PERFORMANCE AND MECHANISMS OF EXCESS SLUDGE REDUCTION IN THE CANNIBAL™ PROCESS

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ABSTRACT

In order to study the performance and mechanisms of excess sludge reduction in the activated sludge that incorporates the Cannibal™ Process, laboratory activated sludge systems incorporating an anaerobic bioreactor into the sludge recycle stream were operated. In this study, the solids production in the Cannibal system was about 35-40% of the conventional system under steady state conditions. The reduction in waste sludge was optimized when the interchange rate, (the ratio of sludge fed from the activated sludge system to the bioreactor compared to the total mass in the activated sludge system) was set at about 10 %. It was found that the release of protein from the anaerobic bioreactor was greater than that from the aerobic bioreactor. The SOUR data suggested that the released protein from the anaerobic bioreactor was easily degraded when the sludge was returned to the activated sludge system. It was also found that when the proportion of sludge added to the anaerobic bioreactor in batch tests was approximately 10%, the protein release was about 30 mg/L. When the proportion of sludge added was increased to 26 to 41%, the release was reduced to 10 and 6 mg/L, respectively. Within 30 hours, the protein release was complete. This suggests that there is an optimum or maximum amount of recycle or interchange (~10%) for the process to function best.

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In particular, I would like to thank Dr. Jin Young Jung and Dr. Suing Il Choi who guided me to study at Virginia Tech and to pursue a right way of life.

DEDICATION

To my family

I would like to thank my parents, Young Bok Chon and Jeong Yeon Hong, for their great guidance, prayer, and sacrifice, and my sister and bother-in law, Eun Young Chon and Seong Won Kim, for their love and support. Without their love and sacrifice, this study would not have been completed.

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INTRODUCTION

Activated sludge is the most widely used biological wastewater treatment process for the degradation of organic matter and removal of nutrients from domestic and industrial wastewaters. The conventional activated sludge process consists of primary clarification, biological treatment in an aeration basin, secondary clarification, sludge recycling and sludge wastage. The activated sludge process mainly relies on bioflocculation, aggregation of microorganisms and microbial products. Microorganisms in bioflocs oxidize organic matter into end products (CO_2, NH_4^+, SO_4, and PO_4) and synthesize new biomass. Dense and large flocs are separated from the liquid in a secondary clarifier by gravity settling.

The SRT has a significant impact on the performance of the activated sludge process and it also controls physical and biological characteristics of activated sludge. Based upon kinetic theory, a longer SRT should provide a greater substrate removal from the wastewaters. However, the characteristics of wastewaters and configuration of the activated sludge process, especially with regard to nutrient removal, should be considered in selecting the SRT (Metcalf and Eddy, 1991).

Anaerobic digestion is the primary process used to reduce the solids content of waste sludge and render it suitable for land application. Anaerobic digestion is basically comprised of three steps. In the first step, hydrolysis, particulate organic matter is converted into soluble substrates. In the second step, acidogenesis, the soluble substrates are broken down into acetic acid, carbon dioxide, and H_2. In the final step, methanogenesis, acetic acid and H_2 are converted into methane and carbon dioxide by H_2-oxidizing methanogens and aceticlastic methanogens. H_2-oxidizing methanogens are all strictly obligate anaerobes so they can obtain energy from the oxidation of H_2 and carbon from carbon dioxide. Through the evolution of methane, the chemical oxygen demand (COD) of wastewaters decrease and the stabilization of the biodegradable organic matter can be achieved (Metcalf and Eddy, 1991 and Grady et al., 1998).

Aerobic digestion has been used to stabilize particulate organic matter and destroy pathogens in wastewaters. As the supply of available substrate is depleted, the microorganisms begin to consume their own protoplasm from cell tissue or internal parts to maintain cell function in the endogenous phase (Metcalf and Eddy, 1991). Advantages of aerobic digestion are a high percentage volatile solids reduction, lower BOD concentrations in centrate or filtrate, production of an odorless, biologically stable end product, and lower capital cost. The major disadvantages of the aerobic digestion process are high power cost for supplying the required oxygen and poorer mechanical dewatering characteristics (Metcalf and Eddy, 1991). For proper operation, temperature, solids reduction, detention time, feed solids concentration, and oxygen requirements are considered to be important (Metcalf and Eddy, 1991).

Exocellular polymeric substances (EPS) are a major part of the floc structure in activated sludge. EPS in activated sludge flocs constitute a matrix in which bacteria and other particles are embedded. Jorand et al. (1995) described the complexity of the activated sludge floc structure using staining and the analysis of polymers released from sonicated sludges and found that the main polymers released from the activated sludge flocs were proteins, polysaccharides and DNA. Frølund et al. (1995) found the components of activated sludge flocs to be protein (46-52% of VS), humic compounds (18-23% of VS) and carbohydrate (17% of VS) using the cation exchange resin extraction technique.

Novak et al. (2003) proposed that there are different mechanisms of floc destruction between aerobic and anaerobic digestion. They quantified the release of biopolymer from sludges under both anaerobic and aerobic conditions and showed that the release of protein into solution was 4-5 times higher under anaerobic digestion than under aerobic digestion, although the VS destruction by anaerobic digestion was no higher than for aerobic digestion. The authors hypothesized that there are two types of biopolymer binding mechanisms in flocs; one fraction associated with divalent cations (Ca^{++} and Mg^{++}) that is degraded primarily under aerobic digestion and another fraction associated with iron that is degraded primarily under anaerobic digestion.

This study was designed to investigate the performance of the Cannibal system, determine sludge reduction mechanisms, and optimize excess sludge reduction in the activated sludge process. For those objectives, laboratory scale activated sludge systems incorporating an anaerobic bioreactor into the sludge recycle stream and batch testes were operated and conducted.

OVERVIEW OF THE ACTIVATED SLUDGE AND DIGESTION PROCESSES

Activated sludge

Activated sludge is the most widely used biological wastewater treatment process for the degradation of organic matter and removal of nutrients from domestic and industrial wastewaters. The conventional activated sludge process consists of primary clarification, biological treatment in an aeration basin, secondary clarification, sludge recycle and sludge wastage. The activated sludge process mainly relies on bioflocculation; aggregation of microorganisms and microbial products. Microorganisms in bioflocs oxidize organic matter into end products (CO_2, NH_4^+, SO_4, and PO_4) and synthesize new biomass. Dense and large flocs are separated from the liquid in a secondary clarifier by gravity settling. Settled solids are recycled or wasted from the sludge settling tank. The purpose of recycling thickened mixed liquor is to maintain the desired concentration of biological suspended solids (MLSS) in the aeration basin and the purpose of sludge wasting is to control the solids retention time (SRT) for reliable treatment.

The reason why activated sludge is currently the most widely used process is that it is flexible, reliable, can produce a high degree of nitrification and can stabilize insoluble organic matter (Grady et al., 1998). However, one of its main disadvantages is the generation of excess sludge. Although the activated sludge process works efficiently, the growth of microorganisms results in the production of excess biomass that requires disposal to control the solids retention time (SRT). To treat this excess sludge, a combination of digestion, chemical treatment, dewatering, and thickening is generally used. The treatment for excess sludge can account for up to 60% of a plant's total operating cost (Horan et al., 1990) and also adds to the capital cost of the facility.

Aerobic digestion

The digestion process plays an important role in the solids handling system of a wastewater treatment process for destruction of organic matter and reduction of pathogens. Digestion may occur either in the presence or absence of molecular oxygen. Very different microbiological and biochemical reactions are used in each digestion. Digestion was considered as a treatment prior to the ultimate disposal of sludge (Metcalf and Eddy, 1991).

Aerobic digestion has been used to stabilize particulate organic matter and destroy pathogens in wastewaters. As the supply of available substrate is depleted, the microorganisms begin to consume their own protoplasm from cell tissue or internal parts to maintain cell function in the endogenous phase (Metcalf and Eddy, 1991).

Advantages of aerobic digestion are a high percentage volatile solids reduction, lower BOD concentrations in centrate or filtrate, production of an odorless, biologically stable end product, and lower capital cost. The major disadvantages of the aerobic digestion process are high power cost for supplying the required oxygen and poorer mechanical dewatering characteristics (Metcalf and Eddy, 1991). For proper operation, temperature, solids reduction, detention time, feed solids concentration, and oxygen requirements are considered to be important (Metcalf and Eddy, 1991).

Mineralization of organic nitrogen during aerobic digestion could be an indicator of aerobic digestion. Bishop and Farmer (1978) and Mavinic and Koers (1982) found that significant rates of nitrification occurred during aerobic digestion. They also reported that mineralized organic nitrogen is equivalent to destroyed VSS. Mavinic and Koers (1982) also observed that the pH of sludge decreased below 4 because of the alkalinity consumption by nitrification.

Novak et al. (2003) investigated biopolymer release in aerobic digestion. The authors measured the accumulation of solution polysaccharide and divalent cations that resulted from degradation while the soluble protein concentration remained fairly low in the supernatant. This indicates that it is the fraction of biopolymer associated with divalent cations (Ca^{++} and Mg^{++}) that is primarily degraded under aerobic digestion. The authors intimated that the low protein concentration was

due to peptidase activity and thus degradation of protein in the floc. The degradation of protein resulted in a weaker flocs, indicating that proteins play a major role in floc formation and floc strength. The authors also showed that aerobic digestion resulted in the greater solids reduction, approximately 50% as compared to 30% for anaerobic digestion for 30 days digestion.

Anaerobic digestion

Anaerobic digestion has been used for the stabilization of organic matter and destruction of pathogens prior to ultimate solids disposal. Anaerobic digestion occurs in the absence of molecular oxygen. The efficiency of anaerobic digestion can be accelerated by heating and mixing the digester. Anaerobic digestion is a slow process and a relatively long detention time is required for slow cell material synthesis (Metcalf and Eddy, 1991).

Anaerobic digestion is basically comprised of three steps. In the first step, hydrolysis, particulate organic matter is converted into soluble substrates. In the second step, acidogenesis, the soluble substrates are broken down into acetic acid, carbon dioxide, and H_2. In the final step, methanogenesis, acetic acid and H_2 are converted into methane and carbon dioxide by H_2-oxidizing methanogens and aceticlastic methanogens. H_2-oxidizing methanogens are all strictly obligate anaerobes so they can obtain energy from the oxidation of H_2 and carbon from carbon dioxide. Through the evolution of methane, the chemical oxygen demand (COD) of wastewaters decrease and the stabilization of the biodegradable organic matter can be achieved (Metcalf and Eddy, 1991 and Grady et al., 1998).

FACTORS THAT AFFECT SLUDGE PRODUCTION

Cell lysis

In the activated sludge system, microorganisms are continually undergoing death and lysis, releasing organic matter to the environment in which they are growing (Grady et al., 1998). This released organic matter is reused in microbial metabolism and a portion of the carbon is discharged as products of respiration. Consequentially, this pathway can result in a reduced

overall biomass production. Therefore, an increase of the lysis efficiency can allow to an overall reduction of sludge generation (Wei et al., 2003).

Solids retention time (SRT)

The SRT has a significant impact on the performance of the activated sludge process and it also controls physical and biological characteristics of activated sludge. Based upon kinetic theory, a longer SRT should provide a greater substrate removal from the wastewaters. However, the characteristics of wastewaters and configuration of the activated sludge process, especially with regard to nutrient removal, should be considered in selecting the SRT (Metcalf and Eddy, 1991).

Bisogni and Lawrence (1971) investigated the impacts of SRT on activated sludge characteristics and found that SRT influences settling properties of activated sludge and effluent biological oxygen demand (BOD). It was observed that bacterial flocs tended to disperse and the effluent suspended solids (SS) concentration was high at the lower values of SRT. As a result of dispersed growth at an SRT of 2 days or less, the settling velocity of activated sludge could be slow and washout could occur. At higher values of SRT, bacterial flocs also tended to deteriorate and disperse. The authors also found an accumulation of polysaccharide at long SRT values.

Reece et al. (1979) found an increase in nonbiodegradable material in activated sludge as the SRT increased, affecting the aerobic digestibility of sludge. Murthy (1998) showed similar results in that as the SRT increased above 10 days, soluble polysaccharide and colloidal protein resulted in an increase in the effluent COD, but the effluent BOD remained constant. This implies that the increased effluent COD was nonbiodegradable organic material.

The S_0/X_0 ratio

It has been shown that the important parameter in batch cultivation of mixed cultures is the ratio of the initial substrate concentration to the initial biomass concentration (S_0/X_0 as COD/biomass). Chudoba et al. (1992) found that cell multiplication did not occur during the exogenous substrate removal when the ratio (S_0/X_0) was sufficiently low. The authors suggested that a biomass increase was mostly due to the synthesis of storage polymers under the sufficiently low ratio (S_0/X_0) and, under the high S_0/X_0 ratio, more energy was spent for cell synthesis, which results in

a greater part of the substrate being oxidized. It was also shown that the observed yield, Y_{obs}, decreases with increasing S_0/X_0 ratio.

Liu et al. (1998) indicated that the excess substrate could impact uncoupling between anabolism and catabolism, resulting in energy spilling. The authors proposed a concept of an energy uncoupling coefficient based on the observed growth yield and they showed an increasing trend of the energy uncoupling with the S_0/X_0 ratio.

FLOC CHARACTERISTICS AND FLOC DESTRUCTION MECHANISMS

Floc structure

Jorand et al. (1995) evaluated the complexity of the activated sludge floc structure using staining and the analysis of polymers released from sonicated sludges. The authors found that the main polymers released from the activated sludge flocs were proteins, polysaccharides and DNA as a function of sonication time. Proteins were the most abundant exocellular polymers released; carbohydrates and DNA released were similar and approximately one-third of the protein released. Frølund et al. (1996) also found the components of activated sludge flocs to be protein (46-52% of VS), humic compounds (18-23% of VS) and carbohydrate (17% of VS) using the cation exchange resin extraction technique. Nielsen and Keiding (1998) confirmed the presence of polysaccharides within the floc matrix by staining with Ruthenium Red

Higgins and Novak (1997) proposed a model of biofloculation in which a lectin-like protein binds polysaccharides that are cross-linked to adjacent proteins. The authors suggested that a majority of the bound protein may be classified as a lectin which is a nonenzymatic protein that binds sugar residues. The authors suggested that cross-linking polysaccharides, or cation bridging play an important role of the stabilization of the biopolymer network.

Murthy et al. (2000) showed that ferric iron has a high affinity for protein and demonstrated that iron (III) salts selectively coagulated solution protein produced from autothermal thermophilic

aerobic digesters. Vilge-Ritter et al. (1999) and Masion et al. (2000) observed that aluminum salts had a high affinity for polysaccharide in natural water.

Floc destruction mechanisms

Higgins and Novak (1997) proposed that the protein content of activated sludge may play a significant role in bioflocculation. They showed that the addition of protein-degrading enzymes resulted in a degradation of proteins and deterioration in dewatering; and the addition of a polysaccharide degrading enzyme resulted in no substantial change in dewatering properties. Novak et al. (2003) proposed that the large protein release during anaerobic digestion is the result of the loss of selective binding between proteins and ferric when iron is induced under anaerobic conditions.

Novak et al. (2003) proposed that there are different mechanisms of floc destruction between aerobic and anaerobic digestion. They quantified the release of biopolymer from sludges under both anaerobic and aerobic conditions and showed that the release of protein into solution was 4-5 times higher under anaerobic conditions than under aerobic conditions, although the VS destruction by anaerobic digestion was no higher than for aerobic digestion. The authors proposed that there are at least two types of biopolymer binding mechanisms in flocs; one fraction associated with divalent cations (Ca^{++} and Mg^{++}) that is degraded primarily under aerobic digestion and another fraction associated with iron that is degraded primarily under anaerobic digestion. The authors hypothesized that the accumulation of much more protein in the anaerobic system is likely due to a much greater amount of released protein from the floc under anaerobic conditions.

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KEY WORDS

Sludge production, Activated sludge, Anaerobic digestion, Aerobic digestion, floc, cations, iron, protein, polysaccharide, volatile solids destruction

INTRODUCTION

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suspended solids (SS) concentration was high at the lower values of SRT. As a result of dispersed growth of bacterial flocs at an SRT of 2 days or less, the settling velocity of activated sludge could be slow and washout could occur depending upon the characteristics of influents. At higher values of SRT, bacterial flocs also tended to deteriorate and disperse. The authors also found an accumulation of polysaccharide at long SRT values. Reece et al. (1979) found an increase of nonbiodegradable material in activated sludge as SRT increases, affecting the aerobic digestibility of sludge. Murthy (1998) showed similar results in that, as the SRT increased above 10 days, soluble polysaccharide and colloidal protein which resulted in an increase in effluent COD, but the BOD in the effluent remained constant. This implies that the increased effluent COD was nonbiodegradable organic material.

The reason why activated sludge is widely used is that it is flexible, reliable, can achieve a high degree of nitrification and can stabilize insoluble organic matter (Grady et al., 1998). However, one of its main disadvantages is the generation of waste sludge. Although the activated sludge process works efficiently, the growth of microorganisms results in the production of excess biomass that requires disposal to control the solids retention time (SRT). To treat the excess sludge, a combination of digestion, chemical treatment, dewatering, and thickening is generally used. The treatment for excess sludge can account for up to 60% of a plant's total operating cost (Horan et al., 1990) and also adds to the capital cost of the facility. Therefore, the reduction of sludge generation in the activated sludge system could be a rational strategy for the sludge problem.

Several strategies and technologies have been proposed to reduce excess sludge generation in biological wastewater treatment processes such as ozonation (Yasui, Shibata, 1994), addition of uncoupling chemical (Ye et al., 2003), and a thermophilic aerobic digestion called S-TE PROCESS (N. Shiota et al., 2002). The oxic-settling-anoxic process (OSA process) using the effect of a reduced oxidation-reduction potential (Saby et al., 2003) has been shown to be successful.

An innovative process, called the Cannibal™ Process, focusing on the further volatile solids (VS) destruction has been developed to reduce excess sludge generation. There are several variations

of the Cannibal™ Process, including the use of a low dissolved re-aeration unit that is coupled with the anaerobic bioreactor prior to return of the sludge to the aeration basin. The basic configuration for this process (Figure 1) utilizes an anaerobic unit through which recycled sludge passes prior to being returned to the aeration basin. This process has been shown in field operations to reduce solids by approximately 60 to 70%.

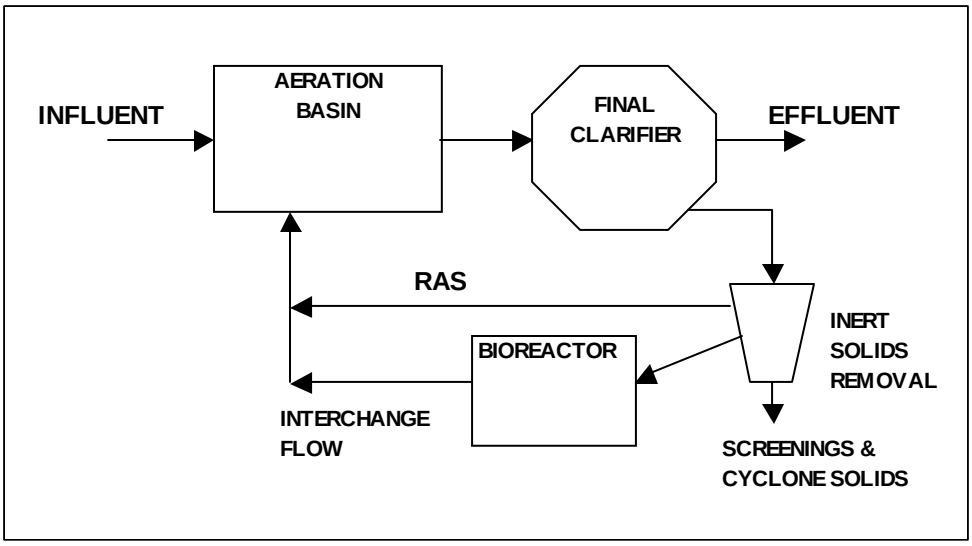

Figure 1: Flow Scheme for the Cannibal™ Process

Currently, anaerobic digestion is the primary process used to reduce the solids content of waste sludge and render it suitable for land application. Anaerobic digestion is basically comprised of three steps. In the first step, hydrolysis, particulate organic matter is converted into soluble substrates. In the second step, acidogenesis, the soluble substrates are broken down into acetic acid, carbon dioxide, and H_2. In the final step, methanogenesis, acetic acid and H_2 are converted into methane and carbon dioxide by H_2-oxidizing methanogens and aceticlastic methanogens. H_2-oxidizing methanogens are all strictly obligate anaerobes so they can obtain energy from the oxidation of H_2 and carbon from carbon dioxide. Through the evolution of methane, the chemical oxygen demand (COD) of wastewaters decrease and the stabilization of the biodegradable organic matter can be achieved (Metcalf and Eddy, 1991 and Grady et al., 1998).

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embedded. Jorand et al. (1995) described the complexity of the activated sludge floc structure using staining and the analysis of polymers released from sonicated sludges and found that the main polymers released from the activated sludge flocs were proteins, polysaccharides and DNA. Frølund et al. (1995) found the components of activated sludge flocs to be protein (46-52% of VS), humic compounds (18-23% of VS) and carbohydrate (17% of VS) using the cation exchange resin extraction technique.

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The main hypothesis in this study is that under anaerobic conditions, ferric ions will be reduced to ferrous ions and as a result degradable protein will be released into solution from flocs in the anaerobic reactor. This protein will then be degraded when the sludge is returned to the aerobic reactor (aeration basin). Through the anaerobic release and aerobic degradation of the released protein, excess sludge generation is reduced.

The purpose of this research is to determine the extent to which the Cannibal system can reduce excess sludge production and to evaluate the mechanisms that occur in the Cannibal™ Process that account for the loss of biomass. To achieve this, side-by-side a conventional and a Cannibal system were operated in the laboratory using sequencing batch reactors (SBR) and a synthetic feed. Items investigated included the influence of the exchange rate between the Cannibal aeration reactor and the Cannibal anaerobic sludge recycle bioreactor, the rate and release of biopolymer in the anaerobic reactor and the aerobic biodegradability of the biopolymer released in the anaerobic bioreactor.

METHODS AND MATERIALS

Experimental set-up

The reactor setup for the Cannibal system and the Conventional system that served as a Control are shown in Figure 2. Sequencing batch reactors (SBR) were used in this study to prevent filamentous bulking. The SBRs had a working volume of 4 liters and were operated at a fixed temperature of 20 ◦C. Mixing inside the reactor was achieved with a paddle mechanical stirrer (110 rpm). Master-flex tubing and connections were used. The SBR was operated at 4 cycles per day. The whole systems were automatically controlled by timers (ChronTrol). Each cycle comprised four steps (Feed, React, Settle, and Decant) for a total of 6 hours. There was a reaction step with aeration and mixing for 5 hours. In the settling step, the mixing and aeration were stopped and the biomass was allowed to settle for 40 min. Finally, in the decant step, the supernatant liquid was discharged from the reactor over a 10 min period. Initially, the Cannibal anaerobic bioreactor was purged with nitrogen to initiate anaerobic conditions. After the color turned black, nitrogen purging was stopped.

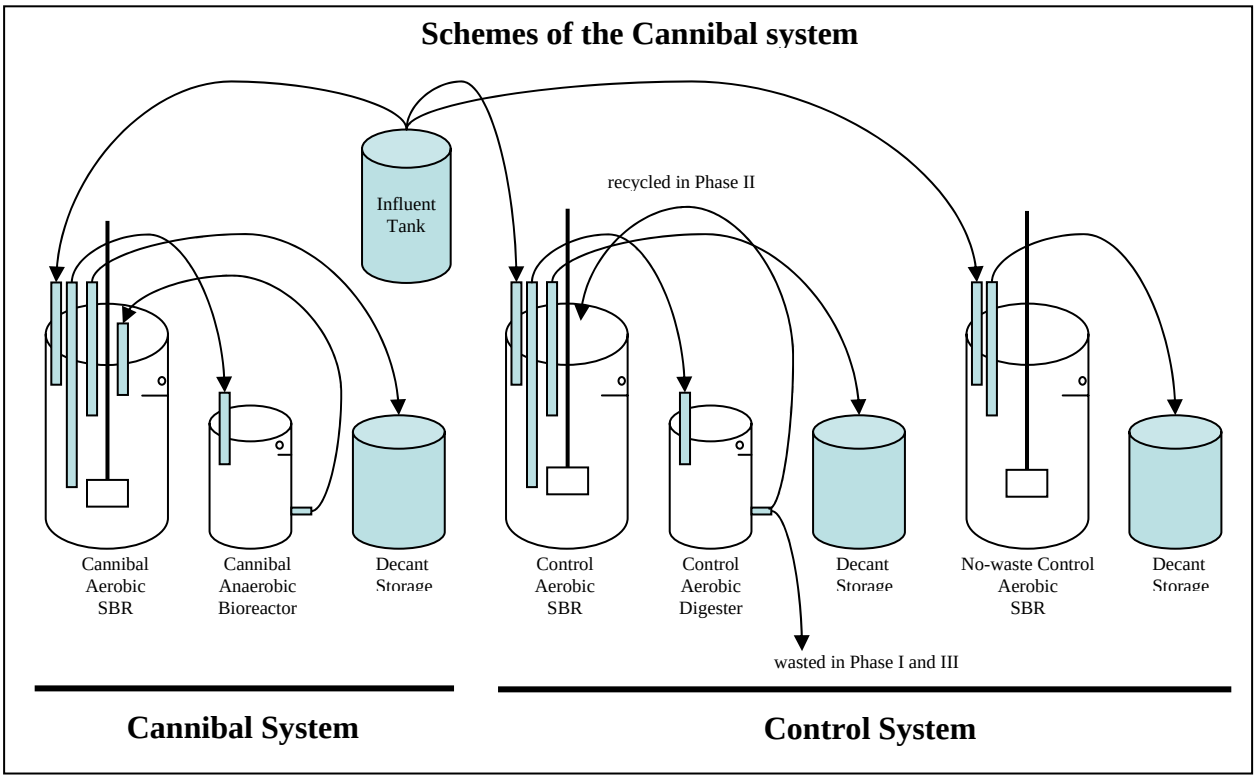


Figure 2: Schematic of the laboratory Cannibal and Control systems

Feed composition

The laboratory Cannibal and Control systems were fed with a synthetic influent containing a source of COD (acetate and Bactopeptone), as shown in Table 1. The synthetic influent was fed from a single feed tank to each reactor. The feed was continuously stirred for homogeneous feeding. In Phase I and II, the feed was made once every two days and in Phase III, the feed was made daily.

Table 1: Medium and trace element solution compositions

Medium Composition	Concentration (mg/L)	Trace element solution	Concentration (g/L)
BactoPeptone	300 mg COD /L	Citric acid	2.73
CH_3COONa	100 mg COD /L	Hippuric acid	2
NH_4Cl	57	$Na_3NTA.H_2O$	0.36
NH_4HCO_3	60	$Na_3EDTA.4H_2O$	1.5
KH_2PO_4	44	$FeCl_3.6H_2O$	1.5
$KHSO_4$	34	H_3BO_3	0.25
$NaHCO_3$	394	$ZnSO_4.7H_2O$	0.15
$CaCl_2.2H_2O$	220	$MnCl_2.4H_2O$	0.12
$MgSO_4.7H_2O$	150	$CuSO_4.5H_2O$	0.06
*$FeCl_3$	20	KI	0.03
*$Al_2(SO_4)_318H_2O$	20	$Na_2MoO_4.2H_2O$	0.03
Allylthiourea	1.4	$CoCl_2.6H_2O$	0.03
Trace element solution	2 ml/L	$NiCl_2.6H_2O$	0.03
		$Na_2WO_4.2H_2O$	0.03

* The concentration of $FeCl_3$ and $Al_2(SO_4)_318H_2O$ was changed from 10 mg/L to 20 mg/L on day 28 in Phase I.

Allythiourea (5 mg/L) was added to the Cannibal SBR, Control SBR and No-waste Control Aerobic SBR from day 12 to day 30. From day 28 in Phase III, the method of making feed was changed. The feed was divided into 4 parts, organic (BactoPeptone and CH_3COONa), inorganic (NH_4Cl, NH_4HCO_3, KH_2PO_4, $KHSO_4$, $NaHCO_3$, and allylthiourea), calcium and magnesium

($CaCl_2.2H_2O$, and $MgSO_4.7H_2O$), $FeCl_3$, $Al_2(SO_4)_3 18H_2O$, and trace elements. Each part of the feed was dissolved in distilled water before being mixed together. This was to prevent precipitation in the feed system.

Analysis

Samples for solution cations and anions analysis were centrifuged at 9000g for 30 minutes. The supernatant liquid was filtered through a 1.5 µm glass fiber filter. Dissolved cations and anions were measured using a Dionex ion chromatograph (IC). Methane sulfonic acid (30 mM) was used as the eluent at a flow rate of 1.0 ml/min.

Total solids (TS), total suspended solids (TSS), total volatile solids (VS), volatile suspended solids (VSS), and soluble COD were measured according to Standard Methods (APHA, 1995). The protein concentration was determined by the Hartree (1972) modification of the Lowry et al. (1951) method using bovine serum albumin as the standard. Polysaccharide was measured by the Dubois et al. (1956) method utilizing glucose as the standard. The pH and oxidation-reduction potential were measured using pH and ORP probes. The oxidation-reduction potential (ORP) was occasionally measured in Cannibal anaerobic bio-reactor. For the ORP measurement, Light's solution was used for calibration.

Phase I

Phase I was conducted for 57 days at 20^oC. During the first phase experiments, two SBRs were operated, a Cannibal and a Conventional system. For the Cannibal system, no solids were wasted purposely and the MLVSS in both systems were maintained at approximately the same MLVSS. The SBRs were fed 500ml each cycle for 10 min in the feeding step and this provided for a hydraulic retention time (HRT) of 2 days. The dissolved oxygen concentration was approximately 5 mg O_2/L using an air pump.

Two bio-reactors were operated for treatment of the biomass removed from the SBRs at the end of the settle phase, one anaerobic and one aerobic. The total volume of these bioreactors was 500ml and they were fed 50 ml per day. The flow from the anaerobic bioreactor was returned to the activated sludge system and the sludge from the aerobic bioreactor was wasted. In effect, the aerobic bioreactor served as an aerobic digester for the control system.

Phase II

In the second phase, the Cannibal and Conventional systems were operated in a manner similar to the first phase except that the contents of the aerobic bioreactor were returned to the activated sludge system. In effect, the Conventional system was converted to an aerobic Cannibal system.

Phase III

During the third phase experiments, three SBRs, Cannibal SBR, Control SBR, and No-waste Control Aerobic SBR were operated (Figure 2). The total volume of each SBR was 4 liter. Two bio-reactors were operated as in Phase I except the feed volume was doubled so the HRT decreased to 1 day. A third reactor was added with no wastage and no bioreactor. Initially, the waste to the bioreactors was kept at 50 ml. After noting that the solids in the activated sludge unit continued to rise, the waste volume was increased to 100 ml per day. The total volumes of the Cannibal anaerobic bioreactor and Control aerobic bioreactor were then changed to a volume of 1 liter.

Batch tests for investigating the solids destruction mechanism

At the end of Phase II, the specific oxygen uptake rate (SOUR) was measured using supernatant liquid from both the aerobic and anaerobic bioreactors. The purpose of these batch tests was to determine if material was released in the bioreactors that could then be readily degraded in the activated sludge system.

SOUR

The specific oxygen uptake rate was conducted to measure and compare the degradability of material released in Cannibal anaerobic bioreactor and Control aerobic bioreactor. The SOUR test was conducted in a 300ml BOD bottle. The test consisted of 200ml biomass from the Cannibal SBR, 50ml solution from the Cannibal anaerobic bioreactor and 50ml deionized water. A sludge sample from the Cannibal anaerobic bioreactor was centrifuged and then 50ml of this liquid was used for the test. After adding 50ml solution from the Cannibal anaerobic bioreactor to 200ml biomass from the Cannibal SBR and 50ml deionized water, the dissolved oxygen was measured for 600 seconds. Three batch tests were conducted as controls. The first control

consisted of 200ml biomass from Cannibal SBR, and 100ml deionized water. The second control consisted of 200ml biomass from Control SBR, 50ml solution from Control aerobic bioreactor, and 50ml deionized water. The third control consisted of 200ml biomass from Control SBR, 50ml solution from Control aerobic bioreactor, and 50ml deionized water. Three controls were measured for 1800 seconds.

Test for release of organic matter in the anaerobic and aerobic bio-reactors

To evaluate the release rate and mechanisms in the bioreactors, samples were taken from Cannibal anaerobic bioreactor and Control aerobic bioreactor at the end of Phase II and then centrifuged for 10 minutes at 10000 rpm. The centrate was then filtered through a 1.5 μm filter. Analyses of protein, polysaccharide, and cations were conducted using filtered centrate.

Batch Test for determining the optimum bioreactor detention time (the second batch test)

One major consideration in this research was how quickly the iron would be reduced in the anaerobic bioreactor and how rapidly organic matter would be released. To accomplish this, at the end of phase III, thickened sludges were added to the sludge in the bioreactors and the change in MLSS, VSS, solution protein, solution polysaccharide, anions and cations were measured over a period of 100 hours.

Table 2: Proportions of sludge types in the batch tests

Batch name	Anaerobic biomass (ml)	Added biomass (ml)	Proportion (%)
ANA 1 (7%)	715	50	7
ANA 2 (13%)	700	100	13
ANA 3 (26%)	500	180	26
ANA 4 (41%)	500	350	41
Batch name	Aerobic biomass (ml)	Added biomass (ml)	Proportion (%)
AER 1 (17%)	500	100	17
AER 2 (17%)	500	100	17

Different amounts (7, 13, 26, and 41%) of thickened sludge from the activated sludge system were added to the anaerobic bioreactor sludge (Table 2). The biomass in the anaerobic bioreactor was used for the anaerobic biomass of ANA 1. The anaerobic biomass of the others (ANA 2, 3, and 4) consisted of biomass that was collected from Cannibal SBR and Control SBR and allowed to sit under anaerobic conditions for 15 days. This was done in order to supply enough anaerobic biomass for the tests. The aerobic biomass was thickened activated sludge from Cannibal SBR and Control SBR. The aerobic biomass was aerated for 15 day prior to being added to the anaerobic biomass. When samples were taken from the anaerobic batch reactor, nitrogen purging was used and the pH was measured.

RESULTS AND DISCUSSION

PERFORMANCE

Phase I

The purpose of Phase I was to compare the performance of the Cannibal system with a Conventional system with an aerobic digester.

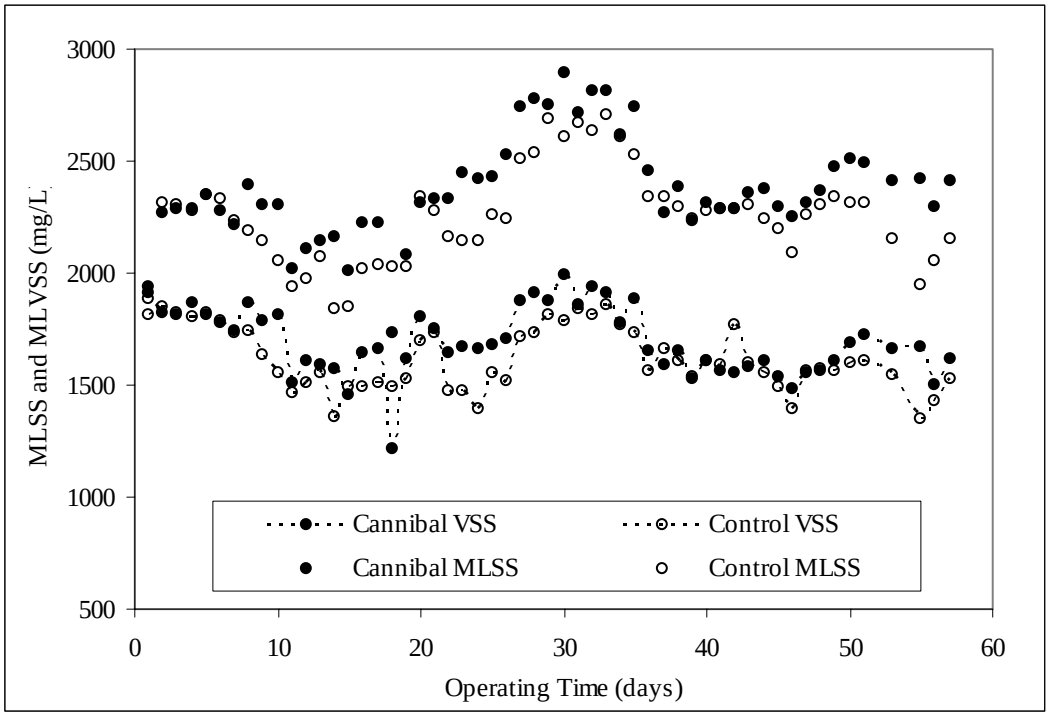


Figure 3: MLSS and MLVSS, Phase I

The MLSS and VSS in Phase I are shown in Figure 3. Both the Cannibal and Conventional systems were maintained at a MLSS of about 2500 and a MLVSS of about 1500 mg/L, respectively. The SRT of both systems was not fixed. There was a one time intentional waste of biomass from both systems on day 10. After increasing the aeration on day 20, the effluent quality improved. Both the Cannibal and Conventional systems produced low effluent suspended solids, especially after day 37.

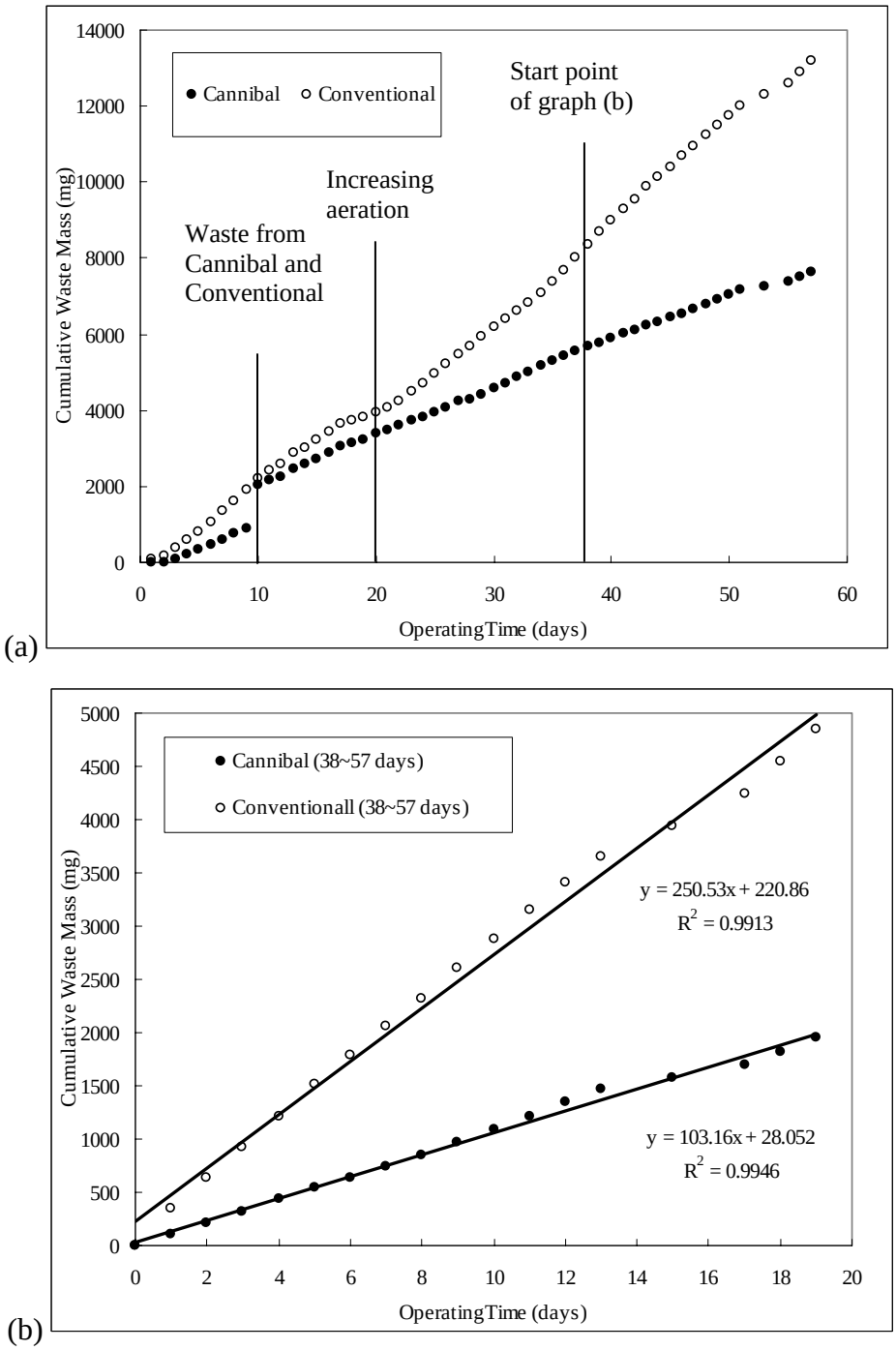


Figure 4: Cumulative solids wastage from the Cannibal and control systems, Phase I, a) entire operating time and b) day 38 to 57.

Data for the comparative cumulative waste mass is shown in Figure 4. Once the reactors reached a steady state operation with regard to MLVSS, the Conventional system produced about 60% more biomass than the Cannibal system. In Phase I, the total influent loading as COD was 33.3 g COD for the Cannibal and 34.0 g COD for the Conventional system. There were three sources of

waste solids; effluent solids, solids removed for analysis and solids intentionally wasted to maintain a constant MLVSS. During the initial period of operation, some deterioration in effluent quality was observed in both of the Cannibal and Conventional systems due to inadequate aeration. As shown in Figure 7, the effluent suspended solids improved greatly, shortly after increasing the aeration on day 20. Another reason of the improvement in the effluent suspended solids might be the increase of the concentration of aluminum and iron from 10 mg/L to 20 mg/L in the influent at day 28. Aluminum and iron were added to make these values similar to conventional municipal wastewater.

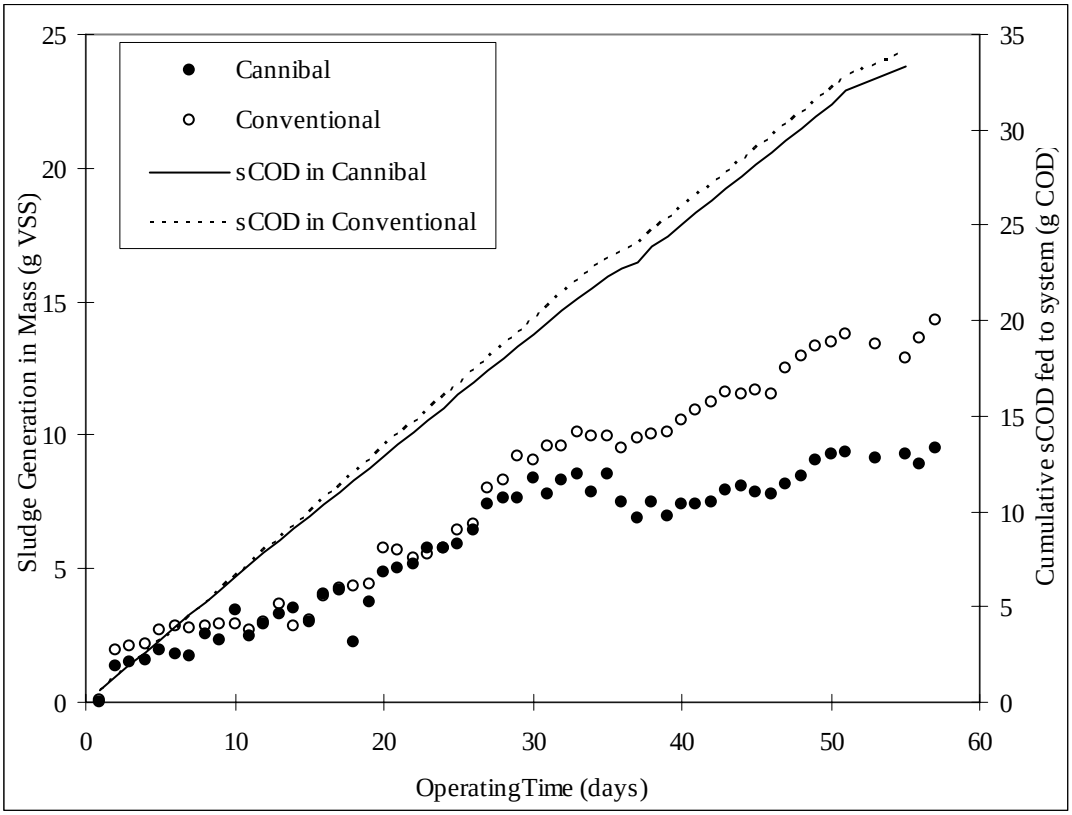


Figure 5: Sludge generation in the Cannibal and control systems, Phase I

Sludge generation from the Cannibal system was less than from the conventional system after day 27 (Figure 5). It could also be that a period of acclimation was needed before the Cannibal system operated effectively. After day 37, the solids production in the Cannibal system was consistently at 40% of the Conventional system (Figure 5).

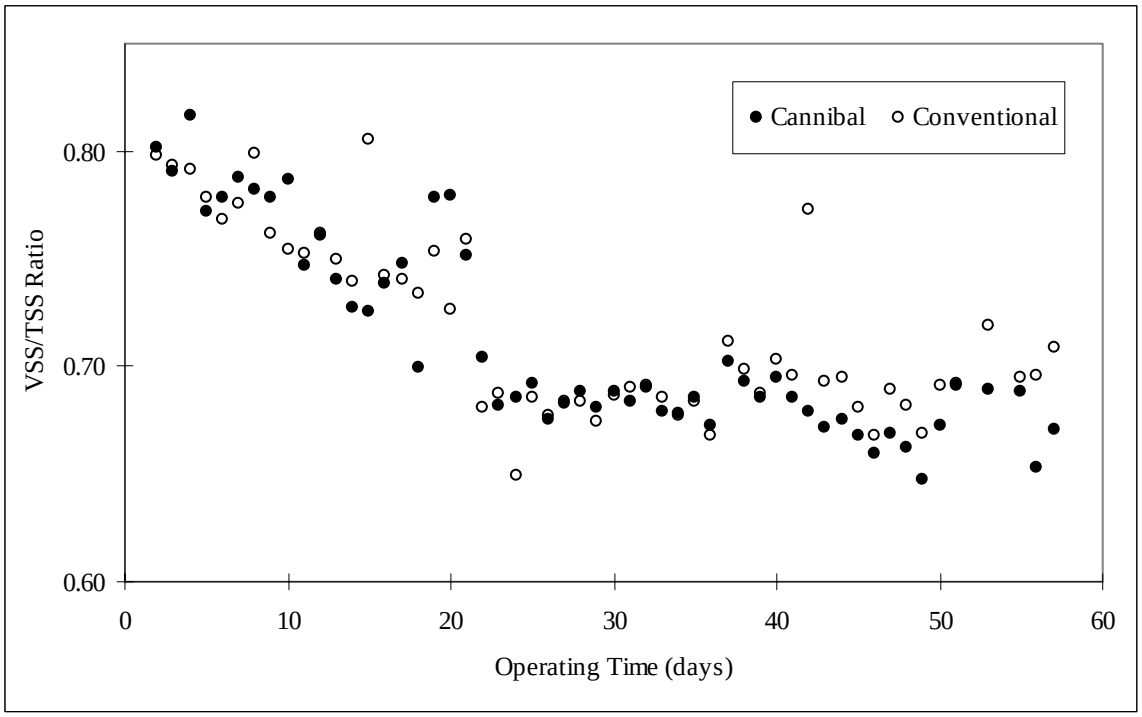


Figure 6: VSS versus TSS ratio, Phase I

The VSS/TSS ratio continued to drop until about day 25 and then leveled off at about 0.67 for the Cannibal and 0.71 for the Conventional system (Figure 6). The decrease of VSS/TSS indicates that inorganic material was accumulating in both systems. These differences reflect the decline in the organic fraction of the sludge and also likely reflect the high SRT for both activated sludge systems. The lower volatile fraction in the Cannibal system possibly reflects the more inert floc material that occurs in the absence of sludge wastage.

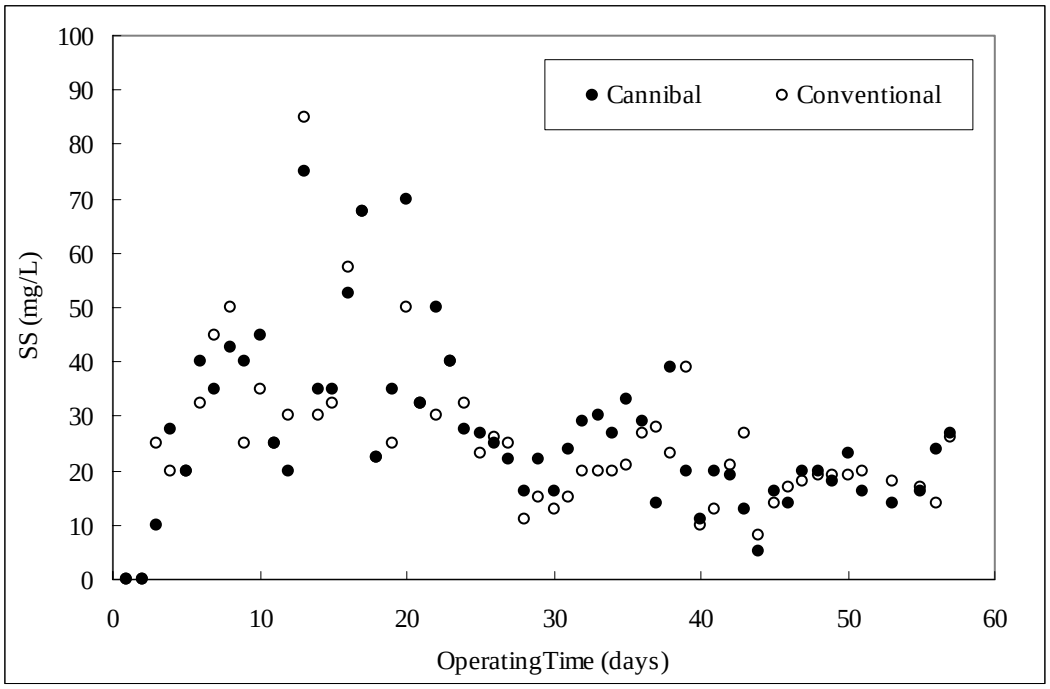


Figure 7: Suspended Solids in Effluent, Phase I

The effluent quality improved after increasing the aeration on day 20 (Figure 7). Both the Cannibal and Conventional systems produced low effluent suspended solids, especially after day 37. The effluent suspended solids of approximately 20 mg/L represents low values, especially for a laboratory reactor system where the settling zone is small and the settling time is less than for a conventional full-scale system.

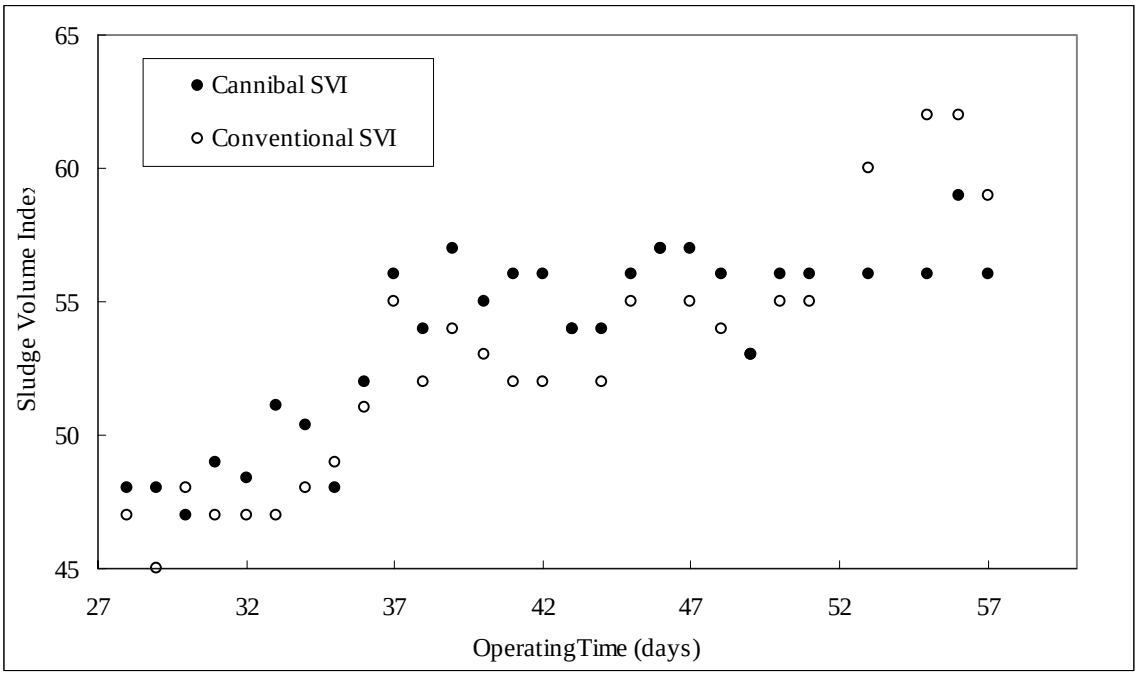


Figure 8: Sludge Volume Index, Phase I

The thickened sludge in both activated sludge systems was fed to the bioreactors when the SBR systems completed the settling cycle. Therefore, the settling characteristics of the sludges impacted the mass interchange rate because the flow rate of settled activated sludge to the bioreactor was fixed without regard to sludge compaction. As with the other parameters, the sludge volume index (SVI) reached a steady state at about day 37 (Figure 8). The Conventional system was slightly better than the Cannibal system with regard to sludge compaction but both sludges were excellent. A SVI of near 60 is considered to be excellent. Both systems were in that range.

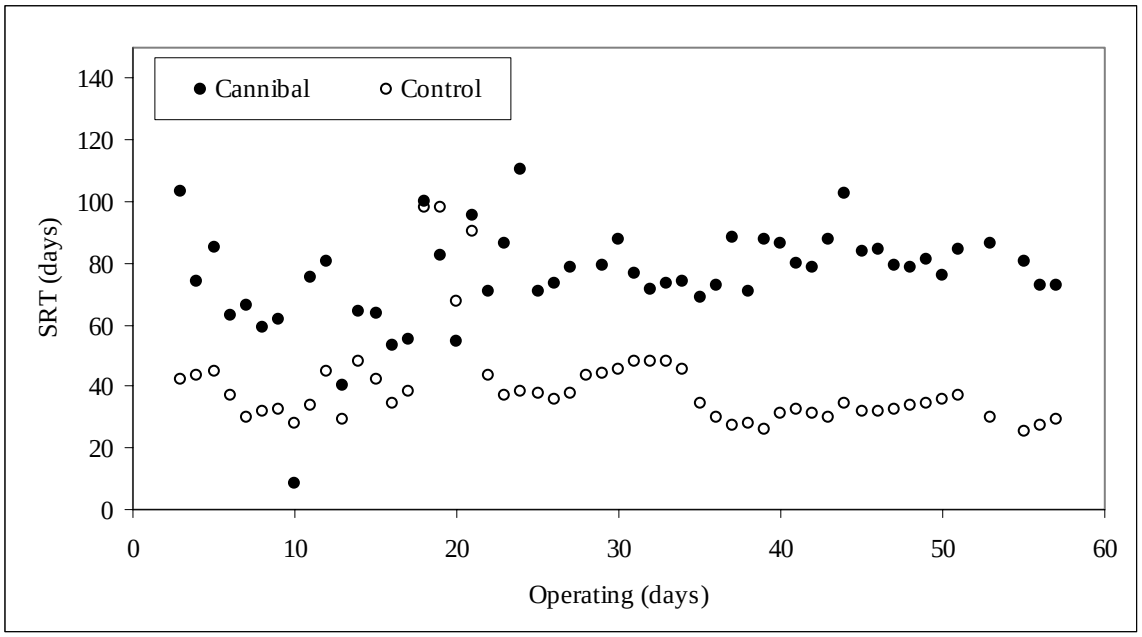


Figure 9: Sludge Retention Time in the Two Systems in Phase I

In Figure 9, the solids retention time (SRT) in Phase I is shown. Due to the lower wastage from the Cannibal system, the SRT for the Cannibal system was about 78 days as compared to the Conventional system about 40 days. At the end of this phase, the color of sludge in Cannibal SBR was darker brown than the Conventional system. This might be because some of the black precipitates (FeS) from the anaerobic bioreactor were not completely oxidized in the activated sludge system.

Phase II

The objective of Phase II was the comparison of performance between the Cannibal system and the "Aerobic Cannibal" system that had sludge returned from the aerobic bioreactor to the activated sludge system.

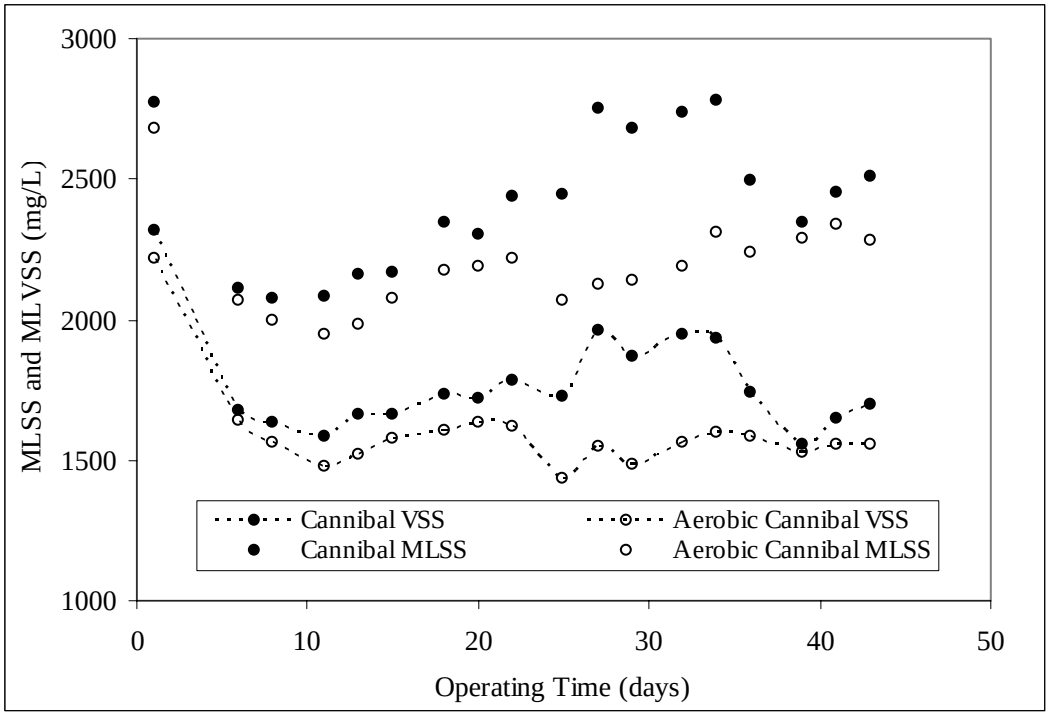


Figure 10: MLSS and MLVSS, Phase II

Phase II was started using thickened biomass from the secondary clarifier at the Blacksburg Wastewater Treatment Plant and operated in the same manner as Phase I. The MLSS and VSS are shown in Figure 10. The MLSS and MLVSS of the Cannibal and Aerobic Cannibal systems in Phase II were maintained at about 2300 and 1500 mg/L, respectively. There was no intentional waste of biomass from either system. Samples were taken 3 times per week.

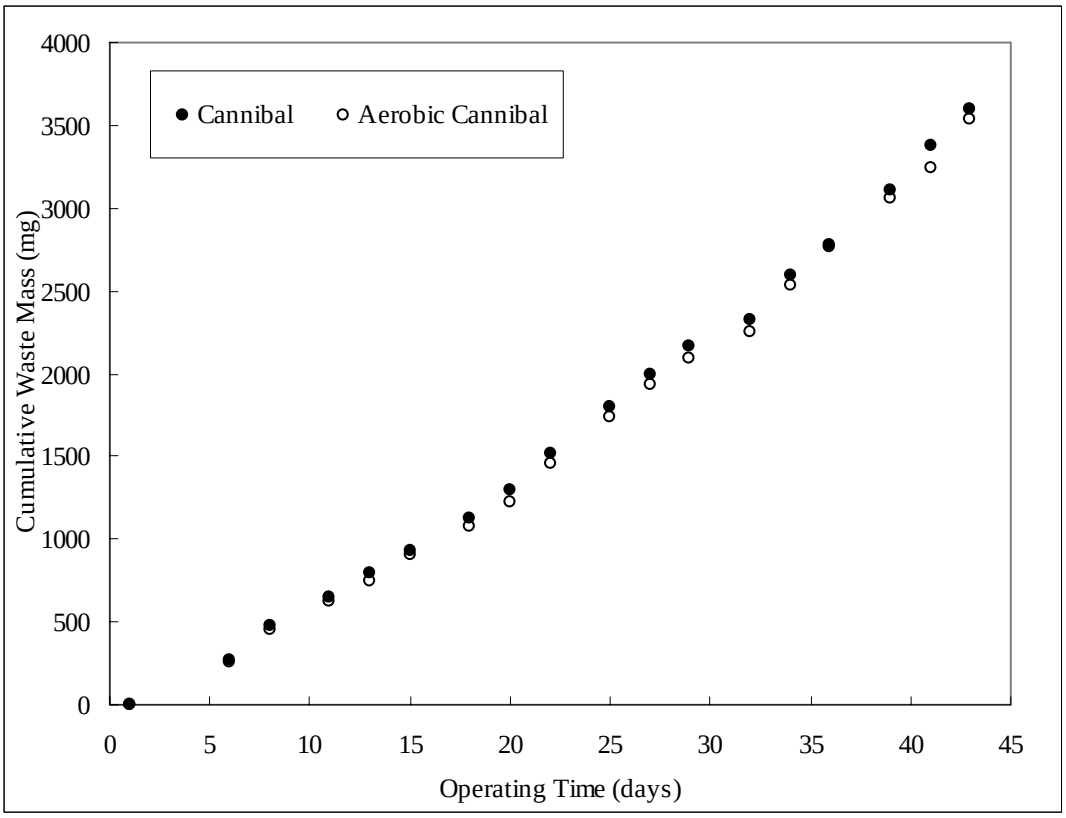


Figure 11: Cumulative solids wastage from the Cannibal and Aerobic Cannibal, Phase II

In Phase II, the total influent load as COD for 43 days was 24.9 g COD and 25.8 g COD into the Cannibal SBR and Control SBR, respectively. As with Phase I, there were three sources of waste solids, effluent solids, solids sampled for analysis and solids intentionally wasted to maintain the MLSS concentration in SBR. The two systems performed similarly, with low amounts of sludge being generated (Figure 11). This indicates that the "Aerobic Cannibal" system also destroys solids as a result of passage through the aerobic bioreactor. It is thought that in the aerobic bioreactor, protein is degraded and polysaccharides are released into solution. These polysaccharides are cycled back to the activated sludge unit where they are likely reincorporated into the flocs rather than degraded. It is thought that the Aerobic Cannibal system is fundamentally different from the Anaerobic Cannibal system. In the Anaerobic Cannibal, iron is reduced and this results in the release of biopolymer from floc that can then be degraded in the activated sludge system. For the Aerobic Cannibal, it is thought that biopolymer is directly degraded in the bioreactor. Therefore, the function of the bioreactor in the Aerobic Cannibal

system is VS destruction and in the Anaerobic Cannibal system it is the release of biopolymer. Research is continuing on this aspect of the study.

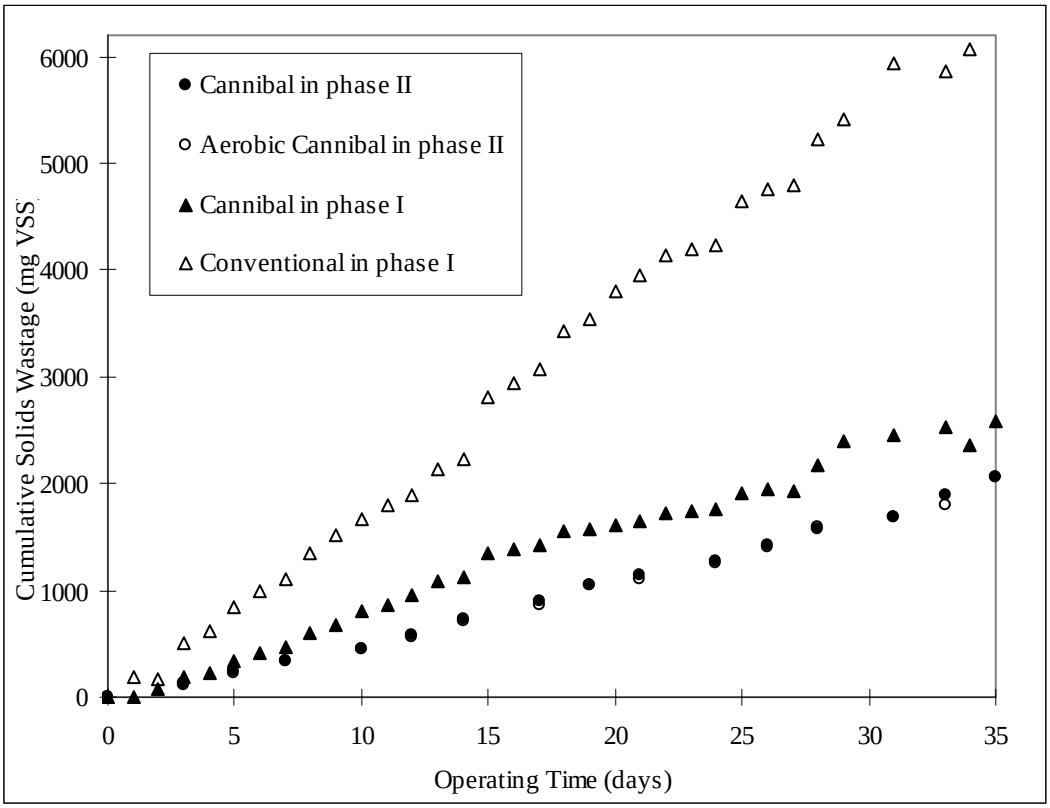


Figure 12: Comparison of phase I and II in cumulative solids wastage

Due to the small amount of sludge generation in the Cannibal system in phase I and II, and the Aerobic Cannibal system as compared to the conventional system, cumulative solids wastages in the Cannibal system in phase I and II, and the Aerobic Cannibal system were about 60% less than for the conventional system in phase I (Figure 12). The operation condition was 2 days of HRT in phase I and II. The solids wastage to maintain the specific MLVSS in phase I and II was only for sampling and effluent SS. After steady state was reached on about day 30 in the Cannibal systems in phase I and II, accumulation of sludge ceased. It is thought that the proper operation of the Cannibal™ Process requires time for anaerobic conditions to develop and the bioreactor to begin operating properly.

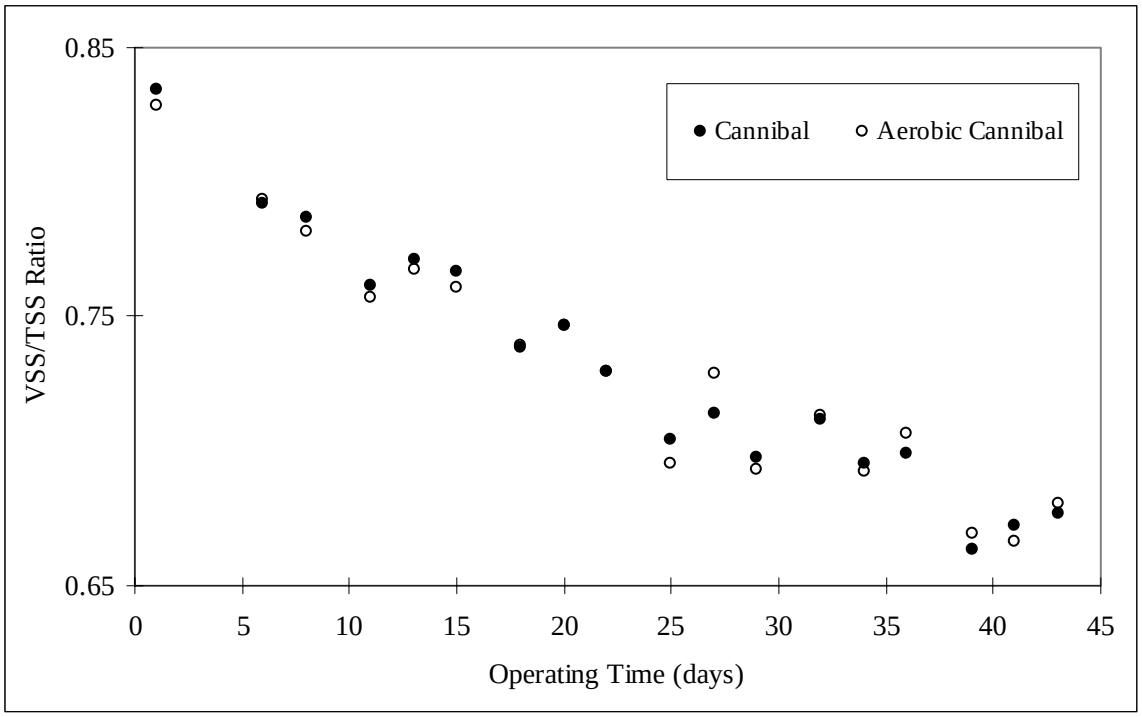


Figure 13: VSS versus TSS ratio, Phase II

After steady state was reached on about day 37, the VSS/TSS ratio of both systems was about 0.68 and reflects decline in the organic fraction of sludge (Figure 13). The similarity of the two ratios indicates that both were destroying solids in an equivalent manner. The lower volatile fraction in both systems possibly reflects the more inert floc material that occurs in the absence of sludge wastage.

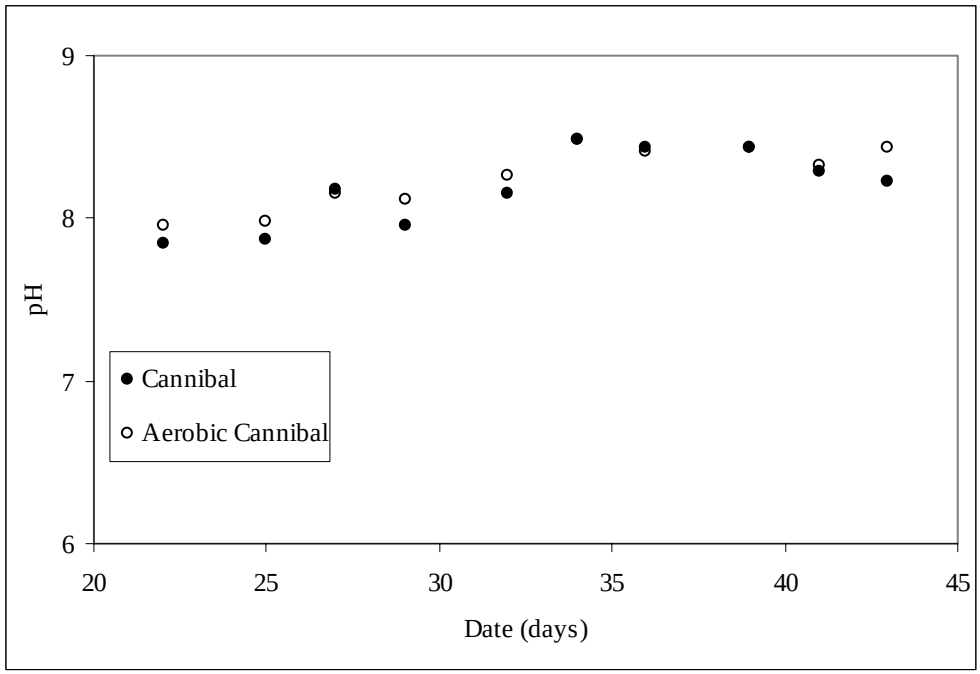


Figure 14: pH, Phase II

The pH data for the reactors is shown in Figure 14. For inhibition of nitrification, allythiourea (10 mg/L) was added to Cannibal SBR and Control SBR from day 20 to day 41. After adding allythiourea, some deterioration in effluent suspended solids was observed. This may have been due to a shift in the microbial population that resulted in the temporary loss of some flocculating bacteria.

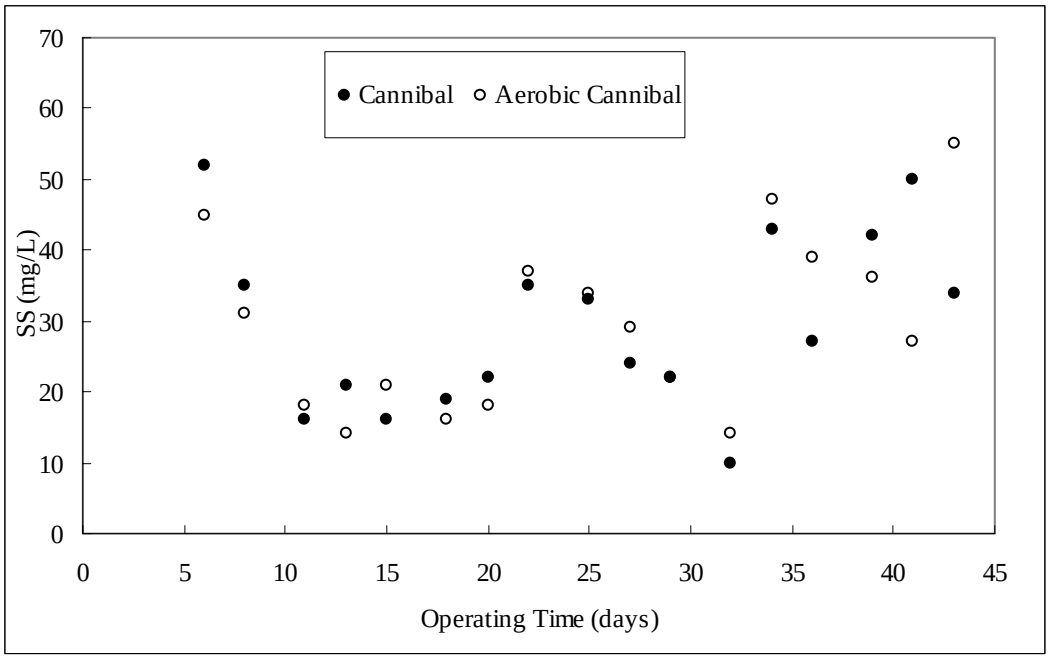


Figure 15: Suspended Solids in Effluent, Phase II

The effluent suspended solids are shown in Figure 15. Nitrification was inhibited in Phase II so the pH would not decline. For most of the experiment, the effluent suspended solids were similar to those from Phase I. However, beginning on day 34, the solids increased. It is not clear why that occurred, but it might be associated with the elevated pH.

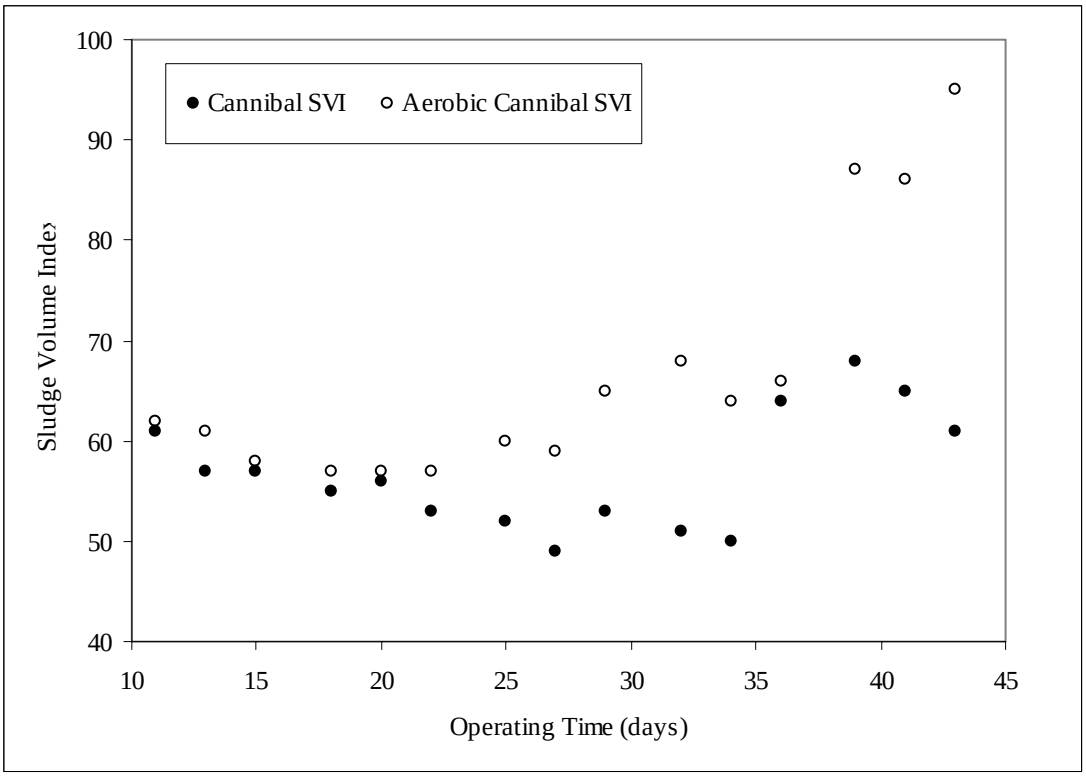


Figure 16: Sludge Volume Index, Phase II

The SVI over time is presented in Figure 16. In a manner similar to the effluent suspended solids, the SVI also increased beginning on day 34. However, in this case, the traditional Cannibal system had the better SVI and the SVI for both sludges was less than 100, indicating a good settling sludge. No explanation for the response was identified.

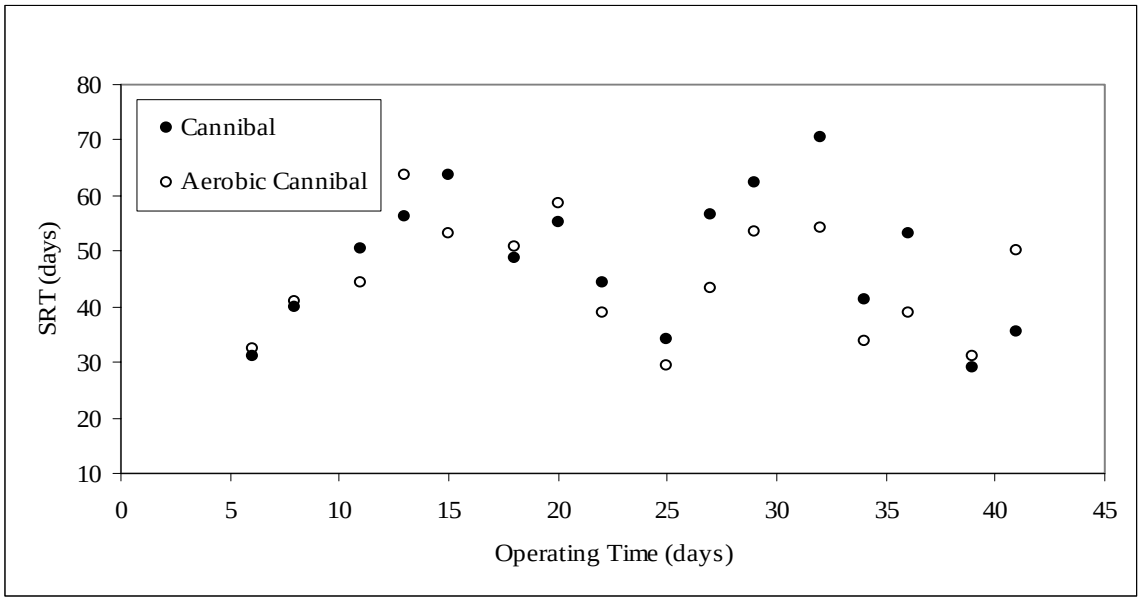


Figure 17: Sludge Retention Time, Phase II

Sludge Retention Time in Phase II is shown in Figure 17. The SRT for both systems was similar, in the range of 30 to 60 days, due to the similar wastage from both systems. At the end of this phase, the color of sludge in Cannibal SBR was also darker brown than in Control SBR.

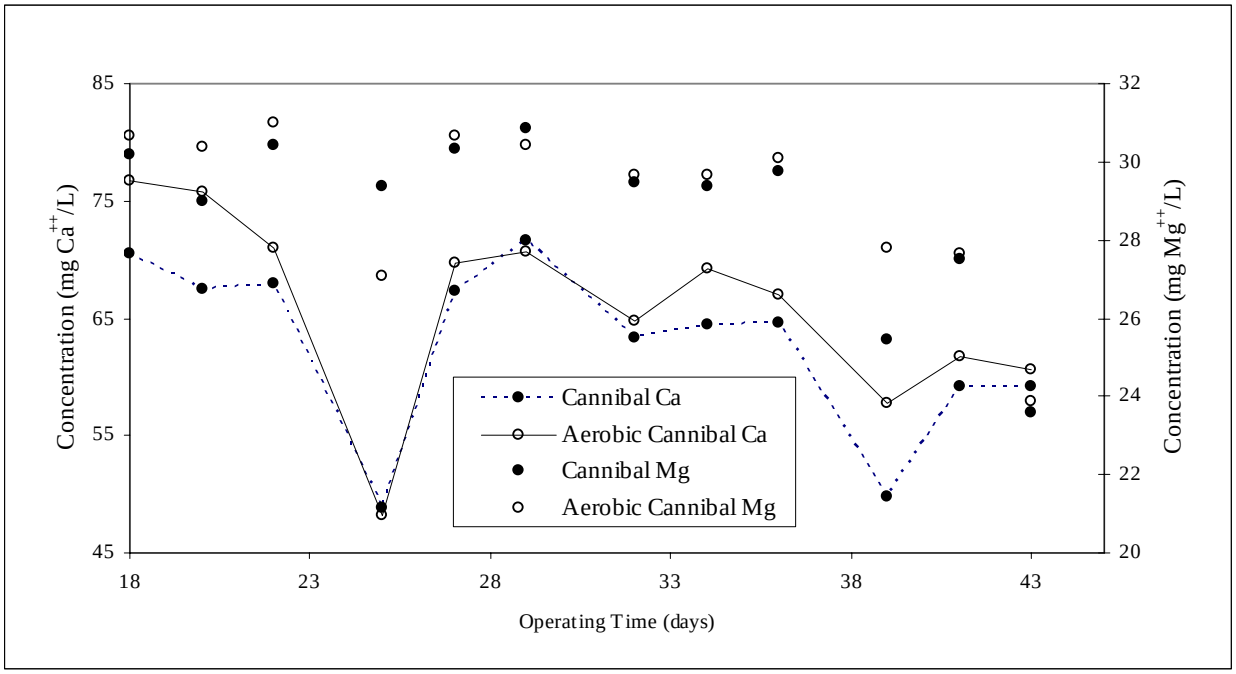


Figure 18: The concentration of Ca^{++} and Mg^{++}, Phase II

The concentration of Ca^{++}, shown in Figure 18, in effluent of control system or Aerobic Cannibal system was slightly higher than that of the Cannibal system. It shows that Ca^{++} was released in the bioreactor and then returned to the aeration basin. Although a relatively high concentration of Ca^{++} was in the bioreactor for the Aerobic Cannibal system, the recycled volume from bioreactor was small compared to the volume of the activated sludge system. Therefore, a slightly higher concentration of Ca^{++} in the effluent of Aerobic Cannibal system results from the dilution between the bioreactor and activated sludge system. It has been shown by Novak et al. (2003) that during aerobic digestion, both polysaccharides and divalent cations (Ca^{++} and Mg^{++}) are released. This indicates that the bioreactor for the aerobic Cannibal system acts like an aerobic digester. It also suggests that there is a fundamental difference in the solids destruction mechanisms between the two types of Cannibal systems.

Phase III

In Phase III, an additional Control system that had no intentional waste or recycle sludge reactor was used. This Control system provides for an additional control to help determine if the decrease in solids production was simply due to the higher SRT of the systems. The HRT was also decreased to 1 day in Phase III. The objectives of Phase III were to study the performance of the Cannibal system at an HRT of 1 day and to determine the effects of the interchange rate of sludge between the activated sludge system and the bioreactor. The interchange rate is the mass fed to the bioreactor to the total mass in the activated sludge system. The interchange rate was about 10% at the start of Phase III.

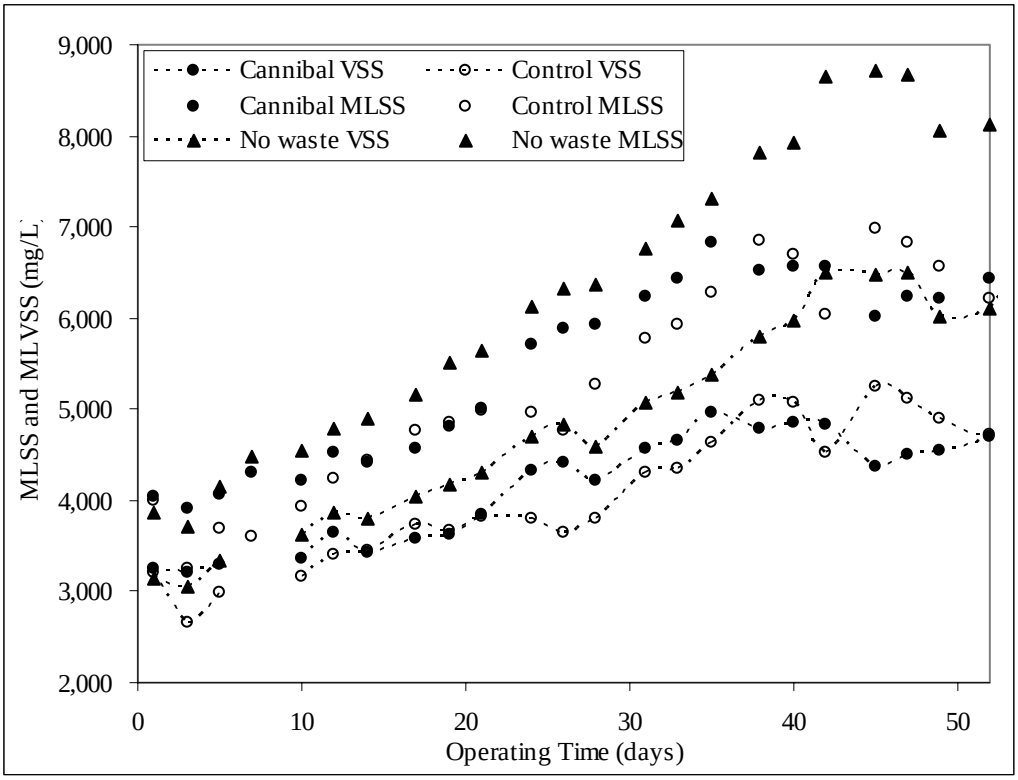


Figure 19: MLSS and MLVSS, Phase III

Due to the reduced HRT, the MLSS and MLVSS increased rapidly (Figure 19). The volume of sludge fed to Cannibal anaerobic bioreactor was initially not changed. The difference between the MLSS and MLVSS in three systems was not great until the interchange rate was increased. After increasing the interchange rate, the sludge stopped accumulating in the Cannibal system

and the difference in sludge production between the Cannibal, the control with an aerobic digester and the no-waste system became clear.

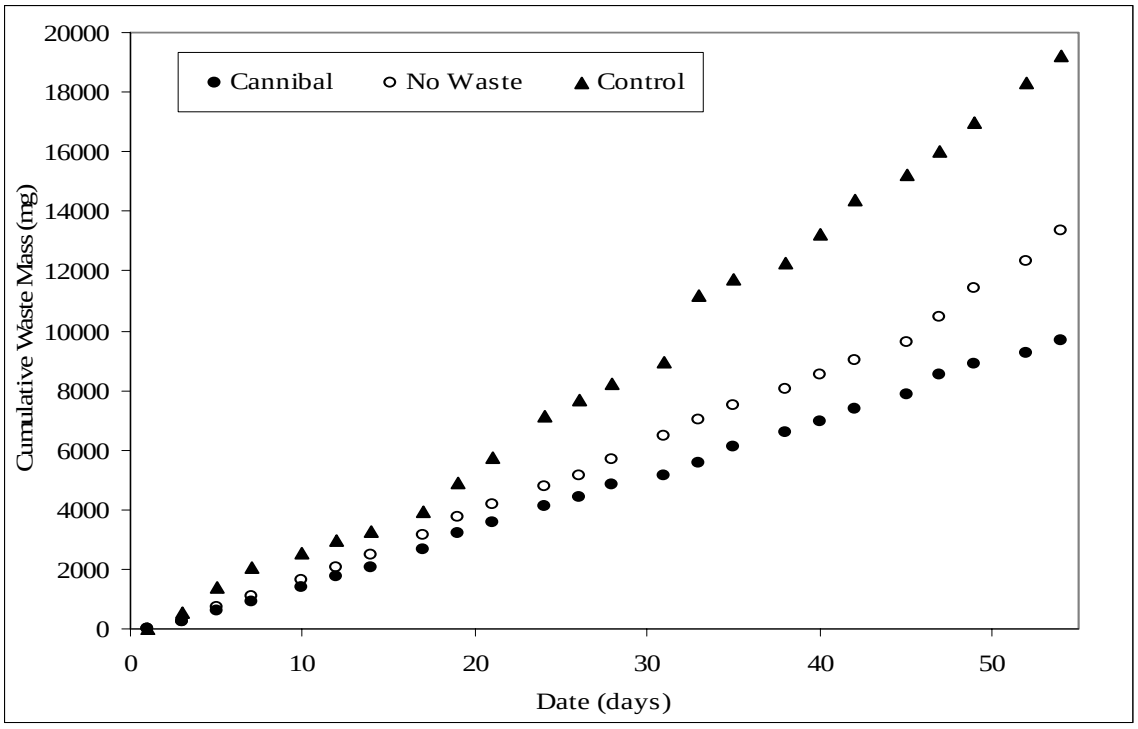


Figure 20: Cumulative solids wastage from the Cannibal and control systems, Phase III

In Phase III, the total influent load as a COD for 54 days were 60.9 g COD, 68.2 g COD and 60.6 g COD into Cannibal SBR, Control SBR, and No-waste Control Aerobic SBR, respectively. In the same manner as with Phase I and II, there were three sources of waste solids; effluent solids, sampling solids for analysis and solids intentionally wasted. The Control system had the most solids wasted and the Cannibal system had the least (Figure 20). It should be noted that in addition to solids wasted, solids were also accumulating in the systems, especially the no-waste system.

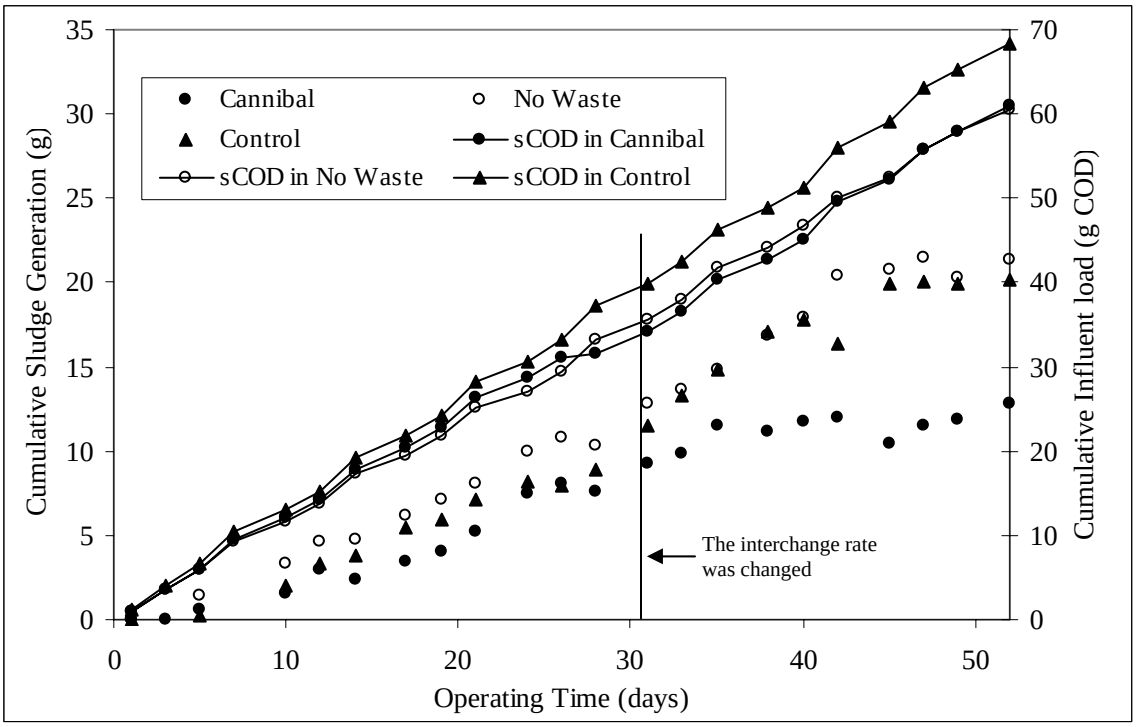


Figure 21: Cumulative VSS generation in Phase III

When the wasted solids were added to the accumulated solids, the total as cumulative sludge generation clearly indicates that the Cannibal system produces the lowest amount of solids (Figure 21). This is important because the no-waste system has a very high SRT and one consideration of this phase of the experiment was to show that the solids reduction was not due to the increase in SRT associated with the Cannibal system.

Due to the doubled COD load, the sludge generation in Phase III was faster than Phase I and II. The volume of sludge going to Cannibal anaerobic bioreactor was initially not changed so the interchange rate, the rate of sludge transferred to the bioreactors system per day as a percent of the total mass in the system, decreased from about 10% in Phase I to 5% in Phase III. As a result, sludge accumulated in all the reactors up to day 32 because the interchange rate between Cannibal SBR and Cannibal anaerobic bioreactor was low. Therefore, the Cannibal system did not realize its potential for solids destruction. Although it produced less solids than the other systems, the difference was not great. After day 32, the volume of the Cannibal bioreactor was increased in a step fashion until the interchange rate reached about 10%. After increasing the interchange rate, the sludge stopped accumulating in the Cannibal system and the difference in

sludge production between the Cannibal, Control with and aerobic digester and the no-waste system became clear. Much less solids were generated in the Cannibal system.

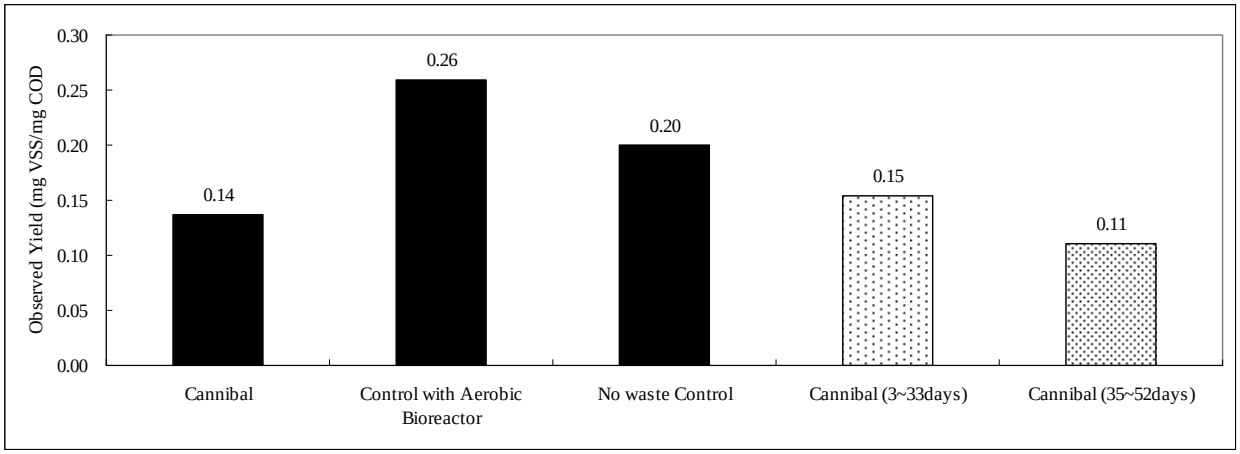


Figure 22: Observed Yields in Phase III

As a result of less sludge generation in the Cannibal system, the observed yield of the Cannibal system was less than for the others systems (Figure 22). This is especially true for the period from 35 to 52 days when the interchange rate was about 15%. The observed yield over that time was 0.11 mgVSS/mgCOD for the Cannibal system.

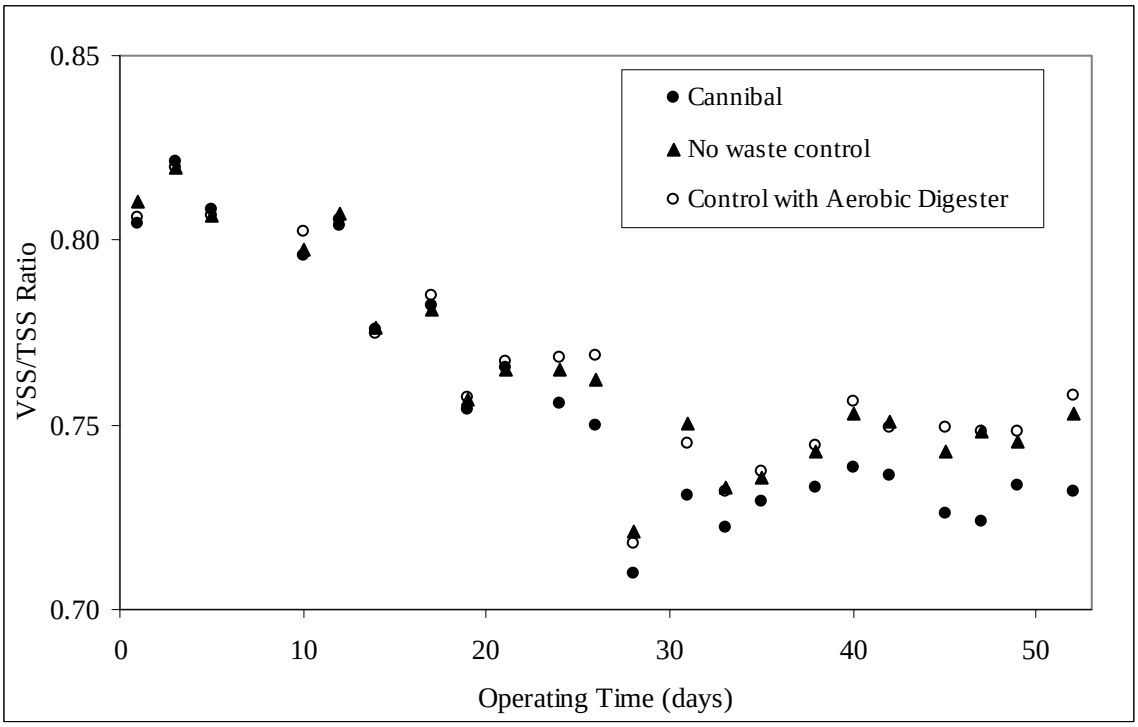


Figure 23: VSS versus TSS ratio in Phase III

As indicated in Figure 23, the VSS/TSS ratio of the Cannibal system was below the other systems. This indicated that inorganic matter was accumulated in Cannibal SBR and effective VS destruction occurred in the Cannibal system. After increasing the interchange rate, the difference in the VSS/TSS ratio became clear. It indicates that VS destruction occurs at the desired level when the optimal interchange rate is adapted to the system. It also suggests that additional VS destruction is possible based on the Phase I VSS/TSS ratio of 0.67.

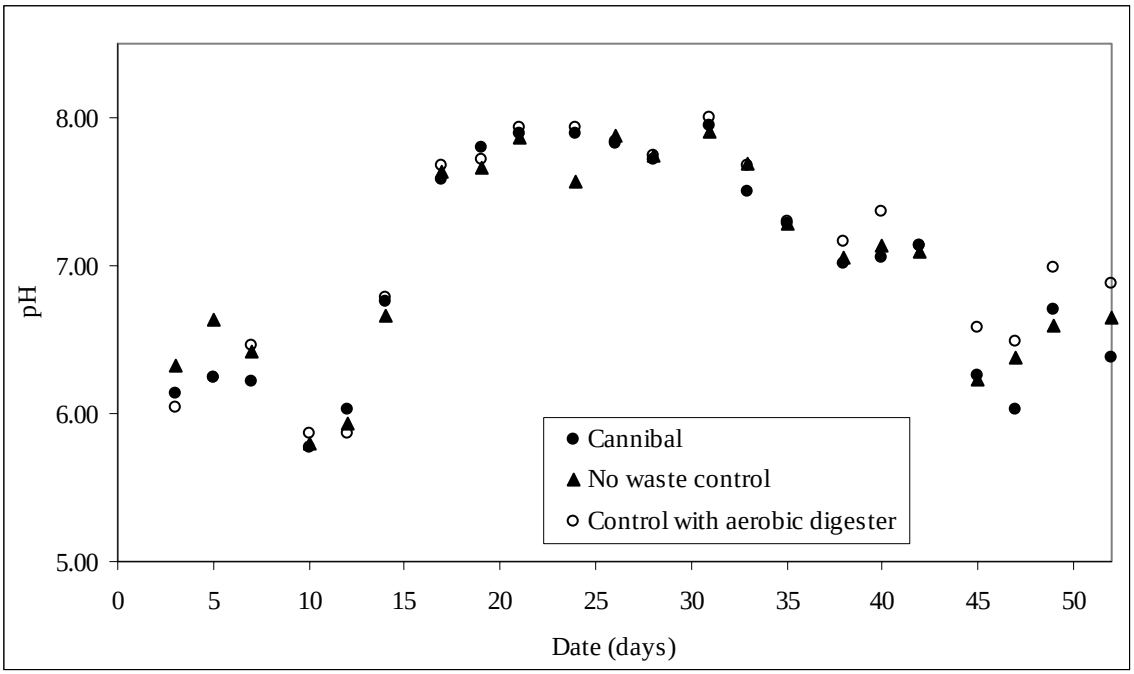


Figure 24: pH, Phase III

Nitrification resulted in low pH as shown in Figure 24 so allythiourea (5 mg/L) was added to Cannibal SBR, Control SBR and No-waste Control Aerobic SBR from day 12 to day 30. From day 28 in Phase III, the method for making feed was changed. The feed was divided into 4 parts, organic (BactoPeptone and CH_3COONa), inorganic (NH_4Cl, NH_4HCO_3, KH_2PO_4, $KHSO_4$, $NaHCO_3$, and allylthiourea), calcium and magnesium ($CaCl_2.2H_2O$, and $MgSO_4.7H_2O$), $FeCl_3$, $Al_2(SO_4)_3 \cdot 18H_2O$, and trace elements. Each part of the feed was dissolved in distilled water before being mixed together to prevent precipitation in the feed system.

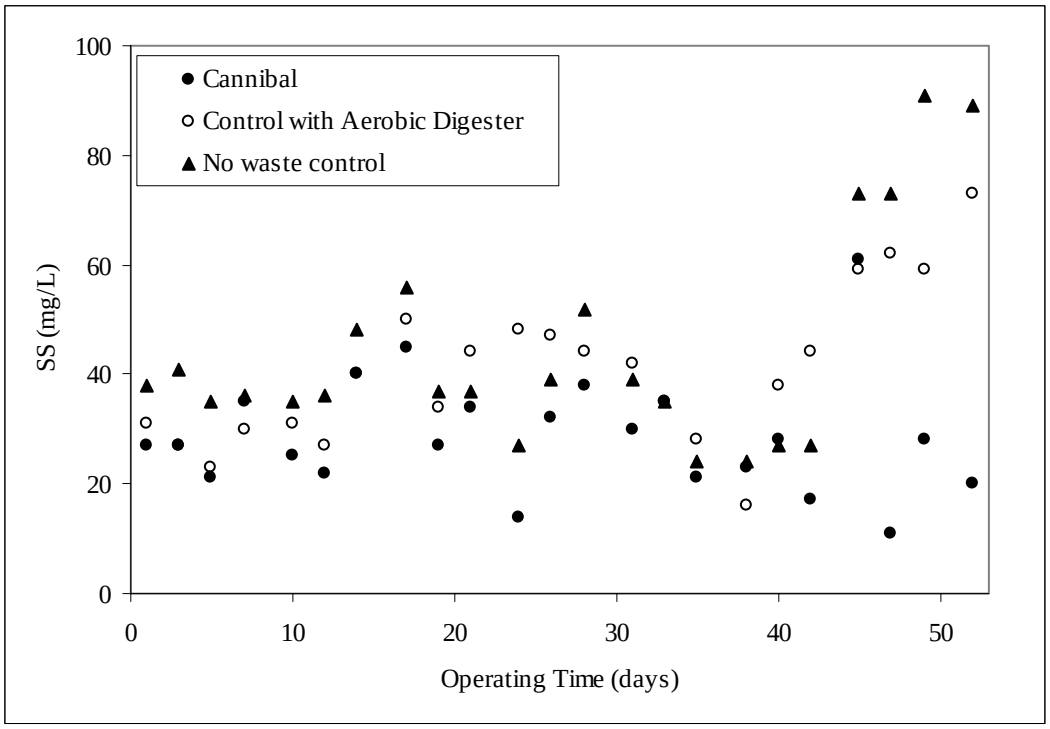


Figure 25: Suspended Solids in Effluent, Phase III

The effluent suspended solids increased for the two Control systems near the end of the run, but the SS for the Cannibal system remained low (Figure 25). It is not clear why this occurred, but it could be associated with the drop in pH to near 6.

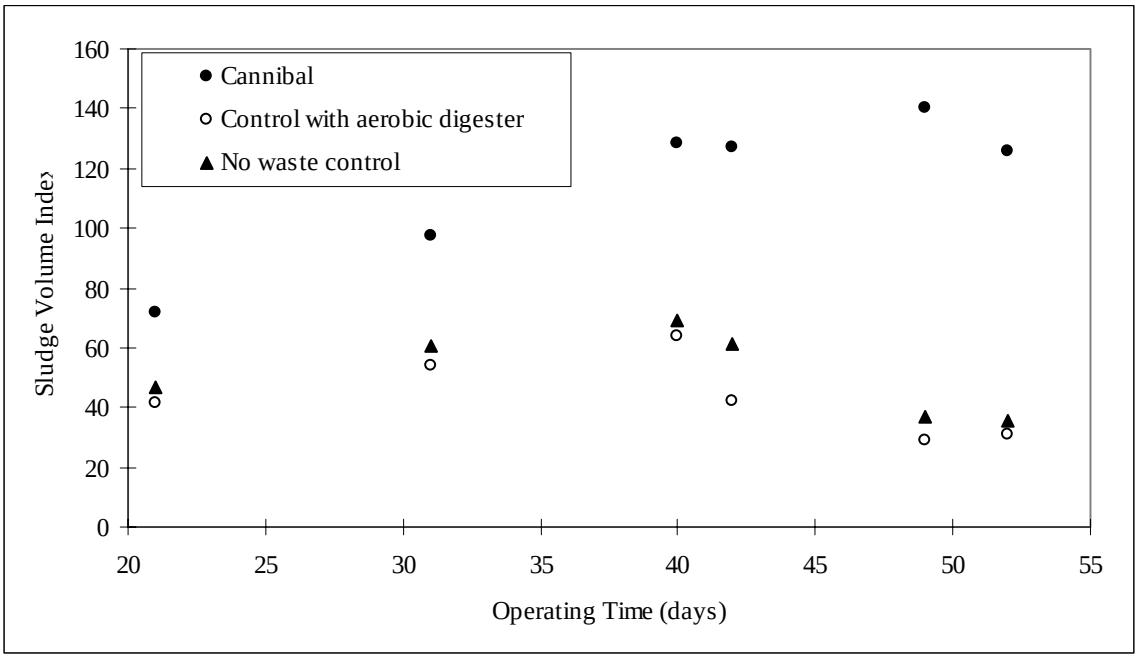
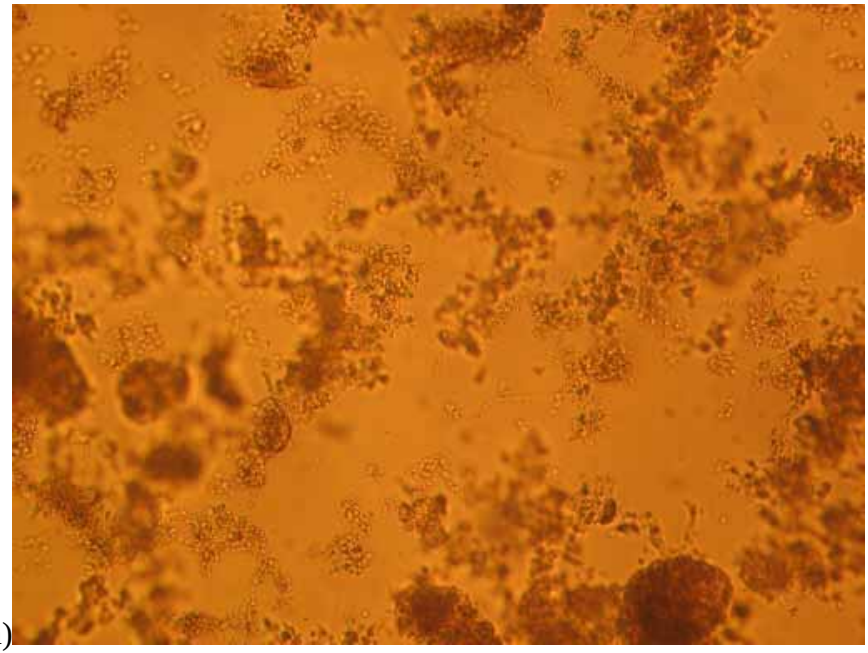


Figure 26: Sludge Volume Index in Phase III

In Figure 26 it can also be seen that the SVI changed late in the run, with the Cannibal system getting worse. Although the sludge did not settle poorly, the settling is of concern. No explanation for the response was identified. For all the reactors, the MLVSS was quite high and the loss of solids and changes in SVI may have been related to the high MLVSS.

(a)



(b)

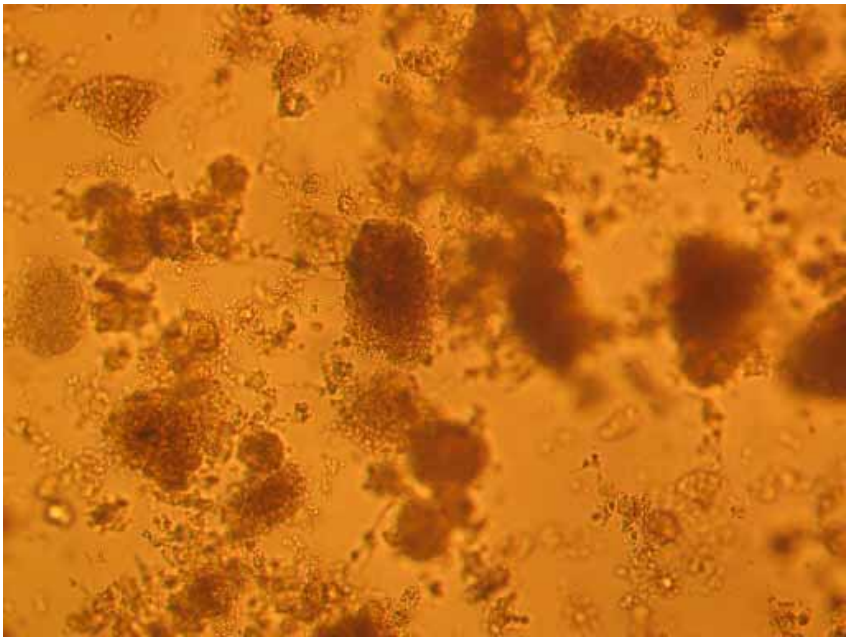


Figure 27: Microscopic analysis of (a) Cannibal SBR and (b) Control SBR in Phase III

Microscopic analyses were conducted on day 40. Filamentous bulking was not observed (Figure 27). The SVIs of Cannibal SBR and Control SBR were 129 and 64, respectively.

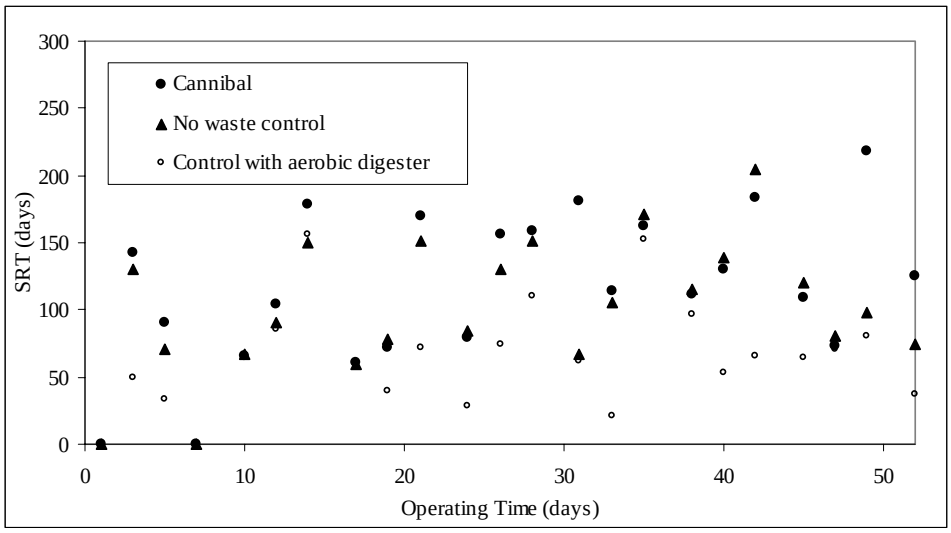


Figure 28: Sludge Retention Time in Phase III

In Figure 28, the SRTs are shown. It can be seen that the Cannibal SRT is very high, in the range of 150 days. Because of the loss of effluent solids, the no-waste system SRT declined after day 40. Prior to that, it was similar to the Cannibal system.

The oxidation-reduction potential (ORP) was occasionally measured in Cannibal anaerobic bio-reactor. For the ORP measurement, Light's solution was used for calibration. The range of ORP in Cannibal anaerobic bioreactor was from +90 and +144 in Phase II to Phase III. According to Saby et al. (2003), when the ORP declines, excess sludge generation will be reduced. Saby et al. showed that the sludge reduction efficiency compared with aeration tank was about 23% with an ORP +100mV. This is similar to the range found in this research.

INVESTIGATION FOR THE SOLIDS DESTRUCTION MECHANISM

Characteristics of bioreactors

The analysis of the soluble protein and polysaccharide was conducted in the end of Phase II to evaluate the mechanisms of Cannibal™ Process. The soluble protein and polysaccharide concentrations in the Cannibal bioreactor were 81 and 10.8 mg/L, respectively and 10.7 and 30.4 mg/L respectively in aerobic bioreactor (Table 3). Novak et al. (2003) proposed that there are different mechanisms of floc destruction between aerobic and anaerobic digestion. They quantified the release of biopolymer from sludges under both anaerobic and aerobic conditions and showed that the release of protein into solution was 4-5 times higher under anaerobic conditions than under aerobic conditions, although the VS destruction by anaerobic digestion was no higher than for aerobic digestion. The authors also found the accumulation of polysaccharide and divalent ion (Ca^{++} and Mg^{++}) in the aerobic digestion. The data shown in Table 3 support the idea that the bioreactor in the traditional Cannibal system results in the release of iron-associated proteins that can be easily degraded in the activated sludge system. In the aerobic bioreactor, polysaccharides and divalent cations are released.

Table 3: Protein and polysaccharides in the activated sludge reactor and bioreactors

	Protein (mg/l)	Polysaccharide (mg/l)	Ca^{++} (mg/l)	Mg^{++} (mg/l)
Cannibal	10.8	4.5	79.5	17.0
Cannibal anaerobic bioreactor	**81.0**	**10.8**	**82.3**	**33.1**
Control	12.6	3.2	88.6	18.1
Control aerobic bioreactor	**10.7**	**30.4**	**321.4**	**61.6**

It is also thought that a high concentration of soluble protein in the return sludge leads to a high ratio of the initial substrate concentration to biomass concentration (S_0/X_0 as COD/biomass) in the Cannibal system. Chudoba et al. (1992) found that cell multiplication did not occur during the exogenous substrate removal when the ratio (S_0/X_0) was sufficiently low. The authors suggested that a biomass increase was mostly due to the synthesis of storage polymers under the

sufficiently low ratio (S_0/X_0) and, under the high S_0/X_0 ratio, more energy was spent for cell synthesis, which results in a greater part of substrate being oxidized. It was also shown that the observed yield, Y_{obs}, decreases with increasing S_0/X_0 ratio. Therefore, data in Table 3 indicates that the released protein from Cannibal anaerobic bioreactor could allow the Cannibal system to have a much higher S_0/X_0 ratio than the Control system.

SOUR

The SOUR was measured in the end of Phase II, and III. The SOUR data resulting from adding thickened sludge from the activated sludge system to supernatant liquid from the Cannibal and control bioreactors was also measured. The traditional Cannibal system had a much higher oxygen uptake rate than the mixed liquor alone and the supernatant liquid from aerobic bioreactor (Figure 29).

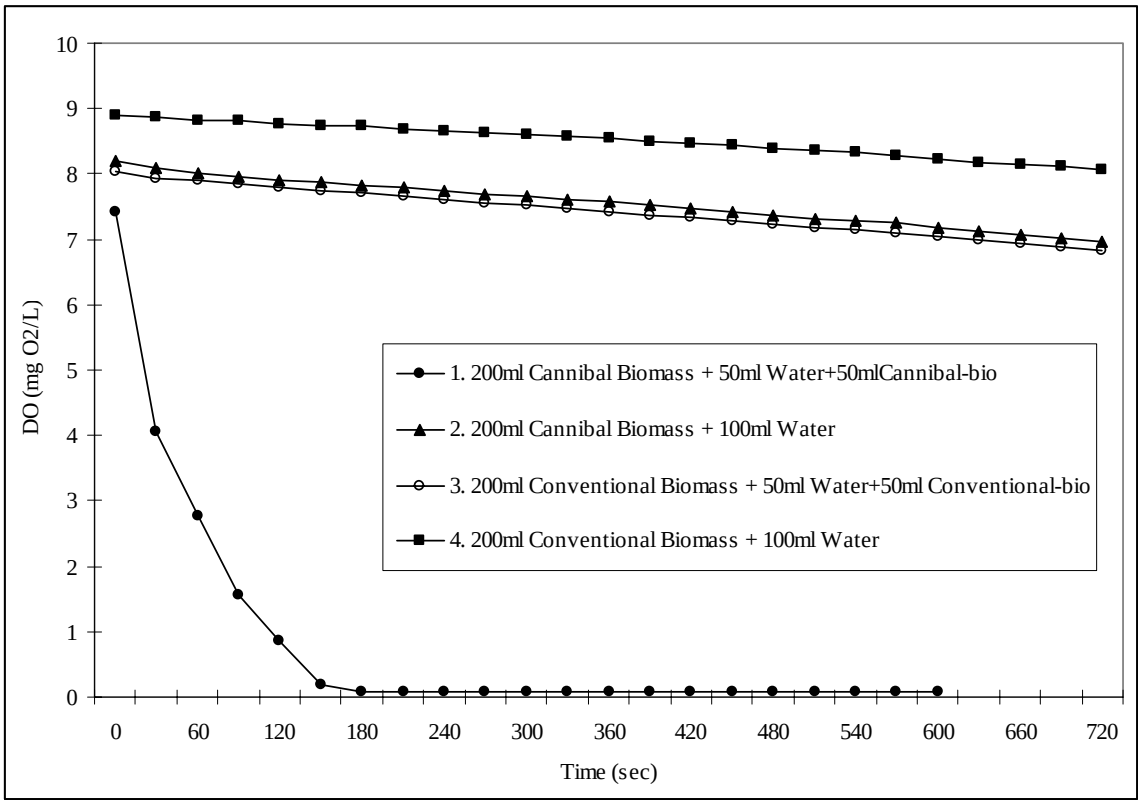


Figure 29: Specific oxygen uptake rate

Table 4: Specific oxygen uptake rate

Batch #	1	2	3	4
SOUR(mg/hr/gVSS)	**111.1**	6.0	6.0	3.7

The SOUR data is shown in Figure 29 and Table 4. Dissolved oxygen concentration in Batch #1 which contained 200ml Cannibal biomass, 50ml distilled water, and 50ml supernatant liquid from Cannibal bioreactor decreased rapidly. This suggests that the mechanism for the traditional Cannibal is to solubilize organic material that is then degraded in the activated sludge system. It is thought that the release of bio-degradable organic maters in the bioreactor and VS destruction in the activated sludge system allow the traditional Cannibal system to reduce excess sludge generation. Substrate is consumed for growth and used for maintenance. As a consequence, substrate is consumed and relatively less biomass is produced due to catabolism through the repeat of Cannibal™ Process mechanisms. For the Aerobic Cannibal, it appears that some of the degradation occurs in the aerobic bioreactor and is the equivalent of aerobic digestion. It is also thought that this degradation in the aerobic bioreactor allows the Aerobic Cannibal system to reduce the excess sludge generation. Therefore, it seems likely that a lower bioreactor SRT may provide good solids destruction for the traditional Cannibal since the major mechanism is release rather than degradation in aerobic bioreactor. For the aerobic Cannibal, a shorter bioreactor SRT would likely lead to less solids destruction because some of the solids destruction is in the bioreactor.

It has been reported that protein has a high affinity for iron and aluminum (Murthy et al. 2000). Biodegradable protein can be bound to both iron and aluminum. Although iron can be reduced from the trivalent to divalent form under anaerobic conditions, aluminum always remains in the trivalent form in flocs under both aerobic and anaerobic conditions. Therefore, further research is necessary to investigate the effect of iron and alum concentrations on the Cannibal system.

DETERMINATION FOR THE OPTIMUM BIOREACTOR DETENTION TIME

Optimization of Cannibal™ Process will be important to insure that the process is cost effective. Cannibal™ Process requires an anaerobic bioreactor. Therefore, it is important to evaluate the impact of detention time in the anaerobic bioreactor on yield.

This batch test was conducted for determination of the optimum bioreactor detention time at the end of Phase III After adding thickened sludge from Cannibal SBR and Control SBR to the aerobic and anaerobic bioreactors, the MLSS, VSS, solution protein, solution polysaccharide, anions, cations, and pH were measured. As shown in Table 2, different amounts (7, 13, 26, and 41%) of thickened sludge from the activated sludge system were added to the anaerobic bioreactor sludge.

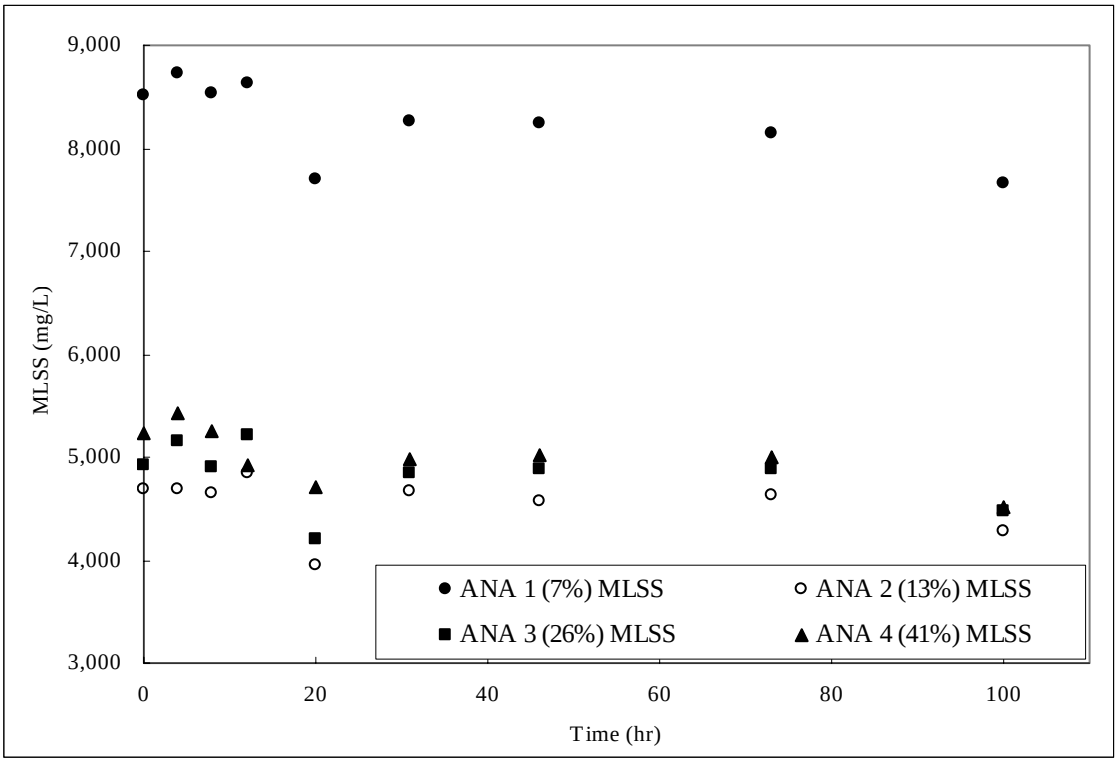


Figure 30: MLSS

The suspended solids are shown in Figure 30. The difference in SS between ANA 1 (7%) and the others (ANA 2, 3, and 4) was because the anaerobic biomass for these tests was from aerobic biomass that had been allowed to become anaerobic over a 15 day period. For ANA 1, the anaerobic biomass was from the anaerobic bioreactor. The small volume of anaerobic biomass in the anaerobic reactor limited the number of tests that could be run using the anaerobic biomass. It was observed that, after 100 hour batch test, the SS in all batch reactors decreased.

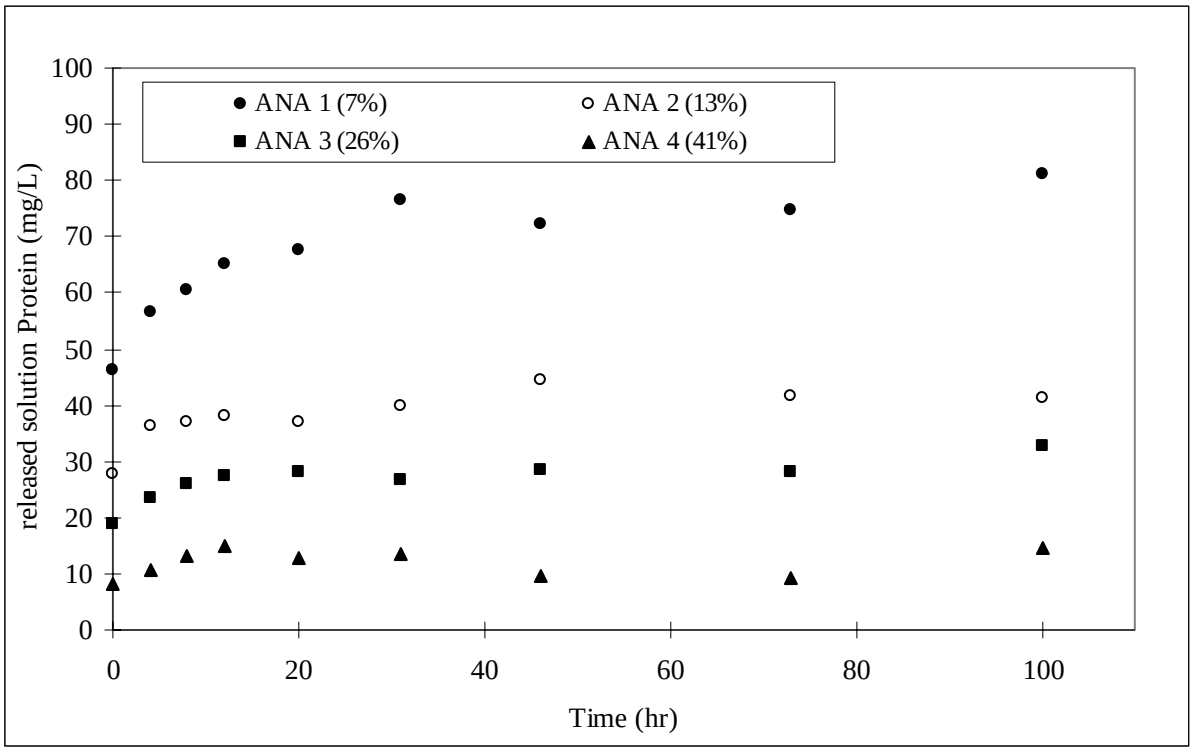


Figure 31: The released solution protein

In Figure 31, the released solution protein is shown. There were four anaerobic batch reactors to which thickened sludge was added in different proportions as shown in Table 2. It can be seen that the smallest proportion of thickened sludge to anaerobic biomass released the most protein. It is not clear why this occurs and warrants further study. What seems clear is that it is possible to overload the bioreactor, decreasing its efficiency in releasing biopolymer.

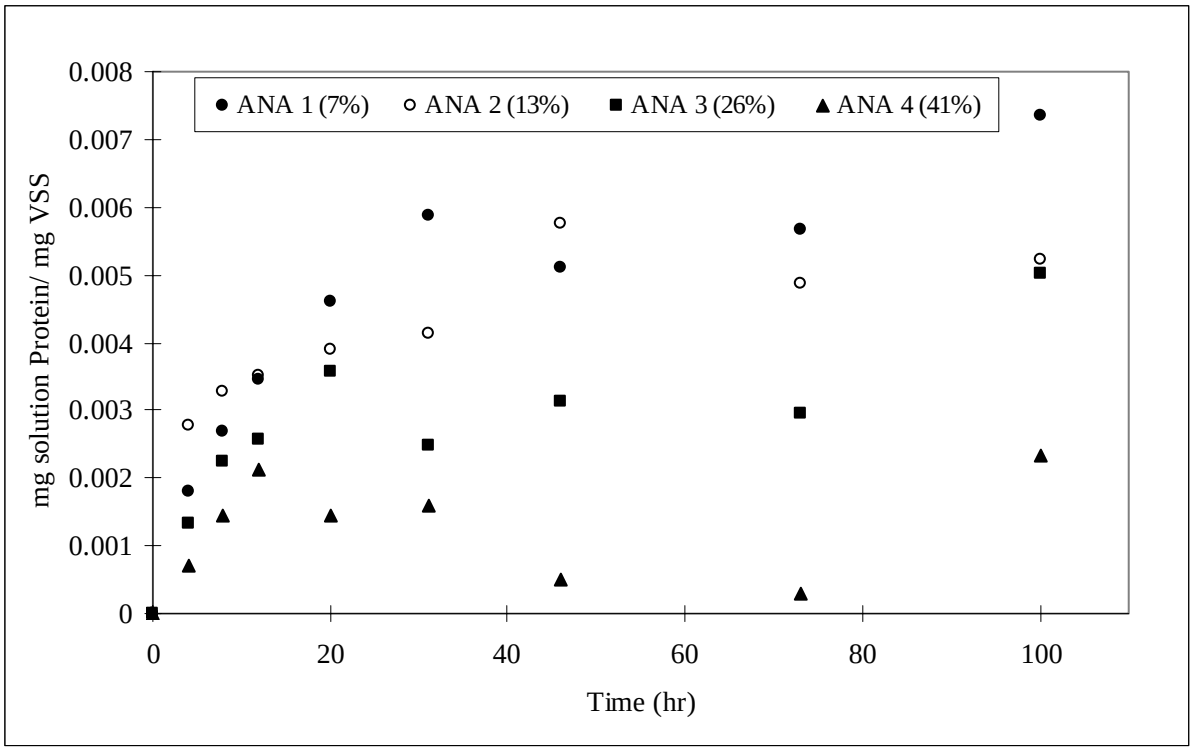


Figure 32: The released solution protein normalized by VSS

The released protein was normalized by VSS as shown in Figure 32. This data indicates that the smallest proportion (7% ~13%) release much more protein than the high proportion (26% ~ 41%). Within 30 hours, the protein release was complete. This suggests that the released protein in the anaerobic bioreactor is the result of a chemical reaction because of the relatively short time needed. This suggests that there might be an optimum or maximum amount of recycle or interchange for the process to function in a successful manner.

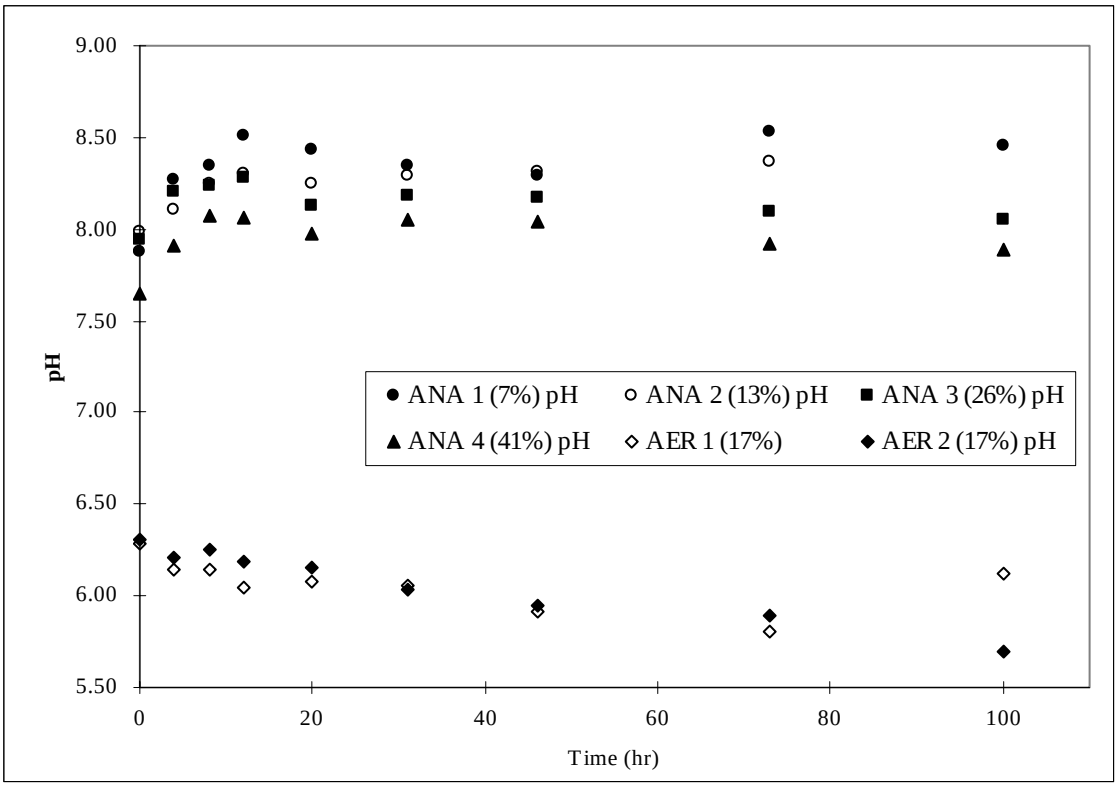


Figure 33: pH

The pH in the second batch test is shown in Figure 33. The pH in the anaerobic batch tests increased for 12 hours. This time is shorter than the time needed to maximize protein release. The pH in the aerobic batch tests steadily decreased after addition of the thickened waste activated sludge.

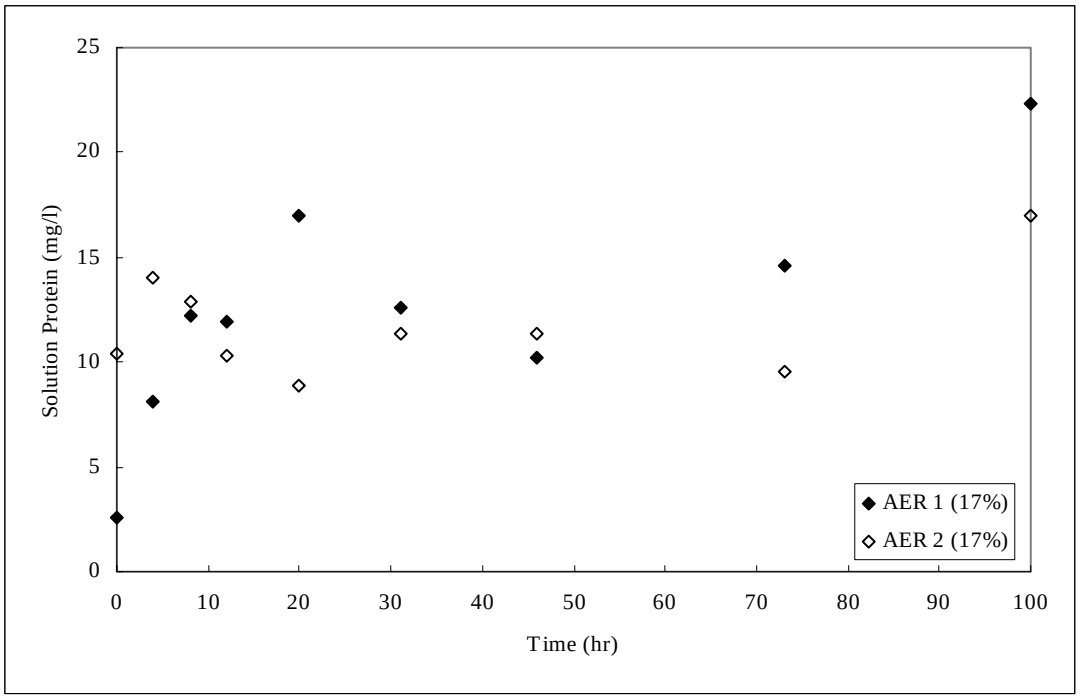


Figure 34: The released solution protein in the aerobic batch tests

A very small amount of protein was released in the aerobic batch tests (Figure 34). Novak, et al. (2003) showed VS destruction and release of divalent cations (Ca^{++} and Mg^{++}) in the aerobic digestion but little protein release. It can be seen that the easily degradable protein associated with divalent cations was most likely degraded in the aerobic batch reactor.

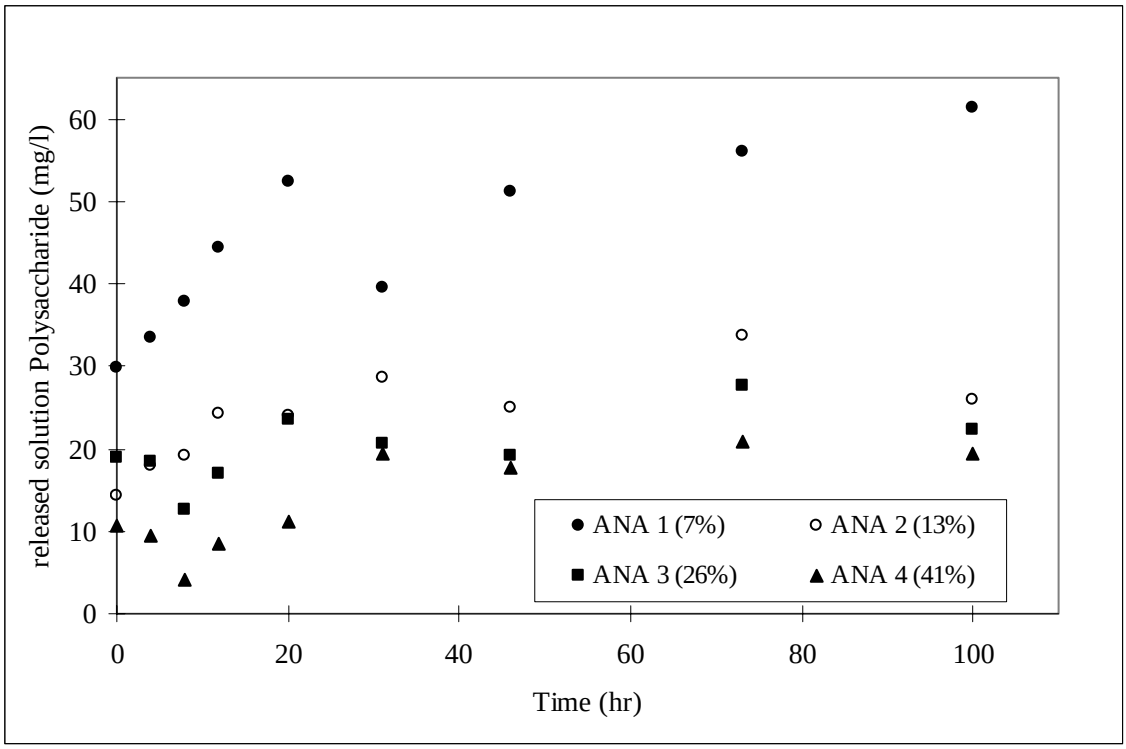


Figure 35: Released solution polysaccharide from anaerobic batch tests

It can be seen that polysaccharide was also released in anaerobic batch reactors (Figure 35). Polysaccharide release during anaerobic digestion has been observed by Novak, et al. (2003), although the polysaccharide concentration is much less than proteins. The polysaccharide release was also rapid, occurring within 20 hour. This indicates that the major function in anaerobic bioreactor is the release of biopolymer, making it bioavailable once the sludge is returned to the aeration basin.

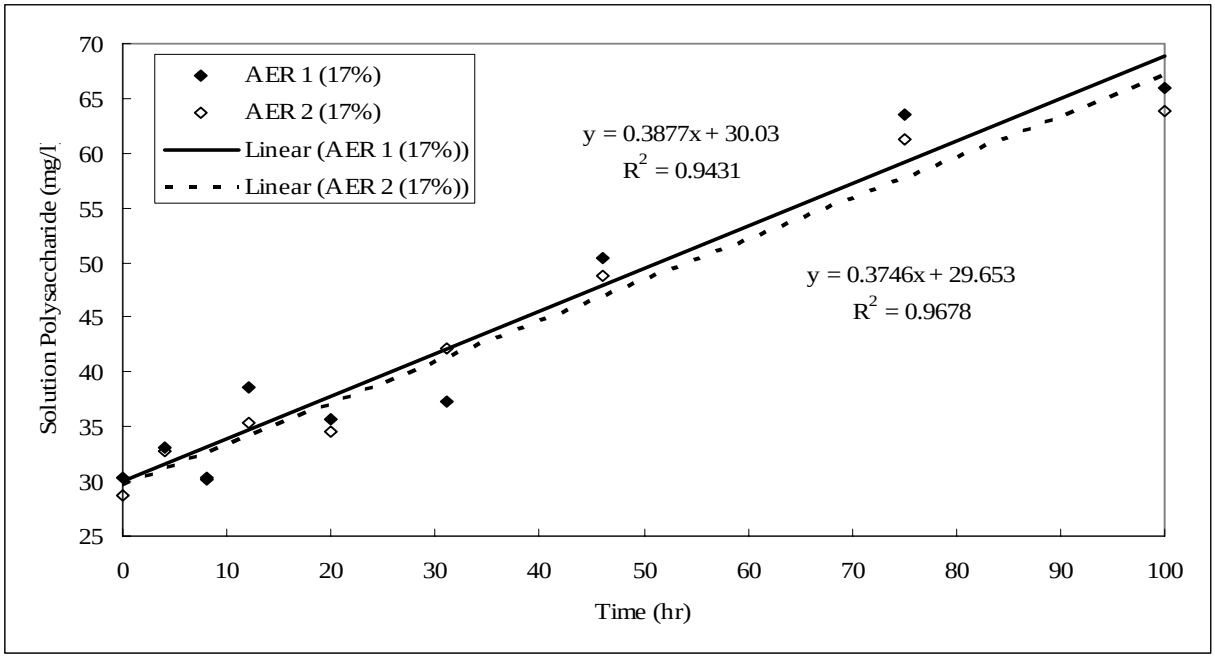
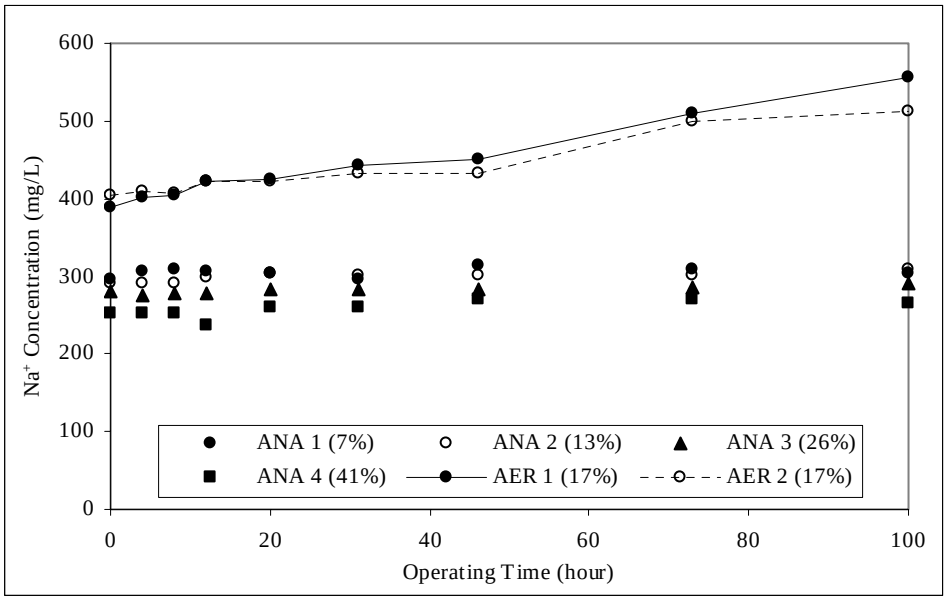
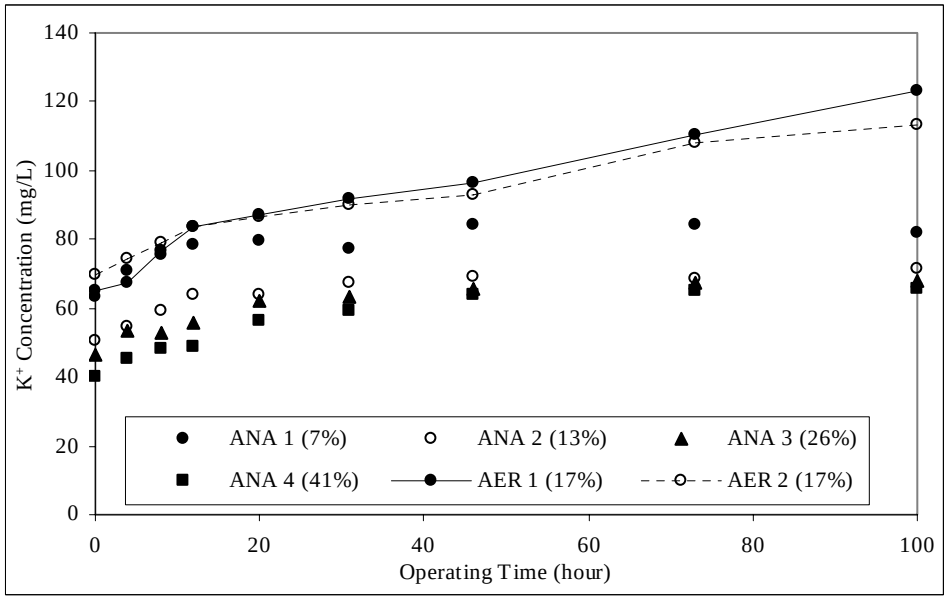


Figure 36: Released solution polysaccharide from aerobic batch tests

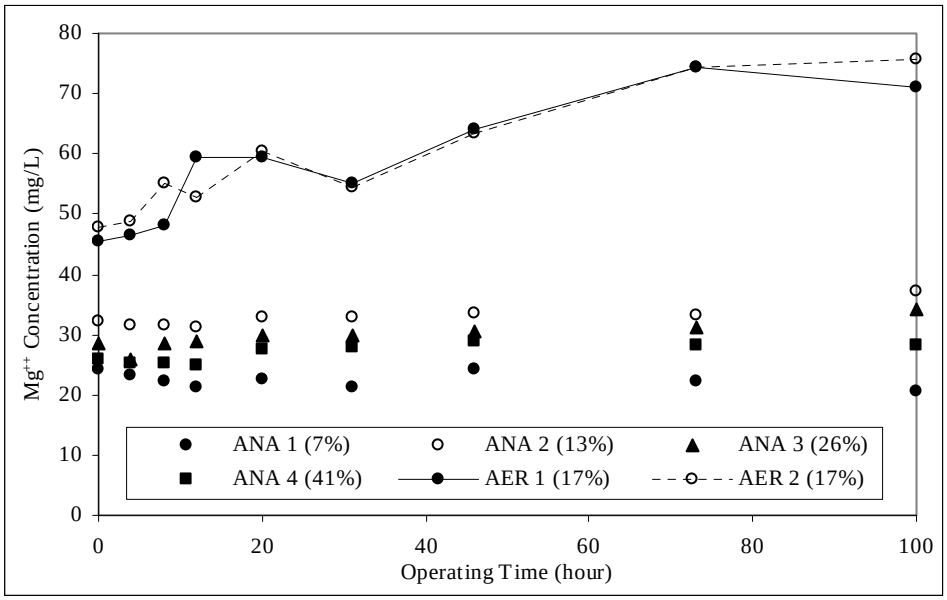
Polysaccharide was released in the aerobic bioreactor (Figure 36). Cations were also released, especially Ca^{++} and Mg^{++}. Novak et al (2003) hypothesized that there are two types of biopolymer binding mechanisms in flocs; one fraction associated with divalent cations (Ca^{++} and Mg^{++}) that is degraded primarily under aerobic digestion and another fraction associated with iron that is degraded primarily under anaerobic digestion. These data in Figures 36 and 37 appear to validate this hypothesis. The data in Figure 36 also indicates that in contrast to the anaerobic reactor, release does not occur rapidly, but rather, continues over 100 hours or more. It was postulated that the Cannibal bioreactor would release protein relatively rapidly and this is what occurred. Within 20 to 30 hours, most of the biopolymer release had occurred.


600
500
400
300
200
100
0
Na+ Concentration (mg/L)
0
20
40
60
80
100
Operating Time (hour)
ANA 1 (7%)
ANA 2 (13%)
ANA 3 (26%)
ANA 4 (41%)
AER 1 (17%)
AER 2 (17%)

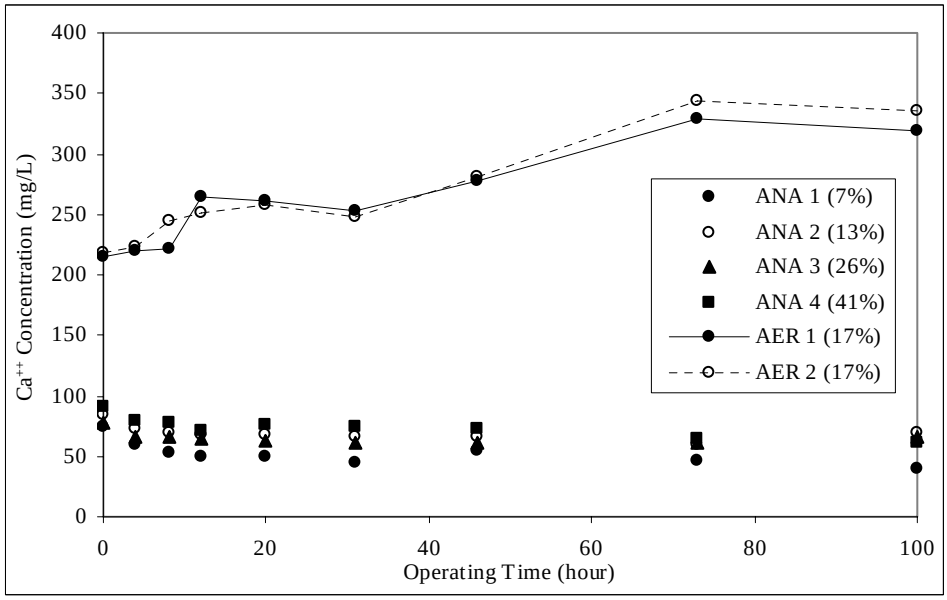
(a)


140
120
100
80
60
40
20
0
K+ Concentration (mg/L)
0
20
40
60
80
100
Operating Time (hour)
ANA 1 (7%)
ANA 2 (13%)
ANA 3 (26%)
ANA 4 (41%)
AER 1 (17%)
AER 2 (17%)

(b)



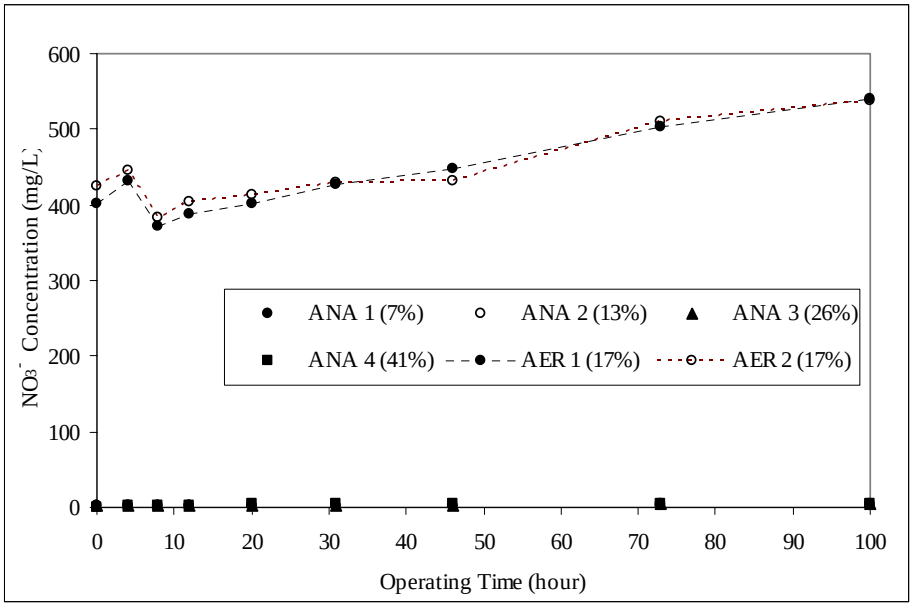
(c)



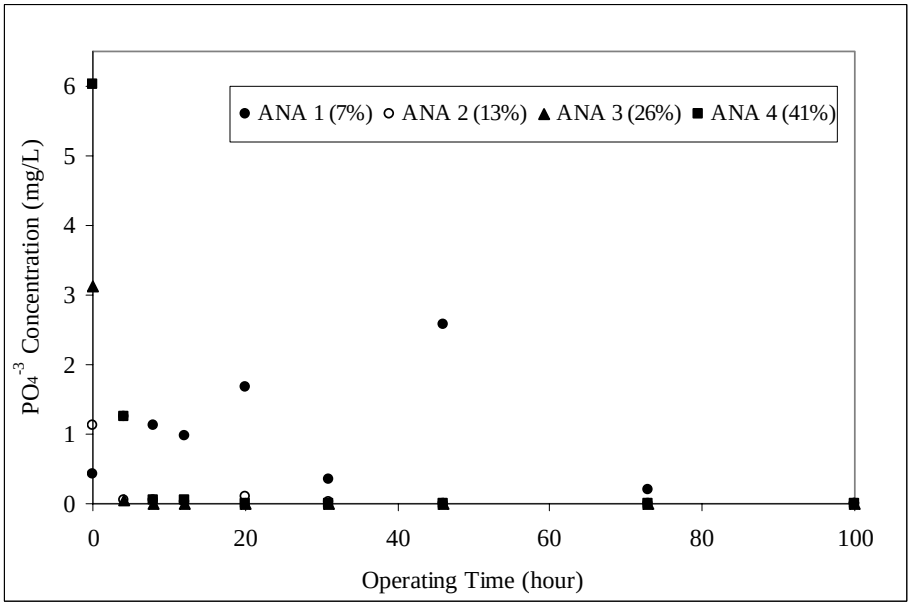
(d)

Figure 37: Released solution Cations from the anaerobic and aerobic batch tests; a) Na^{+}, b) K^{+}, c) Mg^{++} and d) Ca^{++}

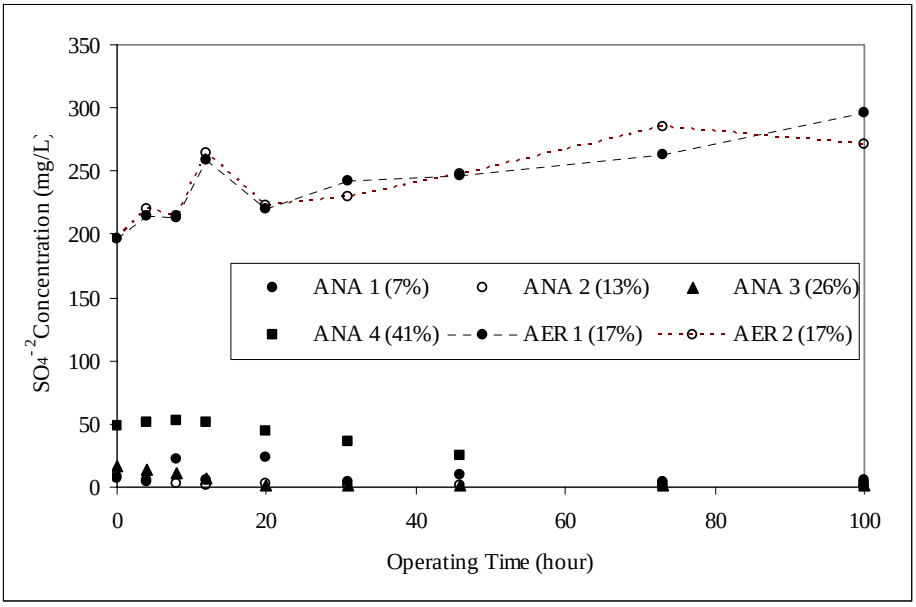
Released solution Cations are shown in Figure 37. It has been shown by Novak et al. (2003) that during aerobic digestion, both polysaccharides and divalent cations (Ca^{++} and Mg^{++}) are released. As shown in Figure 37, cations in the aerobic reactors steadily increased. After adding biomass to the aerobic reactor, the polysaccharide and Ca^{++} steadily increased over 100 hours. This indicates that the aerobic bioreactor acts like an aerobic digester. Therefore, the SRT in the aerobic bioreactor could be critical to the design of the Aerobic Cannibal system.


600
500
400
300
200
100
0
0
10
20
30
40
50
60
70
80
90
100
Operating Time (hour)
NO3- Concentration (mg/L)
ANA 1 (7%)
ANA 2 (13%)
ANA 3 (26%)
ANA 4 (41%)
AER 1 (17%)
AER 2 (17%)

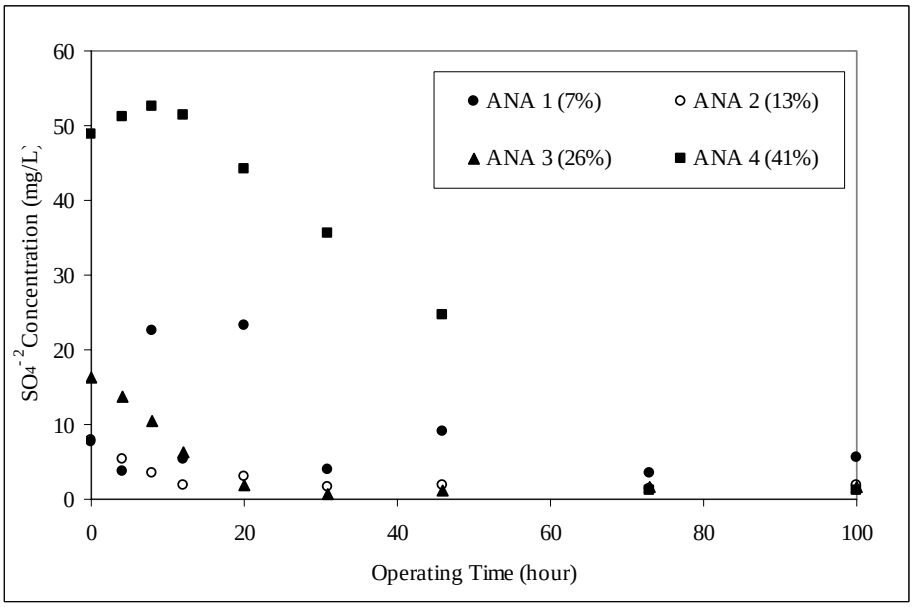
(a)


6
5
4
3
2
1
0
0
20
40
60
80
100
Operating Time (hour)
PO4-3 Concentration (mg/L)
ANA 1 (7%)
ANA 2 (13%)
ANA 3 (26%)
ANA 4 (41%)

(b)



(c)



(d)

Figure 38: Released solution Anions from the anaerobic and aerobic batch tests; a) nitrate, b) phosphate, c) sulfate and d) sulfate in anaerobic batch reactors

The released solution anions from the anaerobic and aerobic second batch tests are shown in Figure 38. Figure 38 (a), (b), (c), and (d) show the changes in NO_3^-, PO_4^{-3}, SO_4^{-2} and SO_4^{-2} in the anaerobic batch reactors. NO_2^- was not detected. The concentration of NO_3^- increased in the aerobic batch test. The data show that nitrification occurred in the aerobic batch reactor and suggest that denitrification occurred in the anaerobic batch reactor. A small amount of PO_4^{-3} was detected in the anaerobic batch reactor and it was not detected in the aerobic batch reactor. The

solution SO_4^{-2}, as shown in Figure c), increased in the aerobic batch reactor, while SO_4^{-2}, as shown in Figure d), in the anaerobic batch reactor decreased. This suggests that ferric ion is reduced to ferrous ion in the anaerobic reactor and then ferrous ion reacts with sulfide forming iron sulfide. This indicates that the major mechanism for the destruction of solids in the Cannibal system is reduction of iron, release of protein and degradation of protein in the activated sludge system. This suggests that the Cannibal bioreactor can be operated successfully at less than 10 days SRT.

CONCLUSIONS

Significant sludge reduction has been demonstrated by incorporating an anaerobic return sludge bioreactor with a 10 day detention time as compared to several types of Control systems. With this sludge reduction, there has been no observation of deterioration in performance.

The main mechanism of Cannibal™ Process appears to be the release of protein in the bioreactor and subsequent degradation in the activated sludge unit. The release is thought to be associated with the reduction of iron. Although there is also polysaccharide in the solution from the anaerobic bioreactor, the protein seems to be the main contributor to the solids destruction. Therefore, the large amount of released protein in the anaerobic bioreactor and easiness of the released protein to be degraded in the aerobic unit may result in less sludge generation and the low yield of biomass. Specific conclusions of this study are:

- The Cannibal system operated with about 60% less solids generation compared to control systems.
- The recycle stream from Cannibal anaerobic bioreactor had a higher oxygen uptake than MLVSS alone
- Increased proteins are found in the Cannibal bioreactor as compared to the aerobic sludge bioreactor unit and more polysaccharides in aerobic unit.
- The interchange rate is critical so as the interchange rates drops below 10%, less solids destruction is found. It appears that the interchange rate is a critical design parameter for the Cannibal system.
- The performance of the Aerobic Cannibal system incorporating an aerobic digester in the sludge recycle stream was similar to the traditional Cannibal system.
- The release of proteins in the bioreactor appears to occur over the first 30 hours. This indicates that a SRT lower than 10 days could be successful.

REFERENCES

Bishop, P.L., and Farmer, M. (1978) Fate of Nutrients During Aerobic Digestion. J. Environ. Eng. Div., Proc. Amer. Soc. Civil Eng., **104,** EE5, 967.

Bruus, JH.; Nielsen, PH.; Keiding, K. (1992). On the Stability of Activated Sludge Flocs with Implications to Dewatering. Wat. Res., **26,** 1597.

Frølund, B., Palmgren, R., Keiding, K., and Nielsen, P.H. (1996) Extraction of Extracellular Polymers from Activated Sludge Using a Cation Exchange Resin. Water Res., **30,** 1749.

Chudoba, P., Capdeville, B., and Chudoba, J. (1992) Explanation of biological meanings of the S_0/X_0 ratio in batch cultivation. Water Sci. Tech., **26,** 743.

Egemen, E., Corpening, J., and Nirmalakhandan, N. (2001) Evaluation of an ozonation system for reduced waste sludge generation. Wat. Sci. Tech., **44,** 445.

Jorand F., Zartarian F., Thomas F., Block J.C., Bottero J.Y., Villemin G., Urbain V., and Manem J. (1995) Chemical and structural (2D) linkage between bacteria within activated sludge flocs. Wat. Res., **29,** 1639.

Grady, C. P. L., Daigger, G. T., and Lim, H.C. (1998) Biological Wastewater Treatment. 2nd Ed., Marcel Dekker, Inc. New York, N.Y.

Higgins, M.J. and Novak, J.T. (1997) Characterization of Exocellular Protein and Its Role in Bioflocculation. J. Environ. Eng., **123,** 479.

Horan, N.J. (1990). Biological Wastewater Treatment Systems. Chichester, London: Wiley.

Liu, Y., Chen, GH., and Paul, E. (1998) Effect of the S_0/X_0 Ratio on Energy Uncoupling in Substrate-Sufficient Batch Culture of Activated Sludge. Water Res.,**32,**2833.

Masion, A., Vilge-Ritter, A., Rose, J., Stone, W.E.E., Teppen, B.J., Rybacki, D., and Bottero, J.Y. (2000) Coagulation-Flocculation of Natural Organic Matter with Al Salts: Speciation and Structure of the Aggregates. Environ. Sci. Technol., **34,** 3242.

Mavinic, D.S. and Koers, D.A. (1982) Fate of Nitrogen in Aerobic Sludge Digestion. J. Water Poll. Control Fed., **54,** 352.

Metcalf and Eddy, Inc. (1991) Wastewater Engineering: Treatment, Disposal, and Reuse. 3rd Ed., McGraw hill Inc., New York, NY.

Murthy, S.N. (1998) Bioflocculation: Implications for Activated Sludge Properties and Wastewater Treatment. Ph.D. Dissertation, VPI & SU, Blacksburg, VA.

Murthy, S.N., Novak, J.T., and Holbrook, R.D. (2000) Optimizing Dewatering of Biosolids from Autothermal Thermophilic Aerobic Digesters (ATAD) Using Inorganic Conditioners. Water Environ. Res., **72,** 714.

Novak, J.T., Sadler, M.E., and Murthy, S.N. (2003) Mechanisms of Floc Destruction During Anaerobic and Aerobic Digestion and The Effect on Conditioning and Dewatering of Biosolids. Water Res., **37,** 3136.

Reece, C.S., Roper, R.E., Jr., and Grady, C.P.L., Jr. (1979) Aerobic Digestion of Waste Activated Sludge. J Environ. Eng. Div., Proc. Amer. Soc. Civil Eng., 105, EE2, 261.

Saby, S; Djafer, M; Chen, G-H (2003). Effect of low ORP in anoxic sludge zone on excess sludge production in oxic-settling-anoxic activated sludge process. Wat. Res., **37,** 11.

Shiota, N., Akashi, A., and Hasegawa, S. (2002). A Strategy in Wastewater Treatment Process for Significant Reduction of Excess Sludge Production. Wat. Sci. Tech., **45,** 127.

Wei, Y., Houtenb, RT., Borgerb, AR., Eikelboomb, DH., and Fana, Y. (2003) Minimization of Excess Sludge Production for Biological Wastewater Treatment. Water Res.,**37,**4453

Yasui, H., and Shibata, M. (1994) An Innovative Approach to Reduce Excess Sludge Production in the Activated Sludge Process. Water Sci Tech.,**30,**11

Ye, F.X., Shen, D.S., Li, Y. (2003) Reduction in Excess Sludge Production by Addition of Chemical Uncouplers in Activated Sludge Batch Cultures. J. Appl. Microbiol., **95**, 781-786.

Vilge-Ritter, A., Masion, A., Boulange, T., Rybacki, D., and Bottero, J.Y. (1999) Removal of Natural Organic Matter by Coagulation-Flocculation: A Pyrolysis –GC-MS Study. Environ. Sci. Technol., **33,** 3027.

VITA

Dong Hyun Chon was born on February 1st, 1975 in Incheon, Korea. He attended Korea University, Korea, in 1993 and received a Bachelors of Science in Environmental Engineering in February of 1997. After his undergraduate work, he served military duty as a section commander and a fire direction officer in Korean Army from March of 1997 to June of 1999. He started his professional life with Ecophile (www.ecophile.com) specializing in soil bioremediation in August of 2000. He was employed with Tyco Thermal Controls Korea (www.tycothermal.com), and his responsibilities included EH&S Management and Quality Assurance from March of 2001 to April of 2002. Before coming to the Virginia Tech, he was working in Korea Institute of Science and Technology (www.kist.re.kr), as an assistance researcher from August of 2002 to April of 2003. In August of 2003, he started his masters work in Environmental Engineering at Virginia Polytechnic Institute and State university, Blacksburg, VA. Following masters work, he plans to work in the same field.