.7        | 838.1        | 464.7        |
| 21/01/2003       | 1031.7        | 896.4        | 665.8        | 908.3        | 520.8        |
| 23/01/2003       | 1310.2        | 907.3        | 691.8        | 1063.4       | 517.2        |
| 28/01/2003       | 941           | 875.6        | 649.1        | 892.8        | 411.3        |
| 30/01/2003       | 1392          | 931.1        | 712.2        | 951.1        | 559          |
| 03/02/2003       | 1225.5        | 875.5        | 596.3        | 821.1        | 479.4        |
| <b>Average</b>   | <b>1224.2</b> | <b>909.5</b> | <b>656.0</b> | <b>876.2</b> | <b>450.0</b> |
| <i>Std. Dev.</i> | <i>198.65</i> | <i>40.14</i> | <i>54.57</i> | <i>79.28</i> | <i>65.64</i> |

## **Appendix B - Data for section 5.0.**

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**Table B - 1. Total, soluble and particulate COD results from all experimental runs.**

| Results COD for all phases |                 |       |       |       |       |       |       |       |       |       |       |       |
|----------------------------|-----------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| SRT                        | Total COD       |       |       |       |       |       |       |       |       |       |       |       |
|                            | NT              | 6T    | 8T    | NM    | 6M    | 8M    | NP    | 6P    | 8P    | NR    | 6R    | 8R    |
| 5                          | 37755           | 68333 | 87937 | 36991 | 69801 | 90220 | 39770 | 71231 | 91183 | 43865 | 79551 | 98467 |
|                            | Soluble COD     |       |       |       |       |       |       |       |       |       |       |       |
|                            | 11045           | 18161 | 23623 | 9686  | 17609 | 24052 | 11028 | 15385 | 19508 | 4494  | 6974  | 9691  |
|                            | Particulate COD |       |       |       |       |       |       |       |       |       |       |       |
|                            | 26710           | 50172 | 64313 | 27305 | 52192 | 66167 | 28741 | 55846 | 71675 | 39371 | 72578 | 88776 |
| SRT                        | Total COD       |       |       |       |       |       |       |       |       |       |       |       |
|                            | NT              | 6T    | 8T    | NM    | 6M    | 8M    | NP    | 6P    | 8P    | NR    | 6R    | 8R    |
| 3.2                        | 32669           | 59499 | 74685 | 32490 | 59963 | 73641 | 33798 | 63093 | 78365 | 38932 | 70561 | 82650 |
|                            | Soluble COD     |       |       |       |       |       |       |       |       |       |       |       |
|                            | 7971            | 13264 | 17437 | 6734  | 11835 | 15815 | 6243  | 11062 | 14433 | 3069  | 5470  | 7071  |
|                            | Particulate COD |       |       |       |       |       |       |       |       |       |       |       |
|                            | 24698           | 46235 | 57247 | 25756 | 48128 | 57827 | 27555 | 52031 | 63932 | 35863 | 65091 | 75579 |
| SRT                        | Total COD       |       |       |       |       |       |       |       |       |       |       |       |
|                            | NT              | 6T    | 8T    | NM    | 6M    | 8M    | NP    | 6P    | 8P    | NR    | 6R    | 8R    |
| 2.2                        | 40227           | 55135 | 73943 | 39792 | 55680 | 73654 | 40744 | 56708 | 75400 | 46454 | 61001 | 78132 |
|                            | Soluble COD     |       |       |       |       |       |       |       |       |       |       |       |
|                            | 10119           | 13740 | 17704 | 9349  | 13509 | 17265 | 8502  | 11680 | 15295 | 5307  | 6621  | 9023  |
|                            | Particulate COD |       |       |       |       |       |       |       |       |       |       |       |
|                            | 30108           | 41394 | 56239 | 30443 | 42170 | 56389 | 32242 | 45029 | 60105 | 41147 | 54380 | 69109 |

N – normal sludge solids content, approx. 3.6% TS (see Figure 5-2)

6 – denotes reactors with target solids content in feed sludge of 6% TS (actual sludge solids content 5.4% TS as in Figure 5-2)

8 – denotes reactors with target solids content in feed sludge of 8% TS (actual sludge solids content 7.8% TS as in Figure 5-2)

R – raw sludge

T – acid digesters operated at thermophilic temperature

M – acid digesters operated at mesophilic temperature

L-M – acid digesters operated at low-mesophilic temperature

**Table B - 2. Average particulate COD established for all experimental phases.**

| % Solids in feed sludge | 3.6% TS         | 5.4% TS      | 7.8% TS      |
|-------------------------|-----------------|--------------|--------------|
| Operating SRT           | Units: mg/l COD |              |              |
| 5                       | 39371           | 72578        | 88776        |
| 3.2                     | 35863           | 65091        | 75579        |
| 2.2                     | 41147           | 54380        | 69109        |
| Average PCOD            | <b>38794</b>    | <b>64016</b> | <b>77821</b> |

**Table B - 3. Fraction of particulate COD remaining in every acid digester at each operating conditions.**

| SRT | Fraction remaining particulate COD in mg/l |      |      |      |      |      |      |      |      |
|-----|--|------|------|------|------|------|------|------|------|
|     | NT   | 6T   | 8T   | NM   | 6M   | 8M   | NL-M | 6L-M | 8L-M |
| 5   | 0.68                                       | 0.69 | 0.72 | 0.69 | 0.72 | 0.75 | 0.73 | 0.77 | 0.81 |
| 3.2 | 0.69                                       | 0.71 | 0.76 | 0.72 | 0.74 | 0.77 | 0.77 | 0.80 | 0.85 |
| 2.2 | 0.73                                       | 0.76 | 0.81 | 0.74 | 0.78 | 0.82 | 0.78 | 0.83 | 0.87 |

All symbols as in Table B-1.

**Table B - 4. Average initial particulate COD and fraction of particulate COD remaining in each acid digester at each operating conditions.**

| Treatment type  | NT               | 6T    | 8T    | NM    | 6M    | 8M    | NL-M  | 6L-M  | 8L-M  |
|-----------------|------------------|-------|-------|-------|-------|-------|-------|-------|-------|
| SRT             | <i>mg PCOD/l</i> |       |       |       |       |       |       |       |       |
| days            |                  |       |       |       |       |       |       |       |       |
| Average initial | 38794            | 64016 | 77821 | 38794 | 64016 | 77821 | 38794 | 64016 | 77821 |
| 5               | 26112            | 43851 | 55930 | 26861 | 45791 | 57549 | 28354 | 49145 | 62786 |
| 3.2             | 27327            | 46536 | 60147 | 27992 | 47992 | 60841 | 29709 | 51496 | 65896 |
| 2.2             | 28002            | 48028 | 62489 | 28621 | 49215 | 62670 | 30462 | 52802 | 67624 |

All symbols as in Table B-1.

**Table B - 5. Average Total and Volatile Solids in semi-continuous acid digesters.**

| SRT 5 days   |            |       |       |       |       |       |       |       |       |       |       |       |
|--------------|------------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
| Sludge Type  | NR         | 6R    | 8R    | NT    | 6T    | 8T    | NM    | 6M    | 8M    | NL-M  | 6L-M  | 8L-M  |
| Units        | <i>g/l</i> |       |       |       |       |       |       |       |       |       |       |       |
| TS average   | 29.24      | 55.12 | 76.13 | 24.19 | 45.96 | 59.76 | 22.22 | 45.84 | 61.46 | 27.61 | 43.68 | 60.04 |
| TS std dev.  | 3.43       | 4.23  | 7.14  | 5.72  | 7.60  | 3.71  | 3.45  | 8.42  | 7.59  | 2.16  | 2.90  | 2.13  |
| VS average   | 22.77      | 42.13 | 60.52 | 17.36 | 34.41 | 44.24 | 16.39 | 33.91 | 45.08 | 20.66 | 33.16 | 44.97 |
| VS std dev.  | 2.46       | 3.33  | 5.12  | 3.22  | 6.05  | 3.10  | 2.42  | 6.58  | 6.30  | 1.52  | 2.88  | 2.34  |
| SRT 3.2 days |            |       |       |       |       |       |       |       |       |       |       |       |
| Sludge Type  | NR         | 6R    | 8R    | NT    | 6T    | 8T    | NM    | 6M    | 8M    | NL-M  | 6L-M  | 8L-M  |
| Units        | <i>g/l</i> |       |       |       |       |       |       |       |       |       |       |       |
| TS average   | 35.99      | 55.27 | 80.95 | 31.33 | 55.60 | 75.16 | 30.48 | 53.97 | 68.19 | 34.02 | 55.14 | 71.65 |
| TS std dev.  | 12.07      | 2.70  | 4.66  | 9.70  | 9.92  | 8.26  | 8.88  | 5.09  | 8.35  | 10.20 | 4.42  | 6.00  |
| VS average   | 22.37      | 33.31 | 49.04 | 18.66 | 32.17 | 42.76 | 17.75 | 30.81 | 38.88 | 20.69 | 33.22 | 42.67 |
| VS std dev.  | 7.29       | 3.76  | 4.83  | 5.53  | 4.70  | 3.72  | 5.28  | 2.82  | 4.94  | 5.98  | 2.05  | 2.65  |
| SRT 2.2 days |            |       |       |       |       |       |       |       |       |       |       |       |
| Sludge Type  | NR         | 6R    | 8R    | NT    | 6T    | 8T    | NM    | 6M    | 8M    | NL-M  | 6L-M  | 8L-M  |
| Units        | <i>g/l</i> |       |       |       |       |       |       |       |       |       |       |       |
| TS average   | 42.52      | 52.03 | 75.75 | 32.94 | 49.97 | 68.14 | 30.03 | 49.62 | 68.95 | 32.79 | 51.59 | 67.30 |
| TS std dev.  | 8.55       | 4.62  | 6.94  | 11.77 | 13.51 | 18.14 | 10.77 | 12.47 | 18.18 | 10.52 | 13.05 | 16.80 |
| VS average   | 25.00      | 30.58 | 43.06 | 18.38 | 27.01 | 36.12 | 16.88 | 27.21 | 37.26 | 18.77 | 28.73 | 36.73 |
| VS std dev.  | 4.41       | 2.09  | 2.90  | 5.73  | 7.94  | 9.84  | 5.82  | 7.24  | 9.97  | 5.92  | 7.43  | 9.50  |

**Table B - 6. Average influent Total Solids in semi-continuous acid digesters from all runs.**

| Sludge    | NR           | 6R           | 8R           |
|-----------|--------------|--------------|--------------|
| Units     | <i>g/l</i>   |              |              |
| SRT, days |              |              |              |
| 5         | 29.24        | 55.12        | 76.13        |
| 3.2       | 35.99        | 55.27        | 80.95        |
| 2.2       | 42.52        | 52.03        | 75.75        |
| Average   | <b>35.92</b> | <b>54.14</b> | <b>77.61</b> |
| Std. Dev. | 6.64         | 1.83         | 2.90         |

**Table B - 7. Total COD results – SRT 5 days.**

| COD Concentration, mg/l |              |              |              |              |              |              |              |              |              |              |              |              |
|-------------------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| Treatment               | NT           | 6T           | 8T           | NM           | 6M           | 8M           | NL-M         | 6L-M         | 8L-M         | NR           | 6R           | 8R           |
| Day/Week                |              |              |              |              |              |              |              |              |              |              |              |              |
| M1                      | 288.5        | 460.1        | 592.7        | 311.9        | 577.1        | 592.7        | 350.9        | 530.3        | 631.7        | 296.3        | 608.3        | 702          |
| W1                      | 288.5        | 514.7        | 600.5        | 319.7        | 584.9        | 647.3        | 350.9        | 483.5        | 631.7        | 319.7        | 553.7        | 709.8        |
| F1                      | 241.7        | 475.7        | 592.7        | 257.3        | 499.1        | 623.9        | 256.6        | 499.1        | 608.3        | 257.3        | 538.1        | 670.7        |
| M2                      | 257.3        | 491.3        | 561.5        | 210.5        | 545.9        | 545.9        | 212.4        | 483.5        | 631.7        | 311.9        | 545.9        | 686.3        |
| W2                      | 327.5        | 460.1        | 553.7        | 265.1        | 374.3        | 545.9        | 288.5        | 452.3        | 662.9        | 311.9        | 514.7        | 647.3        |
| F2                      | 233.9        | 444.5        | 561.5        | 202.7        | 475.7        | 545.9        | 292.3        | 499.1        | 623.9        | 374.3        | 538.1        | 647.3        |
| M3                      | 233.9        | 436.7        | 639.5        | 272.9        | 506.9        | 694.1        | 288.5        | 483.5        | 623.9        | 311.9        | 553.7        | 678.5        |
| W3                      | 249.5        | 569.3        | 561.5        | 265.1        | 514.7        | 577.1        | 343.1        | 530.3        | 608.3        | 272.9        | 530.3        | 702          |
| F3                      | 218.3        | 491.3        | 577.1        | 233.9        | 514.7        | 639.5        | 245.1        | 467.9        | 608.3        | 257.3        | 553.7        | 616.1        |
| M4                      | 265.1        | 475.7        | 569.3        | 265.1        | 491.3        | 592.7        | 275.1        | 530.3        | 655.1        | 288.5        | 499.1        | 639.5        |
| W4                      | 233.9        | 436.7        | 592.7        | 265.1        | 475.7        | 608.3        | 319.7        | 436.7        | 561.5        | 311.9        | 608.3        | 647.3        |
| F4                      | 218.3        | 452.3        | 584.9        | 257.3        | 436.7        | 608.3        | 259.5        | 499.1        | 545.9        | 241.7        | 475.7        | 623.9        |
| M5                      | 413.3        | 249.5        | 577.1        | 233.9        | 405.5        | 545.9        | 267.8        | 421.1        | 584.9        | 257.3        | 522.5        | 639.5        |
| W5                      | 194.9        | 491.3        | 561.5        | 241.7        | 467.9        | 639.5        | 265.1        | 467.9        | 584.9        | 249.5        | 522.5        | 655.1        |
| F5                      | 202.7        | 514.7        | 584.9        | 233.9        | 545.9        | 623.9        | 265.1        | 428.9        | 592.7        | 257.3        | 483.5        | 694.1        |
| M6                      | 226.1        | 436.7        | 592.7        | 233.9        | 522.5        | 647.3        | 265.1        | 460.1        | 600.5        | 280.7        | 499.1        | 647.3        |
| W6                      | 218.3        | 467.9        | 655.1        | 280.7        | 389.9        | 631.7        | 319.7        | 428.9        | 616.1        | 319.7        | 483.5        | 538.1        |
| F6                      | 218.3        | 444.5        | 592.7        | 226.1        | 358.7        | 608.3        | 335.3        | 444.5        | 623.9        | 343.1        | 514.7        | 670.7        |
| Average                 | 251.7        | 455.6        | 586.2        | 246.6        | 465.3        | 601.5        | 265.1        | 474.9        | 607.9        | 292.4        | 530.3        | 656.4        |
| Std. Dev.               | 52.3         | 59.0         | 26.5         | 22.8         | 56.3         | 37.3         | 21.8         | 35.6         | 27.5         | 36.1         | 37.6         | 40.4         |
| Factor                  | 150          |              |              |              |              |              |              |              |              |              |              |              |
| <b>TCOD</b>             | <b>37755</b> | <b>68333</b> | <b>87937</b> | <b>36991</b> | <b>69801</b> | <b>90220</b> | <b>39765</b> | <b>71231</b> | <b>91183</b> | <b>43865</b> | <b>79551</b> | <b>98467</b> |
| Std.Dev.                | 7852         | 8844         | 3978         | 3415         | 8443         | 5594         | 3269         | 5338         | 4126         | 5418         | 5647         | 6063         |

**Table B - 8. Soluble COD results – SRT 5 days.**

| COD Concentration, mg/l |              |              |              |             |              |              |              |              |              |             |             |             |
|-------------------------|--------------|--------------|--------------|-------------|--------------|--------------|--------------|--------------|--------------|-------------|-------------|-------------|
| Treatment               | NT           | 6T           | 8T           | NM          | 6M           | 8M           | NL-M         | 6L-M         | 8L-M         | NR          | 6R          | 8R          |
| Day/Week                |              |              |              |             |              |              |              |              |              |             |             |             |
| M1                      | 264.6        | 388.8        | 482          | 264.6       | 404.3        | 544.1        | 342.2        | 342.2        | 419.8        | 85.99       | 171.4       | 171.4       |
| W1                      | 264.6        | 373.3        | 497.5        | 264.6       | 381          | 497.5        | 233.5        | 326.7        | 419.8        | 78.23       | 155.9       | 202.5       |
| F1                      | 233.5        | 388.8        | 497.5        | 256.8       | 381          | 528.5        | 249          | 326.7        | 450.9        | 78.23       | 194.7       | 218         |
| M2                      | 233.5        | 388.8        | 505.2        | 202.5       | 373.3        | 520.8        | 241.3        | 342.2        | 419.8        | 109.3       | 155.9       | 256.8       |
| W2                      | 233.5        | 388.8        | 497.5        | 218         | 373.3        | 513          | 225.7        | 326.7        | 435.4        | 109.3       | 171.4       | 241.3       |
| F2                      | 233.5        | 357.7        | 466.4        | 202.5       | 396.6        | 528.5        | 225.7        | 334.4        | 427.6        | 132.6       | 186.9       | 264.6       |
| M3                      | 218          | 388.8        | 513          | 194.7       | 388.8        | 528.5        | 218          | 350          | 443.1        | 101.5       | 140.3       | 218         |
| W3                      | 233.5        | 388.8        | 528.5        | 186.9       | 404.3        | 528.5        | 249          | 334.4        | 435.4        | 124.8       | 171.4       | 225.7       |
| F3                      | 233.5        | 419.8        | 536.3        | 202.5       | 404.3        | 559.6        | 233.5        | 381          | 458.7        | 124.8       | 202.5       | 249         |
| M4                      | 233.5        | 404.3        | 520.8        | 210.2       | 404.3        | 536.3        | 218          | 342.2        | 435.4        | 62.7        | 109.3       | 155.9       |
| W4                      | 249          | 365.5        | 419.8        | 186.9       | 326.7        | 435.4        | 186.9        | 280.1        | 357.7        | 47.17       | 109.3       | 171.4       |
| F4                      | 218          | 326.7        | 450.9        | 194.7       | 318.9        | 435.4        | 171.4        | 280.1        | 342.2        | 47.17       | 109.3       | 155.9       |
| M5                      | 194.7        | 318.9        | 482          | 186.9       | 311.1        | 419.8        | 186.9        | 264.6        | 342.2        | 140.3       | 62.7        | 140.3       |
| W5                      | 186.9        | 342.2        | 419.8        | 171.4       | 303.4        | 419.8        | 163.6        | 256.8        | 311.1        | 78.23       | 109.3       | 155.9       |
| F5                      | 171.4        | 334.4        | 466.4        | 171.4       | 326.7        | 435.4        | 171.4        | 249          | 342.2        | 78.23       | 109.3       | 171.4       |
| M6                      | 194.7        | 311.1        | 404.3        | 155.9       | 287.9        | 419.8        | 326.7        | 249          | 342.2        | 47.17       | 93.76       | 179.2       |
| W6                      | 171.4        | 334.4        | 396.6        | 171.4       | 303.4        | 419.8        | 186.9        | 264.6        | 342.2        | 109.3       | 117         | 140.3       |
| F6                      | 194.7        | 342.2        | 419.8        | 186.9       | 326.7        | 435.4        | 171.4        | 287.9        | 342.2        | 62.7        | 140.3       | 171.4       |
| Average                 | 220.9        | 363.2        | 472.5        | 193.7       | 352.2        | 481.0        | 220.6        | 307.7        | 390.2        | 89.9        | 139.5       | 193.8       |
| Std. Dev.               | 28.5         | 32.1         | 44.4         | 31.7        | 42.0         | 53.3         | 49.9         | 40.8         | 49.9         | 30.3        | 39.0        | 41.2        |
| Factor                  | 50           |              |              |             |              |              |              |              |              |             |             |             |
| <b>TCOD</b>             | <b>11045</b> | <b>18161</b> | <b>23623</b> | <b>9686</b> | <b>17609</b> | <b>24052</b> | <b>11028</b> | <b>15385</b> | <b>19508</b> | <b>4494</b> | <b>6974</b> | <b>9691</b> |
| Std.Dev.                | 1427         | 1603         | 2222         | 1586        | 2102         | 2665         | 2495         | 2042         | 2495         | 1514        | 1952        | 2058        |

**Table B - 9. Total COD results – SRT 3.2 days.**

| COD Concentration, mg/l |              |              |              |              |              |              |              |              |              |              |              |              |
|-------------------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| Treatment               | NT           | 6T           | 8T           | NM           | 6M           | 8M           | NL-M         | 6L-M         | 8L-M         | NR           | 6R           | 8R           |
| Day/Week                |              |              |              |              |              |              |              |              |              |              |              |              |
| M1                      | 183.4        | 430.7        | 492.5        | 206.6        | 484.8        | 523.4        | 245.2        | 600.7        | 554.3        | 198.8        | 538.9        | 623.9        |
| W1                      | 183.4        | 484.8        | 446.1        | 183.4        | 507.9        | 430.7        | 183.4        | 453.8        | 569.8        | 191.1        | 477          | 647          |
| F1                      | 183.4        | 469.3        | 461.6        | 214.3        | 477          | 662.5        | 222          | 507.9        | 608.4        | 245.2        | 515.7        | 670.2        |
| M2                      | 183.4        | 446.1        | 600.7        | 183.4        | 399.8        | 569.8        | 191.1        | 492.5        | 554.3        | 191.1        | 392          | 438.4        |
| W2                      | 229.7        | 415.2        | 593          | 198.8        | 507.9        | 600.7        | 252.9        | 422.9        | 531.1        | 229.7        | 446.1        | 562          |
| F2                      | 407.5        | 299.3        | 531.1        | 214.3        | 392          | 515.7        | 245.2        | 507.9        | 461.6        | 222          | 453.8        | 492.5        |
| M3                      | 206.6        | 345.7        | 500.2        | 222          | 415.2        | 461.6        | 237.5        | 353.4        | 492.5        | 291.6        | 461.6        | 554.3        |
| W3                      | 229.7        | 392          | 531.1        | 252.9        | 422.9        | 515.7        | 276.1        | 430.7        | 538.9        | 268.4        | 407.5        | 562          |
| F3                      | 222          | 523.4        | 453.8        | 237.5        | 422.9        | 461.6        | 314.7        | 422.9        | 554.3        | 252.9        | 662.5        | 569.8        |
| M4                      | 307          | 492.5        | 492.5        | 299.3        | 392          | 523.4        | 353.4        | 453.8        | 538.9        | 415.2        | 368.8        | 507.9        |
| W4                      | 399.8        | 415.2        | 554.3        | 345.7        | 384.3        | 515.7        | 368.8        | 407.5        | 523.4        | 330.2        | 384.3        | 515.7        |
| F4                      | 453.8        | 299.3        | 546.6        | 384.3        | 376.6        | 453.8        | 376.6        | 407.5        | 546.6        | 283.8        | 438.4        | 593          |
| M5                      | 237.5        | 477          | 523.4        | 283.8        | 399.8        | 569.8        | 314.7        | 430.7        | 546.6        | 252.9        | 639.3        | 469.3        |
| W5                      | 229.7        | 453.8        | 585.2        | 252.9        | 392          | 507.9        | 307          | 422.9        | 562          | 260.6        | 399.8        | 507.9        |
| Average                 | 217.8        | 396.7        | 497.9        | 216.6        | 399.8        | 490.9        | 225.3        | 420.6        | 522.4        | 259.5        | 470.4        | 551.0        |
| Std. Dev.               | 23.3         | 64.4         | 35.3         | 25.3         | 15.9         | 34.9         | 27.8         | 28.5         | 30.2         | 59.8         | 90.7         | 67.3         |
| Factor                  | 150          |              |              |              |              |              |              |              |              |              |              |              |
| <b>TCOD</b>             | <b>32669</b> | <b>59499</b> | <b>74685</b> | <b>32490</b> | <b>59963</b> | <b>73641</b> | <b>33798</b> | <b>63093</b> | <b>78365</b> | <b>38932</b> | <b>70561</b> | <b>82650</b> |
| Std.Dev.                | 3501         | 9661         | 5299         | 3788         | 2382         | 5236         | 4172         | 4270         | 4529         | 8977         | 13609        | 10092        |

**Table B - 10. Soluble COD results – SRT 3.2 days.**

| COD Concentration, mg/l |             |              |              |             |              |              |             |              |              |             |             |             |
|-------------------------|-------------|--------------|--------------|-------------|--------------|--------------|-------------|--------------|--------------|-------------|-------------|-------------|
| Treatment               | NT          | 6T           | 8T           | NM          | 6M           | 8M           | NL-M        | 6L-M         | 8L-M         | NR          | 6R          | 8R          |
| Day/Week                |             |              |              |             |              |              |             |              |              |             |             |             |
| M1                      | 144.7       | 307.0        | 384.3        | 152.5       | 252.9        | 361.1        | 137.0       | 214.3        | 322.5        | 75.2        | 129.3       | 175.6       |
| W1                      | 152.5       | 307.0        | 353.4        | 137.0       | 260.6        | 322.5        | 121.5       | 268.4        | 307.0        | 90.6        | 129.3       | 160.2       |
| F1                      | 160.2       | 345.7        | 415.2        | 167.9       | 276.1        | 376.6        | 167.9       | 283.8        | 330.2        | 113.8       | 167.9       | 191.1       |
| M2                      | 121.5       | 276.1        | 353.4        | 113.8       | 260.6        | 345.7        | 113.8       | 237.5        | 245.2        | 5.6         | 59.7        | 82.9        |
| W2                      | 113.8       | 268.4        | 330.2        | 82.9        | 291.6        | 322.5        | 98.4        | 222.0        | 283.8        | 28.8        | 90.6        | 98.4        |
| F2                      | 106.1       | 206.6        | 291.6        | 82.9        | 214.3        | 314.7        | 113.8       | 206.6        | 245.2        | 28.8        | 90.6        | 98.4        |
| M3                      | 137.0       | 229.7        | 337.9        | 129.3       | 214.3        | 314.7        | 121.5       | 183.4        | 268.4        | 59.7        | 137.0       | 137.0       |
| W3                      | 191.1       | 260.6        | 353.4        | 214.3       | 252.9        | 314.7        | 137.0       | 214.3        | 314.7        | 113.8       | 106.1       | 229.7       |
| F3                      | 198.8       | 276.1        | 368.8        | 137.0       | 237.5        | 314.7        | 167.9       | 245.2        | 291.6        | 90.6        | 144.7       | 152.5       |
| M4                      | 237.5       | 268.4        | 353.4        | 160.2       | 283.8        | 299.3        | 198.8       | 198.8        | 291.6        | 67.4        | 106.1       | 152.5       |
| W4                      | 222.0       | 252.9        | 361.1        | 183.4       | 237.5        | 307.0        | 206.6       | 229.7        | 291.6        | 75.2        | 98.4        | 137.0       |
| F4                      | 222.0       | 252.9        | 384.3        | 206.6       | 229.7        | 322.5        | 214.3       | 229.7        | 330.2        | 13.4        | 113.8       | 175.6       |
| M5                      | 191.1       | 252.9        | 376.6        | 152.5       | 214.3        | 314.7        | 183.4       | 198.8        | 283.8        | 36.5        | 75.2        | 75.2        |
| W5                      | 183.4       | 252.9        | 392.0        | 129.3       | 222.0        | 291.6        | 160.2       | 222.0        | 322.5        | 59.7        | 82.9        | 113.8       |
| Average                 | 159.4       | 265.3        | 348.7        | 134.7       | 236.7        | 316.3        | 124.9       | 221.2        | 288.7        | 61.4        | 109.4       | 141.4       |
| Std. Dev.               | 42.6        | 34.4         | 30.2         | 39.4        | 26.2         | 23.2         | 37.9        | 27.2         | 28.4         | 34.8        | 29.7        | 44.3        |
| Factor                  | 50          |              |              |             |              |              |             |              |              |             |             |             |
| <b>TCOD</b>             | <b>7971</b> | <b>13264</b> | <b>17437</b> | <b>6734</b> | <b>11835</b> | <b>15815</b> | <b>6243</b> | <b>11062</b> | <b>14433</b> | <b>3069</b> | <b>5470</b> | <b>7071</b> |
| Std.Dev.                | 2130        | 1721         | 1508         | 1971        | 1311         | 1159         | 1896        | 1362         | 1419         | 1742        | 1483        | 2216        |



**Table B - 11. Total COD results – SRT 2.2 days.**

| COD Concentration, mg/l |              |              |              |              |              |              |              |              |              |              |              |              |
|-------------------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|--------------|
| Treatment               | NT           | 6T           | 8T           | NM           | 6M           | 8M           | NL-M         | 6L-M         | 8L-M         | NR           | 6R           | 8R           |
| Day/Week                |              |              |              |              |              |              |              |              |              |              |              |              |
| M1                      | 251.2        | 343.7        | 520.9        | 228.1        | 359.1        | 574.8        | 220.4        | 374.5        | 482.4        | 205          | 436.1        | 551.7        |
| W1                      | 181.9        | 328.3        | 513.2        | 205          | 343.7        | 613.3        | 228.1        | 366.8        | 520.9        | 235.8        | 359.1        | 528.6        |
| F1                      | 228.1        | 366.8        | 482.4        | 220.4        | 328.3        | 513.2        | 228.1        | 366.8        | 497.8        | 274.3        | 405.3        | 520.9        |
| M2                      | 220.4        | 366.8        | 513.2        | 312.9        | 312.9        | 466.9        | 389.9        | 366.8        | 536.3        | 305.2        | 436.1        | 574.8        |
| W2                      | 243.5        | 374.5        | 544          | 251.2        | 359.1        | 544          | 251.2        | 389.9        | 628.7        | 282          | 436.1        | 590.2        |
| F2                      | 243.5        | 474.7        | 513.2        | 258.9        | 389.9        | 597.9        | 266.6        | 420.7        | 605.6        | 251.2        | 474.7        | 551.7        |
| M3                      | 274.3        | 482.4        | 574.8        | 266.6        | 374.5        | 651.3        | 282          | 405.3        | 590.2        | 374.5        | 359.1        | 482.4        |
| W3                      | 305.2        | 451.5        | 567.1        | 282          | 466.9        | 544          | 266.6        | 374.5        | 528.6        | 343.7        | 436.1        | 544          |
| F3                      | 312.9        | 405.3        | 559.4        | 258.9        | 420.7        | 597.9        | 251.2        | 374.5        | 497.8        | 359.1        | 459.2        | 544          |
| M4                      |              |              |              |              |              |              |              |              |              |              |              |              |
| W4                      | 251.2        | 397.6        | 544          | 189.6        | 382.2        | 513.2        | 212.7        | 389.9        | 536.3        | 312.9        | 351.4        | 482.4        |
| F4                      | 266.6        | 389.9        | 482.4        | 228.1        | 389.9        | 528.6        | 235.8        | 389.9        | 490.1        | 235.8        | 382.2        | 482.4        |
| M5                      | 282          | 389.9        | 544          | 251.2        | 389.9        | 428.4        | 289.8        | 382.2        | 505.5        | 328.3        | 451.5        | 482.4        |
| W5                      | 282          | 374.5        | 559.4        | 274.3        | 413          | 520.9        | 297.5        | 389.9        | 497.8        | 297.5        | 320.6        | 497.8        |
| F5                      | 266.6        | 436.1        | 505.5        | 289.8        | 359.1        | 505.5        | 297.5        | 405.3        | 513.2        | 305.2        | 428.4        | 528.6        |
| M6                      | 305.2        | 366.8        | 544          | 343.7        | 397.6        | 497.8        | 328.3        | 413          | 513.2        | 312.9        | 374.5        | 466.9        |
| W6                      | 305.2        | 359.1        | 436.1        | 297.5        | 405.3        | 482.4        | 274.3        | 382.2        | 490.1        | 328.3        | 336          | 497.8        |
| F6                      | 366.8        | 436.1        | 490.1        | 351.4        | 405.3        | 636.4        | 297.5        | 366.8        | 520.9        | 513.2        | 466.9        | 528.6        |
| Average                 | 268          | 368          | 493          | 265          | 371          | 491          | 272          | 378          | 503          | 310          | 407          | 521          |
| Std. Dev.               | 34.6         | 41.3         | 30.4         | 30.2         | 28.6         | 30.9         | 43.4         | 16.0         | 13.0         | 69.6         | 49.1         | 36.0         |
| Factor                  | 150          |              |              |              |              |              |              |              |              |              |              |              |
| <b>TCOD</b>             | <b>40227</b> | <b>55135</b> | <b>73943</b> | <b>39792</b> | <b>55680</b> | <b>73654</b> | <b>40744</b> | <b>56708</b> | <b>75400</b> | <b>46454</b> | <b>61001</b> | <b>78132</b> |
| Std.Dev.                | 5190         | 6194         | 4554         | 4535         | 4293         | 4641         | 6513         | 2406         | 1952         | 10433        | 7368         | 5405         |

**Table B - 12. Soluble COD results – SRT 2.2 days.**

| COD Concentration, mg/l |              |              |              |             |              |              |             |              |              |             |             |             |
|-------------------------|--------------|--------------|--------------|-------------|--------------|--------------|-------------|--------------|--------------|-------------|-------------|-------------|
| Treatment               | NT           | 6T           | 8T           | NM          | 6M           | 8M           | NL-M        | 6L-M         | 8L-M         | NR          | 6R          | 8R          |
| Day/Week                |              |              |              |             |              |              |             |              |              |             |             |             |
| M1                      | 185.4        | 231.7        | 301          | 200.8       | 231.7        | 324.1        | 131.5       | 200.8        | 277.9        | 46.76       | 108.4       | 154.6       |
| W1                      | 162.3        | 247.1        | 324.1        | 162.3       | 247.1        | 324.1        | 139.2       | 224          | 277.9        | 62.17       | 100.7       | 231.7       |
| F1                      | 162.3        | 247.1        | 308.7        | 131.5       | 247.1        | 339.5        | 170         | 231.7        | 277.9        | 131.5       | 139.2       | 200.8       |
| M2                      | 200.8        | 262.5        | 339.5        | 162.3       | 247.1        | 339.5        | 154.6       | 216.3        | 293.3        | 116.1       | 139.2       | 170         |
| W2                      | 177.7        | 285.6        | 393.5        | 193.1       | 285.6        | 385.7        | 154.6       | 224          | 316.4        | 116.1       | 162.3       | 216.3       |
| F2                      | 185.4        | 301          | 385.7        | 177.7       | 293.3        | 401.2        | 154.6       | 247.1        | 370.3        | 123.8       | 185.4       | 177.7       |
| M3                      | 200.8        | 324.1        | 408.9        | 185.4       | 316.4        | 424.3        | 162.3       | 270.2        | 362.6        | 108.4       | 92.99       | 139.2       |
| W3                      | 208.6        | 293.3        | 401.2        | 200.8       | 277.9        | 370.3        | 170         | 239.4        | 524.4        | 108.4       | 123.8       | 139.2       |
| F3                      | 185.4        | 262.5        | 339.5        | 170         | 247.1        | 308.7        | 200.8       | 200.8        | 277.9        | 92.99       | 108.4       | 131.5       |
| M4                      |              |              |              |             |              |              |             |              |              |             |             |             |
| W4                      | 170          | 277.9        | 354.9        | 154.6       | 254.8        | 324.1        | 146.9       | 216.3        | 293.3        | 69.88       | 108.4       | 123.8       |
| F4                      | 262.5        | 277.9        | 378          | 177.7       | 270.2        | 347.2        | 146.9       | 216.3        | 293.3        | 92.99       | 108.4       | 170         |
| M5                      | 193.1        | 277.9        | 362.6        | 216.3       | 262.5        | 339.5        | 170         | 247.1        | 293.3        | 123.8       | 139.2       | 170         |
| W5                      | 231.7        | 285.6        | 385.7        | 231.7       | 270.2        | 354.9        | 200.8       | 239.4        | 324.1        | 123.8       | 131.5       | 262.5       |
| F5                      | 262.5        | 293.3        | 378          | 231.7       | 270.2        | 354.9        | 200.8       | 239.4        | 308.7        | 146.9       | 162.3       | 170         |
| M6                      | 231.7        | 324.1        | 408.9        | 231.7       | 308.7        | 354.9        | 200.8       | 285.6        | 370.3        | 100.7       | 131.5       | 185.4       |
| W6                      | 200.8        | 324.1        | 385.7        | 208.6       | 308.7        | 354.9        | 200.8       | 316.4        | 339.5        | 139.2       | 146.9       | 216.3       |
| F6                      | 208.6        | 285.6        | 385.7        | 185.4       | 277.9        | 331.8        | 200.8       | 239.4        | 324.1        | 100.7       | 162.3       | 208.6       |
| Average                 | 202          | 275          | 354          | 187         | 270          | 345          | 170         | 234          | 306          | 106         | 132         | 180         |
| Std. Dev.               | 29.9         | 19.9         | 20.3         | 25.3        | 21.4         | 31.5         | 23.3        | 31.0         | 63.8         | 20.8        | 27.3        | 40.2        |
| Factor                  | 50           |              |              |             |              |              |             |              |              |             |             |             |
| <b>TCOD</b>             | <b>10119</b> | <b>13740</b> | <b>17704</b> | <b>9349</b> | <b>13509</b> | <b>17265</b> | <b>8502</b> | <b>11680</b> | <b>15295</b> | <b>5307</b> | <b>6621</b> | <b>9023</b> |
| Std.Dev.                | 1496         | 994.6        | 1014         | 1264        | 1071         | 1577         | 1166        | 1552         | 3188         | 1038        | 1367        | 2008        |

**Table B - 13. Gas production from batch acid digestion.**

| Gas production from batch acid digesters |         |         |         |                       |         |         |         |                           |         |         |         |
|--|---------|---------|---------|-----------------------|---------|---------|---------|---------------------------|---------|---------|---------|
| Thermophilic conditions                  |         |         |         | Mesophilic conditions |         |         |         | Low-mesophilic conditions |         |         |         |
| 3.7% TS                                  | 6.0% TS | 8.1% TS | 9.8% TS | 3.7% TS               | 6.0% TS | 8.1% TS | 9.8% TS | 3.7% TS                   | 6.0% TS | 8.1% TS | 9.8% TS |
| STP L/d                                  | STP L/d | STP L/d | STP L/d | STP L/d               | STP L/d | STP L/d | STP L/d | STP L/d                   | STP L/d | STP L/d | STP L/d |
| 0.47                                     | 0.75    | 0.96    | 0.88    | 0.35                  | 0.67    | 0.67    | 0.62    | 0.06                      | 0.37    | 0.46    | 0.35    |
| 0.13                                     | 0.50    | 0.61    | 0.17    | 0.43                  | 0.71    | 0.77    | 0.50    | 0.06                      | 0.60    | 0.71    | 0.56    |
| 0.03                                     | 0.29    | 0.32    | 0.03    | 0.58                  | 0.76    | 0.86    | 0.32    | 0.13                      | 0.61    | 0.56    | 0.40    |
| 0.06                                     | 0.32    | 0.44    | 0.04    | 0.33                  | 0.71    | 0.59    | 0.16    | 0.10                      | 0.34    | 0.35    | 0.14    |
| 0.04                                     | 0.21    | 0.40    | 0.07    | 0.20                  | 0.63    | 0.34    | 0.29    | 0.07                      | 0.23    | 0.26    | 0.17    |

**Table B - 14. Total and volatile solids at the beginning of batch tests.**

| Target TS | Solids measurement # 1 |         | Solids measurement # 2 |         | Solids measurement # 3 |         | Average |         |
|-----------|------------------------|---------|------------------------|---------|------------------------|---------|---------|---------|
|           | TS, g/l                | VS, g/l | TS, g/l                | VS, g/l | TS, g/l                | VS, g/l | TS, g/l | VS, g/l |
| Normal    | 37.61                  | 21.45   | 38.58                  | 19.29   | 35.86                  | 16.9    | 37.35   | 19.21   |
| 6%        | 60.08                  | 33.72   | 64.47                  | 31.41   | 55.42                  | 26.97   | 59.99   | 30.70   |
| 8%        | 81.88                  | 45.74   | 82.60                  | 40.51   | 77.70                  | 36.38   | 80.73   | 40.87   |
| 10%       | 98.58                  | 54.87   | 96.33                  | 47.61   | 99.44                  | 45.98   | 98.11   | 49.49   |

**Table B - 15. Gas composition from batch acid digestion.**

| Gas composition - Thermophilic acid digesters   |      |      |         |      |      |         |      |      |         |      |      |
|---|------|------|---------|------|------|---------|------|------|---------|------|------|
| 3.7% TS   |      |      | 6.0% TS |      |      | 8.1% TS |      |      | 9.8% TS |      |      |
| CH4   | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   |
| 0.0   | 84.1 | 15.9 | 0.0     | 85.3 | 14.7 | 0.0     | 80.8 | 19.2 | 0.0     | 81.4 | 18.6 |
| 0.0   | 88.9 | 11.1 | 0.0     | 84.4 | 15.6 | 0.0     | 84.0 | 16.0 | 0.0     | 80.9 | 19.1 |
| 0.0   | 93.4 | 6.6  | 0.0     | 87.1 | 12.9 | 0.0     | 85.6 | 14.4 | 0.0     | 83.7 | 16.3 |
| 3.9   | 94.1 | 2.0  | 6.4     | 88.6 | 5.0  | 8.0     | 84.4 | 7.6  | 3.4     | 86.5 | 10.1 |
| 7.4   | 91.9 | 0.8  | 8.6     | 88.3 | 3.1  | 15.6    | 81.2 | 3.2  | 11.3    | 85.0 | 3.7  |
| Gas composition - Mesophilic acid digesters     |      |      |         |      |      |         |      |      |         |      |      |
| 3.7% TS   |      |      | 6.0% TS |      |      | 8.1% TS |      |      | 9.8% TS |      |      |
| CH4   | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   |
| 34.8  | 65.2 | 0.0  | 30.8    | 69.2 | 0.0  | 31.2    | 68.8 | 0.0  | 7.7     | 42.3 | 0.0  |
| 39.2  | 60.8 | 0.0  | 38.8    | 61.2 | 0.0  | 33.4    | 66.6 | 0.0  | 33.1    | 66.9 | 0.0  |
| 37.8  | 62.2 | 0.0  | 34.3    | 65.7 | 0.0  | 31.8    | 68.2 | 0.0  | 35.4    | 64.6 | 0.0  |
| 40.1  | 59.9 | 0.0  | 37.0    | 63.0 | 0.0  | 33.3    | 66.7 | 0.0  | 8.2     | 41.8 | 0.0  |
| 40.8  | 59.2 | 0.0  | 42.9    | 57.1 | 0.0  | 35.9    | 64.1 | 0.0  | 8.0     | 42.0 | 0.0  |
| Gas composition - Low-mesophilic acid digesters |      |      |         |      |      |         |      |      |         |      |      |
| 3.7% TS   |      |      | 6.0% TS |      |      | 8.1% TS |      |      | 9.8% TS |      |      |
| CH4   | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   | CH4     | CO2  | H2   |
| 6.6   | 93.4 | 0.0  | 27.5    | 72.5 | 0.0  | 16.5    | 83.5 | 0.0  | 5.3     | 94.7 | 0.0  |
| 8.6   | 91.4 | 0.0  | 31.2    | 68.8 | 0.0  | 25.7    | 74.3 | 0.0  | 21.2    | 78.8 | 0.0  |
| 11.7  | 88.3 | 0.0  | 28.0    | 72.0 | 0.0  | 24.5    | 75.5 | 0.0  | 21.4    | 78.6 | 0.0  |
| 12.7  | 87.3 | 0.0  | 27.1    | 72.9 | 0.0  | 24.8    | 75.2 | 0.0  | 22.4    | 77.6 | 0.0  |
| 13.8  | 86.2 | 0.0  | 26.2    | 73.8 | 0.0  | 24.5    | 75.5 | 0.0  | 21.5    | 78.5 | 0.0  |

**Table B - 16. Influent COD for thermophilic acid batch digesters.**

| Sludge type   | Total COD, mg/l |              |              |              |
|---------------|-----------------|--------------|--------------|--------------|
|               | Normal          | 6%           | 8%           | 10%          |
| Measurement 1 | 33075           | 66249        | 69758        | 106443       |
| Measurement 2 | 30654           | 54751        | 73861        | 84583        |
| Measurement 3 | 27086           | 51314        | 64277        | 82383        |
| Average       | <b>30272</b>    | <b>57438</b> | <b>69299</b> | <b>91136</b> |
| Std. Dev.     | 3013            | 7822         | 4809         | 13302        |

**Table B - 17. Estimation of hydrogen recovery as COD from thermophilic acid batch digesters.**

| Thermophilic acid gas production             |                           |                           |                           |
|--|---------------------------|---------------------------|---------------------------|
| 3.7% TS                                      | 6.0% TS                   | 8.1% TS                   | 9.8% TS                   |
| STP L/day                                    | STP L/day                 | STP L/day                 | STP L/day                 |
| 0.47   | 0.75                      | 0.96                      | 0.88                      |
| 0.13   | 0.5                       | 0.61                      | 0.17                      |
| 0.03   | 0.29                      | 0.32                      | 0.03                      |
| 0.06   | 0.32                      | 0.44                      | 0.04                      |
| 0.04   | 0.21                      | 0.4                       | 0.07                      |
| Hydrogen fraction                            |                           |                           |                           |
| 3.7% TS                                      | 6.0% TS                   | 8.1% TS                   | 9.8% TS                   |
| % H <sub>2</sub>                             | % H <sub>2</sub>          | % H <sub>2</sub>          | % H <sub>2</sub>          |
| 15.9   | 14.7                      | 19.2                      | 18.6                      |
| 11.1   | 15.6                      | 16                        | 19.1                      |
| 6.6  | 12.9                      | 14.4                      | 16.3                      |
| 2  | 5                         | 7.6                       | 10.1                      |
| 0.8  | 3.1                       | 3.2                       | 3.7                       |
| Hydrogen production                          |                           |                           |                           |
| 3.7% TS                                      | 6.0% TS                   | 8.1% TS                   | 9.8% TS                   |
| STP L H <sub>2</sub> /day                    | STP L H <sub>2</sub> /day | STP L H <sub>2</sub> /day | STP L H <sub>2</sub> /day |
| 0.075  | 0.110                     | 0.184                     | 0.164                     |
| 0.014  | 0.078                     | 0.098                     | 0.032                     |
| 0.002  | 0.037                     | 0.046                     | 0.005                     |
| 0.001  | 0.016                     | 0.033                     | 0.004                     |
| 0.000  | 0.007                     | 0.013                     | 0.003                     |
| 0.093  | 0.248                     | 0.374                     | 0.208                     |
| Hydrogen COD = 0.72 g COD / L H <sub>2</sub> |                           |                           |                           |
| <b>67</b>                                    | <b>179</b>                | <b>269</b>                | <b>150</b>                |

## **Appendix C - Data for section 6.0.**

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**Table C - 1. Average Total and Volatile Solids in batch acid digesters.**

| Target Solids content | Actual Solids Content, g/L |                 | Operation Temperature |
|-----------------------|----------------------------|-----------------|-----------------------|
|                       | Total Solids               | Volatile Solids |                       |
| Normal - unadjusted   | 35.9                       | 16.9            | 21 °C                 |
| 6% TS                 | 55.4                       | 27.0            |                       |
| 8% TS                 | 77.7                       | 36.4            |                       |

| Target Solids content | Actual Solids Content, g/L |                 | Operation Temperature |
|-----------------------|----------------------------|-----------------|-----------------------|
|                       | Total Solids               | Volatile Solids |                       |
| Normal - unadjusted   | 38.6                       | 19.3            | 38 °C                 |
| 6% TS                 | 64.5                       | 31.4            |                       |
| 8% TS                 | 82.6                       | 40.5            |                       |

**Table C - 2. Fecal coliforms counts in mesophilic acid digesters.**

| Date       | Day | Fecal coliforms  |                  |                  |          |          |          |                  |                  |                  |          |          |          |                  |                  |                  |          |          |          |
|------------|-----|------------------|------------------|------------------|----------|----------|----------|------------------|------------------|------------------|----------|----------|----------|------------------|------------------|------------------|----------|----------|----------|
|            |     | 3.8% TS          |                  |                  | 6.4% TS  |          |          | 8.3% TS          |                  |                  |          |          |          |                  |                  |                  |          |          |          |
|            |     | Dilution         | Result           | TS               | Results  | Dilution | Result   | TS               | Results          | Dilution         | Result   | TS       | Results  |                  |                  |                  |          |          |          |
| 10/08/2003 | 0   | 10 <sup>-4</sup> | 10 <sup>-6</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-5</sup> | 10 <sup>-5</sup> | 10 <sup>-7</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-5</sup> | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 1.50E+05 | 3.86E+01 | 3.89E+06 | +                | +                | -                | 2.50E+06 | 6.45E+01 | 3.88E+07 | +                | +                | -                | 2.00E+06 | 8.26E+01 | 2.42E+07 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |
| 11/08/2003 | 1   | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 2.50E+04 | 3.66E+01 | 6.83E+05 | +                | +                | -                | 9.50E+04 | 5.37E+01 | 1.77E+06 | +                | +                | -                | 4.50E+04 | 6.82E+01 | 6.60E+05 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |
| 12/08/2003 | 2   | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-5</sup> | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 2.50E+05 | 3.46E+01 | 7.22E+06 | +                | +                | -                | 4.50E+05 | 5.32E+01 | 8.45E+06 | +                | +                | -                | 2.50E+06 | 7.16E+01 | 3.49E+07 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |
| 13/08/2003 | 3   | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 4.50E+04 | 3.52E+01 | 1.28E+06 | +                | +                | -                | 9.50E+03 | 5.43E+01 | 1.75E+05 | +                | +                | -                | 3.00E+05 | 6.86E+01 | 4.37E+06 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |
| 15/08/2003 | 5   | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 9.50E+01 | 3.59E+01 | 2.65E+03 | +                | +                | -                | 2.50E+01 | 5.18E+01 | 4.83E+02 | +                | +                | -                | 4.50E+01 | 6.80E+01 | 6.62E+02 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |

**Table C - 3. Fecal coliforms counts in low-mesophilic acid digesters.**

| Date       | Day | Fecal coliforms  |                  |                  |          |          |          |                  |                  |                  |          |          |          |                  |                  |                  |          |          |          |
|------------|-----|------------------|------------------|------------------|----------|----------|----------|------------------|------------------|------------------|----------|----------|----------|------------------|------------------|------------------|----------|----------|----------|
|            |     | 3.6% TS          |                  |                  | 5.5% TS  |          |          | 7.8% TS          |                  |                  |          |          |          |                  |                  |                  |          |          |          |
|            |     | Dilution         | Result           | TS               | Results  | Dilution | Result   | TS               | Results          | Dilution         | Result   | TS       | Results  |                  |                  |                  |          |          |          |
| 10/08/2003 | 0   | 10 <sup>-5</sup> | 10 <sup>-7</sup> | 10 <sup>-8</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-5</sup> | 10 <sup>-7</sup> | 10 <sup>-8</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-5</sup> | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 4.50E+06 | 3.59E+01 | 1.25E+08 | +                | +                | -                | 9.50E+06 | 5.54E+01 | 1.71E+08 | +                | +                | -                | 9.50E+05 | 7.77E+01 | 1.22E+07 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |
| 11/08/2003 | 1   | 10 <sup>-5</sup> | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-5</sup> | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-5</sup> | 10 <sup>-6</sup> | 10 <sup>-7</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 4.50E+05 | 3.00E+01 | 1.50E+07 | +                | +                | -                | 4.50E+05 | 5.02E+01 | 8.97E+06 | +                | +                | -                | 9.50E+05 | 7.24E+01 | 1.31E+07 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |
| 12/08/2003 | 2   | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 4.50E+04 | 2.88E+01 | 1.56E+06 | +                | +                | -                | 7.50E+04 | 5.10E+01 | 1.47E+06 | +                | +                | -                | 1.50E+05 | 6.62E+01 | 2.27E+06 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |
| 13/08/2003 | 3   | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 1.60E+05 | 2.86E+01 | 5.60E+06 | +                | +                | -                | 4.50E+04 | 5.11E+01 | 8.81E+05 | +                | +                | -                | 2.50E+05 | 5.11E+01 | 4.90E+06 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |
| 15/08/2003 | 5   | 10 <sup>-0</sup> | 10 <sup>-1</sup> | 10 <sup>-2</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-0</sup> | 10 <sup>-1</sup> | 10 <sup>-2</sup> | MPN/ml   | g/l      | MPN/g TS | 10 <sup>-0</sup> | 10 <sup>-1</sup> | 10 <sup>-2</sup> | MPN/ml   | g/l      | MPN/g TS |
|            |     | +                | +                | -                | 0.00E+00 | 2.89E+01 | 0.00E+00 | +                | +                | -                | 0.00E+00 | 4.99E+01 | 1.00E+00 | +                | +                | -                | 0.00E+00 | 4.99E+01 | 1.00E+00 |
|            |     | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        | +                | +                | -                | -        | -        | -        |





**Table C - 7. Fecal coliforms counts – batch experiment #2.**

| Date             | Day              | Fecal coliforms  |                  |                  |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|------------------|------------------|------------------|------------------|------------------|----------|------------------|------------------|------------------|------------------|------------------|----------|------------------|------------------|------------------|------------------|------------------|----------|-------|----------|
|                  |                  | Raw              |                  |                  |          |                  |                  | Batch Meso       |                  |                  |          |                  |                  | Batch Low-Meso   |                  |                  |          |       |          |
|                  |                  | Dilution         | Dilution         | Dilution         | Result   | TS               | Result           | Dilution         | Dilution         | Dilution         | Result   | TS               | Result           | Dilution         | Dilution         | Dilution         | Result   | TS    | Result   |
| 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml           | g/l              | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml           | g/l              | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml           | g/l              | MPN/g TS |       |          |
| 25/08/2004       | Mon              | +                | +                | -                | 4.50E+05 | 29.41            | 1.53E+07         | +                | -                | -                | 2.50E+03 | 27.88            | 8.97E+04         | +                | +                | -                | 2.50E+05 | 28.15 | 8.88E+06 |
|                  |                  | +                | +                | +                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 26/08/2004       | Tue              | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | +                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 27/08/2004       | Wed              | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 28/08/2004       | Thur             | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 29/08/2004       | Fri              | +                | +                | +                | 4.50E+05 | 29.41            | 1.53E+07         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 30/08/2004       | Sat              | +                | +                | -                | 4.50E+05 | 29.41            | 1.53E+07         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 31/08/2004       | Sun              | +                | +                | -                | 4.50E+05 | 29.41            | 1.53E+07         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 01/09/2004       | Mon              | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 02/09/2004       | Tue              | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |

**Table C - 8. Fecal coliforms counts – batch experiment #3.**

| Date             | Day              | Fecal coliforms  |                  |                  |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|------------------|------------------|------------------|------------------|------------------|----------|------------------|------------------|------------------|------------------|------------------|----------|------------------|------------------|------------------|------------------|------------------|----------|-------|----------|
|                  |                  | Raw              |                  |                  |          |                  |                  | Batch Meso       |                  |                  |          |                  |                  | Batch Low-Meso   |                  |                  |          |       |          |
|                  |                  | Dilution         | Dilution         | Dilution         | Result   | TS               | Result           | Dilution         | Dilution         | Dilution         | Result   | TS               | Result           | Dilution         | Dilution         | Dilution         | Result   | TS    | Result   |
| 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml           | g/l              | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml           | g/l              | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml           | g/l              | MPN/g TS |       |          |
| 22/09/2004       | Mon              | +                | +                | -                | 4.50E+05 | 57.86            | 7.78E+06         | +                | -                | -                | 3.00E+04 | 53.51            | 5.61E+05         | +                | -                | -                | 2.50E+03 | 52.81 | 4.73E+04 |
|                  |                  | +                | +                | +                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 23/09/2004       | Tue              | 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | +                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 24/09/2004       | Wed              | +                | +                | -                | 4.50E+05 | 57.86            | 7.78E+06         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 25/09/2004       | Thur             | +                | +                | -                | 4.50E+05 | 57.86            | 7.78E+06         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 26/09/2004       | Fri              | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 27/09/2004       | Sat              | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 28/09/2004       | Sun              | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 29/09/2004       | Mon              | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
| 30/09/2004       | Tue              | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l              | MPN/g TS         | 10 <sup>-1</sup> | 10 <sup>-2</sup> | 10 <sup>-3</sup> | MPN/ml   | g/l   | MPN/g TS |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |
|                  |                  | +                | +                | -                |          |                  |                  |                  |                  |                  |          |                  |                  |                  |                  |                  |          |       |          |

**Table C - 9. Total VFA, pH and un-ionized VFA concentrations – batch experiment #1.**

| Sludge   | Day | Reading |           |           |           |           |           |           |           |         |      | Total  | pH    | Undissociated part of acids |       |       |       |      |       |        |      | Total | % total |
|----------|-----|---------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|---------|------|--------|-------|-----------------------------|-------|-------|-------|------|-------|--------|------|-------|---------|
|          |     | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | mg/l    | mg/l |        |       | mg/l                        | mg/l  | mg/l  | mg/l  | mg/l | mg/l  | mg/l   | mg/l |       |         |
| Low-Meso | 0   | 2008.5  | 776.3     | 82.6      | 327.9     | 100.5     | 87.5      | 46.9      | 9.6       | 3439.8  | 6.1  | 93.2   | 46.5  | 4.6                         | 17.2  | 4.8   | 2.7   | 0.5  | 174.2 | 5.1    |      |       |         |
|          | 1   | 3018.8  | 1160.7    | 139.2     | 570.3     | 189.3     | 170.1     | 87.5      | 20.2      | 5361.1  | 5.9  | 220.7  | 110.5 | 12.1                        | 46.9  | 14.1  | 14.7  | 6.0  | 1.8   | 426.9  | 8.0  |       |         |
|          | 2   | 2894.7  | 1144.1    | 159.5     | 640.3     | 213.7     | 226.0     | 70.8      | 19.7      | 5368.8  | 5.7  | 272.7  | 137.1 | 17.8                        | 67.6  | 20.5  | 25.0  | 8.1  | 2.2   | 551.1  | 10.3 |       |         |
|          | 3   | 3642.8  | 1297.5    | 177.0     | 771.1     | 238.5     | 264.8     | 77.7      | 17.6      | 6466.8  | 5.5  | 568.5  | 252.9 | 32.4                        | 133.8 | 37.9  | 48.0  | 14.6 | 3.3   | 1091.3 | 16.8 |       |         |
|          | 4   | 4370.5  | 1567.4    | 181.7     | 837.7     | 233.2     | 279.8     | 68.3      | 19.4      | 7558.0  | 5.4  | 794.9  | 358.4 | 38.5                        | 168.8 | 43.2  | 58.8  | 14.8 | 4.2   | 1476.7 | 19.5 |       |         |
|          | 5   | 5108.6  | 1893.3    | 200.7     | 953.0     | 250.4     | 317.1     | 83.0      | 19.8      | 8825.9  | 5.3  | 1117.2 | 507.7 | 50.8                        | 229.8 | 55.8  | 79.6  | 21.5 | 5.1   | 2067.4 | 23.4 |       |         |
|          | 6   | 5742.6  | 2177.3    | 214.4     | 1058.4    | 271.1     | 356.5     | 89.4      | 29.1      | 9938.8  | 5.2  | 1421.1 | 653.3 | 61.1                        | 287.7 | 63.2  | 100.7 | 26.0 | 8.4   | 2528.5 | 26.4 |       |         |
|          | 7   | 6687.9  | 2633.2    | 237.1     | 1116.0    | 307.5     | 427.0     | 85.7      | 35.6      | 11590.2 | 5.2  | 1683.9 | 823.6 | 68.6                        | 308.4 | 78.8  | 122.6 | 25.3 | 10.5  | 3121.8 | 26.9 |       |         |
|          | 8   | 6755.1  | 2588.2    | 234.6     | 1087.9    | 301.3     | 389.2     | 80.0      | 31.2      | 11467.4 | 5.2  | 1871.7 | 778.9 | 66.8                        | 295.7 | 75.9  | 109.9 | 23.3 | 9.0   | 3031.2 | 26.4 |       |         |
| Meso     | 9   | 7287.7  | 2248.6    | 234.8     | 902.2     | 296.6     | 374.6     | 71.9      | 50.1      | 11466.9 | 5.3  | 1772.4 | 666.0 | 65.8                        | 241.1 | 73.4  | 104.1 | 20.6 | 14.3  | 2957.6 | 25.8 |       |         |
|          | 0   | 2008.5  | 776.3     | 82.6      | 327.9     | 100.5     | 87.5      | 46.9      | 9.6       | 3439.8  | 6.1  | 93.2   | 46.5  | 4.6                         | 17.2  | 4.8   | 2.7   | 0.5  | 174.2 | 5.1    |      |       |         |
|          | 1   | 1901.1  | 926.8     | 203.1     | 513.6     | 282.0     | 289.2     | 64.1      | 26.7      | 4322.6  | 6.0  | 105.4  | 69.3  | 13.1                        | 31.3  | 15.6  | 18.5  | 4.3  | 1.8   | 259.2  | 6.0  |       |         |
|          | 2   | 3344.2  | 1476.8    | 232.1     | 698.7     | 349.9     | 432.5     | 78.8      | 38.6      | 6651.7  | 5.7  | 321.6  | 186.6 | 26.5                        | 75.3  | 34.4  | 48.9  | 9.2  | 4.5   | 701.0  | 10.5 |       |         |
|          | 3   | 3755.1  | 1666.6    | 194.0     | 649.1     | 272.6     | 347.7     | 60.5      | 29.5      | 6975.1  | 5.5  | 574.7  | 318.9 | 34.8                        | 110.5 | 42.5  | 61.8  | 11.1 | 5.4   | 1159.8 | 16.6 |       |         |
|          | 4   | 4744.3  | 2109.3    | 237.1     | 780.8     | 350.0     | 481.5     | 81.5      | 46.1      | 9030.5  | 5.4  | 895.9  | 539.4 | 52.1                        | 163.2 | 67.3  | 104.9 | 18.3 | 10.3  | 1851.6 | 20.5 |       |         |
|          | 5   | 5906.7  | 2825.6    | 247.9     | 818.8     | 359.1     | 501.6     | 88.5      | 43.6      | 10771.7 | 5.3  | 1291.7 | 757.7 | 62.7                        | 197.4 | 79.9  | 125.9 | 17.7 | 11.2  | 2544.3 | 23.6 |       |         |
|          | 6   | 6371.0  | 3045.5    | 256.5     | 884.5     | 377.9     | 533.5     | 83.5      | 41.8      | 11613.3 | 5.3  | 1469.9 | 869.7 | 68.3                        | 224.6 | 88.7  | 140.9 | 17.3 | 11.3  | 2890.8 | 24.9 |       |         |
|          | 7   | 5122.2  | 2812.2    | 258.7     | 888.8     | 387.1     | 516.1     | 78.7      | 46.3      | 10110.1 | 5.3  | 1181.8 | 792.9 | 68.9                        | 225.7 | 90.9  | 136.3 | 21.4 | 12.5  | 2530.5 | 25.0 |       |         |
| Day 0    | 8   | 5754.5  | 2668.2    | 253.6     | 865.6     | 369.7     | 531.2     | 76.6      | 46.3      | 10863.6 | 5.3  | 1281.2 | 808.9 | 65.3                        | 212.4 | 83.8  | 135.6 | 20.2 | 12.1  | 2619.4 | 24.1 |       |         |
|          | 9   | 5120.3  | 3096.4    | 307.2     | 956.6     | 506.7     | 698.9     | 100.3     | 77.6      | 10864.2 | 5.3  | 1099.7 | 816.4 | 76.4                        | 226.6 | 110.8 | 172.4 | 25.5 | 19.6  | 2547.5 | 23.4 |       |         |
|          | 0   | 2376.5  | 869.6     | 62.5      | 319.8     | 97.7      | 103.9     | 41.5      | 9.6       | 3901.4  | 6.1  | 110.2  | 52.1  | 4.6                         | 16.7  | 4.6   | 2.7   | 0.5  | 196.9 | 5.0    |      |       |         |

**Table C - 10. Total VFA, pH and un-ionized VFA concentrations – batch experiment #2.**

| Sludge   | Day | Reading |           |           |           |           |           |           |           |        |      | Total  | pH    | Undissociated part of acids |       |       |       |      |      |        |      | Total | % total |
|----------|-----|---------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|--------|------|--------|-------|-----------------------------|-------|-------|-------|------|------|--------|------|-------|---------|
|          |     | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | mg/l   | mg/l |        |       | mg/l                        | mg/l  | mg/l  | mg/l  | mg/l | mg/l | mg/l   | mg/l |       |         |
| Low-Meso | 0   | 717.0   | 441.9     | 45.4      | 210.4     | 58.7      | 40.1      | 38.1      | 12.1      | 1563.6 | 6.1  | 31.4   | 25.0  | 2.4                         | 10.4  | 2.8   | 2.1   | 0.6  | 76.6 | 4.9    |      |       |         |
|          | 1   | 1757.0  | 910.5     | 151.5     | 658.0     | 218.3     | 133.0     | 103.3     | 21.5      | 3952.2 | 5.9  | 131.2  | 87.0  | 13.5                        | 55.2  | 16.7  | 11.7  | 9.4  | 2.0  | 326.8  | 8.3  |       |         |
|          | 2   | 2236.9  | 1053.5    | 170.1     | 712.9     | 233.8     | 200.1     | 77.2      | 16.2      | 4700.8 | 5.7  | 238.6  | 142.4 | 21.5                        | 85.1  | 25.5  | 25.0  | 10.0 | 2.1  | 550.2  | 11.7 |       |         |
|          | 3   | 2314.6  | 1113.9    | 168.9     | 785.6     | 233.8     | 248.6     | 75.6      | 20.0      | 4961.0 | 5.5  | 334.0  | 201.5 | 28.6                        | 126.2 | 34.4  | 41.7  | 13.1 | 3.5  | 783.1  | 15.8 |       |         |
|          | 4   | 3226.7  | 1600.3    | 230.0     | 925.3     | 268.5     | 268.3     | 78.6      | 23.8      | 6642.7 | 5.3  | 683.0  | 422.0 | 57.2                        | 219.2 | 63.3  | 66.2  | 20.0 | 6.0  | 1546.9 | 23.3 |       |         |
|          | 5   | 3283.9  | 1785.8    | 228.8     | 970.8     | 285.7     | 284.8     | 73.8      | 26.4      | 6937.9 | 5.3  | 744.3  | 495.2 | 58.4                        | 242.3 | 65.9  | 73.9  | 19.8 | 7.0  | 1707.9 | 24.6 |       |         |
|          | 6   | 3174.4  | 1970.0    | 247.8     | 1096.8    | 306.5     | 346.0     | 64.4      | 29.8      | 7255.8 | 5.3  | 772.0  | 583.4 | 63.4                        | 293.1 | 75.9  | 96.1  | 24.2 | 8.5  | 1922.6 | 26.5 |       |         |
|          | 7   | 3405.3  | 2156.9    | 256.5     | 1103.9    | 324.6     | 339.3     | 78.7      | 30.5      | 7695.6 | 5.2  | 887.3  | 680.9 | 76.7                        | 315.4 | 86.0  | 100.6 | 24.0 | 9.3  | 2180.2 | 28.9 |       |         |
|          | 8   | 4225.4  | 2515.4    | 313.6     | 1246.2    | 391.3     | 404.9     | 86.8      | 33.5      | 9217.3 | 5.2  | 1101.0 | 794.1 | 93.8                        | 356.0 | 103.7 | 120.1 | 26.5 | 10.2 | 2605.3 | 28.3 |       |         |
| Meso     | 9   | 4100.6  | 2246.0    | 236.5     | 1008.3    | 301.7     | 314.3     | 65.0      | 27.1      | 8299.5 | 5.2  | 1032.5 | 686.9 | 68.5                        | 278.7 | 77.3  | 90.2  | 19.2 | 8.0  | 2261.2 | 27.2 |       |         |
|          | 0   | 717.0   | 441.9     | 45.4      | 210.4     | 58.7      | 40.1      | 38.1      | 12.1      | 1563.6 | 6.1  | 31.4   | 25.0  | 2.4                         | 10.4  | 2.8   | 2.1   | 0.6  | 76.6 | 4.9    |      |       |         |
|          | 1   | 1364.3  | 825.9     | 149.0     | 484.4     | 224.0     | 182.5     | 67.0      | 16.4      | 3287.6 | 5.9  | 91.8   | 71.1  | 11.9                        | 35.4  | 15.4  | 14.5  | 4.7  | 1.3  | 245.8  | 7.5  |       |         |
|          | 2   | 1993.8  | 1129.1    | 177.1     | 607.9     | 265.3     | 314.6     | 60.7      | 18.4      | 4566.7 | 5.7  | 217.1  | 155.7 | 22.8                        | 74.0  | 29.5  | 40.1  | 8.0  | 2.4  | 549.7  | 12.0 |       |         |
|          | 3   | 2485.0  | 1609.8    | 203.5     | 693.0     | 290.8     | 337.8     | 59.0      | 18.0      | 5696.9 | 5.4  | 400.6  | 369.4 | 44.0                        | 142.2 | 54.9  | 72.3  | 13.0 | 4.0  | 1164.0 | 20.4 |       |         |
|          | 4   | 3110.2  | 2057.7    | 231.0     | 802.4     | 330.4     | 387.1     | 58.9      | 19.0      | 6996.8 | 5.3  | 705.0  | 570.6 | 60.5                        | 200.3 | 76.2  | 100.5 | 15.8 | 5.1  | 1733.9 | 24.8 |       |         |
|          | 5   | 2784.2  | 2149.3    | 245.8     | 896.3     | 362.2     | 487.6     | 69.2      | 25.5      | 7020.1 | 5.2  | 725.4  | 678.5 | 73.5                        | 256.1 | 96.0  | 106.6 | 21.1 | 7.7  | 2000.0 | 28.5 |       |         |
|          | 6   | 3172.7  | 2264.2    | 227.5     | 844.3     | 332.2     | 426.2     | 68.5      | 24.7      | 7348.6 | 5.1  | 826.7  | 744.8 | 68.1                        | 240.8 | 82.4  | 126.4 | 17.9 | 6.9  | 2201.0 | 29.9 |       |         |
|          | 7   | 3286.5  | 2560.7    | 298.0     | 979.4     | 422.9     | 525.4     | 71.7      | 23.8      | 8168.3 | 5.2  | 841.8  | 795.7 | 87.7                        | 275.2 | 110.2 | 153.3 | 21.5 | 7.1  | 2292.6 | 28.1 |       |         |
| Day 0    | 8   | 3462.8  | 2470.8    | 259.2     | 899.9     | 566.9     | 679.6     | 89.6      | 32.0      | 8461.0 | 5.2  | 871.9  | 755.6 | 75.0                        | 248.7 | 145.2 | 195.1 | 26.5 | 9.4  | 2327.5 | 27.5 |       |         |
|          | 9   | 3262.1  | 2559.2    | 350.7     | 1068.8    | 490.3     | 602.2     | 82.4      | 30.0      | 8425.7 | 5.3  | 779.6  | 745.7 | 91.1                        | 280.8 | 119.2 | 164.5 | 23.2 | 8.4  | 2212.6 | 26.3 |       |         |
|          | 0   | 821.3   | 410.4     | 44.2      | 183.9     | 53.3      | 44.6      | 35.2      | 15.0      | 1418.5 | 6.1  | 27.0   | 23.0  | 2.3                         | 9.5   | 2.4   | 2.3   | 1.9  | 0.8  | 69.2   | 4.9  |       |         |

**Table C - 11. Total VFA, pH and un-ionized VFA concentrations – batch experiment #3.**

| Sludge   | Day | Reading |           |           |           |           |           |           |           |        |      | Total | pH    | Undissociated part of acids |       |      |       |      |      |        |      | Total | % total |
|----------|-----|---------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|--------|------|-------|-------|-----------------------------|-------|------|-------|------|------|--------|------|-------|---------|
|          |     | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | mg/l   | mg/l |       |       | mg/l                        | mg/l  | mg/l | mg/l  | mg/l | mg/l | mg/l   | mg/l |       |         |
| Low-Meso | 0   | 537.7   | 362.4     | 29.1      | 120.5     | 37.6      | 32.7      | 12.0      | 4.7       | 1136.7 | 6.0  | 27.8  | 24.2  | 1.8                         | 7.0   | 2.0  | 0.8   | 0.3  | 65.9 | 5.8    |      |       |         |
|          | 1   | 1181.2  | 881.8     | 116.2     | 570.6     | 160.4     | 180.0     | 33.4      | 16.1      | 3139.7 | 5.6  | 151.3 | 142.2 | 17.5                        | 81.5  | 21.0 | 26.9  | 5.2  | 2.5  | 448.1  | 14.3 |       |         |
|          | 2   | 1473.5  | 990.4     | 121.9     | 667.6     | 164.2     | 176.2     | 31.2      | 14.4      | 3639.4 | 5.6  | 278.3 | 231.3 | 26.3                        | 139.5 | 31.6 | 38.4  | 7.0  | 3.2  | 756.2  | 20.9 |       |         |
|          | 3   | 1424.3  | 1083.5    | 131.7     | 726.5     | 168.4     | 198.9     | 28.3      | 15.5      | 3777.2 | 5.3  | 340.4 | 315.7 | 36.3                        | 190.9 | 41.0 | 54.3  | 8.0  | 4.3  | 990.9  | 26.2 |       |         |
|          | 4   | 1353.0  | 1252.7    | 172.3     | 823.1     | 210.2     | 270.0     | 33.9      | 24.8      | 4139.9 | 5.0  | 463.6 | 508.1 | 66.6                        | 305.9 | 73.1 | 103.7 | 13.3 | 9.7  | 1544.1 | 37.3 |       |         |
|          | 5   | 1835.9  | 1530.7    | 167.7     | 929.2     | 207.6     | 284.4     | 30.9      | 26.5      | 5012.9 | 5.0  | 697.2 | 681.1 | 71.4                        | 381.0 | 80.0 | 120.3 | 15.4 | 11.4 | 2655.8 | 41.0 |       |         |
|          | 6   | 2177.1  | 1713.3    | 176.5     | 963.6     | 213.3     | 289.0     | 28.6      | 28.7      | 5599.2 | 5.1  | 658.4 | 620.3 | 60.8                        | 317.8 | 65.8 | 88.7  | 10.0 | 10.0 | 1841.6 | 32.9 |       |         |
|          | 7   | 2263.9  | 1755.7    | 181.4     | 960.2     | 218.6     | 293.8     | 28.1      | 28.6      | 5730.5 | 5.1  | 684.6 | 635.7 | 62.4                        | 316.7 | 67.2 | 100.4 |      |      |        |      |       |         |

**Table C - 12. Fecal coliforms counts – semi-continuously fed, staged acid digestion with total SRT of 9 days.**

| Date             | Day              | Fecal coliforms  |        |     |          |                  |                  |                  |        |     |          |                  |                  |                  |        |     |          |       |          |
|------------------|------------------|------------------|--------|-----|----------|------------------|------------------|------------------|--------|-----|----------|------------------|------------------|------------------|--------|-----|----------|-------|----------|
|                  |                  | Raw              |        |     |          |                  | Batch Meso       |                  |        |     |          | Batch Low-Meso   |                  |                  |        |     |          |       |          |
|                  |                  | Dilution         |        |     | Result   | TS               | Result           | Dilution         |        |     | Result   | TS               | Result           | Dilution         |        |     | Result   | TS    | Result   |
| 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml | g/l | MPN/g TS | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml | g/l | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-4</sup> | 10 <sup>-5</sup> | MPN/ml | g/l | MPN/g TS |       |          |
| Week 1           | Mon              | +                | +      | -   | 2.50E+05 | 40.97            | 6.10E+06         | +                | +      | -   | 4.50E+02 | 24.12            | 1.87E+04         | +                | +      | -   | 4.50E+03 | 24.99 | 1.80E+05 |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
| Week 1           | Tue              | +                | +      | -   | 9.50E+05 | 26.8             | 3.54E+07         | +                | +      | -   | 2.50E+02 | 32.51            | 7.69E+03         | +                | +      | -   | 4.50E+03 | 32.82 | 1.37E+05 |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
| Week 1           | Wed              | +                | +      | -   | 9.50E+04 | 25.21            | 3.77E+06         | +                | +      | -   | 2.50E+01 | 30.08            | 8.31E+02         | +                | +      | -   | 9.50E+03 | 30.79 | 3.09E+05 |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
| Week 1           | Thur             | +                | +      | -   | 7.50E+04 | 26.61            | 2.82E+06         | +                | +      | -   | 2.50E+02 | 28.29            | 8.84E+03         | +                | +      | -   | 4.50E+03 | 28.73 | 1.57E+05 |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
| Week 2           | Mon              | +                | +      | -   | 9.50E+05 | 26.23            | 3.62E+07         | +                | +      | -   | 9.50E+01 | 25.41            | 3.74E+03         | +                | +      | -   | 9.50E+03 | 24.47 | 3.88E+05 |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
| Week 2           | Tue              | +                | +      | -   | 2.50E+06 | 25.89            | 9.66E+07         | +                | +      | -   | 4.50E+02 | 25.4             | 1.77E+04         | +                | +      | -   | 9.50E+03 | 26.01 | 3.65E+05 |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
| Week 2           | Wed              | +                | +      | -   | 9.50E+05 | 25.99            | 3.66E+07         | +                | +      | -   | 4.50E+01 | 24.1             | 1.87E+03         | +                | +      | -   | 9.50E+02 | 24.72 | 3.84E+04 |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
| Week 2           | Thur             | +                | +      | -   | 4.50E+05 | 24.00            | 1.88E+07         | +                | +      | -   | 4.50E+01 | 23.79            | 1.89E+03         | +                | +      | -   | 9.50E+03 | 23.88 | 3.98E+05 |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |
|                  |                  | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +                | -                | +                | +      | -   | +        | +     | -        |

**Table C - 13. Total VFA, pH and un-ionized VFA concentrations – semi-continuously fed, staged acid digestion with total SRT of 9 days.**

| Sludge     | Day | Reading |        |           |        |         |       |           |      |           |      | pH     | Undissociated part of acids |      |       |       |      |      |     |        | % total |
|------------|-----|---------|--------|-----------|--------|---------|-------|-----------|------|-----------|------|--------|-----------------------------|------|-------|-------|------|------|-----|--------|---------|
|            |     | Acetic  |        | Propionic |        | Ibutric |       | N-valeric |      | I-caproic |      |        | N-caproic                   |      | Total |       |      |      |     |        |         |
|            |     | mg/l    | mg/l   | mg/l      | mg/l   | mg/l    | mg/l  | mg/l      | mg/l | mg/l      | mg/l |        | mg/l                        | mg/l | mg/l  | mg/l  | mg/l |      |     |        |         |
| 1 Low-Meso | 0   | 5702.3  | 2398.6 | 358.4     | 1037.4 | 438.4   | 361.2 | 51.2      | 32.9 | 10380.4   | 5.4  | 1076.9 | 560.3                       | 78.8 | 216.8 | 84.3  | 78.7 | 11.5 | 7.4 | 2114.7 | 20.4    |
|            | 1   | 3912.9  | 1965.5 | 272.1     | 815.2  | 324.5   | 274.5 | 38.4      | 26.3 | 7629.3    | 5.4  | 738.9  | 459.1                       | 59.8 | 170.4 | 62.4  | 59.8 | 8.6  | 5.9 | 1565.0 | 20.5    |
|            | 2   | 3187.6  | 1653.7 | 226.6     | 650.4  | 260.7   | 219.4 | 28.6      | 23.5 | 6268.8    | 5.4  | 602.0  | 385.3                       | 49.8 | 137.6 | 51.7  | 47.8 | 6.4  | 5.3 | 1266.9 | 20.5    |
|            | 3   | 2554.0  | 1232.8 | 168.9     | 549.5  | 194.3   | 191.6 | 24.8      | 20.8 | 4934.9    | 5.4  | 452.3  | 288.0                       | 36.7 | 114.9 | 37.4  | 41.8 | 5.5  | 4.6 | 1011.2 | 20.5    |
|            | 4   | 2602.0  | 1300.7 | 200.5     | 582.5  | 233.9   | 188.5 | 29.5      | 23.6 | 5161.2    | 5.4  | 491.4  | 303.8                       | 44.1 | 121.7 | 45.9  | 41.1 | 6.6  | 5.3 | 1059.0 | 20.5    |
|            | 5   | 3591.3  | 1358.2 | 231.6     | 689.2  | 268.2   | 222.9 | 38.3      | 29.7 | 6429.4    | 5.4  | 678.2  | 317.3                       | 50.9 | 144.0 | 51.6  | 48.6 | 8.6  | 6.6 | 1305.9 | 20.5    |
|            | 6   | 2715.8  | 1155.2 | 204.0     | 593.2  | 234.6   | 190.5 | 34.5      | 25.3 | 5153.1    | 5.4  | 512.9  | 269.8                       | 44.8 | 124.0 | 45.1  | 41.5 | 7.8  | 5.6 | 1051.6 | 20.4    |
| Average    |     | 3468.6  | 1580.7 | 237.1     | 703.6  | 280.4   | 235.5 | 35.9      | 28.9 | 6565.0    | 5.4  | 658.6  | 385.2                       | 52.1 | 147.1 | 53.9  | 51.3 | 7.9  | 5.8 | 1342.8 | 20.5    |
| Std. Dev   |     | 1113.2  | 455.9  | 82.5      | 171.8  | 89.5    | 63.1  | 8.8       | 4.1  | 1931.0    | 5.4  | 210.2  | 105.7                       | 13.7 | 35.9  | 15.5  | 13.7 | 2.0  | 0.9 | 332.6  | 20.5    |
| 2 Low-meso | 0   | 3812.2  | 1748.1 | 303.1     | 718.8  | 361.8   | 262.2 | 43.0      | 21.6 | 7288.6    | 5.2  | 993.3  | 455.0                       | 79.0 | 187.3 | 94.3  | 68.3 | 11.2 | 5.6 | 1893.9 | 26.1    |
|            | 1   | 5901.8  | 1980.9 | 276.8     | 810.0  | 333.1   | 292.6 | 42.4      | 23.2 | 8760.8    | 5.2  | 1003.3 | 516.1                       | 72.1 | 211.0 | 66.8  | 76.2 | 11.0 | 6.0 | 2282.7 | 26.1    |
|            | 2   | 4703.9  | 2008.3 | 295.9     | 805.4  | 345.3   | 288.5 | 41.1      | 25.9 | 8514.2    | 5.2  | 1225.6 | 523.3                       | 77.1 | 209.8 | 90.0  | 75.2 | 10.7 | 6.7 | 2218.4 | 26.1    |
|            | 3   | 4281.2  | 1857.7 | 265.8     | 733.1  | 321.0   | 276.8 | 35.3      | 28.4 | 7799.2    | 5.2  | 1115.5 | 484.0                       | 69.2 | 191.0 | 83.6  | 72.1 | 9.2  | 7.4 | 2032.1 | 26.1    |
|            | 4   | 4116.4  | 1683.1 | 266.7     | 737.5  | 327.4   | 283.9 | 34.1      | 29.6 | 7478.7    | 5.2  | 1072.6 | 438.5                       | 69.5 | 192.2 | 85.3  | 74.0 | 8.9  | 7.7 | 1948.6 | 26.1    |
|            | 5   | 4355.6  | 1823.4 | 275.1     | 780.3  | 331.7   | 295.7 | 36.0      | 32.2 | 7930.0    | 5.2  | 1134.9 | 475.1                       | 71.7 | 203.3 | 86.4  | 77.0 | 9.4  | 8.4 | 2066.2 | 26.1    |
|            | 6   | 2766.4  | 1330.5 | 214.2     | 606.4  | 259.8   | 221.3 | 28.5      | 26.4 | 5453.5    | 5.2  | 720.8  | 346.7                       | 55.8 | 158.0 | 67.7  | 57.7 | 7.4  | 6.9 | 1420.9 | 26.1    |
| Average    |     | 4148.2  | 1775.7 | 271.1     | 741.8  | 325.7   | 274.4 | 37.2      | 26.7 | 7600.7    | 5.2  | 1080.8 | 462.7                       | 70.8 | 193.2 | 84.9  | 71.5 | 9.7  | 7.0 | 1890.4 | 26.1    |
| Std. Dev   |     | 1273.3  | 428.3  | 38.8      | 89.7   | 32.0    | 26.0  | 5.2       | 3.7  | 1085.0    | 5.2  | 187.9  | 59.5                        | 7.5  | 18.2  | 8.3   | 6.8  | 1.4  | 1.0 | 322.7  | 26.1    |
| 1 Meso     | 0   | 2201.7  | 1674.1 | 274.9     | 686.3  | 352.6   | 265.6 | 38.3      | 24.8 | 5518.3    | 5.3  | 464.4  | 353.1                       | 58.0 | 144.8 | 74.4  | 56.0 | 8.1  | 5.2 | 1163.9 | 21.1    |
|            | 1   | 2816.2  | 1774.9 | 299.0     | 757.0  | 393.8   | 330.2 | 42.5      | 32.6 | 6446.1    | 5.3  | 594.0  | 374.4                       | 63.1 | 159.7 | 83.1  | 69.6 | 9.0  | 6.9 | 1359.6 | 21.1    |
|            | 2   | 2897.0  | 1973.7 | 296.8     | 714.9  | 381.9   | 303.2 | 34.4      | 32.5 | 6634.5    | 5.3  | 611.0  | 416.3                       | 62.6 | 150.8 | 80.6  | 63.9 | 7.3  | 6.9 | 1399.4 | 21.1    |
|            | 3   | 3620.9  | 1800.4 | 305.8     | 661.1  | 388.4   | 289.4 | 37.1      | 34.9 | 7344.0    | 5.3  | 807.2  | 379.7                       | 64.5 | 139.4 | 81.9  | 61.0 | 7.8  | 7.4 | 1545.0 | 21.1    |
|            | 4   | 5231.5  | 1796.2 | 290.2     | 668.7  | 359.1   | 264.3 | 33.5      | 31.9 | 8677.3    | 5.3  | 1103.4 | 378.8                       | 61.2 | 141.0 | 75.7  | 56.2 | 7.1  | 6.7 | 1891.2 | 21.1    |
|            | 5   | 3667.2  | 1862.4 | 246.3     | 587.4  | 307.5   | 255.1 | 33.7      | 31.4 | 6451.1    | 5.3  | 773.5  | 287.4                       | 52.0 | 123.9 | 64.5  | 53.8 | 7.1  | 6.6 | 1369.1 | 21.1    |
|            | 6   | 3649.7  | 1510.2 | 279.0     | 657.3  | 340.5   | 268.2 | 39.3      | 34.9 | 6779.1    | 5.3  | 769.8  | 318.5                       | 58.8 | 138.6 | 71.8  | 56.6 | 8.3  | 7.4 | 1429.9 | 21.1    |
| Average    |     | 3470.0  | 1898.8 | 284.6     | 678.1  | 360.5   | 282.6 | 37.0      | 31.9 | 6841.5    | 5.3  | 731.9  | 358.3                       | 60.0 | 142.6 | 76.0  | 59.6 | 7.8  | 6.7 | 1443.0 | 21.1    |
| Std. Dev   |     | 971.8   | 204.3  | 20.7      | 52.7   | 30.6    | 26.8  | 3.9       | 3.4  | 974.8     | 5.3  | 205.0  | 47.1                        | 4.2  | 17.1  | 8.5   | 5.6  | 0.7  | 0.7 | 205.6  | 21.1    |
| 2 Meso     | 0   | 3344.4  | 1886.0 | 305.8     | 690.9  | 405.5   | 361.1 | 44.2      | 28.6 | 7172.4    | 5.3  | 455.8  | 335.9                       | 71.8 | 162.2 | 95.2  | 86.2 | 10.4 | 6.7 | 1684.3 | 23.5    |
|            | 1   | 4384.2  | 2174.4 | 358.0     | 800.4  | 481.0   | 397.3 | 48.2      | 28.5 | 8672.2    | 5.3  | 1029.6 | 510.6                       | 84.1 | 188.0 | 113.0 | 93.3 | 11.3 | 6.7 | 2036.5 | 23.5    |
|            | 2   | 4890.3  | 2319.1 | 360.4     | 855.0  | 497.6   | 406.3 | 45.1      | 32.4 | 9406.1    | 5.3  | 1148.4 | 544.6                       | 84.6 | 200.8 | 116.8 | 95.4 | 10.6 | 7.6 | 2208.9 | 23.5    |
|            | 3   | 4732.4  | 2240.8 | 363.3     | 808.0  | 498.2   | 384.5 | 40.6      | 36.5 | 9104.4    | 5.3  | 1111.3 | 526.2                       | 85.3 | 189.8 | 117.0 | 90.3 | 9.5  | 8.6 | 2138.0 | 23.5    |
|            | 4   | 4164.1  | 2052.8 | 366.9     | 779.9  | 510.0   | 378.1 | 40.3      | 40.4 | 8332.5    | 5.3  | 977.9  | 482.1                       | 86.2 | 183.1 | 119.8 | 88.6 | 9.5  | 9.5 | 1956.7 | 23.5    |
|            | 5   | 4135.4  | 1970.0 | 336.2     | 711.2  | 451.9   | 324.1 | 34.2      | 35.1 | 7992.2    | 5.3  | 971.1  | 462.6                       | 79.0 | 167.0 | 105.9 | 78.1 | 8.0  | 8.2 | 1878.0 | 23.5    |
|            | 6   | 3747.5  | 1796.1 | 314.5     | 692.6  | 432.3   | 317.6 | 34.3      | 37.3 | 7372.3    | 5.3  | 880.0  | 421.8                       | 73.9 | 162.7 | 101.5 | 74.6 | 8.0  | 8.8 | 1731.2 | 23.5    |
| Average    |     | 4242.6  | 2034.2 | 343.6     | 762.6  | 467.9   | 367.9 | 41.0      | 34.1 | 8293.9    | 5.3  | 996.3  | 477.7                       | 80.7 | 179.1 | 109.9 | 86.4 | 9.6  | 8.0 | 1947.7 | 23.5    |
| Std. Dev   |     | 465.5   | 233.0  | 25.0      | 64.6   | 39.1    | 34.6  | 5.3       | 4.5  | 840.0     | 5.3  | 109.3  | 54.7                        | 5.9  | 15.2  | 9.2   | 8.1  | 1.3  | 1.1 | 197.3  | 23.5    |
| Raw        | 0   | 586.7   | 690.5  | 279.9     | 438.2  | 367.3   | 191.9 | 54.2      | 40.4 | 2559.2    | 6.1  | 22.3   | 27.5                        | 11.1 | 17.4  | 12.2  | 7.6  | 2.2  | 1.6 | 101.7  |         |

**Table C - 14. Fecal coliforms counts – semi-continuously fed, staged acid digestion with total SRT of 11 days.**

| Date             | Day              | Fecal coliforms  |        |     |          |                  |                  |                  |        |     |          |                  |                  |                  |        |     |          |       |          |
|------------------|------------------|------------------|--------|-----|----------|------------------|------------------|------------------|--------|-----|----------|------------------|------------------|------------------|--------|-----|----------|-------|----------|
|                  |                  | Raw              |        |     |          |                  |                  | Batch Meso       |        |     |          |                  |                  | Batch Low-Meso   |        |     |          |       |          |
|                  |                  | Dilution         |        |     | Result   | TS               | Result           | Dilution         |        |     | Result   | TS               | Result           | Dilution         |        |     | Result   | TS    | Result   |
| 10 <sup>-4</sup> | 10 <sup>-5</sup> | 10 <sup>-6</sup> | MPN/ml | g/l | MPN/g TS | 10 <sup>-3</sup> | 10 <sup>-2</sup> | 10 <sup>-1</sup> | MPN/ml | g/l | MPN/g TS | 10 <sup>-2</sup> | 10 <sup>-3</sup> | 10 <sup>-4</sup> | MPN/ml | g/l | MPN/g TS |       |          |
| Week 1           | Mon              | +                | +      | -   | 4.50E+05 | 25.65            | 1.75E+07         | +                | -      | -   | 6.00E-01 | 22.55            | 2.66E+01         | +                | +      | -   | 4.50E+03 | 22.93 | 1.96E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 2.50E+02 | 22.45            | 1.11E+04         | +                | +      | -   | 2.50E+03 | 23.18 | 1.08E+05 |
| Week 1           | Tue              | +                | +      | -   | 4.50E+05 | 28.94            | 1.55E+07         | +                | -      | -   | 4.50E+01 | 22.35            | 2.01E+03         | +                | +      | -   | 2.50E+03 | 22.69 | 1.10E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 1           | Wed              | +                | +      | -   | 4.50E+05 | 28.94            | 1.55E+07         | +                | -      | -   | 4.50E+01 | 22.35            | 2.01E+03         | +                | +      | -   | 2.50E+03 | 22.69 | 1.10E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 1           | Thur             | +                | +      | -   | 4.50E+05 | 28.94            | 1.55E+07         | +                | -      | -   | 4.50E+01 | 22.35            | 2.01E+03         | +                | +      | -   | 2.50E+03 | 22.69 | 1.10E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 2           | Mon              | +                | +      | -   | 4.50E+05 | 29.14            | 1.54E+07         | +                | -      | -   | 2.50E+00 | 22.89            | 1.09E+02         | +                | +      | -   | 9.50E+02 | 23.73 | 4.00E+04 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 2           | Tue              | +                | +      | -   | 4.50E+05 | 28.98            | 1.55E+07         | +                | -      | -   | 4.50E+00 | 22.58            | 1.99E+02         | +                | +      | -   | 9.50E+03 | 23.36 | 4.07E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 2           | Wed              | +                | +      | -   | 9.50E+05 | 27.26            | 3.49E+07         | +                | -      | -   | 4.50E+01 | 22.89            | 1.97E+03         | +                | +      | -   | 4.50E+03 | 23.21 | 1.94E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 2           | Thur             | +                | +      | -   | 2.50E+05 | 25.53            | 9.79E+06         | +                | +      | -   | 4.50E+00 | 23.20            | 1.94E+02         | +                | +      | -   | 4.50E+03 | 23.06 | 1.95E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 3           | Mon              | +                | +      | -   | 9.50E+04 | 28.59            | 3.32E+06         | +                | +      | -   | 2.00E+00 | 27.87            | 7.18E+01         | +                | +      | -   | 2.50E+03 | 31.54 | 7.93E+04 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 3           | Tue              | +                | +      | -   | 4.50E+05 | 30.64            | 1.47E+07         | +                | +      | -   | 4.50E+00 | 28.51            | 1.58E+02         | +                | +      | -   | 9.50E+03 | 31.22 | 3.04E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 3           | Wed              | +                | +      | -   | 9.50E+05 | 25.35            | 3.75E+07         | +                | -      | -   | 4.50E+00 | 27.65            | 1.63E+02         | +                | +      | -   | 4.50E+03 | 30.89 | 1.46E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 3           | Thur             | +                | +      | -   | 2.50E+05 | 28.21            | 8.66E+06         | +                | +      | -   | 4.50E+00 | 30.65            | 1.47E+02         | +                | +      | -   | 4.50E+03 | 26.67 | 1.69E+05 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 4           | Mon              | +                | +      | -   | 9.50E+04 | 31.78            | 2.99E+06         | +                | +      | -   | 2.00E+00 | 29.74            | 6.72E+01         | +                | +      | -   | 2.50E+03 | 26.66 | 9.38E+04 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 4           | Tue              | +                | +      | -   | 2.50E+06 | 29.07            | 8.60E+07         | +                | +      | -   | 9.50E+00 | 29.66            | 3.20E+02         | +                | +      | -   | 1.50E+03 | 33.33 | 4.50E+04 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 4           | Wed              | +                | +      | -   | 4.50E+05 | 26.64            | 1.69E+07         | +                | +      | -   | 4.50E+01 | 27.57            | 1.63E+03         | +                | +      | -   | 2.50E+03 | 29.07 | 8.60E+04 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |
| Week 4           | Thur             | +                | +      | -   | 9.50E+04 | 25.36            | 3.75E+06         | +                | +      | -   | 2.50E+01 | 28.14            | 8.88E+02         | +                | +      | -   | 1.50E+03 | 31.2  | 4.81E+04 |
|                  |                  | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +                | +                | +                | +      | +   | +        | +     | +        |
|                  |                  | +                | +      | -   | 9.50E+05 | 28.84            | 3.29E+07         | +                | -      | -   | 1.50E+01 | 22.26            | 6.74E+02         | +                | +      | -   | 2.50E+03 | 22.8  | 1.10E+05 |

**Table C - 15. Total VFA, pH and un-ionized VFA concentrations – semi-continuously fed, staged acid digestion with total SRT of 11 days.**

| Sludge     | Day    | Reading |           |           |           |           |           |           |           |         |        | Total | pH    | Unassociated part of acids |      |      |      |        |        |      |      |      | Total | % total |
|------------|--------|---------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|---------|--------|-------|-------|----------------------------|------|------|------|--------|--------|------|------|------|-------|---------|
|            |        | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | mg/l    | mg/l   |       |       | mg/l                       | mg/l | mg/l | mg/l | mg/l   | mg/l   | mg/l | mg/l | mg/l |       |         |
| 1 Low-Meso | 0      | 3797.7  | 1064.5    | 137.6     | 654.1     | 180.0     | 241.4     | 26.0      | 17.4      | 6118.8  | 845.6  | 290.3 | 35.4  | 160.5                      | 40.8 | 61.6 | 6.9  | 4.6    | 1445.6 | 23.6 |      |      |       |         |
|            | 1      | 4116.7  | 1162.7    | 119.7     | 740.4     | 153.9     | 234.9     | 27.1      | 19.3      | 6581.7  | 917.0  | 317.1 | 30.8  | 181.6                      | 36.0 | 60.0 | 7.1  | 5.0    | 1413.8 | 23.6 |      |      |       |         |
|            | 2      | 4678.3  | 1179.8    | 144.5     | 843.7     | 187.4     | 260.8     | 29.9      | 21.2      | 7545.6  | 1086.2 | 321.7 | 37.2  | 207.0                      | 42.5 | 66.6 | 7.9  | 5.5    | 1774.5 | 23.6 |      |      |       |         |
|            | 3      | 4938.2  | 1169.0    | 78.1      | 727.0     | 116.5     | 229.1     | 26.1      | 19.9      | 7243.9  | 1099.5 | 302.4 | 20.1  | 178.4                      | 26.4 | 58.5 | 6.9  | 5.2    | 1697.4 | 23.4 |      |      |       |         |
|            | 4      | 4523.8  | 1065.3    | 114.8     | 684.6     | 160.7     | 236.1     | 26.6      | 20.1      | 6834.1  | 1007.2 | 290.5 | 29.6  | 168.0                      | 36.4 | 60.3 | 7.5  | 5.3    | 1604.8 | 23.5 |      |      |       |         |
|            | 5      | 4621.5  | 1175.5    | 116.3     | 698.9     | 154.9     | 244.5     | 31.0      | 20.4      | 7062.9  | 1029.0 | 320.6 | 29.9  | 171.5                      | 35.1 | 62.4 | 8.2  | 5.3    | 1661.9 | 23.5 |      |      |       |         |
|            | 6      | 3735.4  | 1063.3    | 118.0     | 626.2     | 160.4     | 225.7     | 32.1      | 18.9      | 5982.9  | 831.7  | 291.3 | 30.4  | 153.6                      | 36.4 | 57.6 | 7.9  | 4.9    | 1413.8 | 23.6 |      |      |       |         |
|            | 7      | 4166.0  | 1200.2    | 111.3     | 840.0     | 147.9     | 234.8     | 30.2      | 19.0      | 6544.0  | 926.3  | 327.3 | 28.6  | 157.0                      | 33.5 | 59.9 | 7.9  | 5.0    | 1545.7 | 23.6 |      |      |       |         |
|            | 8      | 4475.2  | 1355.1    | 107.3     | 651.2     | 140.0     | 226.1     | 29.5      | 18.7      | 7003.1  | 996.4  | 369.5 | 27.6  | 159.8                      | 31.7 | 57.7 | 7.8  | 4.9    | 1655.4 | 23.6 |      |      |       |         |
|            | 9      | 3161.3  | 889.2     | 106.0     | 406.9     | 93.9      | 168.8     | 23.3      | 15.4      | 4863.6  | 703.9  | 242.5 | 27.0  | 99.8                       | 21.3 | 43.0 | 6.1  | 4.0    | 1147.7 | 23.6 |      |      |       |         |
|            | 10     | 3718.2  | 1341.3    | 100.1     | 617.2     | 132.5     | 228.9     | 29.5      | 17.4      | 6185.0  | 827.9  | 365.8 | 25.8  | 151.4                      | 30.0 | 58.4 | 7.8  | 4.5    | 1471.6 | 23.8 |      |      |       |         |
|            | 11     | 3771.6  | 1420.5    | 119.5     | 671.4     | 151.2     | 238.8     | 30.7      | 17.8      | 6421.4  | 839.7  | 387.4 | 30.8  | 164.7                      | 34.3 | 60.9 | 8.1  | 4.7    | 1530.6 | 23.8 |      |      |       |         |
|            | 12     | 4226.5  | 1413.4    | 121.8     | 680.2     | 157.5     | 247.7     | 32.5      | 18.4      | 6897.9  | 941.0  | 385.5 | 31.4  | 166.9                      | 35.7 | 63.2 | 8.6  | 4.8    | 1637.0 | 23.7 |      |      |       |         |
|            | 13     | 3639.0  | 1203.6    | 119.9     | 535.9     | 105.7     | 214.6     | 28.3      | 16.5      | 5863.5  | 810.2  | 328.2 | 30.9  | 131.5                      | 24.0 | 54.8 | 7.4  | 4.3    | 1391.3 | 23.7 |      |      |       |         |
|            | 14     | 4582.3  | 1464.2    | 97.4      | 671.2     | 131.0     | 244.2     | 31.4      | 18.2      | 7240.4  | 1020.2 | 399.3 | 25.1  | 164.7                      | 29.7 | 62.5 | 8.3  | 4.8    | 1714.5 | 23.7 |      |      |       |         |
| 15         | 4045.2 | 1132.4  | 69.6      | 519.5     | 101.7     | 201.8     | 24.5      | 15.0      | 6109.6    | 900.7   | 308.8  | 17.9  | 127.4 | 23.0                       | 51.5 | 6.4  | 3.9  | 1439.8 | 23.6   |      |      |      |       |         |
| Average    |        | 4149.6  | 1202.8    | 111.3     | 648.0     | 142.5     | 229.9     | 28.7      | 18.4      | 6531.2  | 923.9  | 328.0 | 28.7  | 159.0                      | 32.3 | 58.7 | 7.5  | 4.8    | 1542.9 | 23.6 |      |      |       |         |
| Std. Dev.  |        | 496.9   | 186.0     | 19.0      | 99.8      | 27.2      | 21.9      | 2.6       | 1.7       | 672.4   | 116.4  | 43.0  | 4.9   | 24.4                       | 6.2  | 8.4  | 0.7  | 0.5    | 156.6  | 23.6 |      |      |       |         |
| 2 Low-meso | 0      | 4592.5  | 1347.7    | 207.3     | 782.3     | 250.9     | 292.3     | 20.9      | 23.1      | 7512.0  | 1078.5 | 315.3 | 48.7  | 183.7                      | 58.9 | 68.6 | 4.9  | 5.4    | 1764.1 | 23.5 |      |      |       |         |
|            | 1      | 1198.9  | 1677.8    | 156.1     | 1027.4    | 220.9     | 357.2     | 27.4      | 32.4      | 10698.1 | 1690.5 | 394.0 | 36.6  | 241.3                      | 51.9 | 83.9 | 6.4  | 7.6    | 2512.3 | 23.5 |      |      |       |         |
|            | 2      | 5448.1  | 1314.9    | 182.2     | 846.1     | 240.2     | 304.0     | 21.5      | 25.0      | 8381.9  | 1279.4 | 308.8 | 42.8  | 198.7                      | 56.4 | 71.4 | 5.0  | 5.9    | 1968.3 | 23.5 |      |      |       |         |
|            | 3      | 6102.5  | 1376.3    | 147.1     | 884.7     | 203.0     | 248.8     | 29.2      | 27.0      | 8212.4  | 1433.1 | 323.0 | 34.5  | 160.8                      | 48.1 | 57.4 | 6.1  | 6.3    | 2054.4 | 23.5 |      |      |       |         |
|            | 4      | 7167.1  | 1818.2    | 208.3     | 984.1     | 264.1     | 310.2     | 28.0      | 30.7      | 10608.8 | 1683.1 | 379.5 | 48.9  | 231.1                      | 62.0 | 72.8 | 6.6  | 7.2    | 2491.3 | 23.5 |      |      |       |         |
|            | 5      | 5341.0  | 1212.6    | 152.8     | 740.6     | 217.2     | 271.3     | 24.8      | 27.2      | 7987.3  | 1254.2 | 284.8 | 35.9  | 173.9                      | 51.0 | 63.7 | 5.8  | 6.4    | 1875.7 | 23.5 |      |      |       |         |
|            | 6      | 6154.1  | 1295.3    | 139.6     | 719.0     | 187.6     | 233.9     | 23.1      | 26.3      | 8778.8  | 1445.2 | 304.2 | 32.8  | 168.8                      | 44.1 | 54.9 | 5.4  | 6.2    | 2061.6 | 23.5 |      |      |       |         |
|            | 7      | 6978.0  | 1420.3    | 152.4     | 762.2     | 199.9     | 282.9     | 27.2      | 28.8      | 9830.8  | 1638.9 | 333.5 | 35.8  | 179.5                      | 46.9 | 61.7 | 6.4  | 6.8    | 2308.6 | 23.5 |      |      |       |         |
|            | 8      | 6102.5  | 1376.3    | 147.1     | 884.7     | 203.0     | 248.8     | 29.2      | 27.0      | 8212.4  | 1433.1 | 323.0 | 34.5  | 160.8                      | 48.1 | 57.4 | 6.1  | 6.3    | 2054.4 | 23.5 |      |      |       |         |
|            | 9      | 8329.2  | 1515.5    | 137.5     | 703.4     | 179.9     | 260.5     | 28.0      | 27.1      | 9181.2  | 1486.3 | 355.9 | 32.3  | 165.2                      | 42.3 | 61.2 | 6.6  | 6.4    | 2156.0 | 23.5 |      |      |       |         |
|            | 10     | 6128.1  | 1403.8    | 130.2     | 603.0     | 168.6     | 237.5     | 27.3      | 25.0      | 8723.4  | 1439.1 | 329.6 | 30.6  | 141.6                      | 39.6 | 55.8 | 6.4  | 5.9    | 2048.5 | 23.5 |      |      |       |         |
|            | 11     | 6818.0  | 1641.1    | 112.0     | 683.6     | 160.4     | 247.9     | 27.3      | 25.1      | 9715.4  | 1601.1 | 385.4 | 26.3  | 160.5                      | 37.7 | 56.2 | 6.4  | 5.9    | 2281.5 | 23.5 |      |      |       |         |
|            | 12     | 7588.5  | 1803.3    | 98.9      | 881.1     | 161.0     | 290.2     | 29.6      | 24.2      | 10636.8 | 1782.0 | 423.5 | 23.2  | 159.9                      | 37.8 | 58.8 | 6.9  | 5.7    | 2497.9 | 23.5 |      |      |       |         |
|            | 13     | 6489.4  | 1607.7    | 93.8      | 811.9     | 130.6     | 223.5     | 27.1      | 23.3      | 9221.3  | 1510.9 | 377.5 | 22.0  | 143.7                      | 32.8 | 53.7 | 6.4  | 4.7    | 2153.2 | 23.5 |      |      |       |         |
|            | 14     | 5147.3  | 1430.8    | 114.6     | 603.9     | 149.5     | 247.4     | 25.5      | 21.7      | 7740.7  | 1208.7 | 336.0 | 26.9  | 141.8                      | 35.1 | 58.1 | 6.0  | 5.1    | 1817.8 | 23.5 |      |      |       |         |
| 15         | 5307.2 | 1568.6  | 120.7     | 650.3     | 157.8     | 247.3     | 27.4      | 22.5      | 8101.8    | 1246.3  | 368.4  | 28.3  | 152.7 | 37.0                       | 58.1 | 6.4  | 5.3  | 1902.6 | 23.5   |      |      |      |       |         |
| Average    |        | 6169.6  | 1476.7    | 144.4     | 748.1     | 195.0     | 269.0     | 25.9      | 26.1      | 9057.8  | 1448.8 | 347.2 | 33.9  | 175.9                      | 45.8 | 63.2 | 6.1  | 6.1    | 2127.0 | 23.5 |      |      |       |         |
| Std. Dev.  |        | 649.4   | 161.3     | 31.9      | 130.2     | 37.9      | 33.6      | 3.5       | 3.0       | 1018.4  | 199.5  | 37.9  | 8.0   | 30.6                       | 8.9  | 8.7  | 0.8  | 0.7    | 239.7  | 23.5 |      |      |       |         |
| 1 Meso     | 0      | 5453.9  | 2188.0    | 147.7     | 726.2     | 221.3     | 349.5     | 34.0      | 20.1      | 9159.3  | 1280.7 | 513.3 | 34.7  | 170.3                      | 52.0 | 68.5 | 6.8  | 4.5    | 2150.9 | 23.5 |      |      |       |         |
|            | 1      | 4441.6  | 1977.2    | 232.2     | 747.1     | 306.9     | 415.8     | 32.7      | 20.6      | 8174.0  | 1043.0 | 464.3 | 64.5  | 175.4                      | 72.1 | 97.6 | 7.7  | 4.8    | 1919.5 | 23.5 |      |      |       |         |
|            | 2      | 3917.8  | 1503.1    | 208.0     | 684.7     | 272.0     | 284.8     | 37.3      | 38.0      | 6947.6  | 920.0  | 353.5 | 48.8  | 160.8                      | 63.9 | 66.8 | 8.8  | 8.9    | 1631.5 | 23.5 |      |      |       |         |
|            | 3      | 4760.1  | 1872.2    | 246.1     | 842.6     | 308.9     | 309.7     | 36.3      | 32.6      | 8170.4  | 1352.6 | 439.6 | 58.3  | 197.9                      | 72.5 | 72.7 | 6.5  | 7.8    | 2209.9 | 23.5 |      |      |       |         |
|            | 4      | 5191.8  | 1862.0    | 191.8     | 762.7     | 237.8     | 328.0     | 32.8      | 29.2      | 8308.1  | 1333.0 | 503.6 | 56.0  | 181.3                      | 75.3 | 80.3 | 6.0  | 4.7    | 2153.2 | 23.5 |      |      |       |         |
|            | 5      | 5353.3  | 1585.2    | 127.1     | 656.9     | 202.0     | 281.5     | 26.8      | 24.7      | 8039.5  | 1205.9 | 372.3 | 29.8  | 154.3                      | 47.4 | 66.1 | 6.3  | 5.8    | 1887.9 | 23.5 |      |      |       |         |
|            | 6      | 4162.2  | 1441.8    | 131.6     | 547.3     | 191.4     | 248.8     | 26.2      | 20.9      | 6770.3  | 977.4  | 338.6 | 30.9  | 128.5                      | 45.0 | 58.4 | 6.2  | 4.9    | 1589.9 | 23.5 |      |      |       |         |
|            | 7      | 4246.9  | 1525.3    | 144.6     | 581.5     | 204.5     | 271.3     | 28.7      | 21.0      | 7023.8  | 997.3  | 358.2 | 33.9  | 136.6                      | 48.0 | 63.7 | 6.7  | 4.9    | 1649.4 | 23.5 |      |      |       |         |
|            | 8      | 4912.8  | 1774.8    | 188.4     | 671.2     | 237.3     | 294.1     | 32.7      | 22.4      | 8133.6  | 1153.6 | 416.8 | 44.3  | 157.6                      | 55.7 | 69.1 | 7.7  | 5.3    | 1910.0 | 23.5 |      |      |       |         |
|            | 9      | 4246.0  | 1437.4    | 145.1     | 463.3     | 216.6     | 225.1     | 26.7      | 19.8      | 6769.3  | 1015.9 | 377.5 | 22.0  | 143.7                      | 32.8 | 53.7 | 6.4  | 4.7    | 1594.7 | 23.5 |      |      |       |         |
|            | 10     | 5252.4  | 2144.5    | 167.7     | 816.6     | 226.7     | 319.3     | 32.6      | 23.7      | 10256.5 | 1532.4 | 504.6 | 39.4  | 191.8                      | 53.2 | 75.0 | 7.7  | 5.6    | 2408.5 | 23.5 |      |      |       |         |
|            | 11     | 5435.8  | 1738.0    | 90.9      | 574.4     | 144.9     | 241.4     | 27.6      | 19.4      | 6272.4  | 1276.5 | 408.1 | 21.4  | 134.9                      | 34.0 | 56.7 | 6.5  | 4.6    | 1942.6 | 23.5 |      |      |       |         |
|            | 12     | 2914.8  | 1032.4    | 281.0     | 363.7     | 128.3     | 175.5     | 19.0      | 14.1      | 4928.8  | 685.5  | 242.4 | 66.0  | 85.4                       | 30.1 | 41.2 | 4.5  | 3.3    | 1157.4 | 23.5 |      |      |       |         |
|            | 13     | 2653.4  | 1219.7    | 208.2     | 412.8     | 139.9     | 213.8     | 22.8      | 25.0      | 4895.6  | 621.1  | 266.4 | 48.9  | 96.9                       | 32.9 | 50.2 | 5.4  | 5.9    | 1145.6 | 23.5 |      |      |       |         |
|            | 14     | 4411.2  | 1673.6    | 135.5     | 501.1     | 180.5     | 250.0     | 26.9      | 17.3      | 7215.1  | 1035.9 | 393.0 | 31.8  | 119.3                      | 43.8 | 60.1 | 6.3  | 4.1    | 1594.3 | 23.5 |      |      |       |         |
| Average    |        | 4764.3  | 1697.1    | 190.7     | 635.4     | 221.6     | 289.1     | 29.6      | 23.0      | 7780.8  | 1164.7 | 398.5 | 42.4  | 149.2                      | 52.0 | 67.9 | 7.0  | 5.4    | 1827.2 | 23.5 |      |      |       |         |
| Std. Dev.  |        | 1037.6  | 334.4     | 52.4      | 147.3     | 59.3      | 65.2      | 4.8       | 6.0       | 1540.0  | 243.7  | 78.5  | 12.3  | 34.6                       | 13.9 | 15.3 | 1.7  | 1.4    | 361.6  | 23.5 |      |      |       |         |
| 2 Meso     | 0      | 3750.3  | 1177.6    | 160.2     | 594.0     | 240.5     | 255.6     | 23.0      | 18.4      | 6227.8  | 821.9  | 257.5 | 35.0  | 129.9                      | 52.6 | 55.9 | 5.0  | 4.0    | 1361.9 | 21.9 |      |      |       |         |
|            | 1      | 4318.9  | 1409.8    | 165.6     | 697.6     | 289.3     | 314.5     | 23.9      | 24.0      | 7263.6  | 944.5  | 308.3 | 40.8  | 152.6                      | 63.3 | 68.8 | 5.2  | 5.3    | 1585.  |      |      |      |       |         |



**Table C - 17. Total VFA, pH and un-ionized VFA concentrations – three stage acid/gas digestion experiments – raw sludge.**

| Sludge   | Date       | Reading |           |           |           |           |           |           |           |        | pH     | Undissociated part of acids |           |           |           |           |           |           |           |       | Total | % total |
|----------|------------|---------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|--------|--------|-----------------------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-------|-------|---------|
|          |            | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | Total  |        | Acetic                      | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | Total |       |         |
|          |            | mg/l    | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l   |        | mg/l                        | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l  |       |         |
| Raw      | 25/05/2005 | 187.0   | 837.6     | 81.9      | 244.7     | 130.4     | 88.9      | 42.6      | 0.0       | 3302.1 | 65.4   | 37.8                        | 3.4       | 9.6       | 4.6       | 3.6       | 1.9       | 0.0       | 126.2     | 3.8   |       |         |
|          | 27/05/2005 | 220.2   | 988.4     | 110.6     | 312.4     | 166.4     | 95.9      | 53.7      | 0.0       | 3909.4 | 76.6   | 43.7                        | 4.6       | 12.3      | 5.9       | 4.0       | 2.3       | 0.0       | 149.4     | 3.8   |       |         |
|          | 30/05/2005 | 238.1   | 1023.9    | 139.2     | 360.6     | 194.3     | 107.7     | 61.3      | 12.0      | 4262.9 | 82.9   | 46.1                        | 5.9       | 14.2      | 6.6       | 4.5       | 2.6       | 0.5       | 163.1     | 3.8   |       |         |
|          | 01/06/2005 | 3587.2  | 1429.8    | 247.9     | 558.6     | 207.1     | 137.9     | 91.3      | 14.8      | 6364.9 | 124.8  | 64.5                        | 10.4      | 22.0      | 10.6      | 5.7       | 3.1       | 0.6       | 242.5     | 3.8   |       |         |
|          | 03/06/2005 | 2599.0  | 1076.7    | 197.5     | 482.3     | 221.7     | 129.3     | 71.9      | 12.1      | 4790.4 | 90.5   | 48.5                        | 8.3       | 19.0      | 7.9       | 5.3       | 3.0       | 0.5       | 183.1     | 3.8   |       |         |
|          | 06/06/2005 | 1753.5  | 806.1     | 151.9     | 398.6     | 168.2     | 124.2     | 54.5      | 0.0       | 3457.1 | 61.0   | 36.3                        | 6.4       | 15.7      | 6.0       | 5.1       | 2.3       | 0.0       | 132.9     | 3.8   |       |         |
|          | 08/06/2005 | 2381.7  | 934.7     | 191.8     | 469.3     | 219.6     | 134.4     | 56.3      | 12.6      | 4400.5 | 82.9   | 42.1                        | 8.0       | 18.5      | 7.8       | 5.6       | 2.4       | 0.5       | 167.8     | 3.8   |       |         |
|          | 10/06/2005 | 2140.5  | 833.0     | 179.3     | 439.4     | 208.0     | 131.1     | 40.5      | 13.7      | 3991.5 | 74.5   | 37.6                        | 7.5       | 17.3      | 7.4       | 5.4       | 2.0       | 0.6       | 152.2     | 3.8   |       |         |
|          | 13/06/2005 | 2334.0  | 821.6     | 173.8     | 444.6     | 199.3     | 126.1     | 42.3      | 0.0       | 4141.7 | 81.2   | 37.0                        | 7.3       | 17.5      | 7.1       | 5.2       | 1.8       | 0.0       | 151.1     | 3.8   |       |         |
|          | 15/06/2005 | 1764.3  | 653.8     | 146.8     | 355.5     | 172.7     | 111.1     | 33.1      | 0.0       | 3237.3 | 61.4   | 29.5                        | 6.1       | 14.0      | 6.1       | 4.6       | 1.4       | 0.0       | 123.2     | 3.8   |       |         |
|          | 17/06/2005 | 1687.3  | 684.4     | 166.1     | 363.1     | 191.0     | 121.0     | 33.5      | 0.0       | 3246.4 | 58.7   | 30.9                        | 5.9       | 14.3      | 6.8       | 5.0       | 1.4       | 0.0       | 124.0     | 3.8   |       |         |
|          | 20/06/2005 | 1490.0  | 614.0     | 124.1     | 307.3     | 151.6     | 116.0     | 23.8      | 0.0       | 2826.8 | 51.9   | 27.7                        | 6.2       | 12.1      | 5.4       | 4.8       | 1.0       | 0.0       | 108.0     | 3.8   |       |         |
|          | 22/06/2005 | 1438.1  | 633.2     | 141.2     | 323.7     | 160.8     | 130.4     | 24.2      | 0.0       | 2881.6 | 50.1   | 28.5                        | 5.9       | 12.7      | 6.4       | 5.8       | 1.0       | 0.0       | 110.5     | 3.8   |       |         |
|          | 24/06/2005 | 1748.2  | 586.6     | 142.1     | 281.9     | 177.0     | 119.5     | 22.0      | 0.0       | 3075.4 | 60.8   | 26.4                        | 5.9       | 11.1      | 6.3       | 4.9       | 0.9       | 0.0       | 116.4     | 3.8   |       |         |
|          | 27/06/2005 | 1458.5  | 525.5     | 158.1     | 281.0     | 192.8     | 121.7     | 23.7      | 0.0       | 2759.3 | 50.7   | 23.7                        | 6.6       | 11.0      | 6.9       | 5.0       | 1.0       | 0.0       | 105.0     | 3.8   |       |         |
|          | 29/06/2005 | 1105.5  | 401.7     | 98.2      | 187.5     | 112.1     | 80.7      | 15.7      | 0.0       | 2001.3 | 38.5   | 18.1                        | 4.1       | 7.4       | 4.0       | 3.3       | 0.7       | 0.0       | 76.1      | 3.8   |       |         |
|          | 01/07/2005 | 478.4   | 243.0     | 44.6      | 103.7     | 63.2      | 31.0      | 5.0       | 0.0       | 968.9  | 16.6   | 11.0                        | 1.9       | 4.1       | 2.3       | 1.3       | 0.2       | 0.0       | 37.2      | 3.8   |       |         |
|          | 04/07/2005 | 1527.4  | 619.7     | 147.9     | 294.6     | 199.7     | 127.3     | 21.1      | 0.0       | 2928.7 | 53.2   | 27.9                        | 6.2       | 11.6      | 6.8       | 5.3       | 0.9       | 0.0       | 111.8     | 3.8   |       |         |
|          | 07/07/2005 | 256.1   | 116.2     | 15.1      | 37.3      | 37.8      | 9.8       | 0.0       | 0.0       | 472.3  | 8.9    | 5.2                         | 0.6       | 1.5       | 1.3       | 0.4       | 0.0       | 0.0       | 18.0      | 3.8   |       |         |
|          | 08/07/2005 | 394.9   | 172.7     | 20.4      | 51.6      | 30.5      | 12.3      | 0.0       | 0.0       | 682.5  | 13.7   | 7.8                         | 0.9       | 2.0       | 1.1       | 0.5       | 0.0       | 0.0       | 26.0      | 3.8   |       |         |
|          | 11/07/2005 | 315.0   | 151.7     | 19.0      | 46.9      | 31.6      | 12.4      | 0.0       | 0.0       | 576.6  | 11.0   | 6.8                         | 0.8       | 1.8       | 1.1       | 0.5       | 0.0       | 0.0       | 22.1      | 3.8   |       |         |
|          | 13/07/2005 | 687.7   | 316.3     | 42.2      | 104.0     | 60.2      | 24.4      | 0.0       | 2.9       | 1237.7 | 23.9   | 14.3                        | 1.8       | 4.1       | 2.1       | 1.0       | 0.0       | 0.1       | 47.3      | 3.8   |       |         |
|          | 15/07/2005 | 240.0   | 103.6     | 75.1      | 61.7      | 50.0      | 22.7      | 0.0       | 2.7       | 616.2  | 8.4    | 6.7                         | 1.1       | 3.6       | 1.8       | 0.9       | 0.0       | 0.0       | 24.2      | 3.8   |       |         |
|          | 18/07/2005 | 581.5   | 233.8     | 22.0      | 77.2      | 39.1      | 17.8      | 0.0       | 0.0       | 971.4  | 20.2   | 10.5                        | 0.9       | 3.0       | 1.4       | 0.7       | 0.0       | 0.0       | 38.9      | 3.8   |       |         |
|          | 20/07/2005 | 659.4   | 262.2     | 28.7      | 102.6     | 48.0      | 22.7      | 0.0       | 0.0       | 1123.5 | 22.9   | 11.8                        | 1.2       | 4.0       | 1.7       | 0.9       | 0.0       | 0.0       | 42.6      | 3.8   |       |         |
|          | Average    |         | 1406.8    | 623.8     | 110.4     | 281.0     | 136.9     | 96.9      | 32.7      | 7.8    | 2896.3 | 51.1                        | 26.9      | 4.8       | 10.6      | 5.0       | 3.6       | 1.2       | 0.1       | 103.2 |       |         |
| Std. Dev |            | 78.9    | 305.3     | 67.3      | 149.7     | 67.7      | 52.0      | 24.5      | 70.8      | 1399.2 | 30.9   | 15.7                        | 2.9       | 6.3       | 2.7       | 2.1       | 1.1       | 0.2       | 60.9      |       |       |         |

**Table C - 18. Total VFA, pH and un-ionized VFA concentrations – three stage acid/gas digestion experiments – raw, thickened sludge.**

| Sludge        | Date       | Reading |           |           |           |           |           |           |           |         | pH      | Undissociated part of acids |           |           |           |           |           |           |           |       | Total | % total |
|---------------|------------|---------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|---------|---------|-----------------------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-------|-------|---------|
|               |            | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | Total   |         | Acetic                      | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | Total |       |         |
|               |            | mg/l    | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l    |         | mg/l                        | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l  |       |         |
| Raw-Thickened | 25/05/2005 | 1110.84 | 619.61    | 77.09     | 260.62    | 121.48    | 74.49     | 34.74     | 0.00      | 2298.87 | 27.04   | 19.60                       | 2.26      | 7.18      | 3.02      | 2.16      | 1.05      | 0.00      | 62.30     | 2.7   |       |         |
|               | 27/05/2005 | 2328.88 | 993.50    | 126.48    | 469.72    | 201.44    | 120.86    | 63.28     | 0.00      | 4322.16 | 58.63   | 31.42                       | 3.71      | 13.48     | 5.01      | 3.50      | 1.91      | 0.00      | 115.87    | 2.7   |       |         |
|               | 30/05/2005 | 686.10  | 478.38    | 65.14     | 203.33    | 107.62    | 60.21     | 49.69     | 0.00      | 1569.77 | 16.70   | 15.13                       | 2.50      | 5.69      | 2.69      | 1.75      | 1.47      | 0.00      | 45.86     | 2.7   |       |         |
|               | 01/06/2005 | 1734.70 | 883.81    | 157.42    | 422.44    | 178.39    | 99.64     | 70.10     | 0.00      | 3546.49 | 42.22   | 27.95                       | 4.61      | 11.63     | 4.44      | 2.89      | 2.12      | 0.00      | 95.86     | 2.7   |       |         |
|               | 03/06/2005 | 2372.81 | 1065.09   | 215.23    | 553.77    | 229.63    | 132.44    | 83.42     | 0.00      | 4652.38 | 57.75   | 33.68                       | 6.31      | 15.25     | 6.72      | 3.84      | 2.52      | 0.00      | 125.06    | 2.7   |       |         |
|               | 06/06/2005 | 2117.61 | 954.55    | 226.47    | 580.32    | 254.46    | 141.14    | 73.18     | 0.00      | 4347.83 | 51.54   | 30.19                       | 6.64      | 15.98     | 5.33      | 4.09      | 2.21      | 0.00      | 116.98    | 2.7   |       |         |
|               | 08/06/2005 | 1416.13 | 716.73    | 194.75    | 528.85    | 225.31    | 137.97    | 59.26     | 0.00      | 3278.99 | 34.47   | 22.67                       | 5.73      | 14.56     | 5.61      | 4.00      | 1.70      | 0.00      | 86.90     | 2.7   |       |         |
|               | 10/06/2005 | 1691.02 | 698.89    | 0.00      | 433.33    | 203.53    | 138.35    | 56.21     | 0.00      | 3131.33 | 38.97   | 22.10                       | 0.00      | 11.93     | 5.07      | 4.01      | 1.70      | 0.00      | 83.77     | 2.7   |       |         |
|               | 13/06/2005 | 1697.00 | 684.10    | 154.34    | 377.61    | 174.27    | 122.95    | 42.34     | 0.00      | 3252.99 | 41.30   | 21.64                       | 4.52      | 10.40     | 4.34      | 3.56      | 1.28      | 0.00      | 87.04     | 2.7   |       |         |
|               | 15/06/2005 | 916.09  | 587.82    | 149.55    | 279.63    | 160.07    | 83.65     | 36.34     | 0.00      | 2193.34 | 22.30   | 18.59                       | 4.38      | 7.70      | 3.98      | 1.85      | 1.10      | 0.00      | 59.90     | 2.7   |       |         |
|               | 17/06/2005 | 398.33  | 505.07    | 132.92    | 341.03    | 166.57    | 109.67    | 37.08     | 0.00      | 2290.66 | 24.30   | 15.97                       | 3.90      | 9.39      | 4.15      | 3.18      | 1.12      | 0.00      | 62.00     | 2.7   |       |         |
|               | 20/06/2005 | 1489.93 | 638.17    | 135.59    | 347.99    | 167.95    | 134.67    | 26.04     | 0.00      | 2937.43 | 36.26   | 20.12                       | 3.97      | 9.58      | 4.16      | 3.90      | 0.79      | 0.00      | 78.79     | 2.7   |       |         |
|               | 22/06/2005 | 1360.87 | 586.65    | 119.12    | 314.66    | 150.26    | 125.24    | 21.76     | 0.00      | 2678.57 | 33.12   | 18.55                       | 3.49      | 8.67      | 3.74      | 3.63      | 0.66      | 0.00      | 71.86     | 2.7   |       |         |
|               | 24/06/2005 | 1247.46 | 516.95    | 109.22    | 307.21    | 146.56    | 125.51    | 20.97     | 0.00      | 2473.88 | 30.36   | 16.35                       | 3.20      | 8.46      | 3.65      | 3.64      | 0.63      | 0.00      | 66.29     | 2.7   |       |         |
|               | 27/06/2005 | 1133.49 | 520.32    | 118.11    | 322.22    | 155.49    | 133.89    | 20.64     | 0.00      | 2404.16 | 27.59   | 16.46                       | 3.46      | 8.87      | 3.87      | 3.88      | 0.62      | 0.00      | 64.75     | 2.7   |       |         |
|               | 29/06/2005 | 902.06  | 406.69    | 100.16    | 230.53    | 118.22    | 93.84     | 16.77     | 0.00      | 1868.06 | 21.95   | 12.86                       | 2.94      | 6.35      | 2.94      | 2.71      | 0.51      | 0.00      | 50.26     | 2.7   |       |         |
|               | 01/07/2005 | 521.01  | 238.98    | 46.43     | 117.36    | 63.45     | 33.99     | 5.10      | 0.00      | 1026.26 | 12.88   | 7.56                        | 1.36      | 3.23      | 1.50      | 0.88      | 0.15      | 0.00      | 27.55     | 2.7   |       |         |
|               | 04/07/2005 | 1052.81 | 461.08    | 114.77    | 267.60    | 136.55    | 109.13    | 17.16     | 0.00      | 2150.08 | 25.82   | 14.58                       | 3.38      | 7.37      | 3.40      | 3.16      | 0.52      | 0.00      | 58.02     | 2.7   |       |         |
|               | 07/07/2005 | 126.89  | 311.95    | 61.03     | 167.64    | 79.37     | 46.08     | 5.64      | 0.00      | 1398.39 | 17.69   | 9.87                        | 1.79      | 4.62      | 1.98      | 1.34      | 0.17      | 0.00      | 37.44     | 2.7   |       |         |
|               | 08/07/2005 | 604.03  | 284.32    | 38.20     | 136.93    | 60.50     | 27.64     | 4.53      | 2.90      | 1157.05 | 14.70   | 8.99                        | 1.06      | 3.77      | 1.51      | 0.80      | 0.14      | 0.00      | 31.06     | 2.7   |       |         |
|               | 11/07/2005 | 369.22  | 247.62    | 45.01     | 171.60    | 65.40     | 35.48     | 5.46      | 3.08      | 943.07  | 8.99    | 7.83                        | 1.32      | 4.73      | 1.63      | 1.03      | 0.16      | 0.00      | 25.78     | 2.7   |       |         |
|               | 13/07/2005 | 1222.75 | 574.03    | 87.79     | 361.01    | 177.76    | 64.50     | 10.82     | 3.90      | 2482.55 | 29.76   | 16.15                       | 2.57      | 9.84      | 2.93      | 1.81      | 0.33      | 0.00      | 65.67     | 2.7   |       |         |
|               | 15/07/2005 | 607.79  | 342.00    | 47.4      | 103.67    | 58.43     | 34.3      | 4.4       | 0.00      | 1417.68 | 19.66   | 10.97                       | 1.31      | 5.02      | 1.64      | 0.99      | 0.16      | 0.00      | 39.52     | 2.7   |       |         |
|               | 18/07/2005 | 310.72  | 193.65    | 38.10     | 138.44    | 52.31     | 31.18     | 3.49      | 2.58      | 788.47  | 7.56    | 6.12                        | 1.06      | 3.81      | 1.30      | 0.90      | 0.11      | 0.00      | 20.95     | 2.7   |       |         |
|               | 20/07/2005 | 885.21  | 375.60    | 58.40     | 236.50    | 76.68     | 45.58     | 4.17      | 3.10      | 1685.23 | 21.55   | 11.88                       | 1.71      | 6.51      | 1.91      | 1.32      | 0.13      | 0.00      | 45.10     | 2.7   |       |         |
|               | Average    |         | 1281.09   | 616.70    | 112.75    | 353.16    | 147.97    | 97.17     | 32.66     | 3.22    | 2644.73 | 28.90                       | 17.48     | 3.12      | 8.62      | 3.47      | 2.62      | 0.93      | 0.02      | 65.16 |       |         |
| Std. Dev      |            | 664.68  | 297.36    | 63.84     | 198.00    | 74.83     | 49.42     | 23.66     | 6.74      | 1340.72 | 14.00   | 7.72                        | 1.74      | 3.76      | 1.49      | 1.23      | 0.77      | 0.04      | 29.47     |       |       |         |

**Table C - 19. Total VFA, pH and un-ionized VFA concentrations – three stage acid/gas digestion experiments – effluent from 1<sup>st</sup> of three staged mesophilic acid digesters.**

| Sludge            | Date       | Reading |           |           |           |           |           |           |           |       | pH | Undissociated part of acids |           |           |           |           |           |           |           |       | Total | % total |
|-------------------|------------|---------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-------|----|-----------------------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-------|-------|---------|
|                   |            | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | Total |    | Acetic                      | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic | Total |       |         |
|                   |            | mg/l    | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l  |    | mg/l                        | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l  |       |         |
| 1 Acid Mesophilic | 25/05/2005 |         |           |           |           |           |           |           |           |       |    |                             |           |           |           |           |           |           |           |       |       |         |









**Table C - 28. Results of Ascaris suum viability tests – all treatments.**

| Sample        | Eggs                      | Retention time | Larvated | Not larvated | Percent Viable |
|---------------|---------------------------|----------------|----------|--------------|----------------|
| Control       | Free floating             | 9              | 135      | 24           | 84.9           |
|               | Chamber                   | 9              | 192      | 31           | 86.1           |
|               | Chamber                   | 9              | 197      | 46           | 81.1           |
|               | Average chamber           | 9              |          |              | 83.6           |
|               | Free floating             | 12             | 184      | 18           | 91.1           |
|               | Free floating             | 17             | 193      | 22           | 89.8           |
|               | Free floating             | 22             | 134      | 32           | 80.7           |
|               | Chamber                   | 22             | 221      | 113          | 66.2           |
|               | Chamber                   | 22             | 111      | 80           | 58.1           |
|               | Chamber                   | 22             | 122      | 132          | 48.0           |
|               | Average chamber           | 22             |          |              | 57.4           |
| Acid meso     | Free floating - Reactor 1 | 3              | 119      | 29           | 80.4           |
|               | Free floating - Reactor 2 | 6              | 152      | 28           | 84.4           |
|               | Free floating - Reactor 3 | 9              | 81       | 26           | 75.7           |
|               | Chamber                   | 9              | 14       | 386          | 3.5            |
|               | Chamber                   | 9              | 8        | 393          | 2.0            |
|               | Chamber                   | 9              | 7        | 309          | 2.2            |
|               | Average chamber           | 9              |          |              | 2.6            |
| Gas meso      | Free floating             | 12             | 65       | 31           | 67.7           |
|               | Free floating             | 17             | 33       | 55           | 37.5           |
|               | Free floating             | 22             | 2        | 70           | 2.8            |
|               | Chamber                   | 22             | 0        | 1000         | 0.0            |
|               | Chamber                   | 22             | 0        | 1000         | 0.0            |
|               | Chamber                   | 22             | 0        | 1000         | 0.0            |
|               | Average chamber           |                |          |              | 0.0            |
| Acid low-meso | Chamber                   | 12             | 226      | 44           | 83.7           |
|               | Chamber                   | 12             | 235      | 45           | 83.9           |
|               | Chamber                   | 12             | 306      | 66           | 82.3           |
|               | Average chamber           | 12             |          |              | 83.3           |
| Gas meso      | Free floating             | 12             |          |              |                |
|               | Free floating             | 17             |          |              |                |
|               | Free floating             | 22             |          |              |                |
|               | Chamber                   | 22             | 384      | 150          | 71.9           |
|               | Chamber                   | 22             | 316      | 210          | 60.1           |
|               | Chamber                   | 22             | 373      | 138          | 73.0           |
|               | Average chamber           |                |          |              | 68.3           |

**Table C - 29. Fecal coliforms counts – pressure tests – mesophilic digesters.**

| Date       | Day | Fecal coliforms                   |          |                  |                                   |                  |          |                                |          |                  |                                |                  |          |
|------------|-----|-----------------------------------|----------|------------------|-----------------------------------|------------------|----------|--------------------------------|----------|------------------|--------------------------------|------------------|----------|
|            |     | Batch Mesophilic # 1, no pressure |          |                  | Batch Mesophilic # 2, no pressure |                  |          | Batch Mesophilic # 1, pressure |          |                  | Batch Mesophilic # 2, pressure |                  |          |
|            |     | Dilution                          | Result   | Dilution         | Result                            | Dilution         | Result   | Dilution                       | Result   |                  |                                |                  |          |
| 01/08/2005 | 0   | 10 <sup>-5</sup>                  | MPN/ml   | 10 <sup>-5</sup> | MPN/ml                            | 10 <sup>-5</sup> | MPN/ml   | 10 <sup>-5</sup>               | MPN/ml   | 10 <sup>-5</sup> | MPN/ml                         | 10 <sup>-6</sup> | MPN/ml   |
|            |     | 10 <sup>-6</sup>                  |          | 10 <sup>-6</sup> |                                   | 10 <sup>-6</sup> |          | 10 <sup>-6</sup>               |          | 10 <sup>-6</sup> |                                | 10 <sup>-7</sup> |          |
|            |     | 10 <sup>-7</sup>                  |          | 10 <sup>-7</sup> |                                   | 10 <sup>-7</sup> |          | 10 <sup>-7</sup>               |          | 10 <sup>-7</sup> |                                | 10 <sup>-7</sup> |          |
| 02/08/2005 | 1   | +                                 | 7.50E+05 | +                | 4.50E+05                          | +                | 4.50E+06 | +                              | 7.50E+05 | +                | 4.50E+06                       | +                | 4.50E+06 |
|            |     | +                                 |          | +                |                                   | +                |          | +                              |          | +                |                                | +                |          |
|            |     | +                                 |          | +                |                                   | +                |          | +                              |          | +                |                                | +                |          |
| 03/08/2005 | 2   | 10 <sup>-4</sup>                  | MPN/ml   | 10 <sup>-4</sup> | MPN/ml                            | 10 <sup>-4</sup> | MPN/ml   | 10 <sup>-4</sup>               | MPN/ml   | 10 <sup>-5</sup> | MPN/ml                         | 10 <sup>-6</sup> | MPN/ml   |
|            |     | 10 <sup>-5</sup>                  |          | 10 <sup>-5</sup> |                                   | 10 <sup>-5</sup> |          | 10 <sup>-5</sup>               |          | 10 <sup>-5</sup> |                                | 10 <sup>-6</sup> |          |
|            |     | 10 <sup>-6</sup>                  |          | 10 <sup>-6</sup> |                                   | 10 <sup>-6</sup> |          | 10 <sup>-6</sup>               |          | 10 <sup>-6</sup> |                                | 10 <sup>-7</sup> |          |
| 04/08/2005 | 3   | +                                 | 4.50E+04 | +                | 4.50E+04                          | +                | 4.50E+04 | +                              | 2.50E+04 | +                | 2.50E+04                       | +                | 2.50E+04 |
|            |     | +                                 |          | +                |                                   | +                |          | +                              |          | +                |                                | +                |          |
|            |     | +                                 |          | +                |                                   | +                |          | +                              |          | +                |                                | +                |          |
| 05/08/2005 | 4   | 10 <sup>-2</sup>                  | MPN/ml   | 10 <sup>-2</sup> | MPN/ml                            | 10 <sup>-2</sup> | MPN/ml   | 10 <sup>-2</sup>               | MPN/ml   | 10 <sup>-2</sup> | MPN/ml                         | 10 <sup>-3</sup> | MPN/ml   |
|            |     | 10 <sup>-3</sup>                  |          | 10 <sup>-3</sup> |                                   | 10 <sup>-3</sup> |          | 10 <sup>-3</sup>               |          | 10 <sup>-3</sup> |                                | 10 <sup>-4</sup> |          |
|            |     | 10 <sup>-4</sup>                  |          | 10 <sup>-4</sup> |                                   | 10 <sup>-4</sup> |          | 10 <sup>-4</sup>               |          | 10 <sup>-4</sup> |                                | 10 <sup>-4</sup> |          |
| 06/08/2005 | 5   | +                                 | 2.50E+02 | +                | 3.00E+03                          | +                | 2.50E+02 | +                              | 2.50E+02 | +                | 4.50E+03                       | +                | 4.50E+03 |
|            |     | +                                 |          | +                |                                   | +                |          | +                              |          | +                |                                | +                |          |
|            |     | +                                 |          | +                |                                   | +                |          | +                              |          | +                |                                | +                |          |
| 07/08/2005 | 6   | 10 <sup>-0</sup>                  | MPN/ml   | 10 <sup>-0</sup> | MPN/ml                            | 10 <sup>-0</sup> | MPN/ml   | 10 <sup>-0</sup>               | MPN/ml   | 10 <sup>-0</sup> | MPN/ml                         | 10 <sup>-1</sup> | MPN/ml   |
|            |     | 10 <sup>-1</sup>                  |          | 10 <sup>-1</sup> |                                   | 10 <sup>-1</sup> |          | 10 <sup>-1</sup>               |          | 10 <sup>-1</sup> |                                | 10 <sup>-2</sup> |          |
|            |     | 10 <sup>-2</sup>                  |          | 10 <sup>-2</sup> |                                   | 10 <sup>-2</sup> |          | 10 <sup>-2</sup>               |          | 10 <sup>-2</sup> |                                | 10 <sup>-2</sup> |          |
| 08/08/2005 | 7   | +                                 | 9.00E-01 | +                | 3.00E+00                          | +                | 6.50E+00 | +                              | 9.00E-01 | +                | 2.50E+00                       | +                | 2.50E+00 |
|            |     | +                                 |          | +                |                                   | +                |          | +                              |          | +                |                                | +                |          |
|            |     | +                                 |          | +                |                                   | +                |          | +                              |          | +                |                                | +                |          |

**Table C - 30. Fecal coliforms counts – pressure tests – low-mesophilic digesters.**

| Date       | Day | Fecal coliforms                       |          |                  |                                       |                  |          |                                    |          |                  |                                    |                  |          |
|------------|-----|---------------------------------------|----------|------------------|---------------------------------------|------------------|----------|------------------------------------|----------|------------------|------------------------------------|------------------|----------|
|            |     | Batch Low-Mesophilic # 1, no pressure |          |                  | Batch Low-Mesophilic # 2, no pressure |                  |          | Batch Low-Mesophilic # 1, pressure |          |                  | Batch Low-Mesophilic # 2, pressure |                  |          |
|            |     | Dilution                              | Result   | Dilution         | Result                                | Dilution         | Result   | Dilution                           | Result   |                  |                                    |                  |          |
| 01/08/2005 | 0   | 10 <sup>-5</sup>                      | MPN/ml   | 10 <sup>-5</sup> | MPN/ml                                | 10 <sup>-5</sup> | MPN/ml   | 10 <sup>-5</sup>                   | MPN/ml   | 10 <sup>-5</sup> | MPN/ml                             | 10 <sup>-6</sup> | MPN/ml   |
|            |     | 10 <sup>-6</sup>                      |          | 10 <sup>-6</sup> |                                       | 10 <sup>-6</sup> |          | 10 <sup>-6</sup>                   |          | 10 <sup>-6</sup> |                                    | 10 <sup>-7</sup> |          |
|            |     | 10 <sup>-7</sup>                      |          | 10 <sup>-7</sup> |                                       | 10 <sup>-7</sup> |          | 10 <sup>-7</sup>                   |          | 10 <sup>-7</sup> |                                    | 10 <sup>-7</sup> |          |
| 02/08/2005 | 1   | +                                     | 4.50E+05 | +                | 9.50E+05                              | +                | 4.50E+05 | +                                  | 4.50E+05 | +                | 1.50E+06                           | +                | 1.50E+06 |
|            |     | +                                     |          | +                |                                       | +                |          | +                                  |          | +                |                                    | +                |          |
|            |     | +                                     |          | +                |                                       | +                |          | +                                  |          | +                |                                    | +                |          |
| 03/08/2005 | 2   | 10 <sup>-4</sup>                      | MPN/ml   | 10 <sup>-4</sup> | MPN/ml                                | 10 <sup>-4</sup> | MPN/ml   | 10 <sup>-4</sup>                   | MPN/ml   | 10 <sup>-4</sup> | MPN/ml                             | 10 <sup>-5</sup> | MPN/ml   |
|            |     | 10 <sup>-5</sup>                      |          | 10 <sup>-5</sup> |                                       | 10 <sup>-5</sup> |          | 10 <sup>-5</sup>                   |          | 10 <sup>-5</sup> |                                    | 10 <sup>-6</sup> |          |
|            |     | 10 <sup>-6</sup>                      |          | 10 <sup>-6</sup> |                                       | 10 <sup>-6</sup> |          | 10 <sup>-6</sup>                   |          | 10 <sup>-6</sup> |                                    | 10 <sup>-7</sup> |          |
| 04/08/2005 | 3   | +                                     | 9.50E+04 | +                | 2.50E+05                              | +                | 9.50E+04 | +                                  | 9.50E+04 | +                | 1.15E+04                           | +                | 1.15E+04 |
|            |     | +                                     |          | +                |                                       | +                |          | +                                  |          | +                |                                    | +                |          |
|            |     | +                                     |          | +                |                                       | +                |          | +                                  |          | +                |                                    | +                |          |
| 05/08/2005 | 4   | 10 <sup>-3</sup>                      | MPN/ml   | 10 <sup>-3</sup> | MPN/ml                                | 10 <sup>-3</sup> | MPN/ml   | 10 <sup>-3</sup>                   | MPN/ml   | 10 <sup>-3</sup> | MPN/ml                             | 10 <sup>-4</sup> | MPN/ml   |
|            |     | 10 <sup>-4</sup>                      |          | 10 <sup>-4</sup> |                                       | 10 <sup>-4</sup> |          | 10 <sup>-4</sup>                   |          | 10 <sup>-4</sup> |                                    | 10 <sup>-5</sup> |          |
|            |     | 10 <sup>-5</sup>                      |          | 10 <sup>-5</sup> |                                       | 10 <sup>-5</sup> |          | 10 <sup>-5</sup>                   |          | 10 <sup>-5</sup> |                                    | 10 <sup>-5</sup> |          |
| 06/08/2005 | 5   | +                                     | 9.50E+03 | +                | 9.50E+03                              | +                | 1.50E+03 | +                                  | 9.50E+03 | +                | 1.50E+03                           | +                | 1.50E+03 |
|            |     | +                                     |          | +                |                                       | +                |          | +                                  |          | +                |                                    | +                |          |
|            |     | +                                     |          | +                |                                       | +                |          | +                                  |          | +                |                                    | +                |          |
| 07/08/2005 | 6   | 10 <sup>-3</sup>                      | MPN/ml   | 10 <sup>-3</sup> | MPN/ml                                | 10 <sup>-3</sup> | MPN/ml   | 10 <sup>-3</sup>                   | MPN/ml   | 10 <sup>-3</sup> | MPN/ml                             | 10 <sup>-4</sup> | MPN/ml   |
|            |     | 10 <sup>-4</sup>                      |          | 10 <sup>-4</sup> |                                       | 10 <sup>-4</sup> |          | 10 <sup>-4</sup>                   |          | 10 <sup>-4</sup> |                                    | 10 <sup>-5</sup> |          |
|            |     | 10 <sup>-5</sup>                      |          | 10 <sup>-5</sup> |                                       | 10 <sup>-5</sup> |          | 10 <sup>-5</sup>                   |          | 10 <sup>-5</sup> |                                    | 10 <sup>-5</sup> |          |
| 08/08/2005 | 7   | +                                     | 2.50E+02 | +                | 2.50E+02                              | +                | 4.50E+01 | +                                  | 2.50E+02 | +                | 2.50E+02                           | +                | 2.50E+02 |
|            |     | +                                     |          | +                |                                       | +                |          | +                                  |          | +                |                                    | +                |          |
|            |     | +                                     |          | +                |                                       | +                |          | +                                  |          | +                |                                    | +                |          |

**Table C - 31. Pressure generated in pressurized digesters – mesophilic and low-mesophilic reactors.**

| Units | Pressure     |              |                    |                                  |                  |                  |                        |                                  |
|-------|--------------|--------------|--------------------|----------------------------------|------------------|------------------|------------------------|----------------------------------|
|       | Mesophilic 1 | Mesophilic 2 | Mesophilic Average | Fraction of atmospheric pressure | Low-Mesophilic 1 | Low-Mesophilic 2 | Low-Mesophilic Average | Fraction of atmospheric pressure |
|       | PSI          |              |                    | ATM                              | PSI              |                  |                        | ATM                              |
| Day 0 | 0.0          | 0.0          | 0.0                | 1.00                             | 0.0              | 0.0              | 0.0                    | 1.00                             |
| Day 1 | 7.0          | 7.5          | 7.3                | 1.49                             | <5               | <5               | 2.6                    | 1.18                             |
| Day 2 | 15.0         | 14.5         | 14.8               | 2.01                             | 5.0              | 5.5              | 5.3                    | 1.36                             |
| Day 3 | 21.0         | 20.5         | 20.8               | 2.41                             | 9.5              | 10.5             | 10.0                   | 1.68                             |
| Day 4 | 24.0         | 23.5         | 23.8               | 2.62                             | 12.5             | 12.0             | 12.3                   | 1.83                             |
| Day 5 | 24.5         | 25.0         | 24.8               | 2.69                             | 14.0             | 13.5             | 13.8                   | 1.94                             |
| Day 6 | 26.0         | 25.0         | 25.5               | 2.74                             | 14.5             | 14.5             | 14.5                   | 1.99                             |
| Day 7 | 25.5         | 25.5         | 25.5               | 2.74                             | 15.0             | 14.5             | 14.8                   | 2.01                             |



**Table C - 33. Estimation of total VFA concentrations, pH, and concentrations of un-ionized VFA – pressure tests – pressurized digesters (the effect of pressure included).**

| Batch mesophilic acid digesters on average     | Fraction of atmospheric ATM | Day    | Reading |           |           |           |           |           |           |           | Total<br>mg/l | pH     | Undissociated part of acids |           |           |           |           |           |           |           | Total<br>mg/l |
|--|-----------------------------|--------|---------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|---------------|--------|-----------------------------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|---------------|
|  |                             |        | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic |               |        | Acetic                      | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic |               |
|  |                             |        | mg/l    | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      |               |        | mg/l                        | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      |               |
| 1.00   | 0                           | 768.9  | 453.6   | 52.0      | 248.7     | 76.8      | 53.4      | 9.8       | 3.1       | 1666.3    | 6.25          | 23.9   | 18.3                        | 1.9       | 8.8       | 2.4       | 2.0       | 0.4       | 0.1       | 57.9      |               |
| 1.49   | 1                           | 2097.1 | 1232.7  | 183.3     | 948.9     | 288.6     | 277.4     | 19.5      | 6.9       | 5054.4    | 5.86          | 153.3  | 115.4                       | 16.0      | 78.0      | 21.6      | 23.9      | 1.7       | 0.6       | 410.5     |               |
| 2.01   | 2                           | 4076.4 | 2514.7  | 287.9     | 1802.3    | 420.9     | 574.5     | 28.1      | 8.6       | 9511.3    | 5.35          | 829.0  | 629.9                       | 88.0      | 359.9     | 87.2      | 134.4     | 6.3       | 2.1       | 2116.7    |               |
| 2.41   | 3                           | 6539.2 | 4446.6  | 365.8     | 2289.5    | 541.8     | 833.4     | 31.1      | 12.2      | 15070.6   | 5.06          | 2170.2 | 1754.3                      | 145.0     | 826.5     | 192.8     | 311.1     | 11.9      | 4.6       | 5406.6    |               |
| 2.62   | 4                           | 8131.7 | 4893.9  | 435.6     | 2614.4    | 627.2     | 989.1     | 34.6      | 12.9      | 15739.4   | 5.15          | 1780.7 | 1707.3                      | 144.3     | 829.2     | 185.1     | 325.2     | 11.7      | 4.3       | 4987.9    |               |
| 2.69   | 5                           | 8717.3 | 5074.9  | 476.4     | 2881.7    | 669.0     | 1025.9    | 32.2      | 12.6      | 16690.0   | 5.14          | 1982.8 | 1797.1                      | 160.3     | 864.0     | 200.7     | 342.5     | 11.0      | 4.3       | 5362.7    |               |
| 2.74   | 6                           | 7470.4 | 5632.4  | 519.2     | 2930.2    | 727.5     | 1118.4    | 35.1      | 14.5      | 18447.7   | 5.15          | 2169.5 | 1965.0                      | 172.0     | 929.4     | 214.8     | 367.7     | 11.9      | 4.9       | 5835.0    |               |
| 2.74   | 7                           | 6938.0 | 5869.3  | 527.2     | 3004.8    | 771.5     | 1147.9    | 35.7      | 14.6      | 20309.2   | 5.15          | 2585.7 | 2047.6                      | 174.7     | 953.1     | 227.7     | 377.4     | 12.1      | 5.0       | 6383.2    |               |
|  |                             |        |         |           |           |           |           |           |           |           |               |        |                             |           |           |           |           |           |           |           |               |
| Batch low-mesophilic acid digesters on average | Fraction of atmospheric ATM | Day    | Reading |           |           |           |           |           |           |           | Total<br>mg/l | pH     | Undissociated part of acids |           |           |           |           |           |           |           | Total<br>mg/l |
|  |                             |        | Acetic  | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic |               |        | Acetic                      | Propionic | I-butyric | N-butyric | I-valeric | N-valeric | I-caproic | N-caproic |               |
|  |                             |        | mg/l    | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      |               |        | mg/l                        | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      | mg/l      |               |
| 1.00   | 0                           | 768.9  | 453.6   | 52.0      | 248.7     | 76.8      | 53.4      | 9.8       | 3.1       | 1666.3    | 6.3           | 23.9   | 18.3                        | 1.9       | 8.8       | 2.4       | 2.0       | 0.4       | 0.1       | 57.9      |               |
| 1.18   | 1                           | 1544.7 | 766.8   | 114.7     | 470.3     | 158.0     | 150.0     | 23.4      | 3.2       | 3231.0    | 5.8           | 164.4  | 115.5                       | 16.2      | 62.7      | 19.2      | 20.9      | 3.4       | 0.5       | 422.8     |               |
| 1.36   | 2                           | 2435.0 | 1177.5  | 138.2     | 638.9     | 180.7     | 193.8     | 20.6      | 1.8       | 4788.6    | 5.4           | 488.5  | 279.9                       | 30.9      | 136.0     | 35.4      | 43.0      | 4.7       | 0.4       | 998.9     |               |
| 1.68   | 3                           | 3891.1 | 2013.6  | 187.0     | 831.6     | 221.3     | 260.6     | 27.6      | 4.9       | 7337.7    | 5.2           | 1045.6 | 888.8                       | 80.5      | 288.6     | 63.7      | 83.7      | 9.1       | 1.8       | 2239.9    |               |
| 1.83   | 4                           | 4073.2 | 2435.0  | 223.9     | 1150.4    | 269.4     | 319.4     | 29.1      | 1.9       | 8502.2    | 5.1           | 1364.0 | 967.4                       | 84.8      | 418.4     | 91.6      | 120.1     | 11.2      | 0.7       | 3058.2    |               |
| 1.99   | 5                           | 5359.6 | 2959.6  | 255.8     | 1312.2    | 298.1     | 367.9     | 33.3      | 6.3       | 10592.9   | 5.1           | 1794.9 | 1175.8                      | 96.9      | 477.2     | 101.4     | 138.3     | 12.8      | 2.4       | 3799.7    |               |
| 1.99   | 6                           | 4120.4 | 3076.9  | 332.2     | 1702.4    | 452.0     | 531.0     | 43.2      | 7.3       | 10265.3   | 5.1           | 1348.3 | 1197.1                      | 123.1     | 605.5     | 150.2     | 195.4     | 16.3      | 2.7       | 3638.7    |               |
| 2.01   | 7                           | 4283.2 | 3013.9  | 289.3     | 1504.3    | 365.7     | 442.5     | 33.1      | 6.3       | 9938.1    | 5.1           | 1347.9 | 1131.6                      | 103.4     | 515.4     | 118.9     | 156.9     | 12.0      | 2.3       | 3386.4    |               |

## **Appendix D - Data for section 7.0.**



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in feed sludge and multi-phase digestion systems at 5.7% TS in feed sludge. All systems  
at loading rate of 100 tons TS/day and equal volumes. .... 255

**Table D - 1. Gas production – control digester, gas digester of three stage acid mesophilic followed by gas mesophilic system and gas digester of three stage acid low-mesophilic followed by gas mesophilic system.**

| Day       | Date       | Gas production   |   |   |
|-----------|------------|------------------|---|---|
|           |            | Control Digester | Gas digester<br>3-stage acid-meso<br>followed by gas meso | Gas digester<br>3-stage acid low-meso<br>followed by gas meso |
|           |            | std L/d          | std L/d   | std L/d   |
| 1         | 25/05/2005 | 0.71             | 0.70  | 1.06  |
| 2         | 26/05/2005 | 0.21             | 0.99  | 1.53  |
| 3         | 27/05/2005 | 1.09             | 1.18  | 1.68  |
| 4         | 28/05/2005 | 0.73             | 1.14  | 1.25  |
| 5         | 29/05/2005 | 0.68             |   | 1.43  |
| 6         | 30/05/2005 | 0.87             |   | 1.19  |
| 7         | 31/05/2005 | 1.12             |   | 1.20  |
| 8         | 01/06/2005 | 0.10             |   | 1.25  |
| 9         | 02/06/2005 | 0.29             | 1.04  | 1.01  |
| 10        | 03/06/2005 | 1.45             | 1.09  | 1.26  |
| 11        | 04/06/2005 | 0.20             |   | 0.17  |
| 12        | 05/06/2005 |                  | 1.27  | 0.79  |
| 13        | 06/06/2005 | 1.42             | 1.01  | 0.56  |
| 14        | 07/06/2005 | 0.59             | 0.87  | 0.60  |
| 15        | 08/06/2005 |                  | 1.20  | 0.70  |
| 16        | 09/06/2005 |                  | 1.12  | 0.45  |
| 17        | 10/06/2005 | 0.53             | 1.18  | 0.31  |
| 18        | 11/06/2005 | 0.49             | 1.08  | 0.32  |
| 19        | 12/06/2005 | 0.40             | 1.26  | 0.58  |
| 20        | 13/06/2005 |                  | 1.14  | 0.61  |
| 21        | 14/06/2005 |                  | 1.27  | 0.91  |
| 22        | 15/06/2005 |                  | 1.26  | 0.90  |
| 23        | 16/06/2005 | 0.86             | 1.40  | 1.13  |
| 24        | 17/06/2005 | 0.71             | 1.27  | 1.48  |
| 25        | 18/06/2005 | 0.47             | 1.29  | 1.20  |
| 26        | 19/06/2005 | 0.53             | 1.25  | 1.03  |
| 27        | 20/06/2005 | 0.54             | 1.23  | 1.27  |
| 28        | 21/06/2005 | 0.62             | 1.07  | 1.08  |
| 29        | 22/06/2005 |                  | 1.64  | 1.99  |
| 30        | 23/06/2005 | 0.80             |   | 1.23  |
| 31        | 24/06/2005 | 0.66             | 1.67  | 1.83  |
| 32        | 25/06/2005 | 0.47             | 1.00  | 1.37  |
| 33        | 26/06/2005 | 0.76             | 1.69  | 1.82  |
| 34        | 27/06/2005 | 0.50             |   |   |
| 35        | 28/06/2005 | 0.97             |   | 0.95  |
| 36        | 29/06/2005 | 0.19             |   | 1.04  |
| 37        | 30/06/2005 | 0.64             |   | 1.20  |
| 38        | 01/07/2005 | 0.92             | 1.10  | 1.47  |
| 39        | 02/07/2005 | 0.43             | 1.37  | 1.37  |
| 40        | 03/07/2005 | 0.42             | 0.53  | 1.38  |
| 41        | 04/07/2005 | 0.48             | 1.03  | 1.08  |
| 42        | 05/07/2005 | 0.52             | 0.70  | 1.15  |
| 43        | 06/07/2005 | 0.47             | 1.23  | 1.22  |
| 44        | 07/07/2005 | 0.16             | 0.64  | 0.89  |
| 45        | 08/07/2005 | 0.26             | 1.18  | 1.06  |
| Average   |            | 0.66             | 1.16  | 1.07  |
| Std. Dev. |            | 0.28             | 0.26  | 0.38  |

**Table D - 2. Gas production from mesophilic acid digesters.**

| Day       | Date       | Gas production               |                              |                              |
|-----------|------------|------------------------------|------------------------------|------------------------------|
|           |            | 1st mesophilic acid digester | 2nd mesophilic acid digester | 3rd mesophilic acid digester |
|           |            | std L/d                      | std L/d                      | std L/d                      |
| 1         | 25/05/2005 | 0.28                         | 0.03                         | 0.08                         |
| 2         | 26/05/2005 | 0.66                         | 0.27                         | 0.24                         |
| 3         | 27/05/2005 | 0.85                         | 0.42                         | 0.34                         |
| 4         | 28/05/2005 | 0.41                         | 0.07                         | 0.10                         |
| 5         | 29/05/2005 | 0.42                         | 0.14                         | 0.10                         |
| 6         | 30/05/2005 | 0.54                         | 0.10                         | 0.11                         |
| 7         | 31/05/2005 | 0.68                         | 0.15                         | 0.11                         |
| 8         | 01/06/2005 | 0.50                         | 0.23                         | 0.17                         |
| 9         | 02/06/2005 |                              | 0.04                         | 0.02                         |
| 10        | 03/06/2005 | 0.59                         | 0.11                         | 0.01                         |
| 11        | 04/06/2005 | 0.24                         |                              |                              |
| 12        | 05/06/2005 | 0.57                         | 0.37                         | 0.23                         |
| 13        | 06/06/2005 | 0.18                         |                              |                              |
| 14        | 07/06/2005 | 0.55                         | 0.14                         | 0.02                         |
| 15        | 08/06/2005 | 0.80                         | 0.29                         | 0.13                         |
| 16        | 09/06/2005 |                              | 0.13                         | 0.08                         |
| 17        | 10/06/2005 | 0.43                         | 0.11                         | 0.07                         |
| 18        | 11/06/2005 | 0.48                         | 0.14                         | 0.05                         |
| 19        | 12/06/2005 | 0.64                         | 0.19                         | 0.12                         |
| 20        | 13/06/2005 | 0.49                         | 0.09                         | 0.02                         |
| 21        | 14/06/2005 | 0.54                         | 0.16                         | 0.03                         |
| 22        | 15/06/2005 | 0.44                         | 0.12                         | 0.03                         |
| 23        | 16/06/2005 | 0.46                         | 0.05                         |                              |
| 24        | 17/06/2005 | 0.44                         | 0.15                         | 0.02                         |
| 25        | 18/06/2005 | 0.50                         | 0.19                         | 0.11                         |
| 26        | 19/06/2005 | 0.29                         | 0.05                         | 0.03                         |
| 27        | 20/06/2005 | 0.42                         | 0.06                         | 0.02                         |
| 28        | 21/06/2005 | 0.18                         | 0.01                         |                              |
| 29        | 22/06/2005 | 0.60                         | 0.32                         |                              |
| 30        | 23/06/2005 | 0.35                         | 0.08                         | 0.07                         |
| 31        | 24/06/2005 | 0.38                         | 0.14                         | 0.10                         |
| 32        | 25/06/2005 | 0.16                         |                              |                              |
| 33        | 26/06/2005 | 0.52                         | 0.30                         | 0.22                         |
| 34        | 27/06/2005 | 0.32                         | 0.05                         | 0.02                         |
| 35        | 28/06/2005 | 0.28                         | 0.07                         | 0.04                         |
| 36        | 29/06/2005 | 0.30                         | 0.13                         | 0.07                         |
| 37        | 30/06/2005 | 0.46                         | 0.16                         | 0.11                         |
| 38        | 01/07/2005 | 0.45                         | 0.13                         | 0.08                         |
| 39        | 02/07/2005 | 0.44                         | 0.14                         | 0.41                         |
| 40        | 03/07/2005 | 0.39                         | 0.03                         |                              |
| 41        | 04/07/2005 | 0.31                         | 0.01                         | 0.30                         |
| 42        | 05/07/2005 | 0.30                         |                              | 0.23                         |
| 43        | 06/07/2005 | 0.47                         | 0.23                         |                              |
| 44        | 07/07/2005 | 0.32                         | 0.19                         | 0.06                         |
| 45        | 08/07/2005 | 0.39                         | 0.18                         | 0.41                         |
| Average   |            | <b>0.40</b>                  | <b>0.13</b>                  | <b>0.11</b>                  |
| Std. Dev. |            | 0.16                         | 0.09                         | 0.26                         |

**Table D - 3. Gas production from low-mesophilic acid digesters.**

| Day       | Date       | Gas production                   |                                  |                                  |
|-----------|------------|----------------------------------|----------------------------------|----------------------------------|
|           |            | 1st low-mesophilic acid digester | 2nd low-mesophilic acid digester | 3rd low-mesophilic acid digester |
|           |            | std L/d                          | std L/d                          | std L/d                          |
| 1         | 25/05/2005 | 0.12                             | 0.19                             | 0.06                             |
| 2         | 26/05/2005 | 0.29                             | 0.10                             | 0.00                             |
| 3         | 27/05/2005 | 0.25                             |                                  | 0.08                             |
| 4         | 28/05/2005 | 0.24                             | 0.13                             | 0.01                             |
| 5         | 29/05/2005 | 0.23                             | 0.18                             | 0.05                             |
| 6         | 30/05/2005 | 0.19                             |                                  | 0.03                             |
| 7         | 31/05/2005 | 0.23                             | 0.00                             | 0.05                             |
| 8         | 01/06/2005 | 0.01                             |                                  | 0.10                             |
| 9         | 02/06/2005 | 0.36                             | 0.20                             | 0.07                             |
| 10        | 03/06/2005 | 0.22                             | 0.17                             | 0.04                             |
| 11        | 04/06/2005 | 0.18                             | 0.14                             | 0.02                             |
| 12        | 05/06/2005 | 0.17                             | 0.13                             | 0.04                             |
| 13        | 06/06/2005 | 0.09                             | 0.07                             |                                  |
| 14        | 07/06/2005 | 0.16                             | 0.10                             | 0.04                             |
| 15        | 08/06/2005 | 0.22                             | 0.12                             | 0.06                             |
| 16        | 09/06/2005 | 0.26                             | 0.13                             | 0.02                             |
| 17        | 10/06/2005 | 0.23                             | 0.14                             | 0.03                             |
| 18        | 11/06/2005 | 0.27                             | 0.17                             | 0.08                             |
| 19        | 12/06/2005 | 0.25                             | 0.16                             | 0.06                             |
| 20        | 13/06/2005 | 0.24                             | 0.17                             | 0.05                             |
| 21        | 14/06/2005 |                                  |                                  |                                  |
| 22        | 15/06/2005 | 0.19                             | 0.10                             | 0.05                             |
| 23        | 16/06/2005 | 0.00                             | 0.20                             | 0.06                             |
| 24        | 17/06/2005 | 0.34                             | 0.17                             | 0.04                             |
| 25        | 18/06/2005 | 0.25                             | 0.19                             | 0.08                             |
| 26        | 19/06/2005 | 0.24                             | 0.17                             | 0.05                             |
| 27        | 20/06/2005 | 0.24                             | 0.16                             | 0.04                             |
| 28        | 21/06/2005 | 0.22                             | 0.12                             |                                  |
| 29        | 22/06/2005 | 0.34                             | 0.15                             | 0.02                             |
| 30        | 23/06/2005 | 0.27                             | 0.14                             | 0.07                             |
| 31        | 24/06/2005 | 0.12                             | 0.11                             |                                  |
| 32        | 25/06/2005 |                                  |                                  |                                  |
| 33        | 26/06/2005 | 0.10                             | 0.05                             | 0.00                             |
| 34        | 27/06/2005 | 0.10                             | 0.03                             | 0.05                             |
| 35        | 28/06/2005 | 0.24                             | 0.08                             | 0.00                             |
| 36        | 29/06/2005 | 0.30                             | 0.10                             | 0.02                             |
| 37        | 30/06/2005 | 0.19                             | 0.09                             | 0.04                             |
| 38        | 01/07/2005 | 0.07                             | 0.02                             |                                  |
| 39        | 02/07/2005 | 0.23                             | 0.08                             | 0.03                             |
| 40        | 03/07/2005 | 0.13                             | 0.06                             | 0.00                             |
| 41        | 04/07/2005 | 0.08                             | 0.08                             | 0.04                             |
| 42        | 05/07/2005 | 0.13                             | 0.09                             | 0.04                             |
| 43        | 06/07/2005 | 0.15                             | 0.10                             | 0.03                             |
| 44        | 07/07/2005 | 0.22                             | 0.13                             | 0.06                             |
| 45        | 08/07/2005 | 0.23                             | 0.09                             | 0.01                             |
| Average   |            | <b>0.20</b>                      | <b>0.12</b>                      | <b>0.04</b>                      |
| Std. Dev. |            | 0.09                             | 0.05                             | 0.04                             |

**Table D - 4. Gas composition – all digesters.**

| Reactor type    | Gas Composition |          | Date            | Reactor type    | Gas Composition |                 | Date       | Reactor type    | Gas Composition |          | Date       |
|-----------------|-----------------|----------|-----------------|-----------------|-----------------|-----------------|------------|-----------------|-----------------|----------|------------|
|                 | CH4<br>%        | CO2<br>% |                 |                 | CH4<br>%        | CO2<br>%        |            |                 | CH4<br>%        | CO2<br>% |            |
| Control         | 64.48           | 35.52    | 08/05/2005      | Control         | 62.90           | 37.10           | 11/05/2005 | Control         | 63.30           | 36.70    | 17/05/2005 |
| Gas Meso        | 63.29           | 36.71    |                 | Gas Meso        | 63.76           | 36.24           |            | Gas Meso        | 64.92           | 35.08    |            |
| Gas Low-Meso    | 63.86           | 36.14    |                 | Gas Low-Meso    | 64.80           | 35.20           |            | Gas Low-Meso    | 64.33           | 35.67    |            |
| 1 Acid Meso     | 23.36           | 76.64    |                 | 1 Acid Meso     | 19.23           | 80.77           |            | 1 Acid Meso     | 19.56           | 80.44    |            |
| 2 Acid Meso     | 21.19           | 78.81    |                 | 2 Acid Meso     | 20.49           | 79.51           |            | 2 Acid Meso     | 23.10           | 76.90    |            |
| 3 Acid Meso     | 48.91           | 51.09    |                 | 3 Acid Meso     | 48.74           | 51.26           |            | 3 Acid Meso     | 50.66           | 49.34    |            |
| 1 Acid Low-Meso | 34.92           | 65.08    |                 | 1 Acid Low-Meso | 27.34           | 72.66           |            | 1 Acid Low-Meso | 32.13           | 67.87    |            |
| 2 Acid Low-Meso | 23.99           | 76.01    |                 | 2 Acid Low-Meso | 23.15           | 76.85           |            | 2 Acid Low-Meso | 25.90           | 74.10    |            |
| 3 Acid Low-Meso | 16.83           | 83.17    |                 | 3 Acid Low-Meso | 13.32           | 86.68           |            | 3 Acid Low-Meso | 8.31            | 91.69    |            |
| Control         | 62.06           | 37.94    |                 | Control         | 63.02           | 36.98           |            | Control         | 62.24           | 37.76    |            |
| Gas Meso        | 63.03           | 36.97    |                 | Gas Meso        | 68.27           | 31.73           |            | Gas Meso        | 69.62           | 30.38    |            |
| Gas Low-Meso    | 62.32           | 37.68    |                 | Gas Low-Meso    | 67.04           | 32.96           |            | Gas Low-Meso    | 64.06           | 35.94    |            |
| 1 Acid Meso     | 18.49           | 81.51    | 1 Acid Meso     | 22.95           | 77.05           | 1 Acid Meso     | 32.65      | 67.35           |                 |          |            |
| 2 Acid Meso     | 23.68           | 76.32    | 2 Acid Meso     | 25.46           | 74.54           | 2 Acid Meso     | 37.05      | 62.95           |                 |          |            |
| 3 Acid Meso     | 49.22           | 50.78    | 3 Acid Meso     | 44.73           | 55.27           | 3 Acid Meso     | 47.21      | 52.79           |                 |          |            |
| 1 Acid Low-Meso | 28.60           | 71.40    | 1 Acid Low-Meso | 12.38           | 87.62           | 1 Acid Low-Meso | 38.18      | 61.82           |                 |          |            |
| 2 Acid Low-Meso | 26.34           | 73.66    | 2 Acid Low-Meso | 28.53           | 71.47           | 2 Acid Low-Meso | 29.77      | 70.23           |                 |          |            |
| 3 Acid Low-Meso | 17.96           | 82.04    | 3 Acid Low-Meso | 22.81           | 77.19           | 3 Acid Low-Meso | 23.66      | 76.34           |                 |          |            |
| Control         | 61.66           | 38.34    | Control         | 63.97           | 36.03           | Control         | 65.44      | 34.56           |                 |          |            |
| Gas Meso        | 66.78           | 33.22    | Gas Meso        | 67.25           | 32.75           | Gas Meso        | 65.01      | 34.99           |                 |          |            |
| Gas Low-Meso    | 62.83           | 37.17    | Gas Low-Meso    | 54.47           | 45.53           | Gas Low-Meso    | 60.82      | 39.18           |                 |          |            |
| 1 Acid Meso     | 24.44           | 75.56    | 1 Acid Meso     | 20.57           | 79.43           | 1 Acid Meso     | 21.66      | 78.34           |                 |          |            |
| 2 Acid Meso     | 25.29           | 74.71    | 2 Acid Meso     | 24.35           | 75.65           | 2 Acid Meso     | 29.72      | 70.28           |                 |          |            |
| 3 Acid Meso     | 48.29           | 51.71    | 3 Acid Meso     | 37.18           | 62.82           | 3 Acid Meso     | 51.24      | 48.76           |                 |          |            |
| 1 Acid Low-Meso | 24.86           | 75.14    | 1 Acid Low-Meso | 27.56           | 72.44           | 1 Acid Low-Meso | 27.57      | 72.43           |                 |          |            |
| 2 Acid Low-Meso | 28.42           | 71.58    | 2 Acid Low-Meso | 25.75           | 74.25           | 2 Acid Low-Meso | 28.06      | 71.94           |                 |          |            |
| 3 Acid Low-Meso | 26.67           | 73.33    | 3 Acid Low-Meso | 26.94           | 73.06           | 3 Acid Low-Meso | 29.37      | 70.63           |                 |          |            |
| Control         | 67.06           | 32.94    | Control         | 65.87           | 34.13           | Control         | 61.45      | 38.55           |                 |          |            |
| Gas Meso        | 67.10           | 32.90    | Gas Meso        | 58.56           | 41.44           | Gas Meso        | 67.75      | 32.25           |                 |          |            |
| Gas Low-Meso    | 67.31           | 32.69    | Gas Low-Meso    | 75.46           | 24.54           | Gas Low-Meso    | 71.76      | 28.24           |                 |          |            |
| 1 Acid Meso     | 23.88           | 76.12    | 1 Acid Meso     | 29.98           | 70.02           | 1 Acid Meso     | 32.73      | 67.27           |                 |          |            |
| 2 Acid Meso     | 30.89           | 69.11    | 2 Acid Meso     | 29.53           | 70.47           | 2 Acid Meso     | 28.37      | 71.63           |                 |          |            |
| 3 Acid Meso     | 50.85           | 49.15    | 3 Acid Meso     | 47.83           | 52.17           | 3 Acid Meso     | 42.96      | 57.04           |                 |          |            |
| 1 Acid Low-Meso | 29.59           | 70.41    | 1 Acid Low-Meso | 32.20           | 67.80           | 1 Acid Low-Meso | 32.80      | 67.20           |                 |          |            |
| 2 Acid Low-Meso | 23.69           | 76.31    | 2 Acid Low-Meso | 27.19           | 72.81           | 2 Acid Low-Meso | 35.55      | 64.45           |                 |          |            |
| 3 Acid Low-Meso | 26.44           | 73.56    | 3 Acid Low-Meso | 30.01           | 69.99           | 3 Acid Low-Meso | 35.77      | 64.23           |                 |          |            |
| Control         | 62.65           | 37.35    | Control         | 63.72           | 36.28           | Control         | 63.81      | 36.19           |                 |          |            |
| Gas Meso        | 67.79           | 32.21    | Gas Meso        | 69.13           | 30.87           | Gas Meso        | 70.85      | 29.15           |                 |          |            |
| Gas Low-Meso    | 64.84           | 35.16    | Gas Low-Meso    | 71.77           | 28.23           | Gas Low-Meso    | 70.98      | 29.02           |                 |          |            |
| 1 Acid Meso     | 37.30           | 62.70    | 1 Acid Meso     | 38.33           | 61.67           | 1 Acid Meso     | 35.29      | 64.71           |                 |          |            |
| 2 Acid Meso     | 37.77           | 62.23    | 2 Acid Meso     | 43.31           | 56.69           | 2 Acid Meso     | 37.85      | 62.15           |                 |          |            |
| 3 Acid Meso     | 37.55           | 62.45    | 3 Acid Meso     | 49.82           | 50.18           | 3 Acid Meso     | 54.54      | 45.46           |                 |          |            |
| 1 Acid Low-Meso | 38.12           | 61.88    | 1 Acid Low-Meso | 39.73           | 60.27           | 1 Acid Low-Meso | 39.75      | 60.25           |                 |          |            |
| 2 Acid Low-Meso | 35.29           | 64.71    | 2 Acid Low-Meso | 34.31           | 65.69           | 2 Acid Low-Meso | 35.03      | 64.97           |                 |          |            |
| 3 Acid Low-Meso | 34.97           | 65.03    | 3 Acid Low-Meso | 40.80           | 59.20           | 3 Acid Low-Meso | 41.39      | 58.61           |                 |          |            |
| Control         | 63.72           | 36.28    | Average         | Control         | 63.72           | 36.28           | Average    | Control         | 63.81           | 36.19    |            |
| Gas Meso        | 67.10           | 32.90    |                 | Gas Meso        | 67.10           | 32.90           |            | Gas Meso        | 70.85           | 29.15    |            |
| Gas Low-Meso    | 66.48           | 33.52    |                 | Gas Low-Meso    | 66.48           | 33.52           |            | Gas Low-Meso    | 70.98           | 29.02    |            |
| 1 Acid Meso     | 29.07           | 70.93    |                 | 1 Acid Meso     | 29.07           | 70.93           |            | 1 Acid Meso     | 35.29           | 64.71    |            |
| 2 Acid Meso     | 31.78           | 68.22    |                 | 2 Acid Meso     | 31.78           | 68.22           |            | 2 Acid Meso     | 37.85           | 62.15    |            |
| 3 Acid Meso     | 46.56           | 53.44    |                 | 3 Acid Meso     | 46.56           | 53.44           |            | 3 Acid Meso     | 54.54           | 45.46    |            |
| 1 Acid Low-Meso | 31.16           | 68.84    |                 | 1 Acid Low-Meso | 31.16           | 68.84           |            | 1 Acid Low-Meso | 39.75           | 60.25    |            |
| 2 Acid Low-Meso | 30.14           | 69.86    |                 | 2 Acid Low-Meso | 30.14           | 69.86           |            | 2 Acid Low-Meso | 35.03           | 64.97    |            |
| 3 Acid Low-Meso | 30.80           | 69.20    |                 | 3 Acid Low-Meso | 30.80           | 69.20           |            | 3 Acid Low-Meso | 41.39           | 58.61    |            |

**Table D - 5. Total and volatile solids – all sludges and all treatments from three stage acid digesters followed by gas digester experiments.**

| Sludge  | Average Total solids, g/l | Std. Dev. Total Solids, g/l | Average Volatile Solids, g/l | Std. Dev. Volatile Solids, g/l |
|---|---------------------------|-----------------------------|------------------------------|--------------------------------|
| Raw Normal  | 47.94                     | 18.38                       | 24.43                        | 4.93                           |
| Raw-thickened   | 57.02                     | 10.74                       | 31.17                        | 4.17                           |
| Effluent from low-mesophilic acid digesters   | 46.35                     | 2.93                        | 24.44                        | 2.32                           |
| Effluent from gas digester of three stage acid low-mesophilic followed by gas mesophilic system | 39.41                     | 3.98                        | 18.72                        | 1.65                           |
| Effluent from mesophilic acid digesters   | 44.33                     | 2.34                        | 23.04                        | 9.42                           |
| Effluent from gas digester of three stage acid mesophilic followed by gas mesophilic system     | 38.56                     | 2.00                        | 17.62                        | 1.67                           |
| Effluent from control digester  | 25.14                     | 3.73                        | 10.31                        | 1.25                           |

**Table D - 6. Total COD – all sludges and all treatments from three stage acid digesters followed by gas digester experiments.**

| Date            | Total COD  |              |  |              |                  |              |              |
|-----------------|--|--------------|--|--------------|------------------|--------------|--------------|
|                 | Three stage mesophilic acid digestion followed by mesophilic gas digestion |              | Three stage low-mesophilic acid digestion followed by mesophilic gas digestion |              | Control digester | Raw sludge   |              |
|                 | 3 Acid   | Gas          | 3 Acid   | Gas          | Gas              | Normal       | Thickened    |
| 25, May         | 445  | 304          | 437  | 319          | 156              | 319          | 467          |
| 27, May         | 422  | 319          | 385  | 393          | 141              | 630          | 437          |
| 30, May         | 474  | 363          | 467  | 260          | 156              | 349          | 630          |
| 1, June         | 437  | 289          | 393  | 304          | 149              | 334          | 422          |
| 3, June         | 378  | 260          | 408  | 297          | 141              | 378          | 400          |
| 6, June         | 437  | 275          | 415  | 304          | 127              | 230          | 482          |
| 8, June         | 408  | 275          | 393  | 319          | 112              | 201          | 541          |
| 10, June        | 371  | 282          | 385  | 341          | 127              | 260          | 600          |
| 13, June        | 408  | 267          | 422  | 326          | 127              | 186          | 482          |
| 15, June        | 452  | 289          | 422  | 341          | 97               | 260          | 363          |
| 17, June        | 482  | 304          | 452  | 363          | 149              | 245          | 422          |
| 20-Jun          | 408  | 267          | 437  | 349          | 104              | 238          | 378          |
| 24-Jun          | 385  | 282          | 430  | 334          | 119              | 437          | 156          |
| 24-Jun          | 363  | 223          | 334  | 275          | 119              | 304          | 289          |
| 27-Jun          | 312  | 393          | 326  | 297          | 164              | 349          | 326          |
| 29-Jun          | 334  | 289          | 363  | 289          | 141              | 260          | 275          |
| 04-Jul          | 326  | 252          | 356  | 297          | 171              | 467          | 304          |
| 07-Jul          | 326  | 267          | 319  | 282          | 186              | 171          | 371          |
| 07-Jul          | 289  | 230          | 334  | 289          | 178              | 275          | 356          |
| 11-Jul          | 282  | 208          | 312  | 238          | 134              | 201          | 326          |
| 13-Jul          | 282  | 201          | 385  | 238          | 134              | 149          | 408          |
| 15-Jul          | 349  | 245          | 334  | 223          | 127              | 349          | 341          |
| 18-Jul          | 267  | 260          | 267  | 304          | 252              | 252          | 252          |
| 20-Jul          | 297  | 201          | 312  | 193          | 104              | 349          | 482          |
| Factor          | x150   |              |  |              |                  |              |              |
| <b>Average</b>  | <b>55835</b>   | <b>40904</b> | <b>56806</b>   | <b>44833</b> | <b>21349</b>     | <b>44926</b> | <b>59441</b> |
| <i>Std. Dev</i> | <i>9837</i>  | <i>6872</i>  | <i>7899</i>  | <i>6921</i>  | <i>4954</i>      | <i>16077</i> | <i>16446</i> |

**Table D - 7. Soluble COD – all sludges and all treatments from three stage acid digesters followed by gas digester experiments.**

| Date            | Soluble COD  |             |  |             |                  |             |             |
|-----------------|--|-------------|--|-------------|------------------|-------------|-------------|
|                 | Three stage mesophilic acid digestion followed by mesophilic gas digestion |             | Three stage low-mesophilic acid digestion followed by mesophilic gas digestion |             | Control digester | Raw sludge  |             |
|                 | 3 Acid   | Gas         | 3 Acid   | Gas         | Gas              | Normal      | Thickened   |
| 25, May         | 371  | 230         | 378  | 215         | 30               | 112         | 141         |
| 27, May         | 349  | 215         | 334  | 334         | 38               | 122         | 141         |
| 30, May         | 378  | 215         | 378  | 223         | 38               | 156         | 171         |
| 1, June         | 275  | 141         | 349  | 186         | 53               | 127         | 134         |
| 3, June         | 230  | 164         | 297  | 178         | 38               | 171         | 149         |
| 6, June         | 252  | 127         | 238  | 171         | 53               | 92          | 112         |
| 8, June         | 245  | 112         | 260  | 186         | 8                | 97          | 141         |
| 10, June        | 267  | 127         | 289  | 201         | 16               | 90          | 97          |
| 13, June        | 304  | 141         | 297  | 208         | 8                | 62          | 82          |
| 15, June        | 378  | 171         | 378  | 230         | 4                | 67          | 141         |
| 17, June        | 422  | 260         | 378  | 230         | 8                | 63          | 90          |
| 20-Jun          | 363  | 127         | 349  | 193         | 2                | 43          | 53          |
| 24-Jun          | 319  | 127         | 393  | 193         | 12               | 67          | 38          |
| 24-Jun          | 260  | 112         | 245  | 164         | 30               | 97          | 67          |
| 27-Jun          | 171  | 112         | 193  | 97          | 21               | 48          | 60          |
| 29-Jun          | 215  | 75          | 260  | 149         | 16               | 23          | 28          |
| 04-Jul          | 215  | 112         | 260  | 112         | 23               | 60          | 67          |
| 07-Jul          | 208  | 141         | 230  | 156         | 23               | 30          | 97          |
| 07-Jul          | 171  | 149         | 230  | 104         | 75               | 60          | 104         |
| 11-Jul          | 141  | 67          | 208  | 60          | 16               | 46          | 60          |
| 13-Jul          | 171  | 38          | 201  | 67          | 13               | 43          | 60          |
| 15-Jul          | 778  | 104         | 193  | 60          | 20               | 28          | 38          |
| 18-Jul          | 223  | 53          | 289  | 82          | 12               | 23          | 22          |
| 20-Jul          | 215  | 42          | 97   | 30          | 15               | 21          | 12          |
| Factor          | x 50   |             |  |             |                  |             |             |
| <b>Average</b>  | <b>14579</b>   | <b>6781</b> | <b>14402</b>   | <b>8260</b> | <b>1211</b>      | <b>3756</b> | <b>4554</b> |
| <i>Std. Dev</i> | <i>6594</i>  | <i>2774</i> | <i>3346</i>  | <i>3345</i> | <i>901</i>       | <i>2067</i> | <i>2194</i> |



**Table D - 8. Energy calculations for scenario #1 – conventional MAD system at 4.8% TS in feed sludge and multi-phase digestion systems at 5.7% TS in feed sludge. All systems at loading rate of 100 tons TS/day and equal SRT.**

| Parameter   | Unit   | Value            |                    |         |         |                        |         |         |         |         |  |
|---|--|------------------|--------------------|---------|---------|------------------------|---------|---------|---------|---------|--|
|   |  | Conventional MAD | Meso Acid/Meso Gas |         |         | Low-Meso Acid/Meso Gas |         |         |         |         |  |
|   |  |                  | 1 Acid             | 2 Acid  | 3 Acid  | Gas                    | 1 Acid  | 2 Acid  | 3 Acid  | Gas     |  |
| Input   | tons TS/day                                      | 100.0            | 100.0              | 100.0   | 100.0   | 100.0                  | 100.0   | 100.0   | 100.0   | 100.0   |  |
| Sludge thickness  | % TS   | 4.8              | 5.7                | 5.7     | 5.7     | 5.7                    | 5.7     | 5.7     | 5.7     | 5.7     |  |
| Volume  | m <sup>3</sup> /day                              | 2083.3           | 1754.4             | 1754.4  | 1754.4  | 1754.4                 | 1754.4  | 1754.4  | 1754.4  | 1754.4  |  |
| Retention time in reactor                                 | days   | 22.0             | 3.0                | 3.0     | 3.0     | 13.0                   | 4.0     | 4.0     | 4.0     | 10.0    |  |
| Reactor volume  | m <sup>3</sup>                                   | 45833.3          | 5263.2             | 5263.2  | 5263.2  | 22807.0                | 7017.5  | 7017.5  | 7017.5  | 17543.9 |  |
| System volume   | m <sup>3</sup>                                   | 45833.3          |                    |         | 38596.5 |                        |         |         | 38596.5 |         |  |
| Gas Production  | Volume l/(day x digester volume)                 | 0.44             | 0.27               | 0.10    | 0.10    | 0.78                   | 0.13    | 0.09    | 0.03    | 0.71    |  |
| Methane content   | %  | 63.72            | 29.07              | 31.78   | 48.58   | 67.10                  | 31.16   | 30.14   | 30.80   | 66.48   |  |
| Methane production  | Volume CH <sub>4</sub> l/(day x digester volume) | 0.28             | 0.08               | 0.03    | 0.05    | 0.52                   | 0.04    | 0.03    | 0.01    | 0.47    |  |
| Methane production  | m <sup>3</sup> CH <sub>4</sub> /day              | 12849.83         | 413.11             | 167.27  | 245.07  | 11938.97               | 284.27  | 190.38  | 64.85   | 8281.42 |  |
| Energy in biogas @ 35.8 MJ/m <sup>3</sup> CH <sub>4</sub> | GJ/day   | 460.0            | 14.8               | 6.0     | 8.8     | 427.3                  | 10.2    | 6.8     | 2.3     | 296.5   |  |
| Total energy for the system                               | GJ   | 460.0            |                    |         | 456.9   |                        |         |         | 316.8   |         |  |
| Fraction of total energy in system                        | %  | 100.0            | 3.2                | 1.3     | 1.9     | 93.5                   | 3.2     | 2.2     | 0.7     | 93.9    |  |
| Heat requirement calculations                             |  |                  |                    |         |         |                        |         |         |         |         |  |
| Incoming sludge / Digester temperature                    | °C   | 10 / 38          | 10 / 38            | 38 / 38 | 38 / 38 | 38 / 38                | 10 / 24 | 10 / 24 | 10 / 24 | 24 / 38 |  |
| Temperature difference                                    | °C   | 28               | 28                 | 0       | 0       | 0                      | 14      | 0       | 0       | 14      |  |
| Specific gravity of sludge                                | kg/m <sup>3</sup>                                |                  |                    |         |         | 1002.0                 |         |         |         |         |  |
| Specific heat of sludge                                   | J/kg °C  |                  |                    |         |         | 4200                   |         |         |         |         |  |
| Heat requirement for digester                             | GJ/day   | 245.5            | 208.7              | 0.0     | 0.0     | 0.0                    | 103.4   | 0.0     | 0.0     | 103.4   |  |
| Heat requirement for the system                           | GJ/day   | 245.5            |                    |         | 206.7   |                        |         |         | 206.7   |         |  |
| Heat losses calculations                                  |  |                  |                    |         |         |                        |         |         |         |         |  |
| Total digesters volumes                                   | m <sup>3</sup>                                   | 45833.3          | 5263.2             | 5263.2  | 5263.2  | 22807.0                | 7017.5  | 7017.5  | 7017.5  | 17543.9 |  |
| Tank diameter   | m  | 30.0             | 30.0               | 30.0    | 30.0    | 30.0                   | 30.0    | 30.0    | 30.0    | 30.0    |  |
| Tank depth  | m  | 8.0              | 8.0                | 8.0     | 8.0     | 8.0                    | 8.0     | 8.0     | 8.0     | 8.0     |  |
| Tank volume   | m <sup>3</sup>                                   | 5652.0           | 5652.0             | 5652.0  | 5652.0  | 5652.0                 | 4239.0  | 4239.0  | 4239.0  | 5652.0  |  |
| Number of tanks (rounded up)                              | -  | 8.0              | 1.0                | 1.0     | 1.0     | 4.0                    | 2.0     | 2.0     | 2.0     | 4.0     |  |
| Area of walls   | m <sup>2</sup>                                   | 6782.4           | 753.6              | 753.6   | 753.6   | 3040.9                 | 1130.4  | 1130.4  | 1130.4  | 3014.4  |  |
| Area of cover/floor                                       | m <sup>2</sup>                                   | 6358.5           | 706.5              | 706.5   | 706.5   | 2850.9                 | 1413.0  | 1413.0  | 1413.0  | 2826.0  |  |
| Heat transfer coefficients                                |  |                  |                    |         |         |                        |         |         |         |         |  |
| wall + insulation   | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         | 0.054                  |         |         |         |         |  |
| floor with dry earth                                      | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         | 0.025                  |         |         |         |         |  |
| cover   | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         | 0.103                  |         |         |         |         |  |
| Temperatures  |  |                  |                    |         |         |                        |         |         |         |         |  |
| average air   | °C   |                  |                    |         |         | 10                     |         |         |         |         |  |
| earth   | °C   |                  |                    |         |         | 10                     |         |         |         |         |  |
| sludge  | °C   | 38               | 38                 | 38      | 38      | 38                     | 24      | 24      | 24      | 38      |  |
| Heat losses   |  |                  |                    |         |         |                        |         |         |         |         |  |
| walls   | GJ/day   | 10.3             | 1.1                | 1.1     | 1.1     | 4.6                    | 0.9     | 0.9     | 0.9     | 4.6     |  |
| floor   | GJ/day   | 4.4              | 0.5                | 0.5     | 0.5     | 2.0                    | 0.5     | 0.5     | 0.5     | 1.9     |  |
| cover   | GJ/day   | 18.3             | 2.0                | 2.0     | 2.0     | 8.2                    | 2.0     | 2.0     | 2.0     | 8.1     |  |
| Total heat losses for digesters                           | GJ/day   | 32.9             | 3.7                | 3.7     | 3.7     | 14.8                   | 3.4     | 3.4     | 3.4     | 14.6    |  |
| Total heat losses for the system                          | GJ/day   | 32.9             |                    |         | 25.7    |                        |         |         | 24.8    |         |  |
| Heat recovery   |  |                  |                    |         |         |                        |         |         |         |         |  |
| Temperatures  |  |                  |                    |         |         |                        |         |         |         |         |  |
| Incoming sludge   | °C   | 10               | 10                 | 38      | 38      | 38                     | 10      | 24      | 24      | 24      |  |
| Digested Sludge   | °C   | 38               | 38                 | 38      | 38      | 38                     | 24      | 24      | 24      | 38      |  |
| Average temp. in exchanger                                | °C   | 24               | 24                 | 38      | 38      | 38                     | 17      | 24      | 24      | 31      |  |
| Average temp. of cold sludge                              | °C   | 17               | 17                 | 38      | 38      | 38                     | 13.5    | 24      | 24      | 27.5    |  |
| Average temp. of hot sludge                               | °C   | 31               | 31                 | 38      | 38      | 38                     | 20.5    | 24      | 24      | 34.5    |  |
| Total heat recovered                                      | GJ/day   | 122.5            | 103.2              | 0.0     | 0.0     | 0.0                    | 51.6    | 0.0     | 0.0     | 51.6    |  |
| Correction x 0.8  | GJ/day   | 98.0             | 82.5               | 0.0     | 0.0     | 0.0                    | 41.3    | 0.0     | 0.0     | 41.3    |  |
| Total heat recovered for system                           | GJ/day   | 98.0             |                    |         | 82.5    |                        |         |         | 82.5    |         |  |
| Thickening costs  |  |                  |                    |         |         |                        |         |         |         |         |  |
|   | GJ/day   | 0.0              |                    |         | 84.6    |                        |         |         | 84.6    |         |  |
| Energy Demand   |  |                  |                    |         |         |                        |         |         |         |         |  |
|   | GJ/day   | 180.4            | 212.5              | 3.7     | 3.7     | 14.8                   | 150.1   | 3.4     | 3.4     | 76.7    |  |
|   |  | 180.4            |                    |         | 234.5   |                        |         |         | 233.6   |         |  |
| Recoverable Energy  |  |                  |                    |         |         |                        |         |         |         |         |  |
|   | GJ/day   | 368.0            | 11.8               | 4.8     | 7.0     | 341.9                  | 8.1     | 5.5     | 1.9     | 237.2   |  |
|   |  | 368.0            |                    |         | 365.5   |                        |         |         | 252.6   |         |  |
| Net Energy  |  |                  |                    |         |         |                        |         |         |         |         |  |
|   | GJ/day   | 187.6            | -200.6             | 1.1     | 3.4     | 327.1                  | -141.9  | 2.1     | -1.5    | 160.4   |  |
|   |  | 187.6            |                    |         | 131.0   |                        |         |         | 19.1    |         |  |

**Table D - 9. Energy calculations for scenario #2 – conventional MAD system at 4.8% TS in feed sludge and multi-phase digestion systems at 5.7% TS in feed sludge. All systems at loading rate of 100 tons TS/day and equal SRT.**

| Parameter   | Unit   | Value            |                    |         |         |                        |         |         |         |         |
|---|--|------------------|--------------------|---------|---------|------------------------|---------|---------|---------|---------|
|   |  | Conventional MAD | Meso Acid/Meso Gas |         |         | Low-Meso Acid/Meso Gas |         |         | Gas     |         |
|   |  |                  | 1 Acid             | 2 Acid  | 3 Acid  | Gas                    | 1 Acid  | 2 Acid  | 3 Acid  | Gas     |
| Input   | tons TS/day                                      | 100.0            | 100.0              | 100.0   | 100.0   | 100.0                  | 100.0   | 100.0   | 100.0   | 100.0   |
| Sludge thickness  | % TS   | 3.9              | 5.7                | 5.7     | 5.7     | 5.7                    | 5.7     | 5.7     | 5.7     | 5.7     |
| Volume  | m <sup>3</sup> /day                              | 2584.1           | 1754.4             | 1754.4  | 1754.4  | 1754.4                 | 1754.4  | 1754.4  | 1754.4  | 1754.4  |
| Retention time in reactor                                 | days   | 22.0             | 3.0                | 3.0     | 3.0     | 3.0                    | 3.0     | 3.0     | 3.0     | 3.0     |
| Reactor volume  | m <sup>3</sup>                                   | 56410.3          | 5263.2             | 5263.2  | 5263.2  | 22807.0                | 7017.5  | 7017.5  | 7017.5  | 17543.9 |
| System volume   | m <sup>3</sup>                                   | 56410.3          |                    |         | 38596.5 |                        |         |         | 38596.5 |         |
| Gas Production  | Volume (l/day x digester volume)                 | 0.36             | 0.27               | 0.10    | 0.10    | 0.78                   | 0.13    | 0.09    | 0.03    | 0.71    |
| Methane content   | %  | 63.72            | 29.07              | 31.78   | 48.56   | 67.10                  | 31.16   | 30.14   | 30.80   | 65.48   |
| Methane production  | Volume CH <sub>4</sub> (l/day x digester volume) | 0.23             | 0.98               | 0.03    | 0.05    | 0.52                   | 0.04    | 0.03    | 0.01    | 0.47    |
| Methane production  | m <sup>3</sup> CH <sub>4</sub> /day              | 12849.83         | 413.11             | 167.27  | 245.07  | 11936.97               | 284.27  | 190.38  | 64.85   | 8281.42 |
| Energy in biogas @ 35.8 MJ/m <sup>3</sup> CH <sub>4</sub> | GJ/day   | 460.0            | 14.8               | 6.0     | 8.8     | 427.3                  | 10.2    | 6.8     | 2.3     | 296.5   |
| Total energy for the system                               | GJ   | 460.0            |                    |         | 456.9   |                        |         |         | 315.8   |         |
| Fraction of total energy in system                        | %  | 100.0            | 3.2                | 1.3     | 1.9     | 93.5                   | 3.2     | 2.2     | 0.7     | 93.9    |
| <b>Heat requirement calculations</b>                      |  |                  |                    |         |         |                        |         |         |         |         |
| Incoming sludge / Digester temperature                    | °C   | 10 / 38          | 10 / 38            | 38 / 38 | 38 / 38 | 38 / 38                | 10 / 24 | 10 / 24 | 10 / 24 | 24 / 38 |
| Temperature difference                                    | °C   | 28               | 28                 | 0       | 0       | 0                      | 14      | 0       | 0       | 14      |
| Specific gravity of sludge                                | kg/m <sup>3</sup>                                |                  |                    |         |         |                        | 1002.0  |         |         |         |
| Specific heat of sludge                                   | J/kg °C  |                  |                    |         |         |                        | 4200    |         |         |         |
| Heat requirement for digester                             | GJ/day   | 302.1            | 206.7              | 0.0     | 0.0     | 0.0                    | 103.4   | 0.0     | 0.0     | 103.4   |
| Heat requirement for the system                           | GJ/day   | 302.1            |                    |         | 206.7   |                        |         |         | 206.7   |         |
| <b>Heat losses calculations</b>                           |  |                  |                    |         |         |                        |         |         |         |         |
| Total digesters volumes                                   | m <sup>3</sup>                                   | 56410.3          | 5263.2             | 5263.2  | 5263.2  | 22807.0                | 7017.5  | 7017.5  | 7017.5  | 17543.9 |
| Tank diameter   | m  | 30.0             | 30.0               | 30.0    | 30.0    | 30.0                   | 30.0    | 30.0    | 30.0    | 30.0    |
| Tank depth  | m  | 8.0              | 8.0                | 8.0     | 8.0     | 8.0                    | 8.0     | 8.0     | 8.0     | 8.0     |
| Tank volume   | m <sup>3</sup>                                   | 5652.0           | 5652.0             | 5652.0  | 5652.0  | 5652.0                 | 4239.0  | 4239.0  | 4239.0  | 5652.0  |
| Number of tanks (rounded up)                              |  | 10.0             | 1.0                | 1.0     | 1.0     | 4.0                    | 2.0     | 2.0     | 2.0     | 4.0     |
| Area of walls   | m <sup>2</sup>                                   | 7521.4           | 753.6              | 753.6   | 753.6   | 3040.9                 | 1130.4  | 1130.4  | 1130.4  | 3014.4  |
| Area of cover/floor                                       | m <sup>2</sup>                                   | 7051.3           | 706.5              | 706.5   | 706.5   | 2850.9                 | 1413.0  | 1413.0  | 1413.0  | 2826.0  |
| <b>Heat transfer coefficients</b>                         |  |                  |                    |         |         |                        |         |         |         |         |
| wall + insulation   | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         |                        | 0.054   |         |         |         |
| floor with dry earth                                      | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         |                        | 0.025   |         |         |         |
| cover   | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         |                        | 0.103   |         |         |         |
| <b>Temperatures</b>                                       |  |                  |                    |         |         |                        |         |         |         |         |
| average air   | °C   |                  |                    |         |         |                        | 10      |         |         |         |
| earth   | °C   |                  |                    |         |         |                        | 10      |         |         |         |
| sludge  | °C   | 38               | 38                 | 38      | 38      | 38                     | 24      | 24      | 24      | 38      |
| <b>Heat losses</b>  |  |                  |                    |         |         |                        |         |         |         |         |
| walls   | GJ/day   | 11.4             | 1.1                | 1.1     | 1.1     | 4.6                    | 0.9     | 0.9     | 0.9     | 4.6     |
| floor   | GJ/day   | 4.8              | 0.5                | 0.5     | 0.5     | 2.0                    | 0.5     | 0.5     | 0.5     | 1.9     |
| cover   | GJ/day   | 20.3             | 2.0                | 2.0     | 2.0     | 8.2                    | 2.0     | 2.0     | 2.0     | 8.1     |
| Total heat losses for digesters                           | GJ/day   | 36.5             | 3.7                | 3.7     | 3.7     | 14.8                   | 3.4     | 3.4     | 3.4     | 14.6    |
| Total heat losses for the system                          | GJ/day   | 36.5             |                    |         | 25.7    |                        |         |         | 24.8    |         |
| <b>Heat recovery</b>                                      |  |                  |                    |         |         |                        |         |         |         |         |
| <b>Temperatures</b>                                       |  |                  |                    |         |         |                        |         |         |         |         |
| Incoming sludge   | °C   | 10               | 10                 | 38      | 38      | 38                     | 10      | 24      | 24      | 24      |
| Digested Sludge   | °C   | 38               | 38                 | 38      | 38      | 38                     | 24      | 24      | 24      | 38      |
| Average temp. in exchanger                                | °C   | 24               | 24                 | 38      | 38      | 38                     | 17      | 24      | 24      | 31      |
| Average temp. of cold sludge                              | °C   | 17               | 17                 | 38      | 38      | 38                     | 13.5    | 24      | 24      | 27.5    |
| Average temp. of hot sludge                               | °C   | 31               | 31                 | 38      | 38      | 38                     | 20.5    | 24      | 24      | 34.5    |
| Total heat recovered                                      | GJ/day   | 150.8            | 103.2              | 0.0     | 0.0     | 0.0                    | 51.6    | 0.0     | 0.0     | 51.6    |
| Correction x 0.8  | GJ/day   | 120.6            | 82.5               | 0.0     | 0.0     | 0.0                    | 41.3    | 0.0     | 0.0     | 41.3    |
| Total heat recovered for system                           | GJ/day   | 120.6            |                    |         | 82.5    |                        |         |         | 82.5    |         |
| <b>Thickening costs</b>                                   |  |                  |                    |         |         |                        |         |         |         |         |
| Thickening costs  | GJ/day   | 0.0              |                    |         | 84.6    |                        |         |         | 84.6    |         |
| <b>Energy Demand</b>                                      |  |                  |                    |         |         |                        |         |         |         |         |
| Energy Demand   | GJ/day   | 218.1            | 212.5              | 3.7     | 3.7     | 14.8                   | 150.1   | 3.4     | 3.4     | 76.7    |
|   |  | 218.1            |                    |         | 234.5   |                        |         |         | 233.6   |         |
| <b>Recoverable Energy</b>                                 |  |                  |                    |         |         |                        |         |         |         |         |
| Recoverable Energy  | GJ/day   | 368.0            | 11.8               | 4.8     | 7.0     | 341.9                  | 8.1     | 5.5     | 1.9     | 237.2   |
|   |  | 368.0            |                    |         | 365.5   |                        |         |         | 252.6   |         |
| <b>Net Energy</b>   |  |                  |                    |         |         |                        |         |         |         |         |
| Net Energy  | GJ/day   | 150.0            | -200.6             | 1.1     | 3.4     | 327.1                  | -141.9  | 2.1     | -1.5    | 160.4   |
|   |  | 150.0            |                    |         | 131.0   |                        |         |         | 19.1    |         |

**Table D - 10. Energy calculations for scenario #3 – conventional MAD system at 4.8% TS in feed sludge and multi-phase digestion systems at 5.7% TS in feed sludge. All systems at loading rate of 100 tons TS/day and equal volumes.**

| Parameter   | Unit   | Value            |                    |         |         |                        |         |         |         |         |  |
|---|--|------------------|--------------------|---------|---------|------------------------|---------|---------|---------|---------|--|
|   |  | Conventional MAD | Meso Acid/Meso Gas |         |         | Low-Meso Acid/Meso Gas |         |         |         |         |  |
|   |  |                  | 1 Acid             | 2 Acid  | 3 Acid  | Gas                    | 1 Acid  | 2 Acid  | 3 Acid  | Gas     |  |
| Input   | tons TS/day                                      | 100.0            | 100.0              | 100.0   | 100.0   | 100.0                  | 100.0   | 100.0   | 100.0   | 100.0   |  |
| Sludge thickness  | % TS   | 4.8              | 5.7                | 5.7     | 5.7     | 5.7                    | 5.7     | 5.7     | 5.7     | 5.7     |  |
| Volume  | m <sup>3</sup> /day                              | 2093.3           | 1754.4             | 1754.4  | 1754.4  | 1754.4                 | 1754.4  | 1754.4  | 1754.4  | 1754.4  |  |
| Retention time in reactor                                 | days   | 22.0             | 3.0                | 3.0     | 3.0     | 17.1                   | 4.0     | 4.0     | 4.0     | 14.1    |  |
| Reactor volume  | m <sup>3</sup>                                   | 45833.3          | 5263.2             | 5263.2  | 5263.2  | 30043.9                | 7017.5  | 7017.5  | 7017.5  | 24780.7 |  |
| System volume   | m <sup>3</sup>                                   | 45833.3          |                    |         | 45833.3 |                        |         |         | 45833.3 |         |  |
| Gas Production  | Volume (l/day x digester volume)                 | 0.44             | 0.37               | 0.10    | 0.10    | 0.84                   | 0.13    | 0.09    | 0.03    | 0.54    |  |
| Methane content   | %  | 63.72            | 29.07              | 31.78   | 45.56   | 67.10                  | 31.16   | 30.14   | 30.80   | 56.48   |  |
| Methane production  | Volume CH <sub>4</sub> (l/day x digester volume) | 0.28             | 0.08               | 0.03    | 0.05    | 0.43                   | 0.04    | 0.03    | 0.01    | 0.38    |  |
| Methane production  | m <sup>3</sup> CH <sub>4</sub> /day              | 12849.83         | 413.11             | 167.27  | 245.07  | 12819.69               | 284.27  | 190.38  | 64.85   | 6937.07 |  |
| Energy in biogas @ 35.8 MJ/m <sup>3</sup> CH <sub>4</sub> | GJ/day   | 460.0            | 14.8               | 6.0     | 8.8     | 458.9                  | 10.2    | 6.8     | 2.3     | 319.9   |  |
| Total energy for the system                               | GJ   | 460.0            |                    |         | 488.5   |                        |         |         | 338.3   |         |  |
| Fraction of total energy in system                        | %  | 100.0            | 3.0                | 1.2     | 1.8     | 94.0                   | 3.0     | 2.0     | 0.7     | 84.3    |  |
| <b>Heat requirement calculations</b>                      |  |                  |                    |         |         |                        |         |         |         |         |  |
| Incoming sludge / Digester temperature                    | °C   | 10 / 38          | 10 / 38            | 38 / 38 | 38 / 38 | 38 / 38                | 10 / 24 | 10 / 24 | 10 / 24 | 24 / 38 |  |
| Temperature difference                                    | °C   | 28               | 28                 | 0       | 0       | 0                      | 14      | 0       | 0       | 14      |  |
| Specific gravity of sludge                                | kg/m <sup>3</sup>                                |                  |                    |         |         | 1002.0                 |         |         |         |         |  |
| Specific heat of sludge                                   | J/kg °C  |                  |                    |         |         | 4200                   |         |         |         |         |  |
| Heat requirement for digester                             | GJ/day   | 245.5            | 206.7              | 0.0     | 0.0     | 0.0                    | 103.4   | 0.0     | 0.0     | 103.4   |  |
| Heat requirement for the system                           | GJ/day   | 245.5            |                    |         | 206.7   |                        |         |         | 206.7   |         |  |
| <b>Heat losses calculations</b>                           |  |                  |                    |         |         |                        |         |         |         |         |  |
| Total digesters volumes                                   | m <sup>3</sup>                                   | 45833.3          | 5263.2             | 5263.2  | 5263.2  | 30043.9                | 7017.5  | 7017.5  | 7017.5  | 24780.7 |  |
| Tank diameter   | m  | 30.0             | 30.0               | 30.0    | 30.0    | 30.0                   | 30.0    | 30.0    | 30.0    | 30.0    |  |
| Tank depth  | m  | 8.0              | 8.0                | 8.0     | 8.0     | 8.0                    | 6.0     | 6.0     | 6.0     | 8.0     |  |
| Tank volume   | m <sup>3</sup>                                   | 5652.0           | 5652.0             | 5652.0  | 5652.0  | 5652.0                 | 4239.0  | 4239.0  | 4239.0  | 5652.0  |  |
| Number of tanks (rounded up)                              |  | 9.0              | 1.0                | 1.0     | 1.0     | 5.3                    | 2.0     | 2.0     | 2.0     | 4.0     |  |
| Area of walls   | m <sup>2</sup>                                   | 6782.4           | 753.6              | 753.6   | 753.6   | 4005.8                 | 1130.4  | 1130.4  | 1130.4  | 3014.4  |  |
| Area of cover/floor                                       | m <sup>2</sup>                                   | 6358.5           | 706.5              | 706.5   | 706.5   | 3755.5                 | 1413.0  | 1413.0  | 1413.0  | 2826.0  |  |
| Heat transfer coefficients                                |  |                  |                    |         |         |                        |         |         |         |         |  |
| wall + insulation   | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         | 0.054                  |         |         |         |         |  |
| floor with dry earth cover                                | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         | 0.025                  |         |         |         |         |  |
| cover   | MJ/(m <sup>2</sup> °C)                           |                  |                    |         |         | 0.103                  |         |         |         |         |  |
| Temperatures  |  |                  |                    |         |         |                        |         |         |         |         |  |
| average air   | °C   |                  |                    |         |         | 10                     |         |         |         |         |  |
| earth   | °C   |                  |                    |         |         | 10                     |         |         |         |         |  |
| sludge  | °C   | 38               | 38                 | 38      | 38      | 38                     | 24      | 24      | 24      | 38      |  |
| Heat losses   |  |                  |                    |         |         |                        |         |         |         |         |  |
| walls   | GJ/day   | 10.3             | 1.1                | 1.1     | 1.1     | 6.1                    | 0.9     | 0.9     | 0.9     | 4.6     |  |
| floor   | GJ/day   | 4.4              | 0.5                | 0.5     | 0.5     | 2.8                    | 0.5     | 0.5     | 0.5     | 1.9     |  |
| cover   | GJ/day   | 18.3             | 2.0                | 2.0     | 2.0     | 10.8                   | 2.0     | 2.0     | 2.0     | 8.1     |  |
| Total heat losses for digesters                           | GJ/day   | 32.9             | 3.7                | 3.7     | 3.7     | 19.5                   | 3.4     | 3.4     | 3.4     | 14.6    |  |
| Total heat losses for the system                          | GJ/day   | 32.9             |                    |         | 30.4    |                        |         |         | 24.8    |         |  |
| <b>Heat recovery</b>                                      |  |                  |                    |         |         |                        |         |         |         |         |  |
| Temperatures  |  |                  |                    |         |         |                        |         |         |         |         |  |
| Incoming sludge   | °C   | 10               | 10                 | 38      | 38      | 38                     | 10      | 24      | 24      | 24      |  |
| Digested Sludge   | °C   | 38               | 38                 | 38      | 38      | 38                     | 24      | 24      | 24      | 38      |  |
| Average temp. in exchanger                                | °C   | 24               | 24                 | 38      | 38      | 38                     | 17      | 24      | 24      | 31      |  |
| Average temp. of cold sludge                              | °C   | 17               | 17                 | 38      | 38      | 38                     | 13.5    | 24      | 24      | 27.5    |  |
| Total heat recovered                                      | GJ/day   | 31               | 31                 | 38      | 38      | 38                     | 20.5    | 24      | 24      | 34.5    |  |
| Correction x 0.8  | GJ/day   | 98.0             | 82.5               | 0.0     | 0.0     | 0.0                    | 41.3    | 0.0     | 0.0     | 41.3    |  |
| Total heat recovered for system                           | GJ/day   | 98.0             |                    |         | 82.5    |                        |         |         | 82.5    |         |  |
| Thickening costs  | GJ/day   | 0.0              |                    |         | 84.8    |                        |         |         | 84.8    |         |  |
| Energy Demand   | GJ/day   | 180.4            | 212.5              | 3.7     | 3.7     | 19.5                   | 150.1   | 3.4     | 3.4     | 76.7    |  |
| Recoverable Energy  | GJ/day   | 388.0            | 11.8               | 4.8     | 7.0     | 367.2                  | 8.1     | 5.5     | 1.9     | 256.0   |  |
| Net Energy  | GJ/day   | 187.6            | -200.6             | 1.1     | 3.4     | 347.7                  | -141.9  | 2.1     | -1.5    | 179.2   |  |
|   |  | 187.6            |                    |         | 151.6   |                        |         |         | 37.8    |         |  |

**Table D - 11. Energy calculations for scenario #4 – conventional MAD system at 3.9% TS in feed sludge and multi-phase digestion systems at 5.7% TS in feed sludge. All systems at loading rate of 100 tons TS/day and equal volumes.**

| Parameter   | Unit   | Conventional MAD | Value   |         |         |          |         |         |         |         |         |         |
|---|--|------------------|---------|---------|---------|----------|---------|---------|---------|---------|---------|---------|
|   |  |                  | 1 Acid  | 2 Acid  | 3 Acid  | Gas      | 1 Acid  | 2 Acid  | 3 Acid  | Gas     |         |         |
| Input   | tons TS/day                                      | 100.0            | 100.0   | 100.0   | 100.0   | 100.0    | 100.0   | 100.0   | 100.0   | 100.0   | 100.0   | 100.0   |
| Sludge thickness  | % TS   | 3.9              | 5.7     | 5.7     | 5.7     | 5.7      | 5.7     | 5.7     | 5.7     | 5.7     | 5.7     | 5.7     |
| Volume  | m <sup>3</sup> /day                              | 2584.1           | 1754.4  | 1754.4  | 1754.4  | 1754.4   | 1754.4  | 1754.4  | 1754.4  | 1754.4  | 1754.4  | 1754.4  |
| Retention time in reactor                                 | days   | 22.0             | 3.0     | 3.0     | 3.0     | 3.0      | 23.2    | 4.0     | 4.0     | 4.0     | 30.2    | 30.2    |
| Reactor volume  | m <sup>3</sup>                                   | 56410.3          | 5263.2  | 5263.2  | 5263.2  | 40620.8  | 7017.5  | 7017.5  | 7017.5  | 7017.5  | 35357.6 | 35357.6 |
| System volume   | m <sup>3</sup>                                   | 56410.3          |         |         | 56410.3 |          |         |         | 56410.3 |         |         |         |
| Gas Production  | Volume (l/day x digester volume)                 | 0.38             | 0.37    | 0.10    | 0.10    | 0.51     | 0.13    | 0.09    | 0.03    | 0.42    |         |         |
| Methane content   | %  | 63.72            | 29.07   | 31.78   | 45.56   | 67.10    | 31.15   | 30.14   | 30.80   | 60.48   |         |         |
| Methane production  | Volume CH <sub>4</sub> (l/day x digester volume) | 0.23             | 0.08    | 0.03    | 0.05    | 0.34     | 0.04    | 0.03    | 0.01    | 0.28    |         |         |
| Methane production  | m <sup>3</sup> CH <sub>4</sub> /day              | 12849.83         | 413.11  | 167.27  | 245.07  | 13886.07 | 284.27  | 190.38  | 64.65   | 9871.60 |         |         |
| Energy in biogas @ 35.8 MJ/m <sup>3</sup> CH <sub>4</sub> | GJ/day   | 460.0            | 14.8    | 6.0     | 8.8     | 497.1    | 10.2    | 6.8     | 2.3     | 353.4   |         |         |
| Total energy for the system                               | GJ   | 460.0            |         |         | 526.7   |          |         |         | 372.7   |         |         |         |
| Fraction of total energy in system                        | %  | 100.0            | 2.8     | 1.1     | 1.7     | 94.4     | 2.7     | 1.8     | 0.6     | 94.8    |         |         |
| <b>Heat requirement calculations</b>                      |  |                  |         |         |         |          |         |         |         |         |         |         |
| Incoming sludge / Digester temperature                    | °C   | 10 / 38          | 10 / 38 | 38 / 38 | 38 / 38 | 38 / 38  | 10 / 24 | 10 / 24 | 10 / 24 | 24 / 38 |         |         |
| Temperature difference                                    | °C   | 28               | 28      | 0       | 0       | 0        | 14      | 0       | 0       | 14      |         |         |
| Specific gravity of sludge                                | kg/m <sup>3</sup>                                |                  |         |         |         | 1002.0   |         |         |         |         |         |         |
| Specific heat of sludge                                   | J/kg °C  |                  |         |         |         | 4200     |         |         |         |         |         |         |
| Heat requirement for digester                             | GJ/day   | 302.1            | 206.7   | 0.0     | 0.0     | 0.0      | 103.4   | 0.0     | 0.0     | 103.4   |         |         |
| Heat requirement for the system                           | GJ/day   | 302.1            |         |         | 206.7   |          |         |         | 206.7   |         |         |         |
| <b>Heat losses calculations</b>                           |  |                  |         |         |         |          |         |         |         |         |         |         |
| Total digesters volumes                                   | m <sup>3</sup>                                   | 56410.3          | 5263.2  | 5263.2  | 5263.2  | 40620.8  | 7017.5  | 7017.5  | 7017.5  | 35357.6 |         |         |
| Tank diameter   | m  | 30.0             | 30.0    | 30.0    | 30.0    | 30.0     | 30.0    | 30.0    | 30.0    | 30.0    |         |         |
| Tank depth  | m  | 8.0              | 8.0     | 8.0     | 8.0     | 8.0      | 6.0     | 6.0     | 6.0     | 8.0     |         |         |
| Tank volume   | m <sup>3</sup>                                   | 5652.0           | 5652.0  | 5652.0  | 5652.0  | 5652.0   | 4239.0  | 4239.0  | 4239.0  | 5652.0  |         |         |
| Number of tanks (rounded up)                              | -  | 10.0             | 1.0     | 1.0     | 1.0     | 8.0      | 2.0     | 2.0     | 2.0     | 7.0     |         |         |
| Area of walls   | m <sup>2</sup>                                   | 7521.4           | 753.6   | 753.6   | 753.6   | 6028.8   | 1130.4  | 1130.4  | 1130.4  | 5275.2  |         |         |
| Area of cover/floor                                       | m <sup>2</sup>                                   | 7051.3           | 706.5   | 706.5   | 706.5   | 5652.0   | 1413.0  | 1413.0  | 1413.0  | 4945.5  |         |         |
| <b>Heat transfer coefficients</b>                         |  |                  |         |         |         |          |         |         |         |         |         |         |
| wall + insulation   | MJ/(m <sup>2</sup> °C)                           |                  |         |         |         | 0.054    |         |         |         |         |         |         |
| floor with dry earth                                      | MJ/(m <sup>2</sup> °C)                           |                  |         |         |         | 0.025    |         |         |         |         |         |         |
| cover   | MJ/(m <sup>2</sup> °C)                           |                  |         |         |         | 0.103    |         |         |         |         |         |         |
| <b>Temperatures</b>                                       |  |                  |         |         |         |          |         |         |         |         |         |         |
| average air   | °C   |                  |         |         |         | 10       |         |         |         |         |         |         |
| earth   | °C   |                  |         |         |         | 10       |         |         |         |         |         |         |
| sludge  | °C   | 38               | 38      | 38      | 38      | 38       | 24      | 24      | 24      | 38      |         |         |
| <b>Heat losses</b>  |  |                  |         |         |         |          |         |         |         |         |         |         |
| walls   | GJ/day   | 11.4             | 1.1     | 1.1     | 1.1     | 9.1      | 0.9     | 0.9     | 0.9     | 8.0     |         |         |
| floor   | GJ/day   | 4.8              | 0.5     | 0.5     | 0.5     | 3.9      | 0.5     | 0.5     | 0.5     | 3.4     |         |         |
| cover   | GJ/day   | 20.3             | 2.0     | 2.0     | 2.0     | 16.3     | 2.0     | 2.0     | 2.0     | 14.2    |         |         |
| Total heat losses for digesters                           | GJ/day   | 36.5             | 3.7     | 3.7     | 3.7     | 29.3     | 3.4     | 3.4     | 3.4     | 25.6    |         |         |
| Total heat losses for the system                          | GJ/day   | 36.5             |         |         | 40.3    |          |         |         | 35.7    |         |         |         |
| <b>Heat recovery</b>                                      |  |                  |         |         |         |          |         |         |         |         |         |         |
| <b>Temperatures</b>                                       |  |                  |         |         |         |          |         |         |         |         |         |         |
| Incoming sludge   | °C   | 10               | 10      | 38      | 38      | 38       | 10      | 24      | 24      | 24      |         |         |
| Digested Sludge   | °C   | 38               | 38      | 38      | 38      | 38       | 24      | 24      | 24      | 38      |         |         |
| Average temp. in exchanger                                | °C   | 24               | 24      | 38      | 38      | 38       | 17      | 24      | 24      | 31      |         |         |
| Average temp. of cold sludge                              | °C   | 17               | 17      | 38      | 38      | 38       | 13.5    | 24      | 24      | 27.5    |         |         |
| Average temp. of hot sludge                               | °C   | 31               | 31      | 38      | 38      | 38       | 20.5    | 24      | 24      | 34.5    |         |         |
| Total heat recovered                                      | GJ/day   | 150.8            | 103.2   | 0.0     | 0.0     | 0.0      | 51.6    | 0.0     | 0.0     | 51.6    |         |         |
| Correction x 0.8  | GJ/day   | 120.6            | 82.5    | 0.0     | 0.0     | 0.0      | 41.3    | 0.0     | 0.0     | 41.3    |         |         |
| Total heat recovered for system                           | GJ/day   | 120.6            |         |         | 82.5    |          |         |         | 82.5    |         |         |         |
| <b>Thickening costs</b>                                   |  |                  |         |         |         |          |         |         |         |         |         |         |
|   | GJ/day   | 0.0              |         |         | 84.6    |          |         |         | 84.6    |         |         |         |
| <b>Energy Demand</b>                                      |  |                  |         |         |         |          |         |         |         |         |         |         |
|   | GJ/day   | 218.1            | 212.5   | 3.7     | 3.7     | 29.3     | 150.1   | 3.4     | 3.4     | 87.7    |         |         |
|   |  | 218.1            |         |         | 249.1   |          |         |         | 244.5   |         |         |         |
| <b>Recoverable Energy</b>                                 |  |                  |         |         |         |          |         |         |         |         |         |         |
|   | GJ/day   | 368.0            | 11.8    | 4.8     | 7.0     | 397.7    | 8.1     | 5.5     | 1.9     | 282.7   |         |         |
|   |  | 368.0            |         |         | 421.3   |          |         |         | 298.2   |         |         |         |
| <b>Net Energy</b>   |  |                  |         |         |         |          |         |         |         |         |         |         |
|   | GJ/day   | 150.0            | -200.6  | 1.1     | 3.4     | 368.4    | -141.9  | 2.1     | -1.5    | 195.0   |         |         |
|   |  | 150.0            |         |         | 172.3   |          |         |         | 53.6    |         |         |         |