TREATMENT OF A HIGH-SOLIDS SYNTHETIC DOMESTIC WASTEWATER STREAM WITH HIGH-RATE ANAEROBIC DIGESTERS

by

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ABSTRACT

Anaerobic treatment of domestic wastewater greatly reduces the organic load to water bodies with less energy input while generating renewable energy. This study focuses the performance of two different types of high-rate anaerobic digesters (anaerobic migrating blanket reactor [AMBR] and upflow anaerobic sludge blanket [UASB] reactor) treating domestic wastewater at 25°C. A 12 L AMBR and a 5 L UASB were fed with a synthetic colloidal wastewater simulating domestic wastewater at a chemical oxygen demand (COD) concentration of approximatelly 600 mg L^{-1} (this synthetic wastewater consisted of a solution of blended colloidal rice and dog food with trace elements and buffer). Both systems were operated at an identical hydraulic retention time (HRT) of 40 hours and operated for 195 days, however steady-state conditions were not achieved. Although the steaty-state conditions were not reached, data show that full-scale AMBR would be a good choice for domestic wastewater treatment.

ÖZET

Evsel atık suyun oksijensiz arıtımı ile su kaynaklarına karışan organik atık miktarı azaltılmakta ve aynı zamanda yenilenebilir enerji üretimi sağlanmaktadır. Bunun yanı sıra, kullanılan enerji diğer arıtım sistemlerine oranla daha az olmaktadır. Konuya katkısı nedeniyle proje konusu, iki farklı tip anaerobik reaktörün (Oksijensiz Hareketli Bakteri Reaktorü (OHBR) ve Yukarı Akım Oksijensiz Çamur Bakterisi (YAOÇB)) evsel atık suyu 25 °C'de arıtım performansları karşılaştırılması olarak seçilmiştir. 12 litrelik OHBR ve 5 litrelik YAOÇB reaktörleri evsel atık suyu temsil eden yapay koloidal karışım ile beslenmiştir. COD derişimi yaklaşık 600 mg L^{-1} olan sentetik atık su karışımı öğütülmüş pirinç ve kuru köpek maması ile tampon ve mineral çözeltisinden elde edilmiştir. İki sistem de 40 saatlik hidrolik tutuş zamanı 195 gün boyunca eşit şartlarda işletilmiştir, fakat denge durumuna ulaşılamamıştır. Denge durumu elde edilemesine rağmen, deney sonuçları tam boy bir OHBR'nün evsel atık su arıtımında iyi bir seçenek olacağını göstermiştir.

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LIST OF SYMBOLS / ABBREVIATIONS

Symbol	Explanation	Units used
AMBR	Anaerobic Migrating Blanket Reactor	
COD	Chemical Oxygen Demand	($mg\ L^{-1}$)
EBS	Effluent Baffle System	
GSS	Gas-solids Separator	
HOA	Hydrogen Oxidizing Methanogens	
HOM	Hydrogen Oxidizing Acetotrophs	
HRT	Hydraulic Retention Time	(h)
MCOD %	Methane-based COD efficiency	
MLTSS	Mixed Liquor Total Suspended Solids	($g\ L^{-1}$)
MLVSS	Mixed Liquor Volatile Suspended Solids	($g\ L^{-1}$)
SCOD	Soluble COD	($mg\ L^{-1}$)
SMPR	Standard Methane Production Rate	($L\ L^{-1}\ day^{-1}$)
TCOD	Total COD	($mg\ L^{-1}$)
TSS	Total Suspended Solids	($mg\ L^{-1}$)
UASB	Up-flow Anaerobic Sludge Blanket	
VFA	Volatile Fatty Acids	
VSS	Volatile Suspended Solids	($mg\ L^{-1}$)

1. INTRODUCTION

Anaerobic digestion has been successfully applied in tropical and subtropical regions for domestic sewage treatment (van Haandel and Lettinga, 1994). An anaerobic digester can remove up to 80% of the influent organic matter at very limited costs, low sludge production rates, and can remove most of the pathogens. However, post treatment of anaerobic digester effluent is necessary to remove the remaining pathogens and residual organic matter. In the USA, domestic sewage is treated with activated sludge plants, which use significant amounts of energy for aeration and sludge disposal. Such treatment is not sustainable in developing countries.

Rural communities in the developing countries often do not treat their domestic wastewater (Meizen-Dick and Appasamy, 2001). When domestic wastewaters are discharged into water bodies such as rivers etc. without treatment, they tend to contaminate the waters with high concentrations of pollutants of chemical and biological nature. Untreated wastewaters have a wide range of excreted human pathogenic microorganisms which may create big health problems for humanity. In Latin America, 40 million cubic meters of wastewaters are collected daily and poured into rivers, lakes and seas. Less than %10 of the collected sewage water receives treatment before being released into a water body or before its use for the direct irrigation of agricultural products (Moscoso, 1992). This constitutes serious risks of infection to users of these products or contaminated drinking water. In the cities of the developing countries, %80 of the wastewater is used for watering the plants or daily needs of people without treatment (Wahaab, 1995).

Anaerobic digestion has been used as a pretreatment step for the removal of organic compounds, pathogen reduction, and odor control. In addition, it produces a valuable source of energy in the form of methane gas. Since the temperatures of domestic wastewater are relatively low (~ 20 ^{o}C), hydrolysis of the nonsoluble organic matter is typically the rate-limiting step in anaerobic digestion.

This study has focused on organic substrate hydrolysis in the 12 L Anaerobic Migrating Blanket Reactor (AMBR) and the 5 L Upflow anaerobic sludge blanket (UASB) reactor treating domestic wastewater. Performances of anaerobic biological reactor systems were evaluated in terms of process efficiency and stability through estimation of organic matter removal. The parameters such as chemical organic demand (COD), volatile fatty acid concentration (VFA), quantity and composition of biogas produced, etc., were used to compare the AMBR and the conventional UASB reactor.

2. THEORY

The anaerobic digestion process (Figure 2.1) is in many ways ideal for wastewater treatment and has several advantages over the other available methods (Metcalf and Eddy, 2004). The advantages of anaerobic treatment can be indicated by comparing this process with aerobic treatment. In aerobic treatment, the waste (or wastewater) along with the aerobic microorganism are mixed by the introduction of large quantities of air (O_2), which is by itself a drawback of this process in terms of the need for energy to efficiently aerate. Microorganisms convert the organic waste and oxygen into biomass, carbon dioxide and water. Therefore, the carbon source has only changed in form (mostly to new cells), and it needs disposal afterwards.

In anaerobic treatment, air is excluded by definition. Under these conditions, bacteria grow and convert the organic waste to carbon dioxide and methane gas. Unlike aerobic oxidation, the anaerobic conversion to methane gas yields relatively little energy to the microorganisms. Therefore, their rate of growth is slow and only a small portion of the waste is converted to new cells. The major portion of the degradable waste is converted to methane gas.

Good removal efficiency can be achieved in the anaerobic systems, even at high loading rates and low temperatures. The construction and operation of anaerobic systems are relatively simple.

Solids destruction is important in anaerobic digestion of sewage because removal of solids is usually a primary objective. In addition, particulate solids destruction (i.e., hydrolysis of volatile suspended solids) is usually the rate limiting step under relatively low temperatures (El-Mashad et al., 2004). Increasing the temperature of sewage increases the solids degradation rate by improving the bioavailable and kinetics (Makie and Bryant, 1995; Muller et al., 2007), however, sewage cannot be heated due to energy considerations. Making the solids retention time longer than the hydraulic retention time also will improve solids degradation.

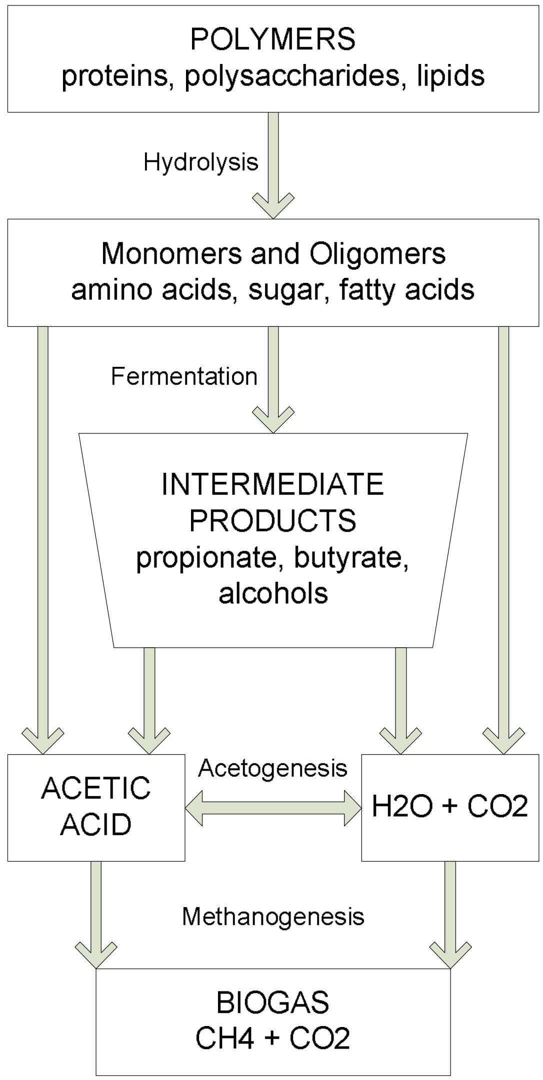

Figure 2.1. Anaerobic food web

The anaerobic degradation of complex organic substances can be divided in four main steps: hydrolysis, fermentation (acidogenesis), acetogenesis and methanogenesis (Figure 2.1). This food web involves six groups of microorganisms (Angenent et al., 2004; Hungate and Stack, 1982):

1. Hydrolytic bacteria functioning mainly breaking the polymers in to oligomers and monomers,
2. Fermentative bacteria functioning converting oligomers and to intermediates (propionate, butyrate, alcohols) , H_2 , CO_2 and acetate,
3. H_2-producing acetogenic bacteria (from intermediates),
4. H_2-utilizing acetogenic bacteria,
5. Hydrogenotrophic methanogenic archaea,
6. Aceticlastic methanogenic archaea.

2.1. Steps of Anaerobic Digestion Process

2.1.1. Hydrolysis

Complex substrates such as proteins, fat, oil etc. need to be broken down into simple components in the process known as hydrolysis before they can be utilized by the micro-organisms. Proteins are hydrolyzed to polypeptides and amino acids. Polysaccharides (e.g. starch, cellulose) are hydrolyzed to oligosaccharides and monosaccharide or simple sugars. Lipids are hydrolyzed to fatty acids and glycerol. These processes are carried out by the extracellular enzymes produced by the groups of hydrolytic bacteria (Hungate, 1982).

McCarty and Lawrence (1969) claimed that particulate solubilization was rate limiting in combined digesters. Eastman and Ferguson (1981) also reported that the hydrolysis of particulate matter in domestic sludge was always the rate limiting step in a separate acid producing reactor. However, O'Rourke (1968) using domestic sludge indicated that hydrolysis of lipids was not the rate limiting step which was opposite of what Kennedy and Van den Berg (1982) claimed. Hydrolysis is considered the rate-limiting step especially during the anaerobic digestion of particulate organic matter (Pavlostathis and Giraldo-Gomez, 1991).

2.1.2. Fermentation (Acidogenesis)

Polymers which were broken in to simple components (amino acids, sugars etc) in hydrolysis processes were converted to organic acids, alcohols, hydrogen and carbon dioxide in the fermantation process by microorganisms. Acetate is the major intermediate product of organic matter converted to biogas. The percentage of the methane produced from acetate is approximately 70% of the total methane produced (Guejer and Zehnder, 1983). Other higher volatile fatty acids are also present in small amounts due to the complex and variable nature of the substrate entering the anaerobic digester.

Fermentative bacteria has a relatively short doubling time, ranging from 33 minutes (Ghosh and Pohland, 1974) to more than 12.5 hours (Andrews and Pearson, 1965). Despite the diversity of the possible products from fermentation, it has never been regarded as the rate limiting step in anaerobic digestion (Eastman and Ferguson, 1981).

2.1.3. Acetogenesis

Acetogenesis is carried out by the hydrogen producing and hydrogen utilizing bacteria. It is the conversion of intermediates to acetate, hydrogen and carbon dioxide. In addition to that the conversion between acetate and hydrogen with carbon dioxide is also a part of the process.

The process is very sensitive, therefore system crash is possible when one group of microorganisms is inhibited or overloaded (Duran and Speece, 1998). For example, accumulation of intermediate products (e.g., propionate, butyrate, or acetate) may hinder acetogenesis and/or methanogenesis by reducing the pH to less than the optimal range for these microorganisms. Propionic acid oxidation is one of the reactions occurring during the acetogenesis phase (Angenent et al., 2004).

$$\underset{\text{Propionic Acid}}{CH_3CH_2COOH} + 2\,H_2O \rightarrow \underset{\text{Acetic Acid}}{CH_3COOH} + CO_2 + 3\,H_2 \tag{2.1}$$

In some stressed anaerobic digestion systems, acetate is converted to methane through a two-step process involving oxidation of acetate to hydrogen and carbon dioxide by homoacetogens and conversion of these products to methane by hydrogenotrophic methanogens. Note that oxidation of acetic acid by homoacetogens is a reversal of the process for which these organisms are best known and is made possible by their syntrophic relationship with the hydrogenotrophic methanogens (Angenent et al., 2004):

$$\underset{\text{Acetic Acid}}{CH_3COOH} + 2H_2O \rightarrow 2\ CO_2 + 4\ H_2 \qquad (2.2)$$

2.1.4. Methanogenesis

Two types of methanogenesis occur in the food web; methanogenesis from acetate and from hydrogen and carbon dioxide.

<u>2.1.4.1. Methanogenesis from Acetate (Acetoclastic).</u> The decarboxylation of acetate was the mechanism found in the 50s to form methane (Pine and Barker, 1956) in the reaction:

$$\underset{\text{Acetate}}{CH_3COO^-} + H_2O \rightarrow \underset{\text{Methane}}{CH_4} + HCO_3^- \qquad (2.3)$$

The carboxyl group is oxidized to carbon dioxide, and the methyl group is reduced to methane in the reaction. The optimum pH range for methanogenesis found as between pH 6 to pH 8. The conversion of acetate to methane will be inhibited if pH drops below this range and acetate will accumulate. As a result of low pH and accumulated acetate, degradation of fatty acids will be inhibited (especially propionic acid) (Fukuzaki et al., 1990).

<u>2.1.4.2. Methanogenesis from Hydrogen and Carbon Dioxide.</u> In anaerobic digesters, nearly all the methane produced from a non-acetate source is the result of the reduction of carbon dioxide/bicarbonate by hydrogen in the reaction:

$$HCO_3^- + 4H_2 + H^+ \rightarrow \underset{\text{Methane}}{CH_4} + 3H_2O \quad (2.4)$$

which yields over four times the energy as that from acetoclastic methane formation. Hydrogen is therefore the most important extracellular intermediate in methane production (Zeikus, 1977).

The hydrogen oxidizing methanogens (HOM) which do not utilize acetate and hydrogen oxidizing acetotrophs (HOA) are in charge of methane formation from carbon dioxide and hydrogen. These bacteria can also use a single carbon substrate such as methanol, methylamine, carbon monoxide and formate (Harper and Pohland, 1986). Zehnder and Wuhrrnann (1977) showed that the HOM were sensitive to pH and temperature variation with the optima at pH 7 and 33-40^{o}C respectively.

The prevalence of high energy yielding and fast growing hydrogen utilizers over acetate utilizers (Non-HOA) may be assumed in an anaerobic process, by looking at the kinetic data. However, in practice, the accumulation of volatile fatty acids due to the lack of acetate utilizers will lower the pH and finally inhibit the H_2 oxidation process. This shows the interdependence between the different groups of microorganism.

2.2. Factors Affecting Anaerobic Digestion of Domestic Wastewater

2.2.1. pH

The stability and value of pH in an anaerobic reactor is very important. Methanogenesis process results with a high efficiency conversion when pH is in the range of 6.3 to 7.8 (van Haandel and Lettinga, 1994). In addition to that a low pH may have inhibatory effects on methanogenesis (Methcalf and Eddy, 2004). Usually pH remain around 6.5 when collodial wastewater such as domestic sweage is treated and there is no need for an additional chemical addition (van Haandel and Lettinga, 1994).

2.2.2. Temperature

The temperature affects the efficiency of anaerobic process significantly (van Haandel and Lettinga, 1994). Optimum range for anaerobic digestion is between 30 and 40 ^{o}C, at the temperatures below optimum range the rate decreases by approximately 11% for each 1 ^{o}C temperature decrease according to the Arrhenius expression. In addition, the amount of overall degraded organic matter decreases as well. At low temperatures the hydrolysis of volatile solids is the rate limiting step (Eastman and Ferguson, 1981). However if the retention of the solids provided, they can be removed from the liquid phase succesfully. Bogte et al. (1993) experimented to accumulate solids during the winter time and provide a degradation during the summer time when the temperature is at the optimum for anaerobic process.

2.2.3. Physical Properties of Wastewater

Domestic wastewater usually contains more particulate organic matter than soluble ones. This affects the overall degradation rate in the reactor (Eastman and Ferguson, 1981). Moreover, hydrolysis of collodial suspended solids to soluble substrates is mostly the rate limiting step. The degradation of suspended substrates is slower than the degradation of soluble substrates which requires a separation of these two phases in the reactor as hydrolysis and methanogenesis. This conclusion leads researchers to multi-stage processing.

2.2.4. Mixing and Flow

Sludge and liquid flow movements affect the performance of the anaerobic process in an upflow reactor (Heertjes et al., 1978). Angenent et al. also proved this fact in the Anaerobic Migrating Blanket Reactor (Angenent and Sung, 2001). The upflow velocity and rising gas bubbles in the reactor are the factors managing the flow pattern in the UASB reactor. However, the AMBR has different features affecting the performance of the process. The AMBR has the advantage of ASBR which has mechanical mixing, biomass retention and granulation. Moreover, pH control by changing the flow direction is possible. These features make the AMBR attractive for low strength wastewaters (Angenent and Sung, 2001).

2.3. Sewage Treatment in the Literature

There are many studies investigating anaerobic treatment of sewage (low strength waste water) which covers domestic wastewater studies as well. Especially, the application of UASB reactor under low temperature conditions has been studied since 1976 (Lettinga et al.,1981; de Man et al.,1986). De Man et al. (1988) reported that anaerobic treatment of raw domestic sewage (COD = 500-700 mg L^{-1}) can be maintained at 18^{o}C applying HRTs of 7-12 h with total COD removal efficiency of 40-60%. It was found that treatment efficiency was affected by the sludge-wastewater contact especially at low temperatures because of the insufficient gas mixing. Notwithstanding, up to 80% removal efficiency was obtained at 10-20 ^{o}C when treating domestic wastewater with a granular bed reactor. The application on expanded granular sludge bed reactor (EGSB) (Kato et al.,1994) characterized with higher upflow velocity showed better removal efficiency of soluble subtrates and sludge-wastewater contact (de Man et al.,1988). Uemura and Harada (2000) reported that high total COD removal efficiency (%70) was obtained at the UASB applicaton at moderatelly low (25 to 13^{o}C) temperatures. However, the hydrolysis of the solids was affected by the temperature adversely. Halalsheh (2002) assessed some interesting results in Jordan with UASB reactors treating strong raw sewage at 18^{o}C in the winter and at 25 ^{o}C in the summer. A comparison was made between one stage and two stage UASB reactor. One stage UASB reactor showed better solids removal than the first stage of the two stage UASB reactor. On the other hand, a compartmentised UASB reactor was investigated by Chemicharo and Cardoso (1999) for the treatment of domestic sewage. The system was composed of three digestion compartments, three gas separation devices and a settler compartment. Eventually, the contact between substrate and biomass , and the overall performance of the reactor were better than one chamber UASB reactors.

There are several pilot plant applications on sewage wastewater treatment. A 120 L UASB reactor was assessed to treat raw sewage with 627 mg COD L^{-1} at ambient temperatures (19-28 ^{o}C) (Barbosa and Sant'Anna, 1989). The reactor operated at an HRT of 4 h throughout the entire 9 months experimental period. COD and Total Suspended Solid (TSS) removal increased during the first 4 months of operation and in the last 5 months, total

Chemical Oxygen Demand (COD) removal reached 74%. The average suspended solids removal during this period was 72%. The results of different applications were promising and gave different design recommendations to upcoming plant installations.

2.3.1 From One Stage Processes to Multi Stage Processes

Two-stage anaerobic processes have been successful and gave the courage of development of multi stage systems to the researchers. In moderate climates, two-stage anaerobic processes have been suggested to hold and degrade suspended solids from sewage (van Haandel and Lettinga, 1994). In the first stage, the collodial solids are hold and partially hydrolyzed into soluble compounds which are digested in the second stage. As a result of the higher removal efficiency of suspended solids in the first reactor, the sludge age is relatively low in the reactor, which reduces methanogenesis to a minimum. Accumulation of biodegradable solids in the first compartment may occur at low temperatures, when hydrolysis becomes the rate limiting step (van Haandel and Lettinga, 1994).

Two-stage UASB reactors were applied in Spain treating domestic sewage at temperatures changing between 9 and 26 ^{o}C (Garcia Encina et al., 1996). The total COD removal reached up to 62% at 14h of HRT. Another study performed to assess the feasibility of a two-stage anaerobic system for sewage treatment. The first part involved two flocculent UASB reactors operated intermittently and the second part was a UASB inoculated with granular sludge (Sayed and Fergala, 1995). The first stage was installed to remove and partially hydrolyze suspended solids and the second stage was removing the soluble organic material. It is claimed that intermittent operation of the first stage provides further stabilization of the removed solids. The study was performed at an ambient temperature of 18-20 ^{o}C and average HRTs of 8-16 h for the overall system and 2 h for the second stage. COD and BOD removal efficiencies up to 80 and 90%, respectively, were achieved.

A two UASB reactors system achieved an average total and suspended COD removal efficiency of 55 and 62%, respectively. The HRT of first reactor was ranging between 8 and 10 hours and the second was 5-6 hours. Mgana (2003) claimed that results from two stage

UASB system treating sewage were promissing under tropical conditions, particularly because sludge washed out from the first reactor was trapped in the second reactor.

Multi-stage anaerobic treatment means separation of the methanogenic and the non-methanogenic digestion phases in separate reactors. Especially two-phase anaerobic systems have been extensively studied in the past. However, the application of two stage systems to raw domestic sewage is a recent application as stated above. Reyes et al. (1999) studied low strength wastewater treatment by a multistage fixed bed reactor. The 6 L reactor consisted of 5 stages and the objective was to purify domestic wastewaters of 1000 mg COD L^{-1} of high microbiological load. The total removal efficiencies varied from 99 % for 4 days to 70 % for 8 h HRT. The results showed that very high hydraulic times are not needed to achieve a good removal efficiency in multi-stage reactors. This allows to use a smaller volume and reactor area or to increase in the capacity for wastewater purification by the reactor. In addition to that, a fixed-bed anaerobic reactor designed with only three could give an efficient treatment of sewage waters with very short HRT, of 7 h or more.

Angenent and Sung (2001) reported separation of the digestion phases in his reserach and proposed the AMBR which fills the need for compartmentalization concept for continously fed systems. However, until this study, there were only few studies which concentrate on domestic wastewater treatment with AMBR.

Hartley and Lant (2006) studied the sewage treatment with an AMBR pilot plant. The reactor was operated at the ambient temperature and consisted of two periods. In the first phase the process operated at 50 hours HRT and at a ranging temperature of 23-35 ^{o}C. In the second phase the operating parameters were 26 days HRT and 12-16 ^{o}C. COD removals in the first and second phase were up to 70% and 28% respectively. The system temperature ranged between 12-35 ^{o}C unlike this study which was performed at 25 ^{o}C fixed.

3. STATEMENT OF THE PROBLEM

Direct discharge to the environment is a common way of dealing with sewage and domestic water, especially in developing countries. This dangers human health significantly. Several options are available today as wastewater treatment systems, including aerobic treatment, activated sludge plants and filters, anaerobic treatment (Angenent and Sung, 2001) and combination of aerobic and anaerobic processes (Jewell, 1996). Through an extensive treatment of domestic waterwater, the quality of the environment can be enhanched immediately. Sufficient wastewater treatment systems must be simple and efficient. Energy consumption and use of complex equipment should be low. These features are required not only for developing countries, but also for all of the countries, where cost and energy use have to be minimized, while the efficiency of treatment systems should be maximized. In this sense, this study investigates an optimum system which covers the features mentioned above.

UASB technology has been used widely for full-scale treatment of low-strength wastewater (Lettinga et al. 1993; Hulshoff et al. 1997). The UASB reactor is a single vessel reactor with a hydraulic upflow pattern and has a gas-liquid-solid separator system and a feed-distribution system, which help retain biomass and distribute influent, respectively. However, previous studies have sugested that there was a need for a simpler configuration that combines compartmentalization (Angenent and Sung, 2001). The AMBR is relatively simple compared to other anaerobic digestion technologies, however, intermittent mixing with paddles is necessary. An additional advantage of the AMBR over other anaerobic bioreactor configurations is the extremely long sludge retentation times due to the compartmentalized configuration. This ensures proper solids and pathogen destruction while the reactor volume can be kept relatively small. There is no need for an unflow pattern, feed distribution system, or gas-solids separation. In this study, the feasibility of AMBR technology for domestic wastewater in rural communities was investigated.

4. MATERIALS AND METHODS

An AMBR and a UASB reactor (a conventional anaerobic digestion configuration as the control) were operated side-by-side in the Angenent Laboratory at Washington University in St. Louis for an operational period of seven months.

4.1. Set-up and Reactor Operation

An AMBR and a UASB reactor were set-up side-by-side (Figure 4.1) and connected to a feed tank through peristaltic pumps programmed to feed the reactors automatically. The AMBR consisted of a rectangular, Plexiglas reactor (inside dimensions: length = 45 cm, hight = 25 cm, width = 15 cm) with an active volume of 12 L and divided into three compartments. The reactor had round openings at the bottom with a diameter of 2.5 cm which enable the migration of the biomass and limit the shortcircuiting of substrate. The UASB was made of glass with an active volume of 5 L and had a water jacket to maintain constant temperature with an external heating recirculator.

Table 4.1. Operating conditions

Operating conditions	Units	UASB	AMBR
HRT	h	40	40
Reactor volume	L	5	12
Temperature	^{o}C	25	25
pH minimum	NA	6.52	6.34
Upflow velocity	$m\ h^{-1}$	0.8-1	NA
No. of reversals in flow	day^{-1}	NA	0.5
COD loading rate	$g\ L^{-1}\ day^{-1}$	0.36	0.36

The operating conditions for the AMBR and UASB are summarized in Table 4.1. The AMBR was operated in an incubator at 25 ^{o}C and the UASB was kept at 25 ^{o}C with water jacket around the reactor. A air fan was used continuously to keep the temperature same in the incubator.

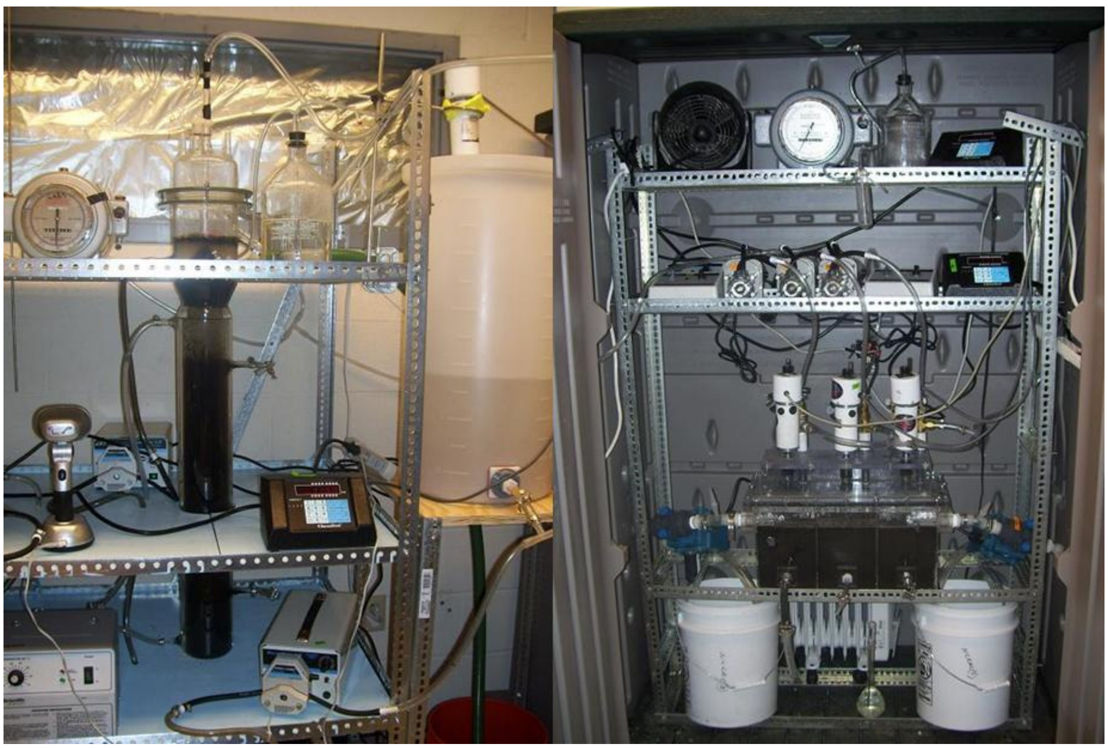

Figure 4.1. Picture of the experimental setup taken in Angenent’s Laboratory

The biogas collection system of each reactor consisted of an bubble observation bottle, a gas sampling port, and a gas meter. Programmable timers were used to control the reactor operation.

The AMBR was seeded with 1.7 L of blended sludge from a mesophilic digestor (Anheuser-Busch, St. Louis, MO) and 0.3 L of biomass present in the rumen of a sheep. The UASB was seeded with 0.7 L of blended sludge and 0.1 L of biomass present in the rumen of a sheep. After inoculation, a 24 h acclimation period was allowed before mixing and another 24 h before feeding.

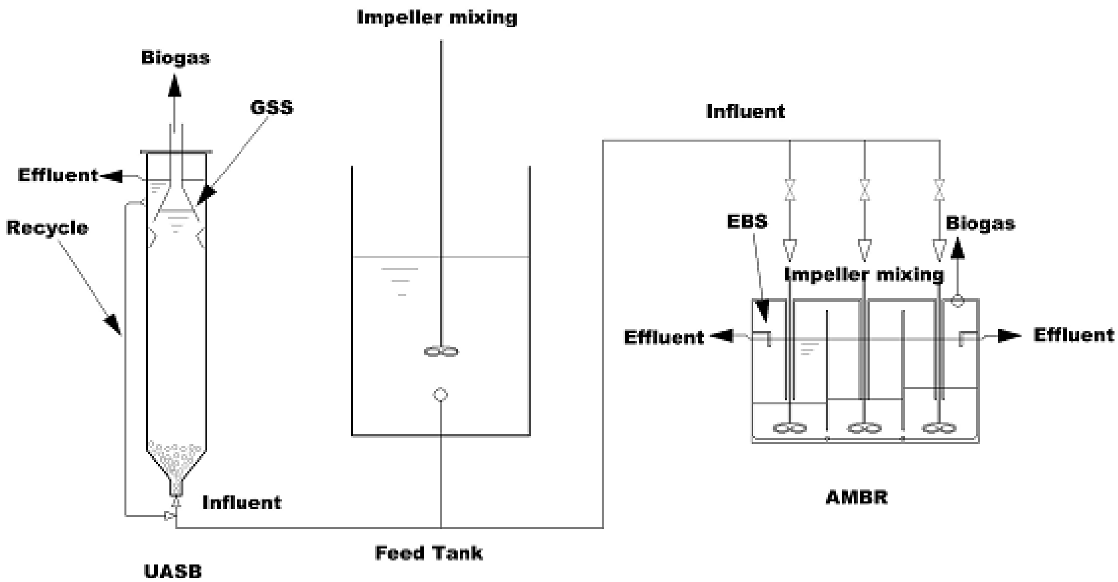

Figure 4.2. Experimental setup

(GSS = Gas-solids separator, EBS = effluent-baffle system)

The synthetic collodial wastewater substrate feed (blended dog food and rice) (Langenhoff et al., 1999) contained per liter of tap water: 0.5 g dry dog food; 0.5 g rice; 1.14 g $NaHCO_3$ and trace elements shown in Table 4.2 (Zehnder et al., 1980). The feed was prepared fresh everyday and mixed by a mechanical agitator with a 6 cm diameter axial flow impeller to stir at approximately 300 rotations per minute (RPM).

The synthetic collodial wastewater substrate feed was fed intermittently every 4 hours with a flow rate sufficient to prevent build up of solids in the feeding tubing. The feed tank was mixed during the intermittent feeding steps to prevent settling. The feed tank was rinsed every day to prevent organic accumulation in the tank.

Table 4.2. Trace element composition (Zehnder et al., 1980)

Chemical	Concentration (mg L^{-1})
$FeCl_3.6H_2O$	10,000
$CoCl_2.6H_2O$	2,000
EDTA	1,000
$MnCl_2.4H_2O$	500
Resazurin	200
$NiCl_2.6H_2O$	142
Na_2SeO_3	123
$AlCl_3.6H_2O$	90
H_3BO_3	50
$ZnCl_2$	50
$(NH_4)_6Mo_7O_{24}.6H_2O$	50
$CuCl_2.2H_2O$	38
HCl (mL L^{-1})	1.0

The AMBR has a special operating feature (Angenent and Sung, 2001). The flow over the horizontal plane of the reactor was reversed every two days (one feeding cycle). After feeding the initial compartment (1^{st}) (Figure 4.3a) for 32 hours (8 feeding steps) the middle compartment (2^{nd}) was fed for 16 hours (4 feeding steps) and effluent was taken from the last compartment (3^{rd}). When one feeding cycle was completed, the flow was reversed by changing the alignment of the compartments (Figure 4.3b) and same feeding procedure was applied as explained above.

Effluent samples were obtained at the midpoint of the time interval between reversals in flow. Therefore, the effluent was sampled after feeding the initial compartment for 1 day (6 feeding steps).

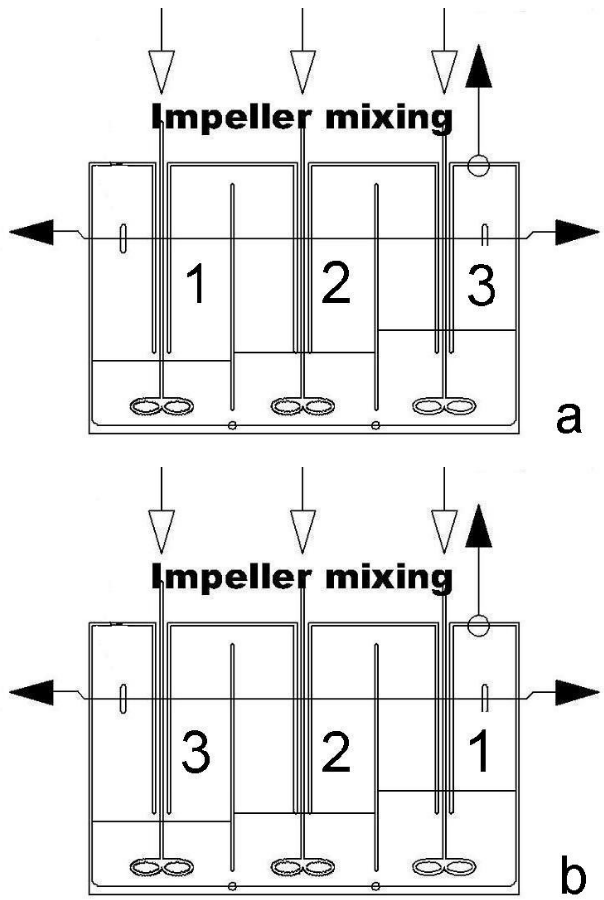

Figure 4.3. Special operating feature of AMBR

4.2. Physical and Chemical Analysis

The reactor performance was monitored with conventional measurements as follows; Daily analyses: pH, biogas production, pressure and temperature of the room, the incubator and the effluents; Weekly or biweekly analyses: Methane composition, TSS, VSS, alkalinity, total ammonia, individual VFA, SCOD and TCOD of the effluents and the methane content in the biogass; Monthly analyses : VFA, biomass concentration (Mixed Liquor Total Suspended Solids (MLTSS), Mixed Liquor Volatile Suspended Solids (MLVSS)) and biomass activity (MAT) were measured.

4.2.1. Analytical Methods

Analyses were in accordance with the procedures outlined in Standard Methods (APHA, 1998).

<u>4.2.1.1. Methane Composition.</u> The analysis was performed by the Gas Chromatograph Gow-Mac Instruments Series 350 equipped with a thermal conductivity detector (Figure 4.4). The GC column was a 3.2 mm OD x 1.2 m length, 20% DC-200 on Chromosorb P AW-DMCS, 80/100 mesh (Varian Inc, Palo Alto, CA). The temperature for the injection port, detection, and column were 50°C, 115 °C, and 25 °C, respectively.

Figure 4.4. Gas Chromatograph Gow-Mac Instruments Series 350

4.2.1.2. Solids Test. The analysis was performed using the Method 2540 B, D and E in Standard Methods (APHA, 1998) and includes TSS, VSS, MLTSS and MLVSS.

4.2.1.3. Volatile Fatty Acids (VFA). The analysis was performed using the Method 5560 C in Standard Methods (APHA, 1998). The distillation setup was installed in the hood of Angenent's Laboratory (Figure 4.5).

4.2.1.4. Chemical Oxygen Demand (COD). This analysis was performed using the Method 5220 C in Standard Methods (APHA, 1998).

Figure 4.5. Distillation setup for VFA analysis

4.2.1.5. Alkalinity. This analysis was performed using the Method 2320 B in Standard Methods (APHA, 1998).

4.2.1.6. Total Ammonia Nitrogen. Total ammonia nitrogen (i.e.,sum of ammonia and ammonium) was measured using an electrode (Model Orion 9512, Thermo Electron Corporation, Bewerly, MA) (Figure 4.6). Standard NH_4Cl solutions were prepared as 0.001, 0.01 and 0.1 M to plot the calibration curve. The pH adjustor (Thermo Scientific, ISA for Ammonia Orion 951211) was added to the standard and sample solutions and the voltage difference of standard and sample solutions were read. The sample calculation is explained in the section 4.2.2.

Figure 4.6. The electrode and the pH adjusting solution used in total ammonia analysis

4.2.1.7. Methanogenic Activity Test (MAT). MAT was adapted from Rinzema et al. (Rinzema, 1988), using the biomass obtained from the operating reactors. The bottles were prepared under an anaerobic hood (Figure 4.7) contained 90% N_2 and 10% H_2. Anaerobic water and trace element solution (Zehnder et al., 1980) were used with 2 g L^{-1} acetate as the organic source. Pure nitrogen was used for flushing the headspace. Before the activity measurement, 1 g L^{-1} acetate was added and pH was adjusted according to the reactor conditions under anaerobic atmosphere (Figure 4.7). Sampling from the headspace was made with a syringe and samples were injected in to the Gas Chromatograph Gow-Mac Instruments Series 350.

Figure 4.7. Anaerobic hood used for the MAT analysis

4.2.1.8. Individual Volatile Fatty Acids (iVFA). Individual volatile fatty acids were measured with the Gas Chromatograph Varian 3400. The GC column was 0.53 mm OD x 15 m length Supelco 2-5326 Nukal (Varian Inc, Palo Alto, CA). The temperature of the injection port, detection, and column were 100^{o}C, 100 ^{o}C, and 250 ^{o}C, respectively. Volatile fatty acid mixture standards were injected before the sample measurements. Samples were prepared according to the procedure prepared by the Angenent et al. The supernatant of the effluent was mixed with %5 formic acid solution at a 1:1 proportion. The standard solution components are acetic, propionic, isobutyric, butyric, isovaleric, valeric, isocaproic, hexanoic and heptanoic acids. The standard solutions were prepared as 0.1, 1, 2, 5, 10 mM and plotted to derive the data equations for each component.

The biogas production rates were measured with gas meters (Model 1 l, Actaris Meter-fabriek, Delft, The Netherlands).

4.2.2. Assessment of the Data

The raw data was converted to relative numbers to be able to interprete accurately.

4.2.2.1. The Standard Methane Production Rate (SMPR). The standard methane production rate was determined according to the following procedure:

1. The methane production rate was calculated from the biogas production rate and the methane content measured by gas chromatography (Series 350, Gow-Mac Instruments, Co., Bethlehem, PA), and adjusted to standard temperature and pressure conditions according to the ideal gas law.
2. The methane production at standard temperature and pressure was converted to SMPR by correcting for the wet volume of the reactor.
3. The dissolved methane present in the effluent was estimated using Henry's law (Perry et al. 1997), and added to the SMPR.

The SMPR was expressed as liters of methane per reactor volume per day ($LL^{-1}day^{-1}$) (Angenent and Sung, 2001). Theoretically, 0.35 L methane is produced per g COD utilized at STP (ignoring biomass growth). The following equation was used to calculate the methane-based COD (MCOD) removal efficiency:

$$MCOD\% = \frac{SMPR}{COD_{loadingrate} x 0.35} 100 \tag{4.1}$$

<u>4.2.2.2. Ammonia Nitrogen Sample Calculation.</u> The values (mV) of different standard NH_4Cl solutions were plotted (Figure 4.8) and a regression equation was derived.

$$\begin{aligned} y &= -25.319 Ln(x) - 141.83 \\ R^2 &= 0.9999 \end{aligned} \tag{4.2}$$

The concentration of the unknown sample was found by putting the voltage reading in to the equation and solving the equation.

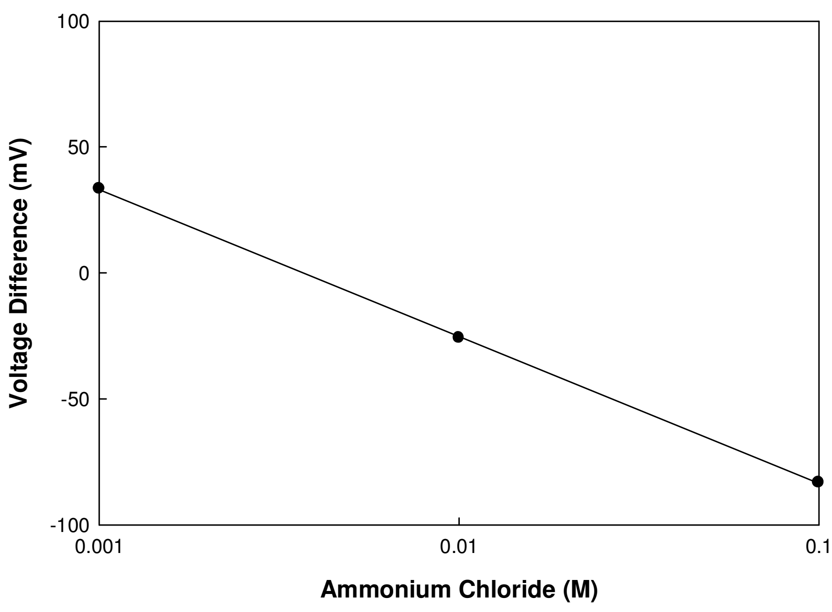

Figure 4.8. Sample plot of NH_4Cl standard solutions for the ammonia nitrogen calculation

5. RESULTS AND DISCUSSION

The aim of this study was to compare the performances of the UASB reactor and AMBR treating low strength wastewater under same operating conditions. Before concentrating on the comparison, results of the reactors are shown separately.

5.1. Up-flow Anaerobic Sludge Blanket Reactor

The UASB reactor was fed by the synthetic wastewater for 195 days of operation. The average COD of the low strenght collodial feed was approximately 600 mg L^{-1}. The feed was prepared everyday according to the recipe writen in the Materials and Methods section. However, even the recipe was followed properly, the COD test results show 56 mg L^{-1} COD standard deviation from the average value of infulent total COD (Figure 5.1). It may be

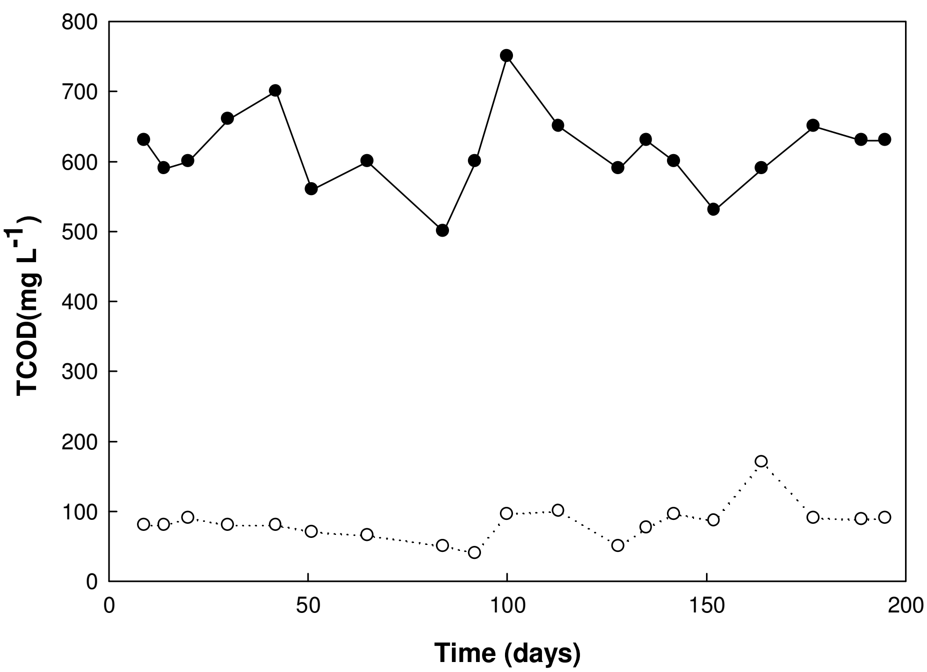

Figure 5.1. Total COD of UASB reactor effluent samples

Symbols represent influent stream(●) and effluent stream (○)

guessed that the deviation was caused by experimental errors occured during the sampling from the feed tank. The homogeneity of collodial feed was difficut to provide during the sampling.

The feed tank was filled with the prepared diluted approximately 17.5 L feed everyday. The daily use of feed was 10.2 L but the experimental error possibilies which might occur during the feeding at night were consired. In addition to that, the outlet of the feed tank was 5 cm higher than the bottom of the feed tank. The feed was prepared more than needed just to tolerate the feeding errors except the daily working hours in the laboratory and to prevent the oxygen escape to the reactors.

Figure 5.2. Experimental setup picture of the UASB reactor

Total and volatile suspended solid tests were performed to asses the solid removal ability of the UASB reactor shown in Figure 5.2. Besides the solids tests, the removal ability was followed by pyhsical appearance of the effluent throughout the 195 days of operation.

The operation period was divided in to phases (days: 0-44, 44-62, 62-80, 80-95, 95-195). Until day 44 it was accepted as the acclimation period. Until the acclimation period was over, the effluent of the UASB reactor contained some washed out sludge and until day approximately 150, it was obvious that the effluent was clear in terms of the sludge.

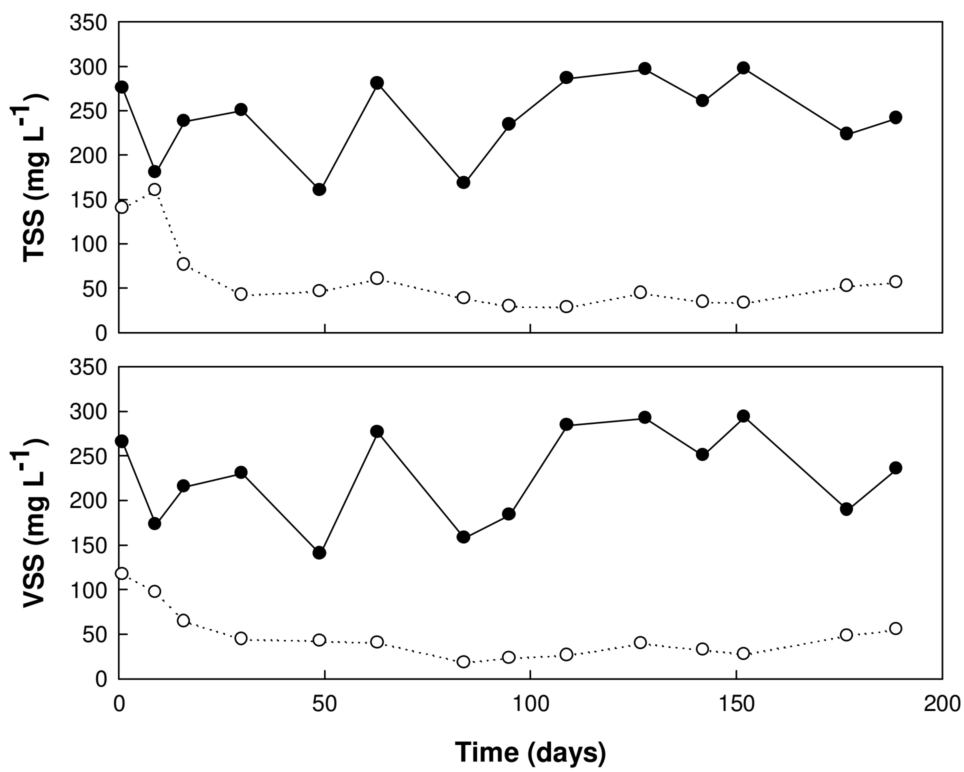

Figure 5.3. Results of total suspended solid and volatile suspended solid analysis of the UASB reactor. Symbols represent influent stream (●) and effluent stream (○)

After day 150, some particals were seen which included some sludge and aerobic bacterial compositions formed on the water surface of the reactor. In Figure 5.9, MLVSS and MLTSS values also prove that the sludge was accumulated in the UASB reactor.

5.2. Anaerobic Migrating Blanket Reactor

The AMBR (Figure 5.6) was fed from the same feed tank which was connected to the UASB reactor. However, the operation of AMBR was very different than UASB reactor as it was explained in the Materials and Methods section.

The physical appearance of the effluent showed a different development in the AMBR. Until approximately day 100 (especially between day 30 and 90) there was a sludge wash out problem in the reactor. The effluent contained significant amount of sludge (Figure 5.5). After operational changes were made, this problem was solved and the solid concentration decreased significantly.

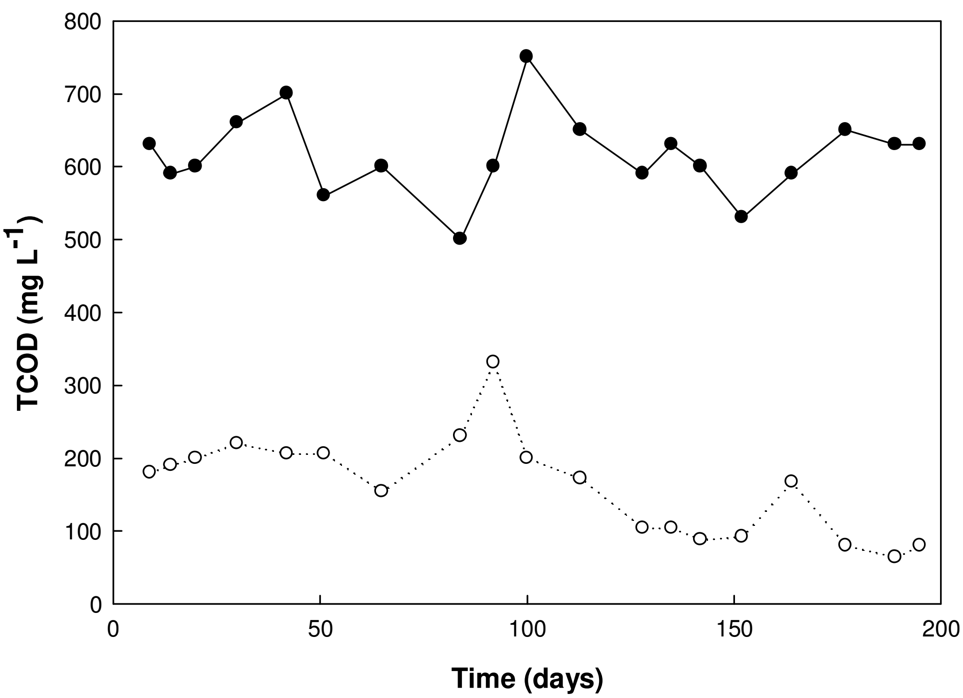

Figure 5.4. Total COD of AMBR effluent samples

Symbols represent influent stream (●) and effluent stream (○)

The solids concentration was high during the wash out period (Figure 5.5). The effluent stream had high solid concentration where influent and effluent solid concentrations were almost same. After day 90, the solid concentrations in the effluent stream decreased. In addition to that, this claim was supported with the physical appearance of the effluent which did not involve solid particles after the operation changes.

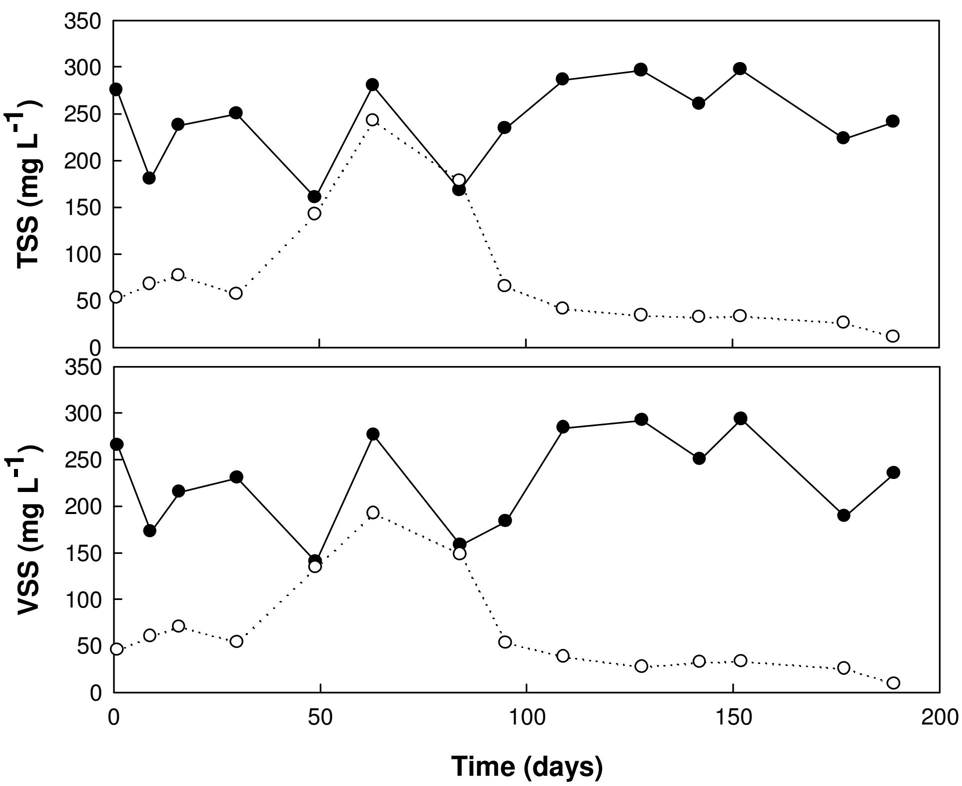

Figure 5.5. Results of total suspended solid and volatile suspended solid analysis of the AMBR. Symbols represent influent stream (●) and effluent stream (○)

5.3. Performance Comparison of UASB Reactor and AMBR

Performance assessment of both reactors was done by conventional parameters. Standard methane production rate, solid removal and COD removal capacities were compared in the figures. Because the configuration, volume and kinetics of the reactors were different, some comparisons were made with the relative numbers calculated from the raw data such as the standard methane production rate (SMPR) and methanogenic activity.

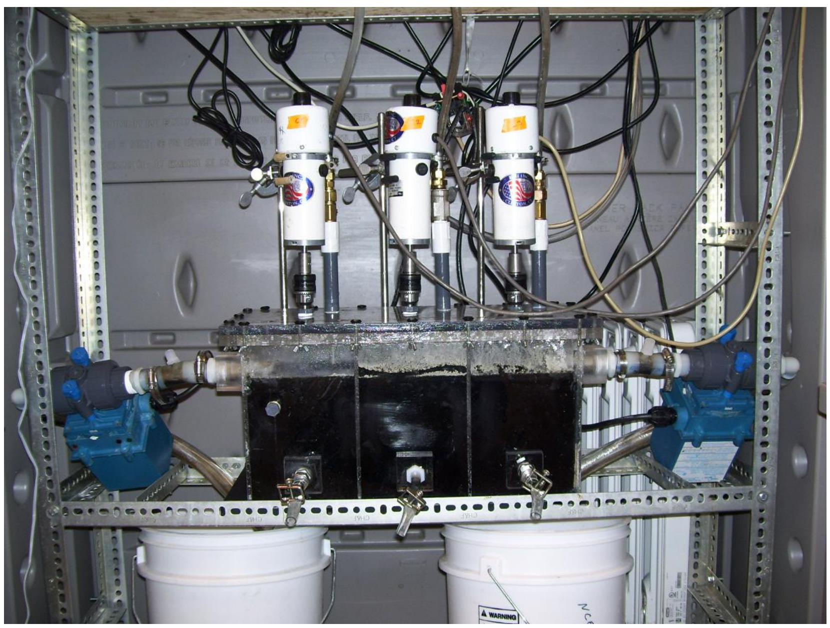

Figure 5.6. Experimental setup of AMBR

5.3.1. Methanogenic Activity Test

Methanogenic activity tests were preformed several times throughout the operating period. The activity tests performed with the synthetic domestic wastewater (regular feed of the reactors) were failed. Therefore, only the tests performed with the acetate solution are presented in this report.

The biomass activity was assessed by dividing the daily methane production by volatile suspended solids concentration in the activity test bottles. This method ensures the accurate comparison between the reactors in case there is a difference between the biomass concentrations of the reactors. The results showed that the biomass activity of the AMBR higher than the UASB reactor, because the biomass concentration of AMBR decreased due to the biomass wash-out problem occurred in first months of the operating period.

Table 5.1. Methanogenic activity test results of UASB and AMBR

Methanogenic Activity Test with Acetate	**Day**	**mL CH_4 g^{-1} VSS day^{-1}**	**SD (n=3)[1]**
UASB	148	6.34	0.34
AMBR	148	15.18	4.29
UASB	195	7.02	1.62
AMBR	195	10.28	0.98

[1] Standard Deviation with three data points

5.3.2. Standard Methane Production Rate

The acclimation period of the biomass to the operating conditions required 44 days. Between day 44 and 62 of the operating period, both reactors showed stability in terms of biogas production rates. A feeding error occured between day 62 and 80 of the operating period, the COD loading rate was higher than the designed loading rate (an organic shock to the reactors). The UASB reactor was able to handle the higher organic loading rate better than AMBR (Figure 5.7). On day 80 of the operationg period, the design COD loading rate was restored again. From day 80 until day 95 of the operating period the reactors recovered from the organic shock load. Between days 95 and 120 of the operating period methane production increased. This may be the effect of a higher pH in the reactors. Starting from day 95, the biogas production was unstable and there was a significant difference between the UASB reactor and AMBR until the last day (195) of the operation .

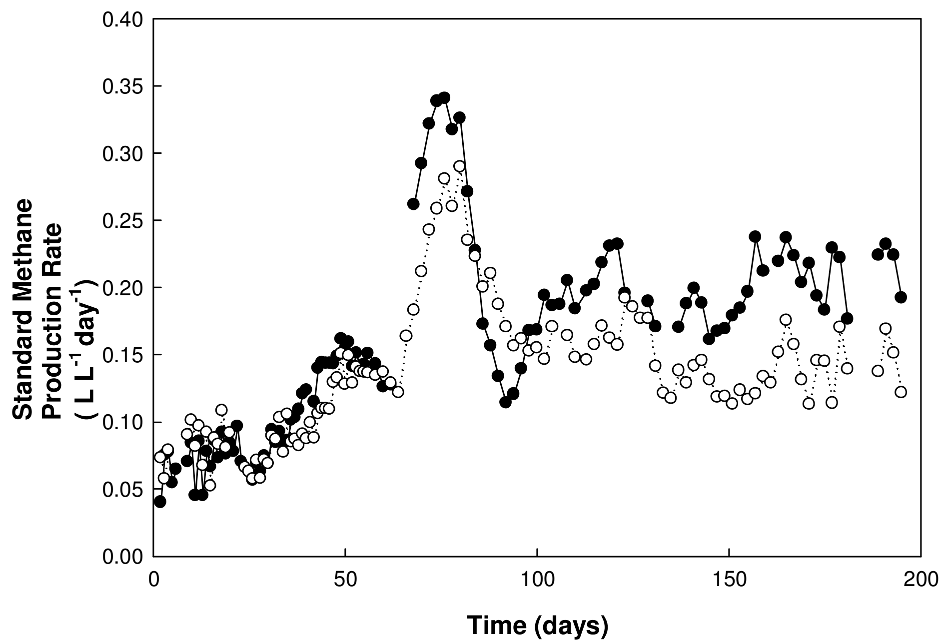

Figure 5.7 Standard Methane Production Rate (SMPR) of the UASB and AMBR. Symbols represent UASB (●) and AMBR (○)

Table 5.2. Average Standard Methane Production Rates (SMPR) of the reactors

	UASB		AMBR	
Operation periods (days)	**SMPR[1]**	**SD[2]**	**SMPR[1]**	**SD[2]**
0-44	0.08	0.02	0.08	0.015
44-62	0.14	0.01	0.13	0.01
62-80	0.30	0.04	0.24	0.04
80-95	0.14	0.02	0.18	0.02
95-195	0.20	0.02	0.15	0.02

[1] $L.L^{-1}day^{-1}$

[2] Standard Deviation

5.3.3. pH Measurements

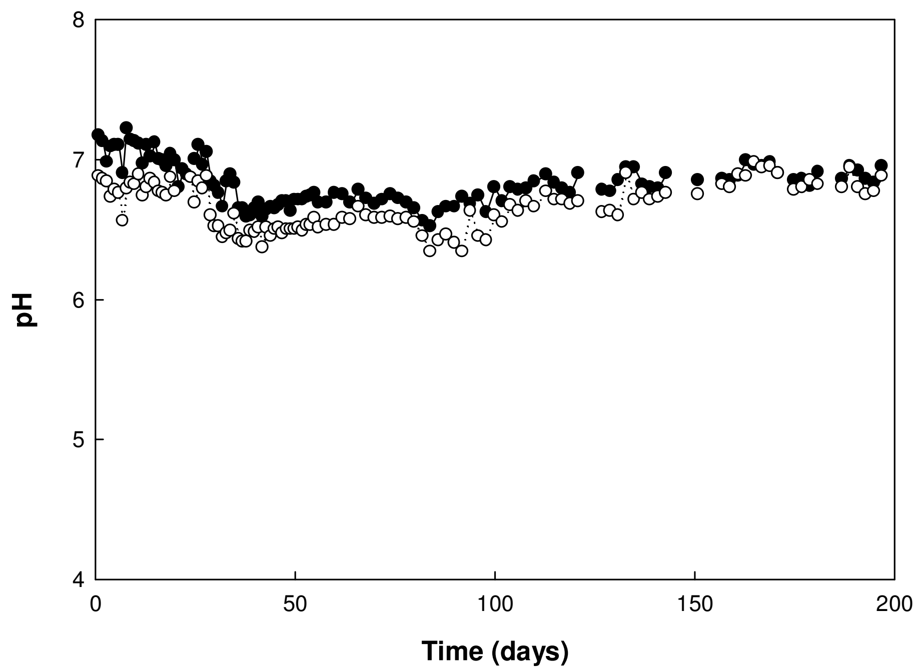

Figure 5.8. pH of UASB and AMBR effluents. Symbols represent UASB (●) and AMBR (○)

At the start of the operation period, the pH of the UASB and AMBR effluent were 7.2 and 6.8, respectively. This value decreased during the start-up period (Days 0-44 of the operating period). Between days 44 and 62 of the operating period, the pH was stable although the buffer concentration was increased slightly. Between days 85-95, the pH values in UASB and AMBR reached the lowest levels of 6.52 and 6.34, respectively. This period was during the time when reactors were recovering from the organic shock load. The buffer concentration of the feed was increased gradually from 0.6 g L^{-1} $NaHCO_3$ to 1.14 g L^{-1} $NaHCO_3$ to increase the pH in the reactors. At the end of the 120-day operating period, the pH of the UASB and AMBR effluents reached 6.8 and 6.7, respectively. The buffer concentration was increased to improve the environmental conditions for the biomass because a low pH may have inhibatory effects on methanogenesis (Methcalf and Eddy, 2004). The higher pH within the neutral ranges correlated with higher methane production rates (Figure 5.7). Until the end of the project (day 195) pH range of the reactors were kept at approximately 6.9.

5.3.4. Biomass Concentration

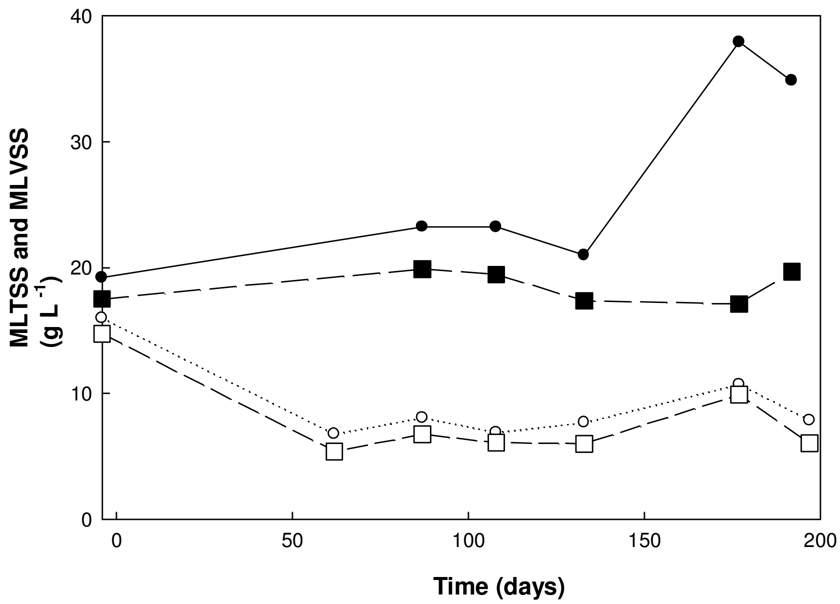

Figure 5.9. Biomass concentration. Symbols represent UASB-MLTSS (●), AMBR-MLTSS (○),UASB-MLVSS (■) and AMBR-MLVSS (□)

The mixed liquor volatile suspended solids (MLVSS) concentrations of AMBR and UASB were 15 g L^{-1} and 17 g L^{-1}, respectively, during the start-up period (Figure 5.9). The initial concentration of biomass increased slightly for the UASB reactor, but decreased for the AMBR. In fact, the MLVSS decreased from 15 g L^{-1} to 5 g L^{-1} due to limitations of feeding solids through thin tubing. Solids were washed out because of the intermittent feeding pattern. On day 92 of operationg period, we decreased the feed flow during a feeding step to prevent excesive washout. In addition, the mixing of the final compartment was terminated. These operational changes decreased sludge wash out considerably and maintained a constant level of the biomass in the AMBR (Figure 5.9). However, the mixed liquor concentrations in the AMBR were much lower compared to the UASB, and this makes a direct comparison between the two systems difficult.

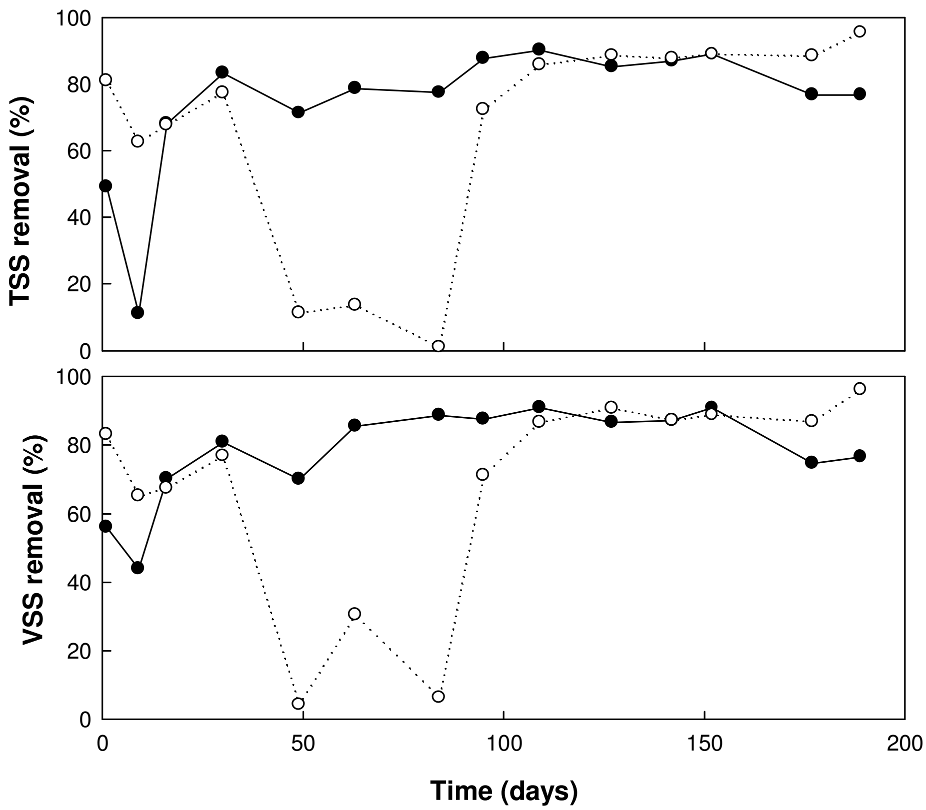

Figure 5.10. Solids removal efficiencies Symbols represent UASB (●) and AMBR (○)

At the end of 110 days of operating period, the MLVSS / MLTSS ratio of the UASB and AMBR are 0.83 and 0.88, respectively. Although the AMBR had a much lower mixed liquor concentration than the UASB, it had a higher MLVSS / MLTSS ratio than the UASB. At the end of the project period (day 195) the MLVSS / MLTSS ratio of the UASB and AMBR became 0.56 and 0.77, respectively. This result showed that there were more solids accumulation in the UASB reactor than AMBR. This may be an advantage for the AMBR in long term operations due to a lower accumulation of total solids. From the literature it is known that accumulation of total solids may become a problem during long term operation.

5.3.5. Solids Removal Efficiencies

The TSS and VSS removal efficiencies for the AMBR were only 10% (average) between day 50 and 85 of the operating period because of the sludge washout problems (Figure 5.10). After day 92 of the operating period, TSS and VSS removal efficiencies for the AMBR became more comparable to the UASB system due to the operational changes explained above. After day 150 until the end of the operating period, the suspended solids removal efficiency of AMBR increased and was more favorable than the UASB reactor.

5.3.6. COD Removal Efficiencies

In both reactors, MCOD removal efficiencies (Angenent and Sung, 2001) were elevated slowly until the high organic load period on day 62 of the operating period after which both reactors needed to achive stable conditions again (Figure 5.11). During the entire operational period, TCOD concentrations of the effluent of AMBR were always higher than UASB because of the presence of washed out biomass in the effluent.

After the operational changes, TCOD removal efficiency of the AMBR reached 80 %. Average TCOD and SCOD removal efficiencies of the UASB reactor were 86.49% (SD = 4.45) and 73.51% (SD = 11.81), respectively. However, steady-state conditions were not reached and it is anticipated that if the operation period were longer than 195 days, steady-state conditions could have been reached. In this case, the statistical values do not explain so much because there is no data at the steady-state conditions.

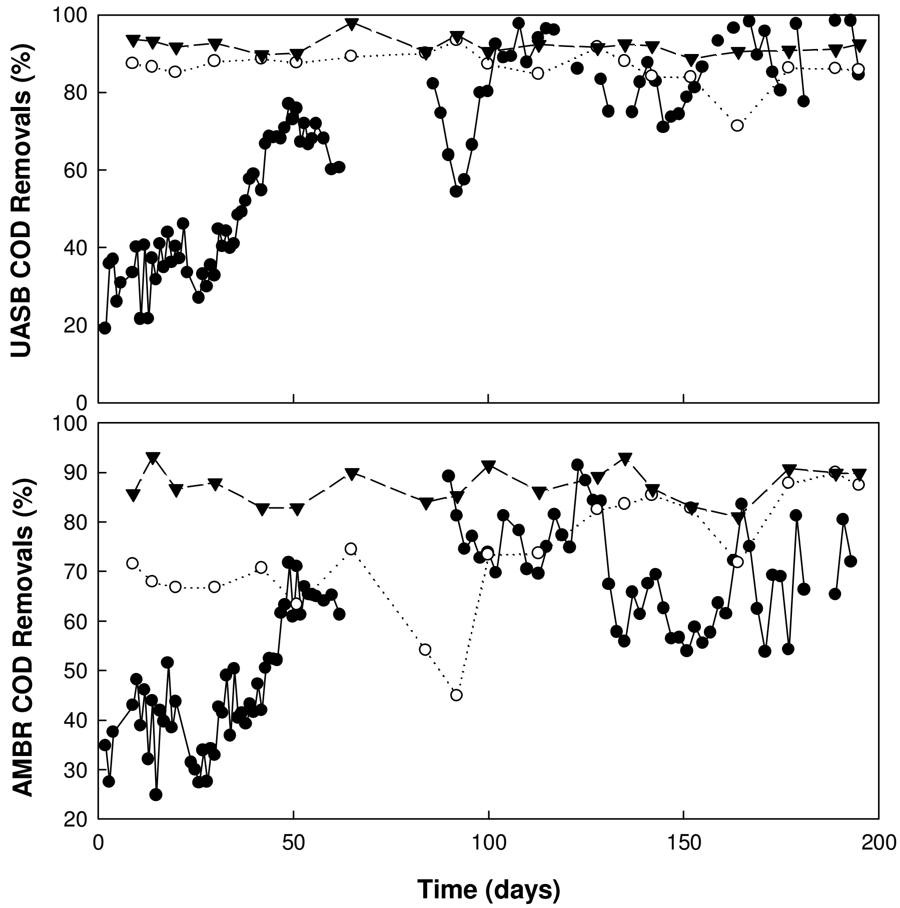

Figure 5.11. COD removal efficiencies in UASB and AMBR. Symbols represent MCOD (●), TCOD (○) and SCOD (▼)

5.3.7. Methane Compositions

The methane composition during the entire operation period showed a consistent pattern with the UASB having a higher methane concentration in biogas than the AMBR (Figure 5.12) (UASB; Average 80 % , SD = 5.2 , AMBR; Average 73 % , SD = 5.2). This may be explained by food to biomass ratio (F/B g COD / g MLVSS.day) concept.

The biomass concentration of the UASB and AMBR shows a distinct difference at the end of 195 days of operating period. On day 195 : F/B $_{UASB}$=0.025 , F/B $_{AMBR}$=0.083. F/B ratio may the reason of lower methane composition in the AMBR. The biomass concentration reached the limit in the UASB reactor. It was predicted that this would lead to lower VSS and TSS removal efficiencies for the UASB. Starting from day 150 the VSS and TSS removal efficiencies in UASB decreased significantly (Figure 5.10).

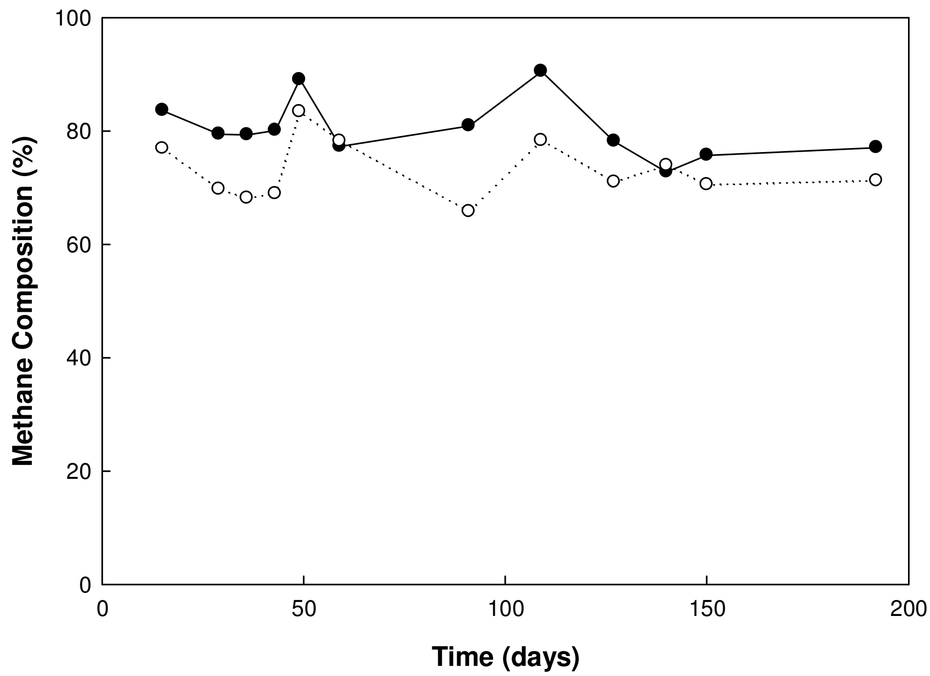

Figure 5.12. Methane composition percentages in UASB and AMBR. Symbols represent UASB (●) and AMBR (○)

5.3.8. Volatile Fatty Acids

Both reactors showed stable total volatile fatty acids (VFA) concentrations at the end of 110 days of operation. (Figure 5.13) VFA concentrations decreased slowly in the UASB reactor. The operational changes, which included decreasing the speed of the AMBR influent pumps and the termination of the final mixing in the compartment helped VFA concentrations to decrease. However, the instability of the reactor could not be worked off. Although no additional changes were made after day 110, there was a problem in both reactors which could not be solved in the entire operation period until day 195.

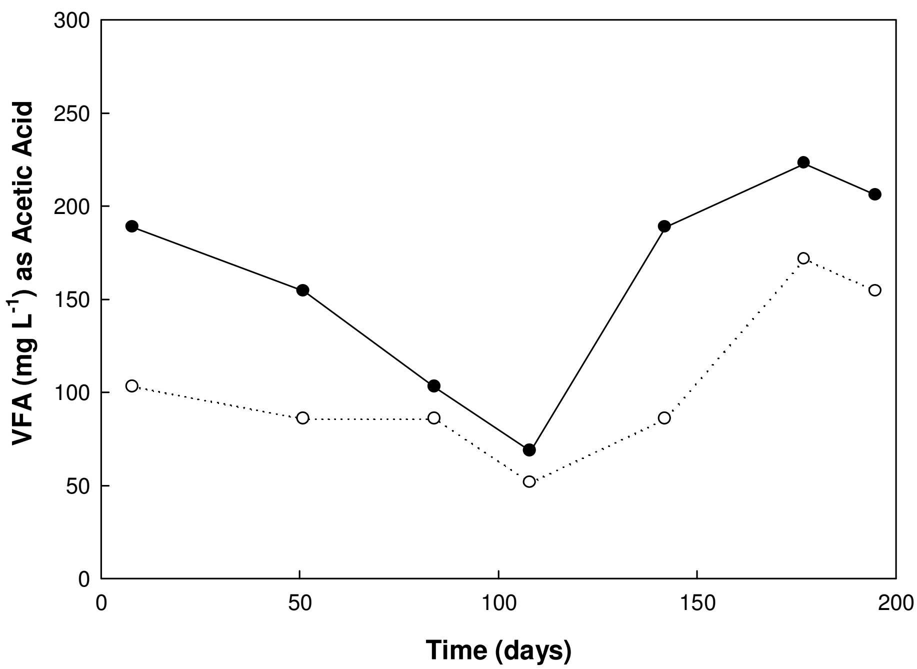

Figure 5.13. Effluent total VFA. Symbols represent UASB (●) and AMBR (○)

Individual volatile fatty acid concentrations were measured with the gas chromatograph as well as the total volatile fatty acid concentrations with the distillation method (Figure 5.14). However, until the end of the period, no significant changes were seen in results of the individual volatile fatty acid test. The acetic acid was the main component. The tests showed that the concentration of other components (propionic, isobutyric, butyric, isovaleric, valeric, isocaproic, hexanoic, heptanoic) were negligible. The results of both tests (Figure 5.14) showed that the trend of the VFA concentration matched in two tests, however, the individual acetic acid concentrations should have been lower than the total VFAs of the both of the reactors where it was in direct contradiction.

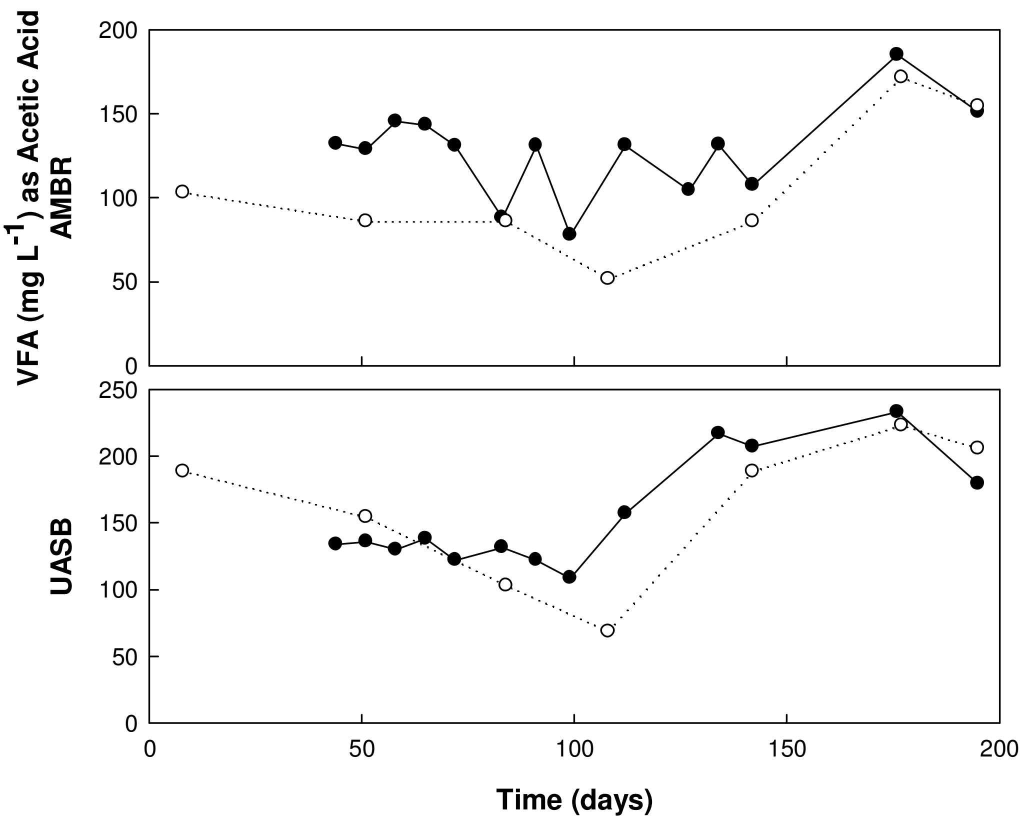

Figure 5.14. The comparison of individual acetic acid and total VFA test results. Symbols represent individual acetic acid concentration (●) and total VFA as acetic acid (○)

6. CONCLUSIONS AND RECOMMENDATIONS

During the start-up period the UASB had shown a better treatment performance of domestic wastewater. The laboratory scale UASB reactor was easier to handle than the laboratory scale AMBR due to a high solids retention. However, steady-state conditions have not been achieved. The MLVSS to MLTSS ratio is more favorable for the AMBR compared to the UASB, which may prevent a long-term problem of total solids accumulation (active biomass crowding) for the AMBR, while the UASB system may accumulate too many total solids. Therefore, a full scale AMBR may be advantageous in that regard.

Synthetic collodial feed caused some problems during the feeding periods. The feed blocked the feeding tubes several times throughout the operation period and this problem disturbed the reactors and prevented the steady-state conditions to be achieved. This problem will be minor if a full scale system is built.

The possible reasons for not reaching the steady-state in the reactors may vary. First of all, the synthetic feed was colloidal and caused blockage in the feeding tubes several times. This stopped the ongoing balanced conditions inside of the reactors. Sometimes, because of the blocked tubes, air went into the reactors during the maintanence. Secondly, the reactors were high-rate and continously fed. A 0.36 g L^{-1} day^{-1} COD loading rate with intermittend feeding might not be proper for these reactors. The loading rate was too low to see the real treating capacity of the reactors, therefore, the comparison of the reactors were more difficult. The initial HRT should have been choosen higher than 40 h. to achieve a higher COD loading rate.

The organic substrate concentration of domestic sewage is generally very low to treat with high-rate anaerobic digesters which have high treatment performance and were used for high loading rates. However, when the conditions of developing countries are considered, it is advantageous to built an AMBR. The temperature in developing countries are usually optimum to stimulate the anaerobic digestion steps and the operational costs of the AMBR are cost effective for these countries.

This study may be repeated and rebuilt by considering these problems that occurred during the 195 days of the operation period. In addition to that, since the acclimation and growth of the bacteria are slow, a longer operation period should be planned to tolerate the possible mistakes in the future studies.

7. REFERENCES

Andrews, J. F., Pearson, E. A., 1965. Kinetics and characteristics of volatile acid production in anaerobic fermentation processes. Air and Water Pollution, 9, 439-461.

Angenent, L. T., Sung, S., 2001. Development of anaerobic migrating blanket reactor (AMBR), a novel anaerobic treatment system. Water Research, 35,7, 1739- 1747.

Angenent, L.T., Karim, K., Al-Dahhan, M. H., Wrenn, B.A., Domingez- Espinosa, R., 2004. Production of bioenergy and biochemicals from industrial and agricultural wastewater. Trends in Biotechnology, 22, 9, 477-485.

APHA, AWWA, WEF, 1998. Standard Methods for the Examination of Water and Wastewater, 20^{th} Edition, Clesceri L. S., Greenberg A. E., Eaton A. D., Washington, D. C.

Barbosa, R. A., Sant'Anna, G. L., 1989. Treatment of raw domestic sewage in an UASB reactor. Water Research, 23, 12, 1483-1989.

Bogte, J. J., Breure, A. M., van Andel, J. G., Lettinga, G., 1993. Anaerobic treatment of domestic wastewater in small scale UASB reactors. Water Science and Technology, 27, 9, 75-82.

Chemicharo, C. A. de L., Cardoso, M. D., 1999. Development and evaluation of a partitioned upflow anaerobic sludge blanket (UASB) reactor for the treatment of domestic sewage from small villages. Water Science and Technology, 40, 8, 107-113.

de Man, A. W. A., Grin, P. C., Roersma, R. E., Grolle, K. C. F., Lettinga, G., 1986. Anaerobic treatment of municipal wastewater at low temperatures, in anaerobic treatment. A grown-up technology, papers of the IAWQ-NVA Conference on Advanced Wastewater Treatment: Aquatech 96, 23-25 September 1996, Amsterdam, The Netherlands, 451-466.

de Man, A. W. A., van der Last, A. R. M., Lettinga, G., 1988., The use of EGSB and UASB anaerobic systems or low strength soluble and complex wastewaters at temperatures ranging from 8 to 30^o C, in Proceedings of the 5th International Symposium on Anaerobic Digestion, Hall, E. R. and Hobson, P. N., eds., Bologna, Italy, 22-26 May, 197-208.

Duran, M., Speece, R.E., 1998. Staging of anaerobic processes for reduction of chronically high concentration of propionic acid. Water Environment Research, 70, 2, 241-248.

Eastman, J.A., Ferguson, J. F., 1981. Solubilization of particulate organic carbon during the acid phase of anaerobic digestion. Journal of Water Pollution Control Federation, 53, 3, Part 1, 352-366

El-Mashad, H.M., Zeeman, G., van Loon, W.K.P., Bot, G. P. A., Lettinga, G., 2004. Effect of temperature and temperature fluctuation on thermophilic anaerobic digestion of cattle manure. Bioresource Technology, 95, 191-201.

Fukuzaki, S., Nishio, N., Shobayashi, M., Nagai, S., 1990. Inhibition of the fermentation of propionate to methane by hydrogen, acetate, and propionate. Applied and Environmental Microbiology, 56, 3, 719-723.

Garcia Encina, P., Rodriguez Flores, R., Fernandez-Polaco, F., van Lier, J., Lettinga, G., 1996. Proceedings of the 4th Latin American workshop and seminar on anaerobic wastewater treatment, Bucaramanga, Colombia, 19-22 November, 159-173.

Ghosh, S., Pohland, F. G., 1974. Kinetics of substrate assimilation and product formation in anaerobic digestion, Journal of Water Pollution Control Federation, 46, 4, 748-759.

Gujer, W., Zehnder, A. J. B., 1983. Conversion processes in anaerobic digestion. Water Science Technology. 15, 127-167.

Halalsheh, M. M., 2002. Anaerobic Pre-treatment of Strong Sewage, Ph.D. thesis, Wageningen University, Wageningen, The Netherlands.

Harper, S.R., Pohland, F. G., 1986. Recent developments in hydrogen management during anaerobic biological wastewater treatment. Biotechnology and Bioengineering, 28, 585-602.

Hartley, K., Lant, P., 2006. Eliminating non-renewable CO_2 emissions from sewage treatment: an anaerobic migrating bed reactor pilot plant study. Biotechnology and Bioengineering, 95, 3, 384-398.

Heertjes, P. M., Kuijvenhoven, L. J., van der Meer, R. R., 1982. Fluid flow pattern in upflow reactors for anaerobic treatment of beet sugar factory wastewater. Biotechnology and Bioengineering 24, 443-459.

Hulshoff Pol, L., Euler, H., Eitner, A., Grohganz, D., 1997. GTZ Sectoral Project; Promotion Anaerobic Technology for the Treatment of Municipal and Industrial Sewage and Wastes, Deutsche Gesellshaft fur Technische Zusammenarbeit GmbH, Internal Report, 13p.

Hungate, R. E., Stack, R. J., 1982. Phenlypropanoic acid: Growth factor for *Ruminococcus albus*. Applied Environmental Microbiology, 44, 1, 79-83.

Jewell, W. J. 1996. Resource-recovery Wastewater Treatments with Biological Systems: Proceedings of the Workshop on Sustainable Municipal Wastewater Treatment Systems, ETC-WASTE, Leusden, The Netherlands, 12-14 November, 67-101.

Kato, M. T., Field, J. A., Versteeg, P., Lettinga, G., 1994. Feasibility of expanded granular sludge bed reactors for the anaerobic treatment of low-strength soluble wastewaters. Biotechnology and Bioengineering, 44, 4, 469-479.

Kennedy, K. J., van den Berg, L., 1982. Anaerobic digestion of piggery waste using a stationary fixed film reactor. Agricultural Wastes, 4, 151-8.

Langenhoff, A. A. M., Narisara, I., Stuckey, D. C., 1999. Treatment of dillute soluble and collodial wastewater using an anaerobic baffled reactor; influence of hydrolic retention time.Water Research, 34,4, 1307-1317

Lant, P., Hartley, K., 2007. Solids characterization in an anaerobic migrating bed reactor (AMBR) sewage treatment system. Water Research, 41, 2437-2448.

Lettinga, G., de Man. A., van der Last, A.R.M., Wiegant, W., van Knippenberg, K., Frijns, J., van Buuren, J.C.L., 1993. Anaerobic treatment of domestic sewage and wastewater. Water Science Technology, 27, 9, 67-73.

Lettinga, G., Roersma, R., Grin, P., de Zeeuw, W., Hulshoff Pol, L., van Velsen, L., Hobma, S., Zeeman, G., 1981. Anaerobic Treatment of Sewage and Low Strength Waste waters, in Proceedings of the 2nd International Symposium on Anaerobic Digestion, Travemunde, Germany, 7 September, 271-291.

Makie, R. I., Bryant, M. P., 1995. Anaerobic-digestion of cattle waste at mesophilic and thermophilic temperatures. Applied Microbiology and Biotechnology, 43, 346-530.

McCarty, P. L., Lawrence, A. W., 1969. Kinetics of methane fermentation in anaerobic waste treatment. Journal Water Pollution Control Federation, 41, 2, R1-R17

Meinzen-Dick, R. Appasamy, P. P., 2001. Urbanization and Intersectoral Competition for Water, http://www.wilsoncenter.org/topics/pubs/popwawa3.pdf

Metcalf and Eddy, Inc, 2004. Wastewater Engineering Treatment and Reuse. McGraw-Hill, New York, USA.

Mgana, S. M., 2003. Towards Sustainable and Robust On-site Domestic Wastewater Treatment for All Citizens, Ph. D. Thesis, Wageningen University, Wageningen, The Netherlands.

Moscoso, J., 1992. Wastewater Use, Web Page of CEPIS Adviser in Wastewater Reuse, http://bvsde.per.paho.org/eswww/proyecto/repidisc/publica/hdt/hdt060.html

Muller, C.D., Abu-Orf, M., Noval, J. T., 2007. Application of mechanical shear in an internal-recycle for the enhancement of mesophilic anaerobic degistion. Water Environment Research, 79, 297-304.

O'Rourke, J. T., 1968. Kinetics of Anaerobic Waste Treatment at Reduced Temperatures. Ph.D. Thesis, Standford University , Stanford, CA.

Pavlostathis, S.G., Giraldo-Gomez, E., 1991. Kinetics of anaerobic treatment: A critical review. Critical Reviews in Environmental Science and Technology, 21, (5 and 6), 411-490.

Perry, R. H., Green D.W., Maloney J.O., 1997. Perry's Chemical Engineer's Handbook, McGraw-Hill, 7th edition, New York, USA.

Pine, M. J., Barker, H. A., 1956. The Pathway of hydrogen in the fermentation. Journal of Bacteriology, 71, 6, 644-648.

Reyes, O., Sanchez, E., Rovirosa, N., Borja, R., Cruz, M., Colmenarejo, M. F., Escobedo, R., Ruiz, M., Rodriquez, X., Correa, O., 1999. Low-strength wastewater treatment by a multistage anaerobic filter packed with waste tyre rubber. Bioresource Technology, 70, 1, 55-60.

Rinzema, A., van Lier, J., Lettinga, G., 1988. Sodium inhibition of acetoclastic methanogens in granular sludge from a UASB reactor. Enzyme and Microbial Technology, 10, 1, 24-32.

Sayed, S. K. I., Fergala, M. A. A. 1995. Two-stage UASB concept for treatment of domestic sewage including sludge stabilization process. Water Science and Technology, 32, 11, 55-63.

Uemura, S., Harada, H., 2000. Treatment of sewage by a UASB reactor under moderate to low temperature conditions. Bioresource Technology, 72, 275-282.

van Haandel, A., Lettinga, G., 1994. Anaerobic sewage treatment: A practical guide for regions with a hot climate. John Wiley and Sons, Chichester.

Vieira, S. M. M., Garcia Jr, A. D., 1992. Sewage treatment by UASB reactor: Operation results and recommendations for design and utilization. Water Science and Technology 25, 7, 143-157.

Wahaab, R. A., 1995. Wastewater treatment and reuse: Environmental health and safety considerations. International Journal of Environmental Health Research, 5, 1, 35-46.

Zehnder, A. J. B., Wuhrmann, K., 1977. Physiology of a *Methanobacterium* strain. Archives of Microbiology, 111, 199-205.

Zehnder, A. J., Huser, B. A., Brock, T. D., Wuhrmann, K., 1980. Characterization of an acetate-decarboxylating, non-hydrogen-oxidizing methane bacterium. Archives Microbiology 124, 1, 1-11.

Zeikus, J. G., 1977. The biology of methanogenic bacteria. Microbiology and Molecular Biology Reviews, 41, 2, 514-541.