

ANAEROBIC TREATMENT OF DILUTE WASTEWATERS

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## **ABSTRACT**

### **ANAEROBIC TREATMENT OF DILUTE WASTEWATERS**

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In this study, domestic wastewater was used to determine the anaerobic treatment performances of a dilute wastewater in one-phase upflow anaerobic sludge blanket (UASB) system and two-phase upflow anaerobic filter (UAF) and UASB system. The acidification performances of domestic wastewater were compared in UAF and UASB reactors. The degree of acidification is higher in UAF reactor (31%) compared to UASB reactor (18%) at 2 h hydraulic retention time (HRT) in the anaerobic treatment of domestic wastewaters. The effluent total volatile fatty acid (tVFA) concentration was higher in UAF reactor than that the UASB reactor at 2 h HRT. The total chemical oxygen demand (tCOD) removal is 40% in winter and 53% in summer in UAF reactor at 2 h HRT. The tCOD removals in two-phase UAF and UASB system and one-phase UASB system were 83% and 63%,

respectively, in 4 h HRT. The conversion rate of COD removed to methane were between 0.005 and 0.067 Nm<sup>3</sup> methane/kg COD removed and between 0.158 and 0.233 Nm<sup>3</sup> methane/kg COD removed in the UAF and the UASB reactor of the two-phase system, respectively. The conversion rate of COD removed to methane varied between 0.029 and 0.199 Nm<sup>3</sup> methane/kg COD removed in one-phase UASB reactor. The results of this study showed that the two-phase reactor system consisting from UAF and UASB reactors provide a good removal of soluble organics variations in acidification reactor and better methane productions in UASB reactor at temperatures 20°C and 35°C through the treatment of domestic wastewater at a HRT of 4 hours.

Key words: Domestic Wastewater, Two Phase, One Phase, Upflow Anaerobic Filter (UAF), Upflow Anaerobic Sludge Blanket (UASB).

## ÖZ

### SEYRELTİK ATIKSULARIN ANAEROBİK ARITIMI

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Bu çalışmada, seyreltik atıksuların anaerobik yöntemlerle tek fazlı yukarı akışlı çamur yatak (YAÇY) sistemi ve iki fazlı yukarı akışlı anaerobik filtre (YAF) ve YAÇY sistemi ile anaerobik arıtım verimlerinin belirlenmesinde evsel atıksu kullanılmıştır. Evsel atıksu asidifikasyon performansları YAÇY ve YAF reaktörlerinde karşılaştırılmıştır. Evsel atıksuyun anaerobik arıtımında, iki saatlik hidrolik bekleme süresinde (HBS) asidifikasyon derecesi YAF reaktöründe (%31), YAÇY reaktörüne (%18) göre daha fazladır. Çıkış suyundaki uçucu yağ asidi (UYA) konsantrasyonu iki saatlik HBS'de YAF reaktöründe YAÇY reaktörüne göre daha yüksek bulunmuştur. Toplam kimyasal oksijen ihtiyacı (tKOİ) giderimi iki saatlik HBS'de YAF reaktöründe kış ayı için %40 ve yaz ayı için %53 oranında bulunmuştur. Dört saatlik HBS'de, iki fazlı YAF ve YAÇY

sisteminde ve tek fazlı YAÇY sisteminde tKOİ giderimleri sırası ile %83 ve %63 oranlarında bulunmuştur. KOİ gideriminin metana dönüşüm oranı ardışık iki-fazlı reaktörün YAF reaktöründe ve iki fazlı sistemin ikinci fazı YAÇY reaktöründe sırasıyla 0.005 ve 0.067 Nm<sup>3</sup> metan/kg KOİ giderim ve 0.158 ve 0,233 Nm<sup>3</sup> CH<sub>4</sub>/kg KOİ giderim aralıklarındadır. Tek-fazlı reaktörde, KOİ gideriminin metana dönüşüm oranı 0.029 ve 0.199 Nm<sup>3</sup> metan/kg KOİ giderim aralığında değişim göstermiştir. Bu çalışmanın sonuçları, 4 saatlik HBS'de, 20°C ve 35°C sıcaklıklarda, evsel atıksu arıtımında, YAF ve YAÇY reaktörlerinden oluşan iki-fazlı reaktör sisteminin asidifikasyon reaktöründe etkili çözülmüş organik madde değişimi ve YAÇY reaktöründe de daha iyi miktarlarda metan verimleri sağlandığını göstermiştir.

Anahtar kelimeler: Evsel Atıksu, İki Fazlı, Tek Fazlı, Yukarı Akışlı Anaerobik Filtre (YAF), Yukarı Akışlı Anaerobik Çamur Yatağı (YAÇY)

*To my mother, father and to the memory of Prof.Dr. Kriton Curi...*

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## ABBREVIATIONS

AAFEB	: Anaerobic attached film expanded bed
ABR	: Anaerobic baffled reactor
AF	: Anaerobic filter
AFBR	: Anaerobic fluidized bed reactor
AH	: Anaerobic hybrid
ASBR	: Anaerobic squenching bed reactor
BOD	: Biochemical oxygen demand
BOD <sub>5</sub>	: 5-day biochemical oxygen demand
BF	: Baffled reactor
COD	: Chemical oxygen demand
CSTR	: Continously stirred tank reactor
tCOD	: Total chemical oxygen demand
sCOD	: Soluble chemical oxygen demand
CSTR	: Completely stirred tank reactor
DO	: Dissolved oxygen
EGSB	: Expanded granular sludge bed
GLS	: Gas liquid seperator
HAc	: Acetic acid
HRT	: Hydraulic retention time
HUSB	: Hydrolysis upflow sludge bed
MLSS	: Mixed liquor suspended solids
MLVSS	: Mixed liquor volatile suspended solids
OLR	: Organic loading rate

RBC	: Aerobic rotating biological reactor
SLR	: Sludge loading rate
SMA	: Specific methanogenic activity
SBR	: Squenching bed reactor
SRT	: Solid retention time
SS	: Suspended solid
tVFA	: Total volatile fatty acids
UASB	: Upflow anaerobic sludge blanket
UAF	: Upflow anaerobic filter
VFA	: Volatile fatty acids
VSS	: Volatile suspended solid
TSS	: Total suspended solids
TS	: Total solids
TVS	: Total volatile solids
PO <sub>4</sub> -P	: Phosphate-phosphorous
TP	: Total phosphorous
NH <sub>4</sub> -N	: Ammonium nitrogen
TF	: Trickling filter.
TKN	: Total kjehdahl nitrogen
WSP	: Waste stabilization pond

## **CHAPTER 1**

### **INTRODUCTION**

#### **1.1. General**

Anaerobic treatment of several wastewaters now seems to be very cost-effective compared to aerobic treatment. Innovations in treatment technology have significantly reduced the size of anaerobic reactors and improved the stability of the operation (Speece, 1996). Increase in cost of sludge disposal and the need to lower energy consumption have focused attention on the potential of incorporating anaerobic processes into wastewater treatment trains. The interest in anaerobic treatment processes for environmental protection and resource preservation is increasing worldwide. It seems that anaerobic treatment systems will become globally the core method of a sustainable integrated low cost environmental protection technology. Anaerobic treatment represents a sustainable and appropriate wastewater treatment system for developing countries and already is becoming an accepted simple and cost-effective technology for the treatment of a variety of wastewaters.

In conventional anaerobic digestion, acidogenic and methanogenic phases proceed in a single reactor. A too rapid acidification of substrate might overload the final stages of conversion of products into methane and carbon dioxide (Cohen et al., 1979). The accumulation of intermediary products might reach concentrations

sufficient to be inhibitory for the methane forming process (Mahr, 1969; Chynoweth & Mah, 1977). Massey and Pohland (1978), Ghosh and Klass (1978), Cohen et al. (1980, 1982), and Anderson et al (1994) have shown improved performances with the implementation of phasing (two-phase configuration) when compared to a single-stage process. Applications and investigations have exhibited the advantages of two-phase anaerobic degradation processes (Ke et al., 2005). Two-phase anaerobic processes have been applied to treat many kinds of wastewater and solid wastes from following sources: distillery, landfill leachate, coffee, cheese whey and dairy, starch, fruit and vegetable solid, food, pulp and paper, olive mill, abattoir, dye, primary and activated sludge and solid (Ke et al., 2005).

The upflow anaerobic sludge blanket (UASB) reactor is the most successfully used as high rate anaerobic treatment system and it becomes possible high volumetric loadings at short retention times and at high temperatures. The performance of the UASB system is limited by slow hydrolysis of entrapped solids especially at low temperatures and a two-phase system can be operated to provide optimal conditions for the microorganisms for greater efficiency in digestion in the treatment of domestic wastewater. At low temperatures, phase separation has been proposed to improve the performance of the anaerobic reactors for the domestic wastewater treatment (Wang, 1994; Elmitwalli et al., 2001).

The two-phase reactors maintain a high solids retention time (SRT: 20-100 d), as keeping the hydraulic retention time (HRT) to a minimum (1.3-20 h) (Langenhoff et al., 2000). In other words, short HRTs are resulted in a smaller reactor configuration. In the aforementioned reactor systems, a variety of dilute soluble and colloidal wastewaters like domestic could be treated economically (Speece, 1996). Finally, two-phase process can be smaller and more cost efficient (Pohland

and Ghosh, 1971; Ghosh and Klass, 1977; Ghosh and Klass, 1982, Ghosh and Conrad, 1975; Fox and Pohland, 1994; Solera et al., 2002).

In the separation of acid and methane fermentation phases, the best environmental conditions can be supplied for each phase in separate reactors. The introduction of an acidogenic phase should allow optimization of required conditions for many complex organic chemicals present in a wastewater to be converted to short-chain volatile fatty acids (VFA) and other simple compounds. Therefore, the slow growing methanogens can be present in the second phase reactor and the risk of inhibition of methanogens by VFA could be eliminated. In the separation of acidogenic and methanogenic phases, it might be provided higher conversion rates and increased stability of the overall process (Cohen et al., 1979). The application of two-stage systems to raw domestic sewage is a rather recent proposition (Wang, 1994, Sayed and Fergela, 1995; Mahmoud, 2002). In two-stage systems, the particulate organic matter is entrapped and partially hydrolyzed into soluble compounds in the first stage and then digested in the second stage while requiring excess sludge to be discharged and stabilized regularly in general (Mahmoud, 2002). Segghezzo (2004) remarked that “two-phase” did not necessarily mean the same as “two-step” or “two-stage”. It was not clear whether a separate acidogenic reactor would be profitable or not in the overall efficiency of the process (Segghezzo, 2004). Lettinga and Hulshoff (1991) stated that a pre-acidification would be beneficial and complete acidification was detrimental in several aspects such as low pH, VFA accumulation and no balance between methanogenesis and acidogenesis.

Domestic wastewater generally contains more particulate organic matters than soluble ones and this influences the overall anaerobic degradation rate (Eastman and Ferguson, 1981). The acid phase is usually the rate limiting step in the degradation of particulate compounds and the degradation rate of these particles in

a reactor is usually slower than the soluble organics. At low temperatures, the hydrolysis step was considered as the limiting step and phase separation has been proposed to improve the performance of the anaerobic reactors treating domestic wastewater (Wang, 1994; Elmitwalli et al., 2001). As a result, it might be worthwhile to investigate the treatment of low strength wastewater with a two-phase system to satisfy the effluent discharge criteria. A separation of the acidogenic and methanogenic phases promotes better degradation of organic carbonaceous and nitrogenous substances. The low pH in the first phase can be used by the acidogenic microorganisms without negatively affecting the methanogens in the second phase. The literature review showed that, all the anaerobic reactor types used recently are not suitable for treating the domestic wastewater containing colloidal compounds. The studies performed on the treatment of dilute colloidal wastewater are limited with fluidized bed (FB) (Sanz and Fdz-Polonco, 1990) and UASB (Barbosa and Sant'Anna, 1989; Kalyuzhnyi et al., 1997) reactors.

The anaerobic treatment of low strength wastewater has been studied using a variety of different reactor configurations. The reactors to treat soluble low strength wastewater that have been most closely examined are those with bacteria attached to carriers, e.g. the anaerobic filter (AF), or the FB and reactors without carrier material, like the UASB, or the expanded granular sludge bed (EGSB), which rely on granule formation to maintain high SRT's (Lagenhoff et al., 2000). The UASB reactor retains a high sludge concentration inside the reactor with simple and low cost equipment (Lettinga et al., 1997) and high biomass concentration allows the reactor operation under high organic loading. In the evolution of high-rate reactors, UASB reactor, the most used alternative, has played an important role. High rate anaerobic wastewater treatment systems like UASB process offer a promising solution for domestic wastewater treatment. Anaerobic filters have been used successfully for the treatment of dilute

wastewater with COD removal efficiencies between 60 and 80% depending on the HRT used (Kobayashi et al., 1983; Matsushige et al., 1990).

In many of developing countries, there is a need for simple and inexpensive domestic wastewater treatment systems. Anaerobic treatment of domestic wastewater has been used successfully in tropical countries, and there are some encouraging results from subtropical and temperate regions (Seghezzi et al., 1998). In a number of tropical countries, full-scale UASB installations have been put in operation for domestic wastewater treatment. Full-scale application of UASB systems for the anaerobic treatment of domestic wastewater is limited so far to the regions with constant and relatively warm temperature conditions (Seghezzi et al., 1998).

In the treatment of low strength wastewater, the possible problems are either wastewater or design related. Wastewater related problems such as low substrate concentration occurring inside the reactor, the possible presence of dissolved oxygen and lower temperatures are inherent to the characteristics of wastewater (Kato et al., 1997). A special problem associated with the fact that at lower substrate concentrations the diffusion rate of substrate to biomass will be lower. The problems associated with low strength wastewater become more significant when biomass population is low. In an anaerobic treatment process, the permissible loading rates are primarily dictated by the sludge retention in the anaerobic reactor. The maintenance of high SRT has been major problem especially for low strength wastewaters (Lettinga et al., 1980). A wastewater treatment process for low strength wastewaters is economical if large volume of wastewater can be treated in a system at a relatively short period of time. This can be provided with the proper application of advanced high rate reactors.

Two-phase anaerobic digestion process has not been extensively studied, before, in the treatment of domestic wastewater. In the other words, there is a lack in the literature in the two-phase treatment of domestic wastewater. Furthermore, no study was found in the literature containing the effect of HRT on two phase reactor system and in which reactor system the acidogenic phase is better provided for optimum reactor volume through anaerobic treatment of domestic wastewater. Therefore, a reactor system in which phase separation performed with an acidified UAF reactor ahead of the UASB reactor for methanogenesis is planned to operate for the treatment of domestic wastewater in this study. The novelties of this study are to determine the anaerobic treatability of domestic wastewater in two-phase system and one-phase system and to compare the effluent characteristics in each phase, separately.

## **1.2. Objective of the study**

The main objective of this study is to contribute to the research area of dilute wastewater treatment by high-rate two phase anaerobic reactor systems in Turkey. In the first continuous reactor studies the possibility of acidification of domestic wastewater was evaluated in UAF and UASB reactors in order to determine the optimum HRT for the acidogenic phase. In the second continuous reactor studies the treatment performances of two-phase UAF and UASB system and one-phase UASB system were compared in domestic wastewaters.

The objectives of this study are summarized below:

- To investigate the effect of HRT and OLR on tCOD, sCOD, sCOD change/VSS ratio, pH, alkalinity, tVFA/sCOD ratio, acidification degree,

solids reduction, gas production, gas composition and nutrient removals in UAF and UASB reactors to determine whether acidification occurred.

- To investigate the treatability of domestic wastewater by two-phase and one-phase systems and to evaluate the performances of two-phase UAF and UASB reactors and one phase UASB reactor treating domestic wastewater.
- To investigate the effects of mesophilic and psychrophilic temperatures on the reactor efficiencies.

## **CHAPTER 2**

### **LITERATURE SURVEY**

The literature survey presented in this study is divided into three sections. In the first section a general review of anaerobic treatment processes is given. This is followed by a summary of studies conducted in relation to high rate reactors for the treatment of domestic wastewaters and the reactor types to be used in this study.

#### **2.1. General Review of Anaerobic Treatment Processes**

The biological treatment processes used for wastewater treatment can be divided into two major groups as aerobic processes and anaerobic processes. In anaerobic processes, the organic wastes breakdown to methane and carbon dioxide while in aerobic processes the organic matter is converted to water and carbon dioxide using the oxygen present in the system.

In anaerobic conditions, the microorganisms convert the organic material to methane and carbon dioxide in the absence of oxygen. In this process the microorganisms take up relatively little energy and their rate of growth is small. In anaerobic treatment a small portion of waste is converted to new cell materials whereas the largest part is converted to methane and carbon dioxide. In anaerobic

treatment, the complex organic compounds must first be hydrolyzed to simple organics after they are fermented to volatile acids by the acidogens (Figure 2.1). The first stage involves acidogenic bacteria which hydrolyze and ferment carbohydrates, proteins and lipids to alcohols, volatile fatty acids, H<sub>2</sub> and CO<sub>2</sub>. The second stage involves acetogenic bacteria which produce acetate, CO<sub>2</sub>, and H<sub>2</sub> from the alcohols and higher fatty acids. Finally, in the third stage, methanogens convert acetate, CO<sub>2</sub>, and H<sub>2</sub> to produce CH<sub>4</sub> and CO<sub>2</sub> (Speece, 1996).

### **2.1.1. The Microbiological Processes in Anaerobic Digestion**

During the anaerobic degradation of organic substances methane is produced through microbial processes. The energy stored in the substrate is predominantly recovered by the methane formed.

The biodegradable organic material is converted to CH<sub>4</sub> and CO<sub>2</sub> by three major groups of bacteria (Fig. 2.1). The fermenting bacteria (group I) converts the organic material to short-chain fatty acids via hydrolysis. Other products of the fermentation process are alcohols, CO<sub>2</sub> and H<sub>2</sub>. The short-chain fatty acids that are longer than acetate are oxidized by the hydrogen producing, acidogenic bacteria (group II) resulting in production of H<sub>2</sub>, formic acid, acetic acid and CO<sub>2</sub>. These products are converted to CH<sub>4</sub> and CO<sub>2</sub> by the methane producing bacteria (group III). Two additional groups of microorganisms are active in methanogenesis. Homoacetogens (group IV) ferments a lot of organics under production of acetic acid. Acetic acid oxidizers (group V) oxidize acetic acid to H<sub>2</sub> and CO<sub>2</sub>. The homoacetogens can produce other types of fatty acids if the concentration of acetate, hydrogen or ethanol is high.

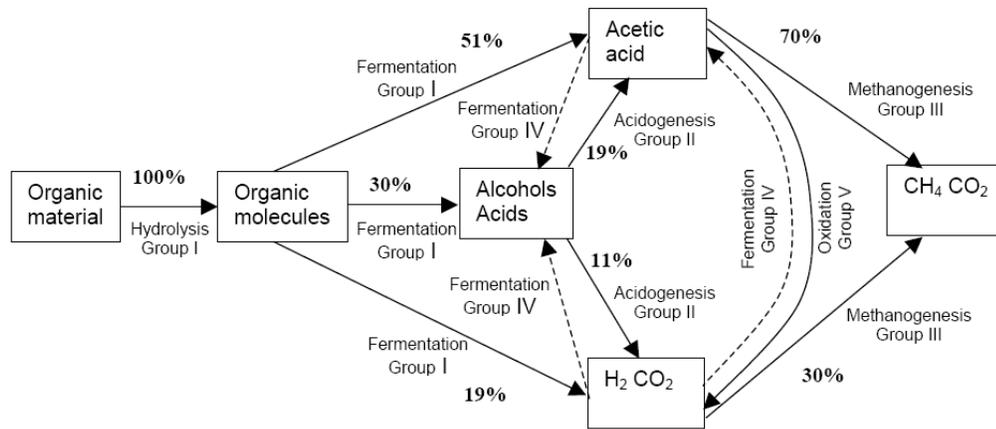


Figure 2.1. Schematic of the processes responsible for degradation of complex organic substances to CH<sub>4</sub> and CO<sub>2</sub> (Poulsen, 2004).

### *Step 1: Hydrolysis*

The hydrolysis step of anaerobic digestion is performed by a mixture of acid formers, which degrade the complex substrate to simpler soluble compounds. The enzymatic hydrolysis occurs in the substrate solution via exocellular enzymes produced by the bacteria cells. The hydrolysis results in the formation of sugars from carbohydrates, amino acids from proteins, and fatty acids from lipids. The intermediate compounds are further broken down to soluble organic end products such as formate, acetate, propionate, butyrate, lactate, ethanol and carbon dioxide gas (Gunaseelan, 1997).

### *Step 2: Acetogenesis*

The intermediates of the hydrolysis are degraded by fermentative bacteria predominantly to volatile fatty acids, mainly acetate along with carbon dioxide. Acetate is the major intermediate in the bioconversion of organic matter to methane and carbon dioxide. Under anaerobic conditions, acetogenic bacteria are active in a wide temperature range of 3 to 70°C, with an optimum at around 30°C.

They need an intensive contact with the substrates while hydrogen is produced as a by-product from this stage (Gunaseelan, 1997; Yılmaz, 2007).

*Step 3: Methane formation*

The third step involves the production of methane by methanogenic bacteria. They are very specific for the temperature ranges and classified as psychrophilic ( $\leq 20^{\circ}\text{C}$ ), mesophilic ( $21-40^{\circ}\text{C}$ ) and thermophilic ( $> 40^{\circ}\text{C}$ ). They convert the intermediate products to methane and carbon dioxide. Approximately 70% of the methane is formed from acetic acid by acidotrophic methane bacteria (Gujer and Zehnder, 1983). The remaining 30% are obtained by the utilization of hydrogen and carbon dioxide by hydrogenotrophic bacteria. If hydrogen concentration increases above a minimal level ( $10^{-4}$  atm), the conversion to acetate by the acetogens will be reduced. Since the main pathway for methane production is the removal of acetate, a decreased rate of biogas production will result (Vandevivere et al., 2002). Since the methanogenic bacteria are sensitive and growing slow, it is important to maintain the optimum temperature and pH. Methanogenic bacteria are strictly anaerobic, the presence of molecular oxygen is toxic for them, and even inorganic sources of oxygen, (e.g. nitrates) may inhibit their growth.

Anaerobic treatment of compatible wastes seems to be very cost-effective compared to aerobic treatment. New advances in treatment technology have significantly reduced the size of anaerobic reactors and improved the stability of the operation. The main advantages and drawbacks of anaerobic treatment is given in Table 2.1 (Speece, 1996).

### **2.1.1.1. Key Factors Affecting Anaerobic Degradation**

The operating conditions significantly affect anaerobic digestion process. The loading rate and the influent strength are very important in anaerobic treatment because of excess accumulation of acids would result in the failure of the digester. Temperature and pH are important variables in anaerobic treatment because of the sensitivity of methane producing bacteria to these. Some important factors affecting anaerobic degradation in domestic wastewater can be given as temperature, pH, HRT, suspended solids, reactor design and operation (Seghezzi, 1998).

#### *a. Effect of Temperature*

Anaerobic digestion is highly dependent on the reactor temperature (Bogte et al., 1993; van Haandel and Lettinga, 1994) and can be grouped based on the temperature (Pol, 1995): psychrophilic, mesophilic and thermophilic (42-75 °C). The optimum range for mesophilic digestion is between 30°C to 40°C and for temperatures below the optimum range the digestion rate decreases. Temperature affects the final degradation extent and at low temperatures, more organic matter will remain undegraded at a given hydraulic retention time due to slow hydrolysis of volatile solids. Thus, for a given degradation degree, the lower the temperature, the longer is the degradation time. In the mesophilic range, the bacterial activity and growth decreases by one half for each 10°C drop below 35°C (Pol, 1995). Therefore, for a given degree of digestion to be attained, the lower the temperature, the longer is the digestion time.

Table 2.1. Positive features and possible disadvantages of anaerobic treatment  
(Speece, 1996)

Positive features of anaerobic biotechnology
<ul style="list-style-type: none"> <li>•provision of process stability</li> <li>•reduction of waste biomass disposal cost</li> <li>•reduction of nitrogen and phosphorus supplementation costs</li> <li>•reduction of installation space requirements</li> <li>•conservation of energy, ensuring ecological and economical benefits</li> <li>•minimization of operation attention requirement</li> <li>•elimination of off-gas air pollution</li> <li>•avoidance of foaming with surfactant wastewaters</li> <li>•biodegradation of aerobic non-biodegradables</li> <li>•reduction of chlorinated organic toxicity levels</li> <li>•provision of seasonal treatment</li> </ul>
Possible disadvantages of anaerobic treatment
<ul style="list-style-type: none"> <li>•long startup requirement for development of biomass inventory</li> <li>•insufficient inherent alkalinity generation potential in dilute or carbohydrate wastewater</li> <li>•insufficient effluent quality for surface water discharge in some cases</li> <li>•insufficient methane generation from dilute wastewaters to provide for heating to the 35 °C optimal temperature</li> <li>•sulfide and odor generation from sulfate feed stocs</li> <li>•nitrification not possible</li> <li>•greater toxicity of chlorinated aliphatics to methanogens vs aerobic heterotrophs</li> <li>•low kinetic rates at low temperatures</li> <li>•high NH<sub>4</sub> concentrations (40-70 mg/L) required for maximum biomass activity</li> </ul>

The effect of temperature on the hydrolysis and acidogenesis processes of anaerobic digestion is not very significant because of the availability of some bacteria having their optimum range among the mixed population. The second and third stages of decomposition can only be performed by certain specialized microorganisms (acetogenic and methanogenic bacteria), which are much more

sensitive to temperature shift (Mudrak and Kunst, 1986, Rajeshwari et al., 2000). Domestic wastewater is a complex type of wastewater, characterized by a high fraction suspended solids and relatively low temperatures (Seghezzi et al., 1998; Lier et al., 2001). When using UASB Reactors, the SRT should be long enough to provide methanogenic conditions, which imposes a long HRT at low temperatures. The reduction in operational temperatures leads to a significant decrease in the maximum growth and substrate utilization rates (Lettinga et al., 2001).

However, an important characteristic of anaerobic bacteria is that their decay rate is very low at temperatures below 15°C. Thus, it is possible to preserve the anaerobic sludge for long periods without losing much of its activity for seasonal treatment (Rajeshwari et al., 2000).

#### *b. Effect of pH*

In anaerobic treatment, the value and stability of the pH is very important because methanogenesis only proceeds at a high rate when the pH is maintained in the neutral range (6.3 to 7.8) (van Haandel and Lettinga, 1994). The influence of pH on the acidogenesis of soluble substrate range from 4.0 to 7.9, and recommended the pH range of 5.7- 6.0 for the acid reactor to provide a stable and most favorable substrate for the methane reactor. In general, the pH of a domestic sewage is in the optimum range without the need for chemical addition due to the buffering capacity of the most important acid-base system in an anaerobic digester (van Haandel and Lettinga, 1994). Elefsiniotis and Oldham (1994) reported that the variation in pH between 4.3 and 5.2 did not affect VFA production and COD solubilization, but higher pH levels (5.9-6.2) affected both parameters in acidogenesis. Variations in higher pH levels from 6.0-8.0 affect the dominant microbial populations in the acid reactor (Demirel and Yenigun, 2002).

The pH of conventional anaerobic system is typically maintained between methanogenic limits, to prevent the predominance of the acid forming bacteria, which may cause VFA accumulation. It is essential that the reactor contents provide enough buffer capacity to neutralize any eventual VFA accumulation, and thus prevent build-up of localized acid zones in the conventional anaerobic digestion. While in two-phase anaerobic degradation system, each stage can use different pH value so that acidogenesis and methanogenesis processes proceed in optimal conditions, respectively. pH controlling is very important, especially to the methanogenic stage.

The distribution of effluent products was also substantially influenced by pH, and VFA since last one was strongly dependent on pH. Acetate, butyrate, and *i*-butyrate predominated above pH 6.0, whereas propionate predominated below pH 5.0, the region between pH 5.0 and 6.0 was the transition zone. Since the significant changes in product distribution occurred between pH 4.0–7.0, pH control should be important for the production of a stable effluent composition from an acidogenic reactor (Yu et al. 2003). Ren et al. (1997) stated that the operating pH, plays a major role on the effluent composition of the acidogenic reactor.

The optimum pH for the acidogenic activity was found to be close to 5.9 in the acidogenesis of gelatin-rich wastewater in an upflow reactor (Yu and Fang, 2003). Yu et al. (2002) stated that as pH increased, partial pressure of hydrogen decreased accompanied by the increase of methane production. At pH 6.5, the biogas contained 31% methane and became free of hydrogen. This indicates that most of the hydrogen produced was consumed by the hydrogenotrophic methanogens at this pH. In order to effectively separate the acidogenic phase from the methanogenic phase, it is important to keep the pH low, 5.5 or less.

Breure and Van Andel (1984) and Bull et al. (1984) stated that pH value varied during acidogenesis, which caused the system tend to buffer itself toward a pH value to the range of 5-7 if no control was carried out. In the treatment of a complex wastewater like domestic sewage, pH is usually in the optimum range without the need for chemical addition, due to the buffering capacity of the most important acid-base system in an anaerobic digester: the carbonate system (van Haandel and Lettinga, 1994; Segghezzo, 1998). Alexiou et al. (1994) concluded that the final pH value mainly depended on the nature of wastewater while secondly on the environmental conditions existing in the reactor.

*c. Effect of Suspended Solids (SS)*

The SS present in the wastewater can affect the anaerobic treatment in the following ways (Lettinga and Hulshoff Pol, 1991):

- Reduce the specific methanogenic activity of the sludge due to adsorption and entrapment of poorly or non-biodegradable SS
- Formation of scum layers
- Counteract the formation of granular sludge
- Spontaneous and sudden washout of the sludge in case of prolonged continuous entrapment of SS in a granular sludge bed.

The adsorption of finely dispersed SS to the sludge granules were detected during anaerobic digestion (Lettinga et al., 1983; Grin et al., 1985). This phenomenon may wrap the active biomass particles with a film of increasing thickness of non-biomass matter and results in difficulty the substrate transport through the active biofilm.

*d. Effect of Organic Loading Rate (OLR)*

In anaerobic wastewater treatment, the organic loading rate plays an important role. The chemical oxygen demand (COD) is generally used to measure the content of organic matter in wastewater thus the OLR for biological systems is stated in terms of COD per reactor volume per unit time (i.e., kg COD/m<sup>3</sup>.day). The OLR can be varied by changing the influent concentration and by changing the HRT. In the nonattached biomass reactors, where the hydraulic retention time is long, overloading results in biomass washout and leads to process failure. Fixed film, expanded and fluidized bed reactors can withstand higher organic loading rate and the systems can be rapidly restored to normal in the case of a failure because of shock load. In comparison to a continuous stirred tank reactor (CSTR) system, fixed film and other attached biomass reactors have better stability.

*e. Effect of Hydraulic Retention Time (HRT)*

The microorganisms require a certain amount of time to digest the organic matter and to achieve the desired level of treatment. The HRT can be defined as the reactor working volume divided by the daily influent flow rate. It is important the control of the HRT for preventing cell washout of slow-growing methanogenic microorganisms (Shieh et al., 2000).

Dinopoulou et al. (1988) and Chang et al. (1989) stated that HRT was a key parameter for the performance of a hydrolysis-acidification reactor. The maximum process efficiency is usually obtained by operating the acidogenic step at short HRT, thus preventing methanogens development (Guerrero et al., 1999). Kim et al. (2002) observed that VFA concentrations and distributions changed as a function of HRT. Elefsiniotis and Olham (1994) reported that VFA

concentrations increased with HRT up to 12 h and then decreased at an HRT of 15 h with an increase in gas production during the acidogenesis of primary sludge indicating the stimulation of methanogenesis. Demirer and Chen (2004) stated that the pH drop was inversely proportional with the increase in HRT of the reactor and volatile solid removal during acidification showed an increase with increase in HRT at constant organic loading rate. The hydrolysis/acidogenesis of wastewater was greatly influenced by the HRT (Zoetemeyer et al., 1982; Henry et al., 1987). The control of HRT was found critical to the successful enrichment of hydrolytic/acidogenic bacteria in the first reactor of a two-phase system (Fang and Yu, 2001).

Domestic wastewater is a complex type of wastewater, characterized by a high fraction of suspended solids and mostly of relatively low temperatures (Segghezzo et al., 1998; Lier et al., 2001). When using one step UASB reactors, the solid retention time (SRT) should be long enough to provide methanogenic conditions, which imposes a long HRT at low temperatures (Zeeman and Lettinga, 1999). In the investigations of Castillo et al. (1997), the importance of HRT was emphasized on a pilot scale UASB reactor. In the treatment of domestic wastewater, it was observed that the removal efficiencies of sCOD and tCOD increased with an increase in HRT but the removal efficiencies became constant at HRTs higher than 6 h. Their results showed that the removal efficiencies strongly depended to the variations of HRT (changing OLR imposed by increasing HRT) in domestic wastewater.

## **2.2. High-rate bioreactors**

The importance of the sludge retention concept in reducing the reactor size began to be recognized in the 1950's (McCarty, 2001). One of the most important successes in the development of anaerobic wastewater treatment was introduction of high-rate reactors in which biomass retention and liquid retention became independent from each other (Lettinga et al., 2001). The term "high-rate" is now widely used to refer to anaerobic treatment systems meeting at least the following two conditions: a) high retention of viable sludge under high loading conditions, and b) proper contact between incoming wastewater and retained sludge (Lettinga et.al, 1987). In high-rate systems, wastewater flows through the anaerobic sludge where purification takes place and organic matter is converted into biogas and sludge. High-rate bioreactors provide a high reaction rate per unit volume thus reducing reactor volume allows the application of high volumetric loading rates (Borja et al., 1994; Barber and Stucky, 1999; Rebac et al., 1999) and low energy requirements (Lettinga et al., 1997; Rajeshwari et al., 2000). Table 2.2 summarizes some of the important features of high rate reactors.

Anaerobic treatment in high rate reactors is recognized as the core method of an advanced technology for environmental protection and resource preservation, and represents a sustainable and appropriate wastewater treatment system for developing countries (Lettinga et al.,1987; Lettinga,1996). Lettinga and co-workers introduced UASB and investigated and developed several modified versions of UASB system such as, the Expanded Granular Sludge Bed (EGSB) system (Man et al., 1988), the UASB-septic tank reactor (Bogte et al., 1993), the Hydrolysis Upflow Sludge Bed HUSB (Wang, 1994), the Staged Multi Phase

Anaerobic Reactor (Lier, 1999) and the two-stage Anaerobic Filter- Anaerobic Hybrid (AF-AH) system (Elmitwalli, 2000).

Table 2.2. Characteristics of different reactor types (Stronach et al 1986, Kaul & Nandy 1997) adapted from Rajeshwari et al., 2000

Anaerobic reactor type	Start-up period	Channeling effect	Effluent recycle	Gas solid separation device	Carrier packing	Typical loading rates (kgCOD/m <sup>3</sup> day)	HRT (d)
CSTR	-	NP	NR	NR	NE	0.25-3	10-60
Contact	-	NEx	NR	NR	NE	0.25-4	12-15
UASB	4-16	Low	NR	Essential	NE	10-30	0.5-7
AF	3-4	High	NR	Beneficial	Essential	1-40	0.5-12
AAFEB	3-4	Less	Required	NR	Essential	1-50	0.2-5
AFB	3-4	NEx	Required	Beneficial	Essential	1-100	0.2-5

NP= Not present, NR=Not Required, NE=Not essential, NEx=Not existent

Yu and Anderson (1996) stated that the new advanced (high-rate or biomass retained) anaerobic reactors such as UASB, anaerobic filter (AF), anaerobic fluidized bed (AFB), anaerobic attached film expanded bed (AAFEB) and expanded bed (EB) reactors had an excellent biomass retention and significant difference between the HRT and SRT. These systems offer great opportunities for the treatment of a large variety of medium and low strength wastewaters and these systems might also overcome the other reactor design related problems of conventional anaerobic treatment systems such as unstable operation (Yu and Anderson, 1996). The application of high rate reactors has enhanced the recognition of anaerobic digestion as a cost effective and efficient technology for

environmental protection (Parawira, 2004). High-rate bioreactors include the UASB, packed-bed and fluidized-bed reactor, based on the mechanism used to achieve biomass retention within the bioreactors.

### **2.2.1. Anaerobic Packed Bed (fixed film) Reactor**

In these reactors microorganisms grow as a biofilm on the surface of inert carriers. In stationary fixed film reactors (Figure 2.2), the reactor has a biofilm support structure (media) such as activated carbon, PVC (polyvinyl chloride) supports, or ceramic rings for biomass immobilization and the wastewater is distributed from above/below the media. In fixed film reactors, the advantages were given as: simplicity of construction, elimination of mechanical mixing, better stability at higher loading rates, and capability to withstand large toxic shock loads (Van den Berg et al., 1983) and organic shock loads (Lettinga, 1995). The reactors can recover very quickly after a period of starvation (Van den Berg et al., 1983). The main limitation of the reactor design is the reactor volume is relatively high compared to other high rate processes due to the volume occupied by the media. Another constraint is clogging of the reactor due to increase in biofilm thickness and/or high suspended solids concentration in the wastewater (Rajeshwari, 2000).

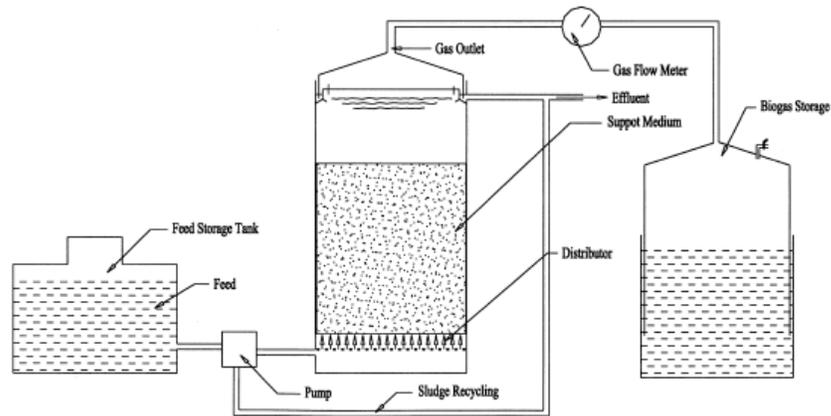


Figure 2.2. Configurations of Fixed Film Reactor, Rajeshwari (2000).

The early anaerobic filters consisted of a submerged column packed with rocks, which provided a support surface for the attachment of microorganisms. Coulter et al. (1957) developed a two stage system for the treatment of sewage. This system consisted of a sludge contact chamber and a packed rock contact chamber. In the investigations of Pretorius (1970) it was used an anaerobic filter preceded by a modified upflow digester. Pretorius (1970) reported that most of the gas production and COD reduction occurred in the filter and not the upstream digester in contrast to the findings of Coulter et al.(1959). Pretorius (1970) also showed that dilute wastewaters could be treated with high efficiencies provided that the SRT was carefully controlled. Anderson et al.(1994) observed mainly unattached biomass was retained in the voids of a nonporous medium. Young and Dahab (1983) found that most of the COD was removed in the lower end of column by suspended biomass of an upflow anaerobic filter. Sludge accumulation is often encountered in anaerobic filters. Periodic solids removal would be necessary if the reactor were to operate under high hydraulic loadings and this could be accomplished without causing problems (Genung et al., 1980). Manariotis and

Grigoropoulos (2006) observed that the anaerobic filter process was a promising method for the treatment of raw municipal wastewater.

### **2.2.2. Upflow Anaerobic Sludge Blanket Reactor (UASB)**

The UASB reactor is a high-rate methane bioreactor with a sludge bed, or blanket of settled microorganisms through which the wastewater flows upwards (Yan et al., 1990). The main advantage of the UASB process is that no support material is required for retention of the high-density anaerobic sludge (Lettinga et al., 1980; Elias et al., 1999; Zoutberg and Eker, 1999). However, the absence of carriers necessitates the availability and maintenance of highly settleable biomass either as flocs or as dense granules (0.5-2.5 mm in size) (Callander and Barford 1983; Lettinga,1995). The UASB does not require the expense and energy consumption of pumps for recirculation of effluent (Lettinga and Hulshorff 1991; Rajeshwari et al., 2000; Wentzel et al., 1995). The dense structure and high settleability of the sludge (60-80m/h), allow upflow anaerobic reactors to be operated at very high upflow liquid velocities, without loss of granules (Lettinga and Hulshoff, 1991; Wentzel et al., Zoutberg and Eker,1999). The channelling problems may occur only at low loading rates and when the distribution by the feed inlet is poor.

There are certain disadvantages of the UASB design. The bed can be disrupted if the influent flow rate is too fast, or if gas production is too vigorous. The bioreactor may not treat particulate in the bed, thus reducing its effectiveness per unit volume. Another disadvantage is that the reactor requires granular seed sludge for faster start-up. A major advantage is that the technology has comparatively less investment requirements when compared to an anaerobic filter or a fluidized bed system. Among notable disadvantages, it has a long start-up period along with the requirement for a sufficient amount of granular seed sludge

for faster startup. Moreover, significant wash-out of sludge during the initial phase of the process is likely and the reactor needs skilled operation. A UASB reactor (Figure 2.3) essentially consists of gas-solids separator (to retain the anaerobic sludge within the reactor), an influent distribution system and effluent draw of facilities. Significantly higher loading rates can be accommodated in granular sludge UASB reactors as compared to flocculent sludge bed reactors (Lettinga, 1982). In the latter, the presence of poorly degraded or non-biodegradable suspended matter in the wastewater results in an irreversible sharp drop in the specific methanogenic activity because the dispersed solids are trapped in the sludge. Moreover, any significant granulation does not occur under these conditions.

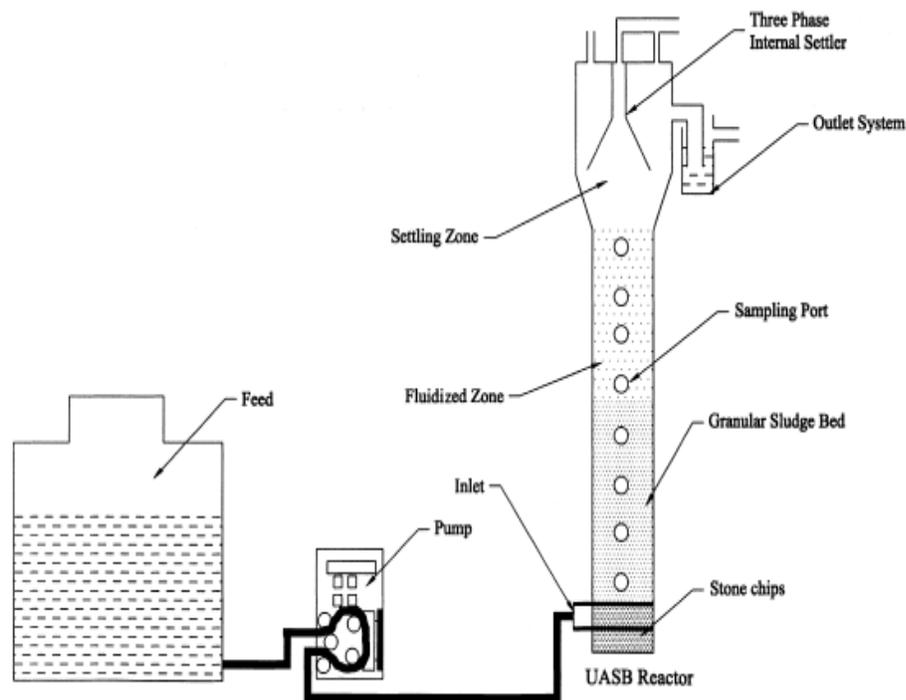


Figure 2.3. Configuration of Anaerobic Sludge Blanket Reactor (Rajeshwari, 2000).

Another high rate digester, EGSB, is a modified form of UASB in which a slightly higher superficial liquid velocity is applied ( $5\pm 10$  m/h as compared to 3 m/h for soluble wastewater and  $1\pm 1.25$  m/h for partially soluble wastewater in an UASB) (Lettinga, 1995). Because of the higher upflow velocities, mainly granular sludge will be retained in an EGSB system. A significant part of granular sludge bed will be in an expanded or even in a fluidized state in the higher regions of the bed and the contact between the wastewater and sludge is excellent. Moreover, the transport of substrate into the sludge aggregates is much better as compared to situations where the mixing intensity is much lower (Lettinga, 1995). The maximum achievable loading rate in EGSB is slightly higher than that of an UASB system.

One of the most serious problems associated with expanded-bed digesters is the instability of the granular conglomerates during continuous operation. This also applies, though to a much lesser extent, to UASB reactors and loss of biomass might occur due to: (i) granule disintegration, (ii) wash-out of hollow granules, (iii) occurrence of fluffy granules, and (iv) scaling due to inorganic precipitates (Rajeshwari et al., 2000).

### **2.2.3. Anaerobic Fluidized-Expanded Bed Reactor**

Fluidized bed or expanded bed reactors consist of a reactor filled with granular material to which biomass adheres in thin film, a wastewater distributor, head space for collection of methane gas and effluent–recycling systems. Wastewater flowing up through these reactors fluidizes, or at least expands the bed of particles to which the microorganisms are attached by 20-50%, so good contact between wastewater and biomass is ensured (Chen et al., 1985); Lettinga et al., 1984; Parawira, 2004). The principle used for the expansion is also similar to that for the fluidized bed, i.e. by a high upflow velocity and recycling.

In the anaerobic fluidized bed (Figure 2.4), the media for bacterial attachment and growth is kept in the fluidized state by drag forces exerted by the upflowing wastewater. The media used are small particle size sand, activated carbon, etc. Under fluidized state, each media provides a large surface area for biofilm formation and growth. It enables the attainment of high reactor biomass hold-up and promotes system efficiency and stability. This provides an opportunity for higher organic loading rates and greater resistance to inhibitors. These reactors have several advantages over anaerobic filters such as elimination of bed clogging, a low hydraulic head loss combined with better hydraulic circulation (Collivingnarelli et al., 1991) and a greater surface area per unit of reactor volume. As a result of these factors, the capital cost is lower due to reduced reactor volumes. However, the recycling of effluent may be necessary to achieve bed expansion. Because the principle used for fluidization and expansion is a high upflow velocity usually provided by recycling.

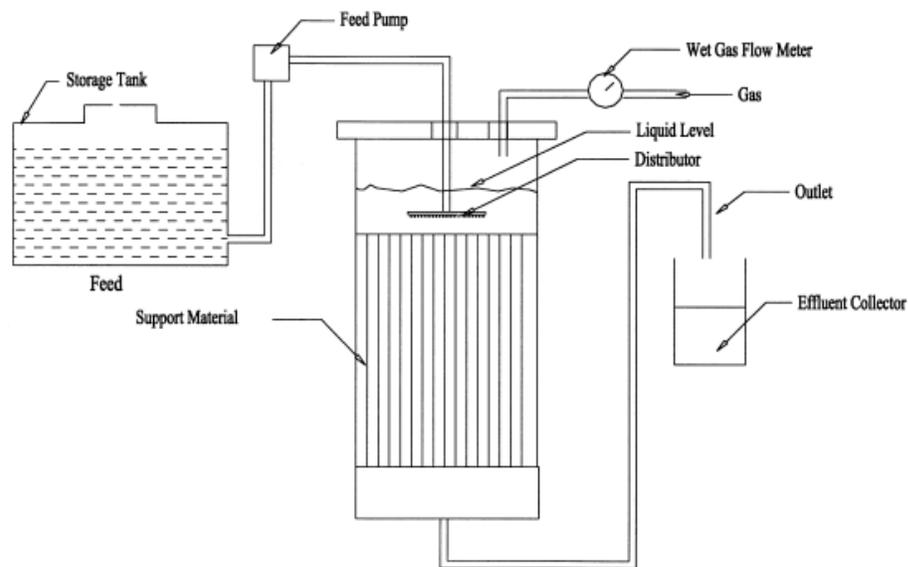


Figure 2.4. Configuration of Anaerobic Fluidized Bed Reactor (Rajeshwari, 2000).

#### 2.2.4. Factors Governing the Reactor Choice for Anaerobic Treatment

Rajeshwari et al. (2000) considered an anaerobic digestion system more reliable less capital, less land area and being able to run at high organic and hydraulic loading rates with minimum operation and maintenance requirements. The organic and hydraulic loading potential of reactor depended on; (1) Amount of active biomass that can be retained by a reactor per unit volume, (2) Contact opportunity between the retained biomass and the incoming wastewater; (3) Diffusion of substrate within the biomass. Based on the comparisons of various reactor types, the orders recommended for reactor choice are summarized in Table 2.3 (Rajeshwari et al., 2000).

The biomass retention is provided by biofilm attachment to the packing in the downflow filters while biofilms are provided on high settling particles in the fluidized and expanded bed reactors. The biomass retention in the UASB reactors is provided by high settling velocity dense granules. In the upflow filter configuration, a minor fraction of the biomass is attached to the packing surface and most of the biomass remains unattached (Speece,1996).

Table 2.3. Comparisons of various reactor types (Rajeshwari et al., 2000)

Parameters	Rating
Operating skills	Fixed film < UASB < RBC < fluidized bed
Energy consumption	UASB < fixed film < EGSB < fluidized bed < RBC
Capital cost, land requirement, operation and maintenance	RBC < fixed film < UASB < EGSB < fluidized bed

Based on the characteristics of the different reactors COD reduction, biomass retention and other factors like cost, operation and maintenance requirements, UASB and fixed film configurations appear to be the most suitable for the anaerobic treatment of wastewaters.

The UASB reactor has less investment requirements when compared to an anaerobic filter or a fluidized bed system. Among notable disadvantages, it has a long start-up period along with the requirement for a sufficient amount of granular seed sludge for faster startup. Moreover, significant wash-out of sludge during the initial phase of the process is likely and the reactor needs skilled operation. The UASB reactor can be considered as the best choice with the only limitations being the tendency of granules to float and shearing of granules at high loading rates. These constraints are also valid to a lesser degree for attached biomass reactors such as fixed film, fluidized bed and rotary biological contactor. In addition, due to the space occupied by the media, the attached biomass reactors possess comparatively lower capacity for biomass retention per unit volume of the reactor (Rajeshwari et al., 2000).

The upflow anaerobic filter (UAF) reactor is simply of constructed, the mechanical mixing is not required and is capable to prevent the toxic shocks (Van den Berg et al.,1983) and organic shock loads (Lettinga, 1995). The reactors can recover very quickly after a period of starvation (Van den Berg et al., 1983).

#### **2.2.5. Two-Phase Reactors**

In addition to various anaerobic reactor design configurations, anaerobic systems can be operated as single-phase or two-phase systems. Single-phase systems involve only one reactor for the microorganisms to digest the organic matter,

whereas two-phase systems separate the hydrolysis and acidogenic, and methanogenic organisms into two separate reactors. Since the nutrient and growth requirements of the acidogenic and methanogenic organisms may be different, the two-phase system can be operated to provide optimal conditions for the microorganisms in each phase for greater efficiency in digestion. In the first phase of the two-phase system, the acid fermentation phase, acidogenic organisms digest organic solids and complex soluble organics, converting them to Volatile Fatty Acids (VFAs). In the second phase, methane-producing microorganisms (methanogens) utilize the VFAs to produce methane and carbon dioxide.

Two phase anaerobic digestion means the separation of the nonmethanogenic and methanogenic digestion phases in separate reactors. Anaerobic digestion is performed by distinct groups of bacteria that differ with respect to physiology, nutritional requirements, growth characteristics, metabolic characteristics, environmental optima, and sensitivity to environmental stress (Stafford et al.,1980). The relative growth rates of methanogenic and hydrolytic/acidogenic bacteria are different and dominant populations of either of these two bacterial groups can be promoted by controlling digester operation conditions. The different growth rate and pH optima for acidogenic and methanogenic organisms has led to the development of the two phase AD processes (Ghosh, 1987).

The acidogenic bacteria perform the hydrolysis and acidogenesis step of anaerobic digestion whereas the optimum pH is 5.2-6.5 at acidogenesis step (Demirer and Chen, 2004). At the second step, the products of the first step which can not be metabolized by methanogenic bacteria such as propionate and butyrate are degraded to acetate and H<sub>2</sub> at an optimum pH of 6.6-7.6 (Speece,1996: Solore et al.,2002). Finally the optimum pH environment for methanogens is within the range 7.5-8.5 (Hobson and Wheatly, 1993).

Treating wastewater in two phases will refer to the development of unique biomass in each reactor, first an acid forming phase, followed by a methanogenic phase. Separate phasing optimizes environmental conditions for each phase because in single phase processes both classes of organisms are forced to operate in a common environment (Speece et al., 1996).

Demirer and Chen (2005) summarized two phase systems advantages over one stage processes as: 1) allows the selection and enrichment of different bacteria in each phase, in the first phase, complex pollutants are degraded by acidogenic bacteria into VFA, which are subsequently converted to CH<sub>4</sub> and CO<sub>2</sub> by acetogenic and methanogenic bacteria in the second phase, 2) increases the stability of the process by controlling the acidification phase in order to prevent overloading and the built up of toxic materials, 3) the methanogenic phase buffered by the prior acid phase and prevented pH shock to the methanogenic population. Additionally, two-phase systems can be smaller and more cost efficient because of the acidogenic phase conditions being at low pH, a high organic loading and a short hydraulic retention time (Ghosh and Klass, 1977; Fox and Pohland, 1994; Solera et al., 2002).

Cohen et al. (1980) evaluated the influence of phase separation on the anaerobic digestion of glucose using single-phase and two-phase UASB reactors. Cohen et al. (1980) observed that under shock loading conditions, single and two phase systems accumulated volatile acids. Two-phase system returned to typical operating conditions in 1/5 of the time required by the single phase system. The acid forming stage can handle overloading and will recover rapidly from failure due to toxic substances. As a result methanogenic stage can be operated safely near the maximum loading (van den Berg and Kennedy, 1983).

In the past, two-phase anaerobic digestion has been studied on; municipal sludges by a system consisting of an upflow acid-phase digester and a CSTR methane digester (Ghosh., 1987, 1991), cellulosic wastes by an acidogenic and fixed film reactor (Girard et al., 1986), sewage sludge by a CSTR acid-phase digester and an upflow anaerobic reactor (Lin and Quang,1993), waste activated sludge by a complete mixing tank and a fluidized bed (Shimizu et al., 1993), dairy wastewater by an acidogenic CSTR and an UAF reactor (Anderson et al., 1994), cheese whey by an acidogenic CSTR and an UAF reactor (Yilmazer and Yenigün,1999), diluted molasses by a CSTR and a fluidized bed reactor (Romli et al., 1994), acidified glucose media by an acidification and anaerobic upflow reactor (Cohen et al.,1980), sewage sludge by an acid phase A-UASB and a methane phase UASB (Fongsatitkul et al., 1995), dye waste by an acidogenic CSTR and UASB reactor (Chinwekivanich, 2000), unscreened dairy manure by a continuously-mixed acidogenic anaerobic reactor and a methanogenic reactor (Demirer and Chen, 2004 and 2005). In some of the two-phase studies, the obtained COD removal efficiencies were about; 85% for cane-molasses alcohol stillage (Yeoh,1997), 90% for landfill leachate (Lin,1991), 87% for slaughterhouse wastes (Banks and Wang,1999), 90%-98% for cheese whey and dairy wastewater (Malaspina et al.,1996; Strydom et al., 1997; Ince, 1998), 84% for pulp and paper (He et al., 1995) and volatile solids removal efficiencies were; 30-62% for unscreened dairy or cattle manure (Demirer and Chen, 2004) and 43% for primary and mixed activates sludges (Ghosh and Taylor, 1999).

Zhang and Noike (1991) found that the substrate degradation patterns of the two systems were very different and concluded that the conventional single-phase system could not be regarded as just the sum of the acidogenic and methanogenic phases of the two-phase system. In addition, they found that the number of acetate-utilizing methanogens in the methanogenic reactor of the two-phase system were 2 to 10 times higher than in the single-phase system. The two-phase

system was more stable during changes in pH than the single-phase system and the methanogenic reactor of the two-phase system produced a higher methane percentage at a greater rate compared with the single-phase system. Jeyaseelan and Matsuo (1995) studied the effects of phase separation on two different synthetic substrates prepared from baby food formula and powdered skimmed milk by two-phase anaerobic digestion at 20°C. The two-phase system consisted of an anaerobic continuously stirred tank reactor (CSTR) for acidogenesis and an UAF reactor for methanogenesis. A single-phase UAF reactor was previously operated under similar conditions using the same synthetic feed for comparison. Compared with the single-phase system, the two-phase system produced better digestion efficiencies based on biogas production. The authors concluded that phase separation is a definite advantage in waste stabilization especially when lipids are present in the wastewater.

The significance of phase separation is still unclear as the overall efficiency of two-phase systems is similar to or slightly better than single-phase systems when treating real wastewaters with high COD and solids concentrations. Several researchers comparing the single-phase and two-phase anaerobic digestion processes of various synthetic wastewaters have concluded that phase separation can significantly improve the performance of the methanogenic reactor as a result of the optimized conditions for acidogenesis and methanogenesis in each reactor. However, few of the comparison studies were conducted using actual raw wastewater as the substrate and therefore have limited impact in the design of full-scale treatment systems.

### **2.3. Treatment of Low Strength (Dilute) and Municipal Wastewaters**

The application of anaerobic technologies for the treatment of concentrated wastewaters dates back over 100 years (McCarty and Smith, 1986) and the

simplest and most widely used process is the septic tank (Jewell, 1987). The feasibility of anaerobic processes for the treatment of low strength wastewater was firstly demonstrated by the development of anaerobic contact processes and anaerobic filter. In these processes, the key concept relates to the ability to control the mean cell retention time. Before the development of these processes, the requirement of long mean cell retention time and no knowledge on the technique to obtain mean cell residence time independent of HRT resulted in very large anaerobic reactor volume requirements.

A lot of industrial wastewaters in developing countries, originated from the food processing sectors, are frequently found with low strength since the water management is not always very efficient and large volumes of dilute wastewaters are produced (Kato et al., 1997). Low strength wastewaters can be characterized as dilute industrial effluents of less than 2000 mg/L COD, which may contain a variety of biodegradable compounds (Lettinga and Hulshoff Pol, 1991) e.i. effluents from alcoholic and soft drink bottling industries, paper recycle and papermaking mills. Mergeart et al. (1992) and Ndon and Dague (1994) define the low strength wastewaters as those with COD value less than 1000 mg/L as municipal sewages.

Some problems have arisen from anaerobic treatment of low strength wastewaters. The problems are either wastewater or reactor design related. Wastewater related problems such as low substrate concentration the presence of dissolved oxygen and lower temperatures, are inherent due to the characteristics of wastewater (Kato et al., 1997). At lower substrate concentrations the diffusion rate of substrate to biomass will be lower. The problems become more pronounced when biomass population is low.

High organic loading rates in an anaerobic treatment process could be tolerated by the sludge retention in the anaerobic reactor. The maintenance of a high solid retention time (SRT) has been the major problem especially for wastes with a COD below about 3000 mg/L (Lettinga et al., 1980). A waste treatment process for low strength wastes is economical if large volumes of waste can be forced through the system in a relatively short time period. Therefore, the biomass retention of processes can be controlled of the wastewater flow rate. Conventional anaerobic treatment processes of the flow-through type are therefore inadequate to treat low strength wastes. Therefore, an anaerobic process that can overcome the problems between biomass and substrate is a system that is able to retain the granular biomass and to grow at low substrate concentrations (Ndon and Dague, 1994). As high biomass concentrations, could be maintained by slow growing methanogenic bacteria in the reactor. These organisms are necessary for removing the organic pollutants from the wastewater in the anaerobic digestion process (Van Der Last and Lettinga, 1992).

Since 1980, a considerable amount of research has been carried out on treatment of municipal wastewater by using various anaerobic reactor types. The studies relevant to the treatability of low strength wastewaters in varied advanced anaerobic reactors are given in Table 2.4. In that table, the studies are given in a historical evidence and it may be concluded that the treatment efficiencies decreased with the decrease in the strength of the influent wastewater. It was observed that full scale reactors were built only in tropical countries and subtropical countries. Some of the important studies performed under high, and low temperatures are summarized below:

The studies performed with the treatment of low strength wastewaters, the COD removal efficiencies at studied HRTs and temperatures are tabulated in Table 2.4.

In this table it was observed that the COD removals varied between 30% and 85% in one phase lab-scale UASB reactors and the COD removals affected by HRT, OLR and temperature applied to the reactors. At high HRT and temperature values it was observed high COD reduction percentages. At high to moderate temperatures, it was observed 47-79% COD removals at 3-18 h HRTs in pilot-scale reactors. Temperature affected the COD removals and at low temperatures such as 4°C resulting in decreases in COD removals from 54% to 24% in pilot-scale UASB reactors (Man et al., 1987). In full-scale applications of UASB reactors, the COD removals were between 60-74% at lower temperatures (16.5°C) and low HRTs (6 h). In low strength wastewater, the strength of influent domestic wastewater affected the COD reductions. Decreases in influent COD values were resulted decreases in COD removals (Collivignarelli, 1990; Segghezzo, 2002).

In Table 2.4, it was depicted that, the COD removals were between 33-85% at temperatures and HRTs between 10-30°C and 6-30 h, respectively in AF reactors. In anaerobic sequencing bed reactor (ASBR), it was obtained high COD removals (90%) at high HRTs as 105 h (Imura et al., 1990). In Anaerobic Fluidized Bed Reactor (AFBR), it was possible to obtain COD removals between 62% and 76% at short HRTs varied between 1 h and 2.8 h.

Two-stage anaerobic processes have been proposed to degrade and retain suspended solids from sewage at lower temperatures like those prevailing in moderate climates (van Haandel and Lettinga, 1994; Wang, 1994). In the first stage, the particulate organic matter was entrapped and partially hydrolyzed into soluble compounds, which are digested in the second stage. In two-stage reactors, excess sludge of the first reactor needs to be discharged because of the suspended solids removal efficiency of the first reactor was higher and than that organic matter and excess sludge needs to be discharged regularly (Wang, 1974). In the

effluent of the first reactor the organic matter is dominantly present as dissolved compounds and accumulation of biodegradable solids may occurs at low temperatures, when the hydrolysis rate becomes rate limiting. At low temperatures, the excess sludge of such a reactor required stabilization in a separate sludge digester (van Haandel and Lettinga, 1994; Wang, 1994; Mahmoud, 2002). The results given by Wang (1994) were 71% COD and 83% SS removal efficiencies at temperatures above 15°C in a system of hydrolysis upflow sludge bed (HUSB) and EGSB reactor combined with a sludge stabilization tank. In two stage reactors such as UASB+UASB and expanded granular sludge blanket (EGSB) + EGSB reactors, the COD removal efficiencies were considerably high. Sayed and Fergela (1995) studied the feasibility of two-stage anaerobic system for sewage treatment in UASB+UASB reactors resulting with 80% COD removal efficiency (at HRTs of 8-16 h +2 h). In two stage EGSB reactors, the COD reductions were 90% for influent COD values varying between 500 and 900 mg/L at low temperatures (3-8°C) (Lettinga et al., 1999). In EGSB reactors, decreases in temperatures were better tolerated compared to UASB reactors. In two-stage UASB reactors, the COD removals were 85% at an influent COD value of 630 mg/L at a HRT of 8 h (Fatma el Ghory, 1999).

Table 2.4. Low strength wastewater treatment studies in high rate reactors

Reactor Type	T (°C)	HRT (h)	Influent COD (mg/L)	COD removal (%)	Reference
AAFEB lab	20	2-24	119-320	57-84	Jewel ,1981
UASB-lab	8-18	4-8	420-920	45-75	Lettinga et.al. 1981
UASB-lab	8-20	-	140-1100	50-85	Lettinga et.al.1983
AF-lab	35	24	288	73	Kobayashi et.al. 1983
UASB-lab	8-20	8	400-500	30-75	Grin et.al., 1983
UASB-pilot	24-26	3-3,5	144-730	75-82	Schellinkout et.al.1985
AF-lab	10	6	467-700	53.7	Deryche and Verstraete,1986
UASB-lab	20-35	4	341-424	60-65	Vieira et.al., 1986
UASB-lab	7-8	9-14	467-700	57	Man et al., 1987
UASB-pilot	4-18	8.7-15	370-400	24-54	Man et al., 1987
UASB-pilot	23-27	5	426	66	Jakma et al.,1987
UASB-lab	12-20	7-8	190-1180	30-75	Man et al., 1988
EGSB-lab	12-20	2-3	70-250	24-53	
UASB-pilot	12-18	18	465	65	Monroy et al.,1988
UASB-lab	18-28	4	627	74	Barbosa,1989
UASB-pilot	25	6	-	50-75	Haskoning,1989
AF-UASB-pilot	-	12	400	49-78	Joshi et al.,1987
UASB-pilot	>20	6	370	62-70	Alaerts et al., 1993
Reactor Type	T (°C)	HRT (h)	Influent COD (mg/L)	COD removal (%)	Reference
UASB-pilot	14	42	205	47	Collivignarelli, 1990
AFBR-lab	5-20	2.8	475	76	Sanz et al.,1990
AF-lab	5-30	7.5-30	66	33-85	Matssushige et.al.,1990
UASB-full 1200 m3 -	20-30	6	563	74	Draijer et.al. 1992
EGSB-lab	>13	1.5- 3	250-350	42-51	Last and Lettinga,1992
AFB-pilot (silversand)	19	2	372	76	Last and Lettinga, 1992
AFB-lab (sand)	13-31	1-1.5	552-700	62-71	Marango-Campos (1992)
UASB-pilot	10-32	5-15	113-595	60	Vieira&Garcia, 1992
UASB-pilot	24-27.5	18	286-394	58-73	Kriyama et.al.1992
ASBR-lab	35-15	12-48	400-1000	80-90	Udeme and Dague(1992)
UASB-pilot	-	5-19	-	66-72	Schellinkhout& Collazes, 1992

Table 2.4. Low strength wastewater treatment studies in high rate reactors (Cont'd)

Reactor Type	T (°C)	HRT (h)	Influent COD (mg/L)	COD removal (%)	Reference
SBR-pilot	16.5	105	45-190	90	Imura et al., 1993
UASB-pilot	14±4	44-103	976±126	33-60	Bogte et.al. 1993
UASB-full 3360 m <sup>3</sup>	16.5	5.2	380	60	Schellinkhout 1993
UASB pilot	30	3	450	90	Gnanadipathy, 1993
ASBR-lab	25-35	24-4	400-1000	90	Udema et.al. 1994
USBR-lab	15-35	12-16	400-600	85	
UASB-full	16-23	7	402	74	Vieira et al. 1994
HUSB+EGSB-lab	17	3+2	697	71	Wang, 1994
AFBR-lab	10	1.5	480	70	Sanz et.al., 1990
UASB+	18-20	(8-	200-700	80	Sayed and
UASB-lab		16)+2			Fergala, 1995
UASB+EGSB-lab	20	9+9	536 (raw & brewery)	80	Tang et al., 1995
ABR-lab	18-28	4-10	386-405	67.8-83.5	Yu & Andersen., 1996
UASB-semi pilot scale	20-35	3	500	90-92	Singh et al., 1996
UASB+Aerobic SBR-lab	30	4	422	58	Sousa & Foresti 1996
EGSB-lab	30	0.5	579-772	56-82	Kato, 1997
UASB-full 477 m <sup>3</sup>	-	13	600	68	Chernicharo & Borges., 1997
AAEBR-lab (diatomaceous earth)	20	10	196	76	Collins et.al., 1998
UASB-lab	20	10	350-500	60-75	Singh-Virarhavan 1998
EGSB+ EGSB-lab	8	3.1-4	550-1100	90	Van Lier et.al., 1997
UASB+	-	8	630	79	El-Ghory and Nasr, 1999
UASB-lab					
UASB+	-	8	630	85	El-Ghory and Nasr, 1999
UASB-pilot					
EGSB+EGSB-lab	3-8	1.5+1.5	500-900	90	Lettinga et.al. (1999)
EGSB+EGSB-lab	10-15	3.5	500-800	67-78	Rebac 1999

Table 2.4. Low strength wastewater treatment studies in high rate reactors  
(Cont'd)

Reactor Type	T (°C)	HRT (h)	Influent COD (mg/L)	COD removal (%)	Reference
UASB-lab	25	$V_{up}:0.6$ -1.3 m/h	500	75	Jeisson and Champy, 1999
EGSB-lab	25	$V_{up}:1.8$ -10.8 m/H	500	80	Jeisson and Champy, 1999
ABR-lab	35-20-10	6-10	500	80-70-60	Langenhoff et al., 2000
Membrane. Coupled -lab*	15-25	4-6	100-2600	97	Cheng Wen et.al. 1999
AFBR-lab Carrier:sand		4.8-12	100-2000	85-92	Marin et.al.1999
UASB+Aerate d biofilter-pilot	Brazil	4-16	463	73	Gonçalves et.al.,1999
UASB-pilot	Brazil	7.5	712	79	Chernicharo et.al. 1999
UASB-lab		12	310	38-44	
ASBR-lab	9-15	10-46	500-640	56-84	Badik et al 2000
UAF-lab		10-20	490-690	46-87	
UASB-lab	29	6.2	320	65	Kalogo et al., 2001
UASB-full	30	9.7	563	67	Florencio et.el. 2001
UASB+Aerobi c SBR	21	6	569	71	Torres&Foresti ,2001
UASB+TF- pilot	Brazil	4 (UASB)	521	70 (UASB)	Chernicharo- Nascimento 2001
UASB-lab	21.6	6	152.6	55	Seghezze et.al. 2002
UASB+UASB Digester	15+35 (digester)	6 (UASB)	721	66	Mahmoud 2002
UASB	15	6	721	44	
AF-lab	25.4-15.5	-	325-403	72-80	Manariotis- Grigoropoulos, 2003
AFBR-full	26-28	3.2	96-854	71	Mendonça et.al.2004
UASB-pilot	27	1-6	92-816	57-60	Leitao...Lettinga 2005
UASB+UASB Hybrid-lab	28	24-3	200-1300	72-82	Lew et.al.2004
UASB-lab	14	24-	200-1300	48-70	Lew et.al.2004
UASB Hybrid-	10-14	24-3	200-1300	38-60	Lew et.al.2004

Table 2.4..Low strength wastewater treatment studies in high rate reactors

(Cont'd)

Reactor Type	T (°C)	HRT (h)	Influent COD (mg/L)	COD removal (%)	Reference
EGSB-membrane coupled	15	3.5-5.7	383-849	90	Chu et.al. 2004
UASB-pilot	23	5.6	131	63.2	Chu et al. 2004
UASB+UASB pilot	23	6.4+5.6	472	89	Segghezzo, 2004
AF-lab	25.4	12	253 (TSS)	80 (TSS)	Manariotis-Grigoropoulos, 2006

\*Membrane module consisted of bundle of hollow fibre polyethylene membranes as packing medium excellent COD effluents <20 mg/L

\*\*UASB Reactor installed after a conventional full-scale sedimentation tank (settler)

\*\*\*WSP treatment applied to the effluent of UASB

-: not indicated; ABR: Anaerobic baffled reactor; UASB: Upflow anaerobic sludge blanket; EGSB: Expanded granular sludge bed; AH: Anaerobic hybrid; ASBR: Anaerobic squenching bed reactor; AFBR: Anaerobic fluidized bed reactor; AAFEB: Anaerobic attached film expanded bed; HUSB: Hydrolysis upflow sludge bed; AF: Anaerobic filter; BF: Baffled reactor; RBC: Aerobic rotating biological reactor; WSP: Waste stabilization pond; TF: Trickling filter.

### **2.3.1. High Temperatures**

The first application of UASB reactor for sewage treatment in tropical countries was experienced with the construction of a pilot plant in 1983. Schellinkhout et al. (1985) studied the feasibility of UASB reactors by using a 64 m<sup>3</sup> pilot plant for raw sewage treatment in Cali, Colombia in tropical conditions. They reported that COD and BOD<sub>5</sub> removal efficiencies were 75-82% and 78-85% on the basis of raw influent and filtered effluent samples respectively with an HRT of 3-3.5 hours. The UASB reactors were reported as being a very feasible, attractive and economical option for treating highly septic and low strength domestic wastewater for tropical areas. The UASB Process was found to be economically more attractive than facultative ponds and oxidation ditches, especially when capital costs were considered (Alaerts et al., 1993).

Barbosa et al. (1989) reported the results of the operation of a 120 litre UASB reactor for raw domestic sewage treatment. The reactor operated without inoculation and reached satisfactory performances after 4 months of operation with an HRT of 4 hour at temperatures ranging 19 to 28°C. The removal efficiencies of 78%, 74% and 72% BOD<sub>5</sub>, COD and TSS respectively, were obtained. It was explained the high suspended solids content of the sewage used in this work and the reactor solids retention potential helped to the success of self-inoculation. The self-inoculation is very important since it been a costly operation and usually difficult to execute due to the lack of conventional treatment plants those have good quality anaerobic sludge availability.

Full-scale application of the UASB process has been successfully implemented in several tropical countries like India, Brazil, Mexico and Colombia (Draijer et al., 1992; Vieira et.al., 1994; Schellinkhout, 1993; Tare et al., 1997). A full-scale treatment plant with a wastewater treatment capacity of 5000 m<sup>3</sup>/day was

constructed at Kanpur, India in 1989. The UASB reactor is used in biological treatment for the municipal wastewater treatment of Kanpur. It was observed that the sewage temperature decrease to 20°C caused a decline in gas production while treatment efficiencies remaining constant and no change in the sludge quality. As soon as the wastewater temperature increased, the biogas production increased consequently. Draaijer et al (1992) concluded that the process stability was good and anaerobic treatment of sewage by using the UASB concept was feasible with COD, BOD and TSS reductions of respectively 74, 75 and 75% at a HRT of 6 hours. A post treatment was required to meet the Indian discharge standards.

Schellinkhout (1993) discussed the experience with the one of the largest anaerobic sewage treatment plant using UASB system Bucaramanga, Colombia. The design capacity was 160.000 inhabitants with 31.000 m<sup>3</sup>/day flow or 5000 kg BOD per day in Bucaramanga. The biological units of the system consisted of UASB reactors and facultative lagoons. Firstly the system was designed only with facultative lagoons. Changing some part of the system from anaerobic lagoons to UASB reactors showed a decrease of 90% on flat land requirement, thus UASB + facultative lagoon system was constructed. The effluent standards to be met required a total BOD removal efficiency of approximately 85%. The UASB reactors should have a BOD removal efficiency of 70% and the lagoon another 15%. The UASB technology has proven to be feasible in warm climates. It was accepted as an attractive solution for sewage treatment for its low power consumption, land requirement and good effluent quality in warm climates, especially when combined with a trickling filter or a lagoon for post treatment. In Brazil, Vieira et al (1986) used settled sewage to test the UASB reactor at ambient temperature in winter and 22°C in summer period while the average temperatures inside the reactor being 20°C in winter and 23°C in summer. The reactor used was 106 L in capacity and HRT was 4 hours. It was observed non-filtered BOD

removal efficiencies were 72% in summer and 69% in winter. Non-filtered COD removal efficiencies were also 65% during summer and 60% during winter. The results indicated that it was possible to scale up the system to a fairly large digester and a 120 m<sup>3</sup> reactor constructed by using this semi-pilot scale reactor. This system was operated for four years for demonstration and technology development purposes, following by two years modified operation period in Sao Paulo. HRTs ranging from 5-15 hours resulted in an effluent with 50 to 150 mg COD/L and 40 to 85 mg BOD/L as accepted good results. In the operation of the 120 m<sup>3</sup> UASB reactor inoculated with digested sewage sludge, at a temperature of 21-25°C and HRT of 4.7 h, non-filtered BOD efficiency was 61% and COD 50%.

Collivignarelli et al. (1990) constructed a pilot plant to treat municipal wastewater in a capacity of 500 to 2500 population equivalent (p.e.) in Italy. Different configurations of UASB, fluidized bed and aerobic fixed bed reactors were tested. The UASB reactor had a capacity of 1500 to 2000 p.e., and the fixed and fluidized bed reactors had capacities of about 500 p.e. The plant was designed so that the fluidized and fixed bed reactors would be utilized both aerobically and anaerobically depending on the treatment lines by inserting the reactors in different sequences at ambient temperatures (7 to 27°C).

The UASB reactor could be considered to be a reliable and easily managed process. Kalogo and Verstraete (2000) proposed a system composed of a primary sedimentation tank to remove suspended solids from raw sewage and a UASB reactor. The supernatant was treated in the UASB reactor and the primary sludge is digested separately in a conventional sludge digester. The effluent from the UASB reactor was disinfected with ozone and used in agriculture. In Tropical countries, the UASB reactor appears as the most robust of all the anaerobic

treatment processes, and “is by far the most widely used high-rate anaerobic system for sewage treatment” (van Haandel and Lettinga, 1994; Seghezzi, 2004).

### **2.3.2. Moderate to Low Temperatures**

The results of several studies on lab scale and pilot scale reactors operated at low temperatures have opened new perspectives (Table 2.4) but there is no full scale reactor put in operation (Zeeman and Lettinga, 1999; Lettinga et al., 2001; Zakkour et al., 2001). The application of UASB reactors to sewage treatment under low temperature conditions has been studied in The Netherlands since 1976 (Lettinga et al., 1981; Grin et al., 1983; de Man et al., 1986). Lettinga et al. (1981) studied the treatment of raw domestic sewage by using a 6 m<sup>3</sup> UASB pilot reactor at 20°C and a HRT of 8-48 h. It was obtained non-filtered COD removal efficiencies of 75% with a HRT of 8 hours.

The results obtained on 120 L lab scale UASB reactor were reported by Lettinga et al. (1983) and its applicability was investigated at ambient temperatures first with flocculant, later with granular sludge. The results by using digested sewage of a rather poor specific activity as seed in the UASB reactors was similar to the above mentioned studies. The results also demonstrated that, to achieve the similar effluent values at lower temperatures below 10°C, the HRT must be almost doubled. The reason was explained as the accumulation of suspended solids becoming significant at lower temperatures due to very slow hydrolysis of the entrapped solids. The highest overall treatment efficiencies were obtained with sewage having a high fractional content of suspended matter and for influent suspended matter with influent COD values exceeding 400-500 mg/l. In that case 65-85% COD reduction could be achieved in treating raw sewage in temperatures 8-20°C. It was concluded by Lettinga et al. (1983) that anaerobic treatment was effective at temperatures from 8°C to 10°C by providing a high quality-

preferentially a granular-seed sludge application. Regarding the results obtained anaerobic treatment presents an attractive option also for moderate climates.

De Man et al. (1986) investigated raw domestic wastewater (COD=500-700 mg/L) at 12-18°C with HRT's of 7-12 h and obtained total COD and BOD removal efficiencies of 40-50% and 50-70%, respectively. The reason of low removal efficiency was found as the sludge and wastewater contact being low because of the gas mixing being poor. In later investigations of de Man et al. (1988) the removal efficiency of soluble substrates was increased with the application of higher upflow velocities and providing a better sludge-wastewater contact in EGSB Reactors.

Last and Lettinga (1992) conducted experiments using presettled domestic sewage in 120 L EGSB reactor and 205 L FB reactor using the granular sludge seed from a full scale UASB reactor. In this study, higher superficial velocities were used to cause the granular sludge bed to expand/fluidize and resulted in a better sludge-wastewater contact and less accumulation of flocculent excess sludge between the granular sludge. In the UASB case, the superficial liquid velocity was kept as 1 m/h while as the reactor working as EGSB, the superficial velocity kept as 6 m/h. In FB systems a mobile heavy carrier material, silversand, was employed to immobilize the biomass in thin attached biolayers/ biofilms. In FB reactor, the initial superficial velocity was 24 m/h and gradually reduced to 12 and 10 m/h respectively. The maximum possible removal efficiencies obtained for soluble COD for UASB and EGSB systems were similar, but the maximum values for total COD removal efficiency was significantly higher in UASB systems (65 %) compared to 42 % in EGSB systems in batch treatment at 20°C. The reason for this relatively big difference was explained as the poor efficiency for EGSB systems for removing suspended solids (SS). Also it became possible to obtain

soluble COD removal efficiency of 84% at T=19°C and HRT= 2 hours in an EGSB system seeded with granular sludge and combined with SS removal step.

It was concluded that a one step FB system did not offer any prospect for sewage treatment and these systems could be useful for cultivating granular sludge providing the system operating in EGSB mode and at an HRT of 3.3- 5.8 hrs.

Kato et al. (1997) evaluated the feasibility of anaerobic treatment of low strength soluble industrial wastewater and the applicability of UASB or EGSB reactors for the treatment of such wastewater. It was observed that methanogens located in granular sludge have a high tolerance to oxygen and lowering temperature from 30°C to 15°C showed that anaerobic treatment may be feasible at low temperatures. It was obtained COD removal efficiencies as 95 % at organic loading rates up to 6.8 g COD/ L.d with influent COD concentrations ranging from 422 to 722 mg/l at 30°C in UASB reactors. In EGSB reactors, efficiencies were above 80% at OLRs up to 12 g COD/L.d with COD as low as 100 to 200 mg/l at 30°C.

Elmitwalli et al. (1999) investigated UASB and anaerobic hybrid (AH) reactors at a temperature of 13°C and at HRTs of 8 hours. Vertical polyurethane foam sheet media was used in AH reactors and sheets were oriented with two different spacings in two different reactors. The use of sheets (3 sheets) in the AH reactors increased suspended COD removal efficiencies 8% as compared to the UASB reactors and reached to 87% removal for presettled sewage treatment. It was also observed the treatment efficiency of pre-settled sewage was better than raw sewage in AH reactors. No sludge flotation was observed when the UASB reactor treated settled sewage. At steady state for presettled sewage treatment, the total COD removal for UASB and AH reactors were 60% and 64% respectively.

El-Gohary et al (1999) compared the performances of one stage and two stage UASB reactors by using domestic wastewater. The performance of one stage UASB at 8 hrs HRT was satisfactory and 77% and 83% removal efficiencies were obtained for total COD and BOD respectively. The COD removal efficiency increased 2% while BOD removal efficiency stayed constant and soluble BOD removal increased from 74% to 76% in two stage UASB reactor (reactors having same volumes) in 8 hour HRT. A slight improvement in the quality of the effluent was observed in two-stage UASB reactors.

Rebac et al. (1999) reported low strength synthetic and malting wastewater treatment by using a single and two module expanded granular sludge bed reactor system at 10-12°C and indicated COD removal efficiencies over 90% could be achieved at organic loading rates up to 12 kg COD/m<sup>3</sup> day and HRTs as low as 1.6 hour at 10-12°C using influent COD concentrations of 500-800 mg/l. The COD removal efficiencies exceeding 90% were also achieved at 8°C and 4°C at organic loading rates of 12 and 5 kg COD/m<sup>3</sup>.day and at HRTs of 2.0 and 4.0 hours while treating VFA mixture by a two module EGSB set-up as the influent and it required at least 4 months to get good quality effluent.

### **2.3.3. Reactor Types Used in the Treatment of Domestic Wastewaters**

Conventional anaerobic treatment processes are inadequate to treat low strength wastewaters and processes are required in which the biomass retention time can be controlled independently of the wastewater flow rate. The solution for the biomass retention problem resulted in the development of upflow anaerobic reactor configurations. Among these configurations, Upflow Anaerobic Sludge

Blanket (UASB) reactors have found the highest application (Pol and Lettinga, 1986).

### **2.3.2.1. UASB Reactor**

The success of the UASB concept relies on the establishment of dense sludge bed in the bottom of the reactor, in which all biological processes take place. The sludge bed is basically formed by accumulation of incoming suspended solids and bacterial growth. In upflow anaerobic reactors, under certain conditions, it was observed that bacteria naturally aggregate in flocs and granules (Hulshoff Pol, 1989). These aggregates have good settling properties and retention of active sludge, either granular or flocculent, enables good treatment performance at high loading rates. Higher organic loads can be applied in UASB systems than in aerobic systems (Kato, 1994).

In the design of the UASB reactors basically, the influent is distributed in the bottom of the reactor, passed through the bed of sludge by forming granules and leaves the reactor from its upper part (Figure 2.5). During passage, suspended solids are entrapped and degraded within the granules. The biogas produced provides a gentle mixing of the content of the reactor, which favours the contact substrate microorganisms. Good settleability, high biomass concentration (30000 to 80000 mg/L) and excellent solid liquid separation are possible with proper granulation in UASB reactors. High biomass concentration and granular structure of biomass ensure the performance of the UASB reactor not easily disturbed by sudden high loadings and toxic shocks compared to the suspended growth systems (Maat and Habets, 1987; Speece, 1996).

In order to achieve highest possible sludge hold up under operational conditions, it is necessary to equip the UASB reactor with a gas liquid separation (GLS) device. The main objective of this design is to facilitate the sludge return without help of any external energy and control device. The GLS should be designed to meet the requirements such as, provision of enough gas-water interface inside the gas dome, sufficient settling area outside the dome to control surface overflow rate; and sufficient aperture opening at bottom to avoid turbulence due to high inlet velocity of liquid in the settler, to allow proper return of solid back to the reactor.

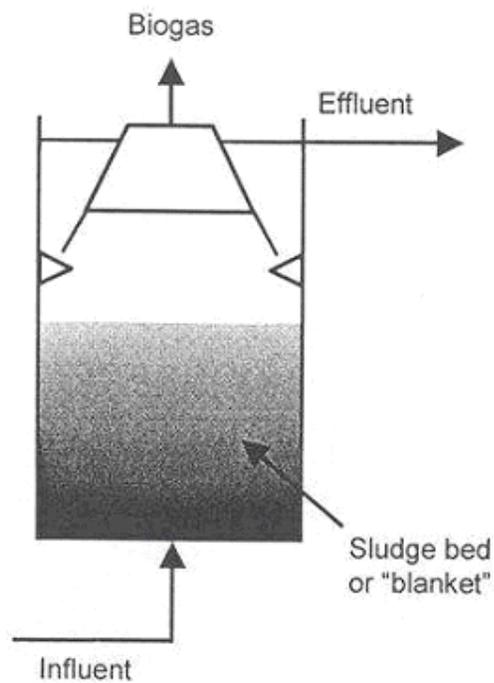


Figure 2.5. Schematic diagram of UASB reactor. Modified from van Haandel and Lettinga (1994).

The loading rates permissible in an anaerobic waste treatment process are mainly depends on the sludge retention in the reactor. Effluent recycle (to fluidize the sludge bed) is not necessary as sufficient contact between wastewater and sludge is guaranteed even at low organic loads with the influent distribution system (Lettinga, 1982).

#### **2.3.2.2. UAF Reactor**

The principle of the operation is that wastewater is passed upwards at low velocities through a column filled with packing material. The packing material acts as a surface for the attachment of microorganisms and as an entrapment mechanism for unattached flocs of organisms. This attached and entrapped anaerobic biomass will convert soluble and organic matter to the methane and carbon dioxide as the wastewater flow passes upwards through the column (Figure 2.6).

The essential features of the anaerobic filter design was listed by Switzenbaum (1983) as: a distributor in the bottom of the column, a media support structure, inert packing material, a free board above the packing material, effluent draw-off and optional features such as, recycle facilities, backwashing facilities or a sedimentation zone below the packing material. The distributor is designed for the even distribution of the incoming waste stream over the whole cross sectional area of the anaerobic filter to avoid short-circuiting. The packing material is generally an inert medium such as rocks or pall rings. The free board above the packing material is designed as a head space to allow the accumulation and capture of methane gas. Recycle of effluent back to the influent is not usually practiced in the operation of the anaerobic filter.

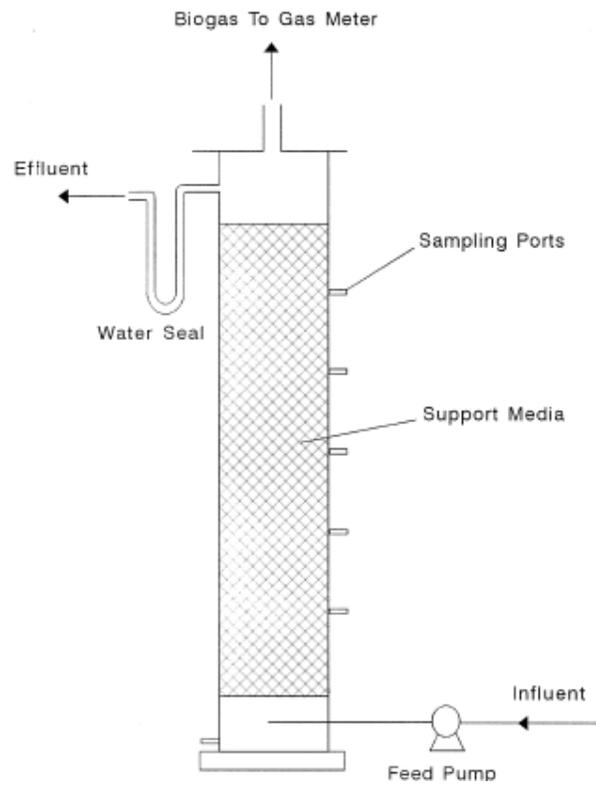


Figure 2.6. Schematic Diagram of UAF (Show and Tay, 1998).

## **CHAPTER 3**

### **MATERIALS AND METHODS**

Characterization of seed cultures and substrate used in the experiments and the methods used in this study are presented in the sections given below.

#### **3.1. Seed Culture**

Mixed anaerobic digester sludge and anaerobic granular sludge were used as the seed cultures in the experiments. The characteristics of seed cultures used in both sets of experiments are explained in the following sections.

##### **3.1.1. Seed Culture Used in Set 1 Experiments (Acidogenic Study)**

Mixed anaerobic digester sludge was used as seed in the UAF reactor and it was obtained from the anaerobic sludge digesters of Ankara Municipal Wastewater Treatment Plant. The mixed anaerobic digester sludge was concentrated by settling before being used as inoculum. Its characteristics are depicted in Table 3.1.

Table 3.1. Characterization of the seed inoculum used in the UAF Reactor

Parameter	Unit	Concentration
MLSS	$\text{g l}^{-1}$	$51.800 \pm 0.150$
MLVSS	$\text{g l}^{-1}$	$22.200 \pm 0.100$
Density	$\text{g l}^{-1}$	1018

Anaerobic granular sludge, which was used in the UASB reactor as seed, was obtained from the UASB reactors of the Wastewater Treatment Plant of Efes Pilsen Beer Factory located in Kazan-Ankara. The volume of granular sludge to be placed in the reactors was measured by waiting 18 hours for settling of the granules in order to determine the density. The granules were washed three times with tap water then they placed to the reactor.

The characteristics of granular sludge used in the UASB reactor in acidogenic study are given in Table 3.2.

Table 3.2. Characterization of granular sludge used in UASB reactor

(acidogenic study)

Parameter	Unit	Concentration
MLSS	$\text{g l}^{-1}$	$34.545 \pm 1.645$
MLVSS	$\text{g l}^{-1}$	$31.250 \pm 1.510$
Density	$\text{g l}^{-1}$	1017.6

### 3.1.2. Seed Culture Used in UASB reactors in Set 2 Experiments (two-phase system/ one-phase system)

The granular sludge used in UASB reactors for Set 2 experiments were also obtained from the UASB reactors of the Wastewater Treatment Plant of Efes Pilsen Beer Factory located in Kazan-Ankara. The characteristics of granular sludge used in the UASB reactors in Set 2 are given in Table 3.3.

Table 3.3. Characterization of granular sludge used in the UASB reactors  
(Set 2 experiments)

Parameter	Unit	Concentration
MLSS	g l <sup>-1</sup>	42.472 ± 2.317
MLVSS	g l <sup>-1</sup>	35.603 ± 0.080
Density	g l <sup>-1</sup>	1018

### 3.3. Substrate

The substrate was obtained from the output of the fine screening units (pore size of 3 mm) of the domestic wastewater collection point of dormitories and academic village (ODTÜ KENT) of Middle East Technical University. The characteristics of the wastewater are shown in Table 3.4. The substrate was kept in a refrigerator to minimize any variation in its characteristics. The domestic wastewater is continuously mixed in a refrigerator at 6° C and fed to the reactors by means of a peristaltic pump/ pumps (Figure 3.1).

Table 3.4. The characteristics of the domestic wastewater used in the experiments

SET 1	Parameter	Unit	Range	Average $\pm$ SD	n*
	pH		7.45- 7.85	7.71 $\pm$ 0.12	28
	Alkalinity	mg l <sup>-1</sup> as CaCO <sub>3</sub>	130- 237	189 $\pm$ 33	28
	sCOD	mg l <sup>-1</sup>	125- 237	260 $\pm$ 74	26
	tCOD	mg l <sup>-1</sup>	312- 786	536 $\pm$ 130	26
	BOD <sub>5</sub>	mg l <sup>-1</sup>	94- 410	229 $\pm$ 101	23
	TS	mg l <sup>-1</sup>	750-1086	953 $\pm$ 110	27
	TVS	mg l <sup>-1</sup>	183- 364	285 $\pm$ 54	27
	SS	mg l <sup>-1</sup>	126- 272	195 $\pm$ 82	27
	VSS	mg l <sup>-1</sup>	32- 156	139 $\pm$ 59	27
	PO <sub>4</sub> -P	mg l <sup>-1</sup>	5- 18	8 $\pm$ 4	18
	TP	mg l <sup>-1</sup>	10- 21	13 $\pm$ 11	08
	NH <sub>4</sub> -N	mg l <sup>-1</sup>	28- 42	41 $\pm$ 33	18
	TKN	mg l <sup>-1</sup>	31- 46	42 $\pm$ 2	08
	Protein-COD**	mg l <sup>-1</sup>	185- 219	204 $\pm$ 15	11
	VFA- COD	mg l <sup>-1</sup>	0-53	25 $\pm$ 21	17
SET 2	Parameter	Unit	Range	Average $\pm$ SD	n*
	pH		7.43- 7.68	7.56 $\pm$ 0.09	10
	Alkalinity	mg l <sup>-1</sup> as CaCO <sub>3</sub>	106- 161	125 $\pm$ 13	10
	sCOD	mg l <sup>-1</sup>	78- 189	146 $\pm$ 43	08
	tCOD	mg l <sup>-1</sup>	340- 633	368 $\pm$ 117	10
	BOD <sub>5</sub>	mg l <sup>-1</sup>	65- 150	128 $\pm$ 62	0 6
	TS	mg l <sup>-1</sup>	800- 1075	907 $\pm$ 140	10
	TVS	mg l <sup>-1</sup>	135- 276	214 $\pm$ 54	10
	SS	mg l <sup>-1</sup>	144- 310	262 $\pm$ 138	10
	VSS	mg l <sup>-1</sup>	20- 136	86 $\pm$ 71	10
	PO <sub>4</sub> -P	mg l <sup>-1</sup>	7- 19	13 $\pm$ 4	10
	TP	mg l <sup>-1</sup>	19- 36	24 $\pm$ 10	08
	NH <sub>4</sub> -N	mg l <sup>-1</sup>	28- 46	33 $\pm$ 8	10
	TKN	mg l <sup>-1</sup>	30- 46	41 $\pm$ 3	08
	Protein-COD**	mg l <sup>-1</sup>	33- 147	79 $\pm$ 45	16
	VFA- COD	mg l <sup>-1</sup>	10-96	60 $\pm$ 15	10

\* number of measurements; \*\*1 g protein assumed as (C<sub>4</sub>H<sub>6.1</sub>O<sub>1.2</sub>N)<sub>x</sub> is equivalent to 1.5 g COD (Sanders, 2001; Mahmoud, 2002).

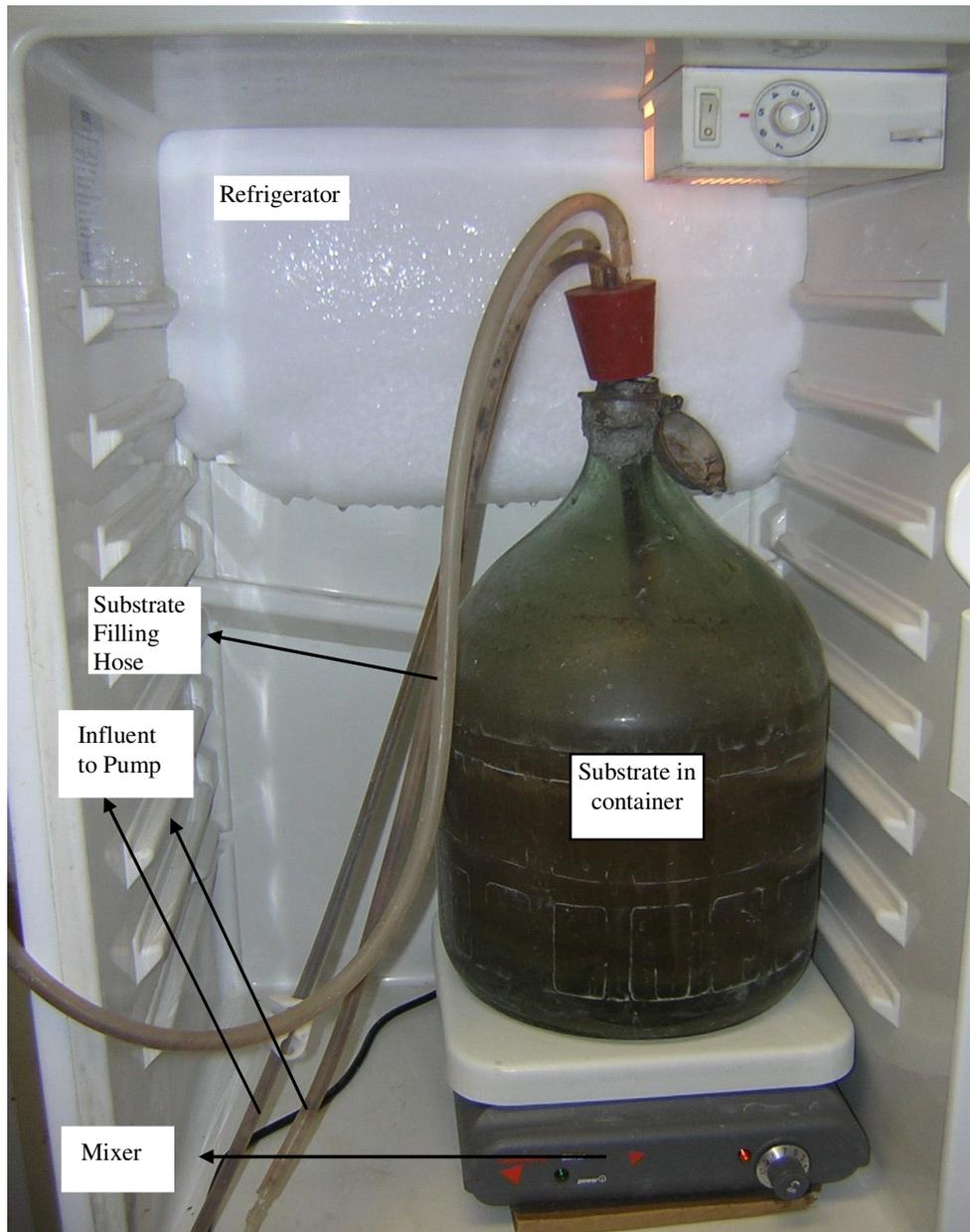


Figure 3.1. The substrate mixed in the refrigerator

### 3.4. Experimental Set-up

Two different experimental set-ups were envisaged. A schematic representation of laboratory-scale anaerobic digestion systems used in the experiments namely Set 1 and Set 2 are depicted in Figure 3.2.

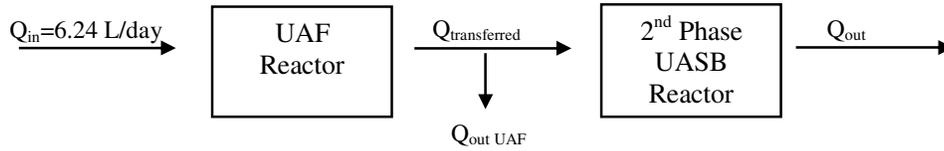
The aim of the first continuous reactor experiments (Set 1) was to evaluate the possibility of acidification of domestic wastewater. In the first part (Set 1) of the study, the optimum reactor and hydraulic retention time leading to maximum acidification was investigated. For this purpose, two plug flow reactors namely UAF and UASB were operated to compare the acidification degrees of the reactors and to optimize the acidification conditions. The operational period for acidification experiments (Set 1) can be described in three periods; Period 1, Period 2 and Period 3 which the hydraulic retention times are equal to 24 hours, 8 hours, and 2 hours, respectively. The UAF reactor was selected as the acidification reactor for the second continuous reactor experiments at a HRT of 2 h (Set 2). The experiments performed in this study and their objectives are summarized in Table 3.5.

The aim of the second continuous reactor experiments was to compare the COD, BOD<sub>5</sub>, solid, nutrient treatment efficiencies and gas productions in two-phase and one-phase reactor systems in domestic wastewater treatment. In Set 2 experiments, the effect of acidification was investigated in domestic wastewater treatment by comparing the treatment performances of two-phase UAF and UASB reactor system with one-phase UASB reactor system at different HRTs and temperatures. The effect of HRT/OLR on the COD, BOD<sub>5</sub>, pH, VFA and alkalinity, gas production, nutrient and solids removals were monitored.

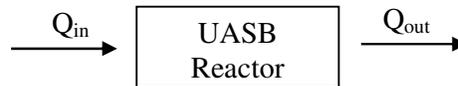
Furthermore, the effects of mesophilic and psychrophilic temperatures on the reactors efficiencies were investigated.



Set 1- Acidogenic UAF and UASB Reactors



Set 2-a) Two Phase UAF and UASB System



Set 2-b) One Phase UASB System

Figure 3.2. Experimental set-up used in the study (experiments are performed in Set 1-Periods 1-3 and Set 2-Periods 1-4 at 35°C and in Set 2 Period 5 at 20°).

Table 3.5. Set-specific experimental targets

Laboratory set-up	Objective of experiments
Set 1	Investigation the acidification of domestic wastewater, selection the reactor for optimum removal efficiencies and HRT for acidification of domestic wastewater
Set 2	Comparison of treatment efficiencies and effluent characteristics of domestic wastewater in two-phase and one-phase systems.

### 3.4.1. Acidogenic Reactor Experiments

Acidogenic reactor experiments were performed in plug-flow reactors namely, Upflow Anaerobic Filter (UAF) reactor and Upflow Anaerobic Sludge Blanket (UASB) reactors. The UAF and UASB Reactors operated in the temperature-controlled room are illustrated in Figure 3.3. The experiments were performed in two cylindrical glass columns with an inner diameter of 4.4 cm and a height of 67 cm. The active volume (liquid volume) of each glass reactor was 0.79 L. The domestic wastewater was completely mixed with a Corning Heavy Duty Magnetic Stirrer (230 VAC- 92 watts- 50/60 Hz) in a refrigerator at 6 °C while being pumped to the reactors using a peristaltic pump (Master Flex L/S Standard Drive Pump, P-07521, Cole-Parmer, USA). The reactors were set-up in a constant temperature room at 35±2°C. The wastewater was pumped to the reactors from the refrigerator which was placed outside of the hot room.

The total volume of the granular sludge used in UASB reactor was 0.52 L (See Table 3.2 in subsection 3.2.1). In order to prevent the escape of the granules to the

inlet pipe a few glass beads were placed to the bottom part of the column. A spiral cable was inserted inside the granular sludge of the UASB to prevent the collection of gas pockets inside the granules. The details of gas solids separator and effluent structures of UASB reactor are shown in Figure 3.4.

In the UAF Reactor, filter material and glass beads were placed at the bottom part of the reactor. Bioballs were located as the filter material (support media) in UAF reactor (Figure 3.5). Bioballs had an approximate diameter of 2 cm and a volume of 3.3 mL/each. The bioballs were splitted into two pieces where it was required to increase the surface area and the amount of the support media in the UAF reactor. At the bottom part of the reactor, 0.5 cm diameter glass beads were placed to the inlet of the reactor to provide the equal distribution of the influent flow. The upper part of the plastic filter material was covered by fine cloth screen and 14 glass beads with a diameter of 1 cm to prevent the flotation of the filter material.

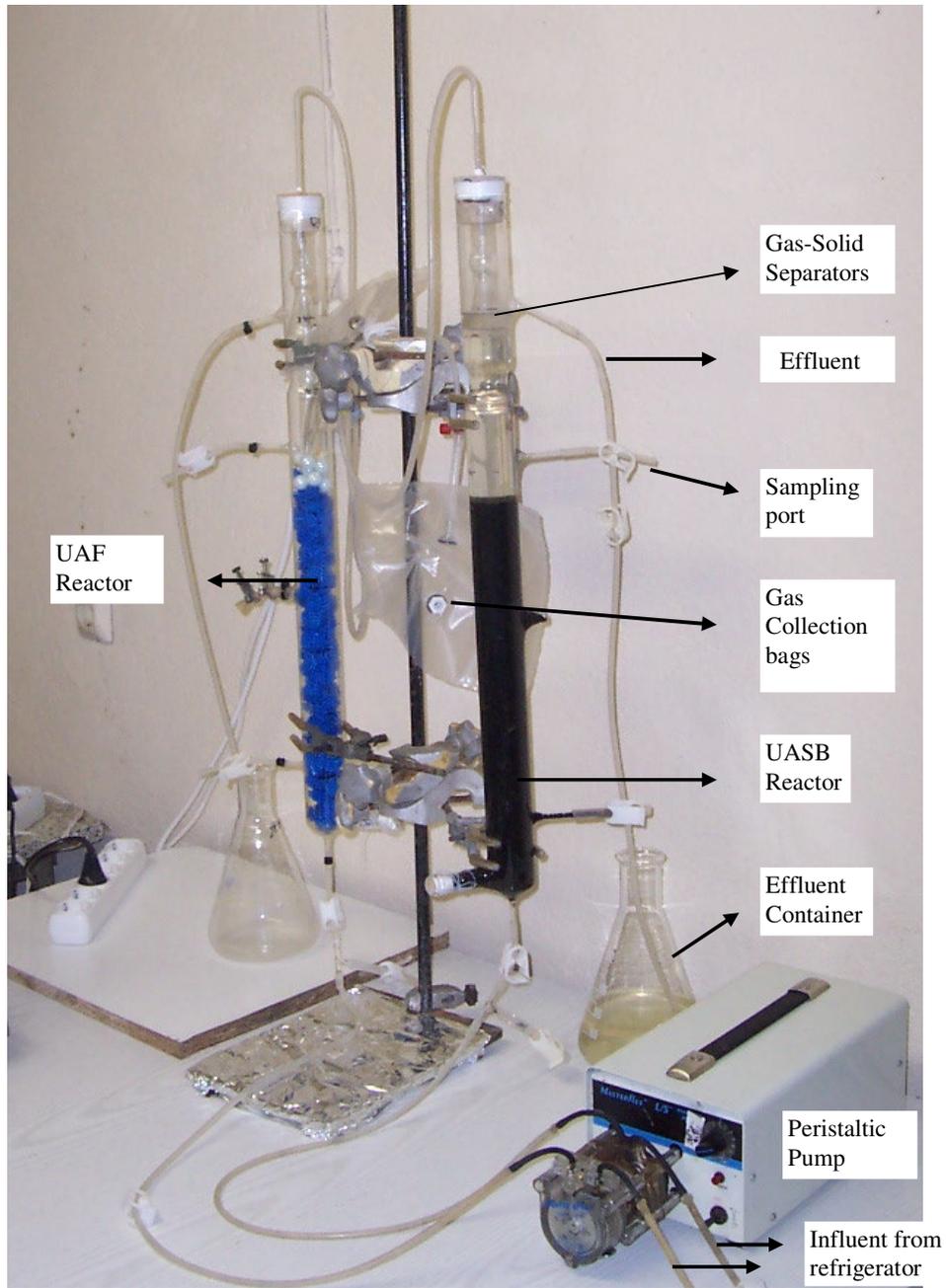


Figure 3.3. UAF and UASB Reactor set-up for acidification study in temperature- controlled room during start-up period.

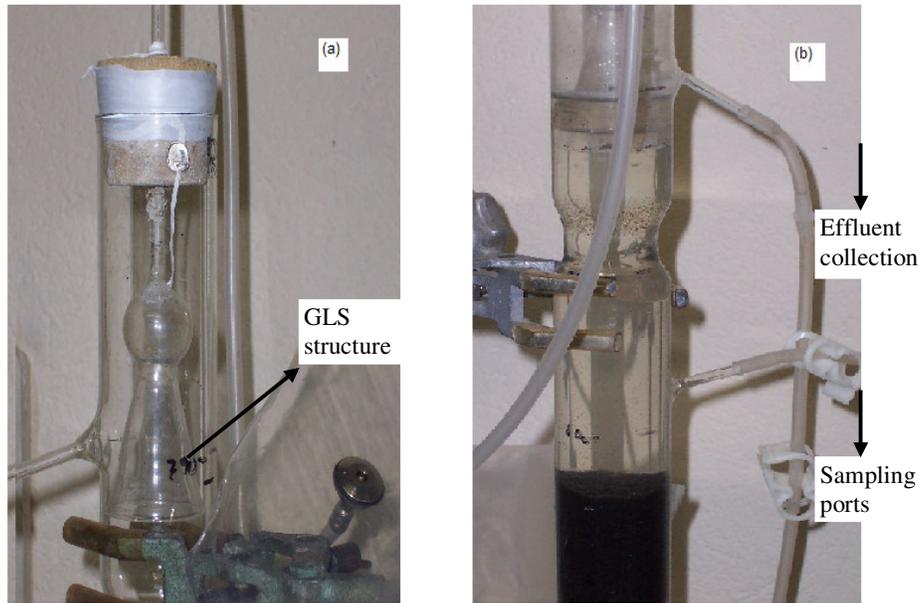


Figure 3.4. Details of UASB reactor, (a) Gas Liquid Separator (GLS) and (b) GLS structure, effluent collection and sampling ports.



Figure 3.5. Picture of bioballs as filter material in UAF reactor (half of total bioballs splitted into two).

The effective volume of the UAF reactor was 520 mL while the volume of the digester sludge used in the UAF reactor was 390 mL and the total volume of bioballs was 130 mL at the start up of the acidogenic study. The characteristics of the sludge used in the UAF reactor have given in Table 3.1 in Subsection 3.2.1. The sludge was added from the top of the reactor after the placement of the filter material.

The initial HRT of the UAF and UASB reactors was 24 hour. This HRT value was selected, since it was known that the COD treatment efficiencies ranged between 33% and 85% for HRTs between 7.5 h and 30 h (Matsushige, 1990) in domestic wastewater treatment. Furthermore, COD treatment efficiency was observed as 73% at an HRT of 24 h for UAF reactor (Kobayashi, 1983). The treatment efficiencies for UASB reactor were between 66% and 72% at HRT's between 5 h and 19 h (Schellinkout and Collazes, 1992) and between 45% and 68% at an HRT of 10 h (Forster and Wase, 1983). Therefore, both reactors were operated at 24 h HRT in the start up period of acidogenic phase.

During the start-up period (4 months), feed rate, pH and ORP of the reactors were observed daily. The pH and ORP analyses were performed with the grab sample taken from the upper second port of the reactor to monitor the variations. Alkalinity, Volatile Fatty Acids (VFA), COD, solids (TS, TVS, TSS and VSS) were analyzed one day in a week.

The UAF and UASB reactors were operated concurrently in three periods in acidogenic study (Table 3.6). In Period 1, the HRT was 24 h and the start-up operating parameters and loading strategy were the same as applied in start-up period. In Period 2, the HRT of the reactors were set-up to 8 hour. In Period 3, the HRT of the reactors were adjusted to 2 hours. The operational conditions for

acidogenic UAF and UASB reactor are seen in Table 3.6. In Period 1, Period 2 and Period 3, the performances of reactors were monitored by the determination of BOD<sub>5</sub>, TKN, NH<sub>4</sub>-N, TP, PO<sub>4</sub>-P and protein concentration once in two weeks. Gas samples were collected with gas collection bags located at the outlet of the GLS structure in Period 1 and Period 2. It was noticed that a certain amount of gas from the effluent collection structure was lost. Therefore effluents of the reactors were collected via calibrated gas holders (See Figure 3.6). Gas samples was collected by the displacement of water in calibrated gas holders. Initially, it was filled with water up to the top and the volume of biogas was measured daily by taking the water level readings in the cylinder. The gas holders were connected to effluent collection chambers so that the gas volumes could be measured at atmospheric pressure.

Table 3.6. Operational conditions for acidogenic study

	UAF and UASB Reactors			UAF Reactor	UASB Reactor
	T (°C)	HRT (h)	Flow rate (L/d)	OLR*	F/M**
Start-up	35±2	24	0.52	0.21	0.02
Period 1	35±2	24	0.52	0.21	0.02
Period 2	35±2	8	1.56	0.56	0.05
Period 3	35±2	2	6.24	1.52	0.38

\*OLR= g COD/ L.day: \*\* F/M= kg BOI<sub>5</sub>/kg MLVSS.day

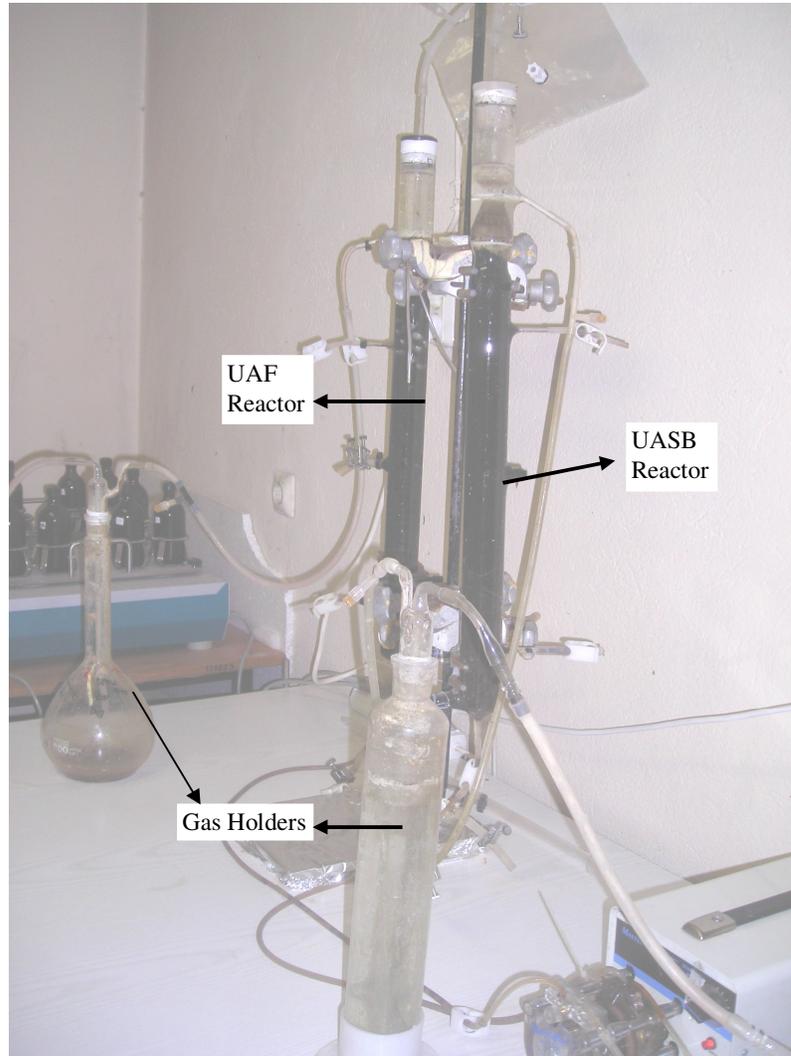


Figure 3.6. Gas collection system in acidogenic study- Period 3 (HRT=2).

The list of the samples taken from the reactors during acidification experiments is shown in Table 3.7.

Table 3.7. Sampling outline during operation period of acidification reactors  
(Set1)

	Influent	UAF Reactor	UASB Reactor
Flowrate	Daily	Daily	Daily
Gas Production Rate		Daily	Daily
pH	Daily	Daily	Daily
ORP	Daily	Daily	Daily
tCOD	Once a week	Once a week	Once a week
sCOD	Once a week	Once a week	Once a week
BOD <sub>5</sub>	Once in two weeks	Once in two weeks	Once in two weeks
NH <sub>4</sub> -N	Once a week	Once a week	Once a week
PO <sub>4</sub> -Phosphate	Once a week	Once a week	Once a week
TKN	Once in two weeks	Once in two weeks	Once in two weeks
TP	Once in two weeks	Once in two weeks	Once in two weeks
Alkalinity	Once a week	Once a week	Once a week
VFA	Three times a week	Three times a week	Three times a week
Total Solids	Once a week	Once a week	Once a week
Total Volatile Solids	Once a week	Once a week	Once a week
Volatile Suspended Solids	Once a week	Once a week	Once a week
Suspended Solids	Once a week	Once a week	Once a week
Gas Composition		Twice a week in Period 3	Twice a week in Period 3
Protein	(1-4) times in each Period	(1-4) times in each Period	(1-4) times in each Period

### 3.4.2. Two-phase UAF and UASB system and one-phase UASB system

The UAF reactor was selected as the acidification reactor and operated continuously at 2 hour HRT without stopping after the completion of Set 1 (See Figure 3.7). The UAF reactor is followed by the UASB reactor as methanogenic reactor in two-phase UAF and UASB system.

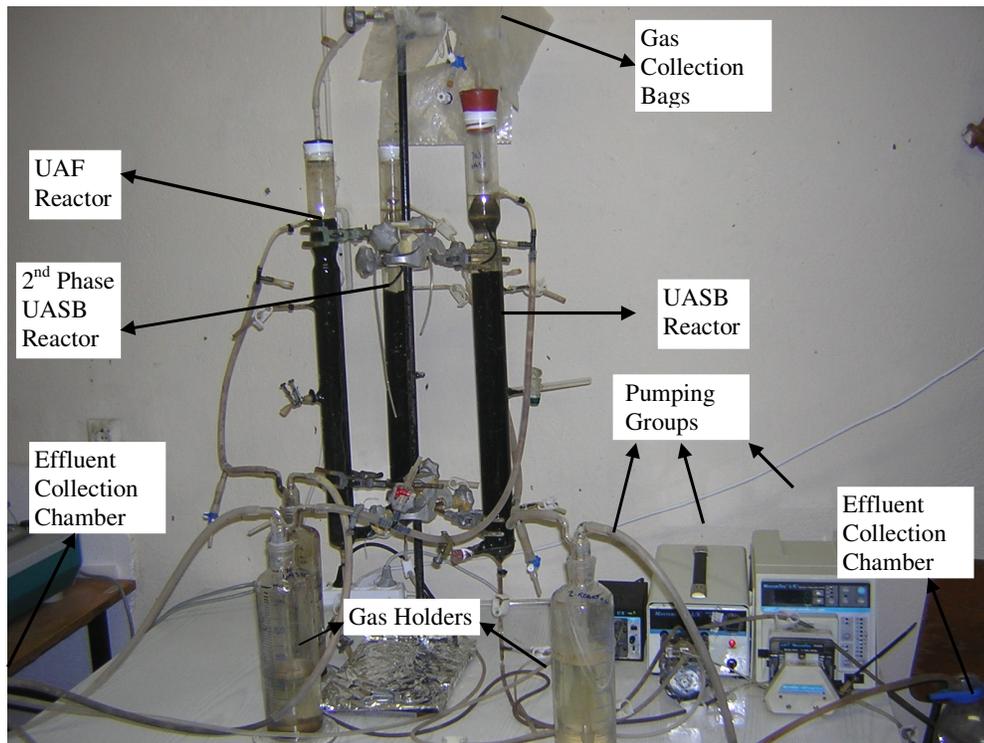


Figure 3.7. General view of two-phase UAF and UASB system and one-phase UASB system.

### **3.4.3. Operational Conditions for Two-phase UAF and UASB System and One-Phase UASB System**

The operational conditions for two-phase UAF and UASB system and one-phase UASB system used in this study are given in Table 3.8. The volume of granular sludge of UASB reactor of two-phase reactor system was 0.416 L and the volume of granular sludge of one-phase UASB reactor was 0.520 L. In periods 2, 3 and 4, the HRTs of the reactors were adjusted by changing the flow rate of the pumps. It was inserted one or two more pumps (masterflex pumps) for flow adjustments when needed. The experiments were performed as the same way mentioned in Section 3.3.1.

The outline of the samples taken from the reactors during experimentation is shown in Table 3.9. At the end of the operational period, in addition to the tests presented in that table the specific methanogenic activity (SMA) of granular sludges of UASB reactors were tested to assess activity of granular sludge during methanification phase.

### **3.4.4. Start-up and Operation**

#### **3.4.4.1. Start-up and Operation of Two-Phase UAF and UASB System**

The aim of the first continuous reactor system (Set 1) was to evaluate the possibility of acidification of domestic wastewater. For this purpose, two plug flow reactors namely UAF and UASB were operated to compare the acidification degree of the reactors and to optimize the acidification conditions.

The mean HRT during start up was kept at  $24\pm 1.6$  h. Start-up was completed, when the pH values ranged less than  $\pm 5\%$  and VFA production had become steady with a change less than 20%. After the start-up, the HRT was lowered stepwise from the initial 24 h to 8 h and finally to 2 h. The operational parameters are given in Table 3.6 in Subsection 3. 4. 1. The start-up operational parameters are given in Table 3.6.

Table 3.8. Operational conditions in two-phase UAF and UASB system and one-phase UASB system

Period	T (°C)	One-phase UASB System				Two-phase UAF and UASB System							
		UASB Reactor				UAF Reactor				UASB Reactor (2nd Reactor)			
		HRT (h)	Flow rate (L/d)	OLR *	F/M**	HRT (h)	Flow rate (L/d)	OLR	F/M	HRT (h)	Flow rate (L/d)	OLR	F/M
1	35±2	10	1.25	0.54	0.014	2	6	2.57	0.128	8	1.25	0.28	0.006
2	35±2	8	1.56	0.67	0.018	2	6	2.57	0.128	6	1.66	0.51	0.011
3	35±2	6	2.08	0.76	0.024	2	6	2.18	0.128	4	2.50	0.35	0.005
4	35±2	4	3.12	0.78	0,036	2	6	1.5	0.094	2	4.99	0.70	0.010
5	20±3	4	3.12	0.78	0.036	2	6	1.5	0.094	2	4.99	0.70	0.010

\*OLR= g COD/ L.day; \*\* F/M= kg BOI<sub>5</sub>/kg MLVSS.day

Table 3.9. Sampling outline during operation period of the reactor

Parameter	Influent	Two-phase UAF and UASB system	One-phase UASB system
Flowrate	Daily	Daily	Daily
Gas Production Rate		Daily	Daily
pH	Daily	Daily	Daily
ORP	Daily	Daily	Daily
tCOD	Three times a week	Three times a week	Three times a week
sCOD	Three times a week	Three times a week	Three times a week
BOD <sub>5</sub>	Once a week	Once a week	Once a week
NH <sub>4</sub> -N	Once a week	Once a week	Once a week
PO <sub>4</sub> -Phosphate	Once a week	Once a week	Once a week
TKN	Once a week	Once a week	Once a week
TP	Once a week	Once a week	Once a week
Alkalinity	Once a week	Once a week	Once a week
VFA	Three times a week	Three times a week	Three times a week
Total Solids	Once a week	Once a week	Once a week
Total Volatile Solids	Once a week	Once a week	Once a week
Volatile Suspended Solids	Once a week	Once a week	Once a week
Suspended Solids	Once a week	Once a week	Once a week
Gas Composition		Twice a week	Twice a week
Protein	Once a week	Once a week	Once a week

In the case of acidification of domestic wastewater, HRT is the most easily controllable operational parameter. HRT is a key parameter for the performance of the hydrolysis in acidification reactor, since it determines the solid solubilization efficiencies and the degree of acidification of the influent. It was planned to encourage the growth of acid formers and suppress the growth of methane producers by reducing the HRT. Therefore, the HRT of the UAF and UASB reactors are adjusted to 24 h, 8 h and 2 h during the acidification study period (Set 1) to make a comparison to select the acidification reactor to be used in Set 2 as the acidification reactor at optimum HRT prior to the UASB reactor.

The influences of HRT on the performance of acidogenic phase of anaerobic digestion were investigated by the degree of acidification, effluent composition and performances of UAF and UASB Reactors. During the operation period of 240 days, the performances and effluent composition of the reactors were monitored by measuring pH, ORP, VFA, alkalinity, sCOD, tCOD, BOD<sub>5</sub>, TVS, TS, NH<sub>4</sub>, TKN, PO<sub>4</sub> and TP.

#### **3.4.4.2. Start-up and Operation of Two-Phase UAF and UASB Reactor System and One- Phase UASB Reactor System**

In the start-up of two-phase systems, UAF Reactor was followed by UASB Reactor and the UAF reactor continuously studied without stopping after the acidification study. The UASB reactors of two-phase system and one-phase system were started at the beginning of Set 2 Experiments. The operational conditions are given in Table 3.8 in Subsection 3.4.3. The operational conditions at start-up period were the same with Period 1 (Table 3.8). The start-up period

lasted for about 40 days. The parameters monitored in start-up and during the operation of two-phase UAF and UASB reactor system and one-phase UASB reactor system are given in Table 3.9.

### **3.5. Analytical Methods**

The influent samples were obtained from the inlet port of UAF and UASB reactors. The 24-h composite effluent samples were used for all analysis with the exception of pH and ORP. The pH and ORP measurements were performed immediately with grab samples taken from the second port below the effluent port to monitor the reactor conditions.

#### **3.5.1. pH and ORP measurement**

pH in samples was measured by a bench-top pH meter (Jenway Ltd., Essex, UK) and a general purpose pH electrode (Cole Parmer, Niles, IL, USA). ORP was also measured by a bench-top pH/ORP meter with an ORP electrode.

#### **3.5.2. Bicarbonate alkalinity measurement**

Bicarbonate alkalinity (as mg/L CaCO<sub>3</sub>) was measured according to titration procedure given by Anderson and Yang (1992).

### **3.5.3. Suspended Solids (SS), volatile suspended solids (VSS), mixed liquor volatile suspended solids (MLVSS), total solids (TS) and total volatile solids(TVS) measurement**

These parameters were measured according to Standard Methods (APHA, 1998).

### **3.5.4. Total Volatile Fatty Acids (VFA) measurement**

VFA's determinations were conducted by GC analysis. Liquid samples were prepared by filtering with a 0.22  $\mu\text{m}$  filter paper (Whatman Co.) and acidifying with 99% formic acid to a pH less than 3 to convert the fatty acids to their undissociated forms (i.e., acetic acid, propionic acid, butyric acid, etc.). A gas chromatograph (GC) (Thermo Electron Co.) equipped with a flame ionization detector and a 30 m column was used for VFA analyses. The column temperature was started at 100°C with 2 min holding time and then increased to 250°C with 8°C/min ramping, and the injector/detector temperature was kept at 200/350°C with nitrogen as the carrier gas and a flow rate of 30 mL/min. The gas flow rates were gauged at 350 mL/min for air and 35 mL/min for hydrogen. 1  $\mu\text{L}$  of the acidified sample was injected into the GC.

### **3.5.5. Gas Analysis**

Headspace gases of reactors were analyzed by gas chromatography (GC) for methane, nitrogen and carbon dioxide in a Trace GC Ultra (Thermo Electron Corporation) equipped with FID (Flame Ionization Detector) and TCD (Thermal Conductivity Detector). Gas samples for gas composition analysis were taken by a

100  $\mu$ L Hamilton gas-tight glass syringe from gas sampling port. The gas composition was determined by a (GC) unit (Thermo Electron Co.) equipped with thermal conductivity detector. Methane, nitrogen and carbon dioxide were separated through a 15 m Porapak Q, 5 mm I.D.column. Column was operated with helium as the carrier gas at a constant pressure of 20 kPa at 40°C. The injector was maintained at 100°C, and the detector temperature was set to 100°C. The calibration was carried out by using an individual standard gas for each of the gas measured.

### **3.5.6. Chemical Oxygen Demand (COD) Measurement**

COD concentrations were determined according to an EPA approved reactor digestion method (for a COD range of 0- 1500 mg/L) as given in Hach Water Analysis Hand Book (1989). A Hach COD reactor and a Hach DR/2000 spectrophotometer operated at a wavelength of 620 nm were used. For soluble COD (sCOD) analysis, the samples were firstly filtered through a 45 $\mu$ m sized glass fiber filter.

In Set 2 Experiments (two- phase UAF and UASB reactor system and one-phase UASB reactor system) experiments most of the COD measurements were performed using 5220 B. Open Reflux Method (APHA 1998).

### **3.5.7. Five-Days Biochemical Oxygen Demand (BOD<sub>5</sub>) Measurement**

BOD<sub>5</sub> is analyzed in parallel and as replicates according to procedure described in the Standard Methods (APHA; 1998).

### **3.5.8. Total Kjeldahl Nitrogen (TKN) measurement**

TKN values of samples were measured by following the Standard Methods (4500-Norg B. Macro Kjeldahl Nitrogen) (APHA, 1998).

### **3.5.9. Ammonium nitrogen (NH<sub>4</sub>-N) measurement**

NH<sub>4</sub>-N was measured with an Aqualytic Photometer and with Aqualytic HR Ammonium Nitrogen Reagent Sets. The analyses were performed according to Aqualytic PC Multi Direct Instruction Manual (Method numbers: 60 for ammonium nitrogen).

### **3.5.10. Total Phosphorous (TP) measurement**

TP were measured by following the standard methods (4500-P E. Ascorbic Acid Reduction Method) (APHA, 1998).

### **3.5.11. Ortho-phosphate (PO<sub>4</sub>-P) measurement**

PO<sub>4</sub>-P was measured with an Aqualytic Photometer and Aqualytic Ortho-Phosphate Reagent Sets, respectively. The analyses were performed according to Aqualytic PC Multi Direct Instruction Manual (Method numbers: 323 for ortho-phosphate).

### **3.5.12. Protein measurement**

Folin-ciocalteu protein measurement method is used (Lowry et al. 1951). In this method the reagents given below were used:

Reagent A : 2 % W/V sodium carbonate in 0.1 N NaOH

Reagent B : 1 % W/V sodium potassium tartarate in 0.5 % W/V cupric sulphate

Reagent C : 1 mL of Reagent B + 49 mL of Reagent A

Reagent D : Folin-Ciocalteu's phenol reagent (Diluted by the ratio of 10:9 with distilled water).

In the protein measurement procedure, 3 mL of reagent C was added to the solution having a volume of 0.6 mL containing 0-200 µg protein and mixed and stood at room temperature for 10 minutes. 0.3 mL of Reagent D was added and mixed well immediately. Then it is allowed to stand for 30 minutes. The intensity of the blue color at 750 nm against a reagent blank was read. The protein calibration curves are given in Appendix A.

### 3.5.13. Specific Methanogenic Activity (SMA)

Specific methanogenic activity (SMA) test was used to determine the methane production potential rates of anaerobic sludges. SMA was defined as the substrate dependent methane production rate per unit mass of volatile solids biomass. The presence of viable methanogens of initial and final sludge samples of UASB reactors were determined with SMA experiments.

The acidogenic UASB reactor was stopped after a period of 237 day study and emptied. SMA experiments were conducted in order to determine the methanogenic activities of discharged granular sludges of acidogenic UASB Reactor and granular sludges which is planned to be used in methanogenic UASB reactor.

After the day 309, the SMA's of the UASB Reactors of one-phase and two-phase systems were determined following the switch off the reactors. Reactors having a volume of 110 mL were seeded with the sludge (the MLVSS concentration of the mixture sample was 2000 mg/L) to be tested and basal medium was added (50 mL effective volume). Glucose was fed to the reactors to achieve COD concentration of 600 mg/L. pH of reactors was adjusted to 7 with the addition of HCl and/or NaOH. The basal medium was added to provide the necessary volume of micro and micro nutrients and heavy metals. This basal medium was consisted of the following inorganic compounds (in mg/L):  $\text{NH}_4\text{Cl}$ , (1200);  $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$  (400);  $\text{KCl}$  (400);  $\text{Na}_2\text{S} \cdot 9\text{H}_2\text{O}$  (300);  $(\text{NH}_4)_2\text{HPO}_4$  (80);  $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$  (50);  $\text{FeCl}_3 \cdot 4\text{H}_2\text{O}$  (40);  $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$  (10);  $\text{KI}$  (10);  $(\text{NaPO}_3)_6$  (10); L-cysteine (10);  $\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$  (0.5);  $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$  (0.5);  $\text{CuCl}_2$  (0.5);  $\text{ZnCl}_2$  (0.5);  $\text{NH}_4\text{VO}_3$  (0.5);  $\text{NaWO}_4 \cdot 2\text{H}_2\text{O}$  (0.5);  $\text{H}_3\text{BO}_3$  (0.5);  $\text{NiCl}_2 \cdot 6\text{H}_2\text{O}$  (0.5);  $\text{NaMoO}_4 \cdot 2\text{H}_2\text{O}$  (0.5);  $\text{Na}_2\text{SeO}_3$  (0.5) (Speece, 1996). About 0.01 g sodium thioglucollate per one liter was added to

reduce the redox potential and sustain the anaerobic conditions in the reactors having a volume of 110 mL. After flushing the reactors with a gas mixture of N<sub>2</sub>/CO<sub>2</sub> (70/30) for 3-4 minutes, the reactors were sealed with rubber septa. Reactors were incubated in the temperature-controlled room at 35±2 °C. Cumulative gas production and daily gas production were measured for 4 days by a liquid displacement method. Furthermore the biogas methane (CH<sub>4</sub>) content was determined using GC. The end of assay is defined with the cessation of the gas production. SMA was calculated by considering the average methane produced (gram COD equivalent) per gram of volatile suspended solids during the assay (Punal et al., 1999).

#### **3.5.14. Statistical Analysis**

The Statistical analyses were performed by Analysis of Variance (ANOVA), Statistical Package for Social Sciences 11.5 (SPSS 11.5) and a sub-program Microsoft Office Software Excel 2003.

ANOVA analysis of variance between experimental data was performed to detect *F* and *p* values. In the other words Anova test is used to determine the differences among dependent and independent groups. The comparison between the actual variation of experimental data averages and standard deviation is expressed in terms of *F* ratio. *F* is equal to found variation of data averages (between groups variance) / expected variation of the data averages (within group variance). *p* report the significance level, *dF* indicates the number of degrees of freedom. One way ANOVA test (at a significance level of 0.05) was performed to determine whether e.i. COD removals, VFA production and solids removals of reactors or removals at different HRTs are statistically different.

Bivaried correlation analysis was applied to determine whether as A scores (OLR and HRT) increase (decrease), B scores (sCOD removal efficiency (%)/ g VSS ratios) decrease or increase. in UAF and UASB reactors.

Multiple regression was performed to determine the correlation between y (independent) variables and x (dependent) variables such as y (sCOD removal efficiency, VSS removal) and x (VFA production, alkalinity, methane percentage, methane production).

## CHAPTER 4

### RESULTS AND DISCUSSION

#### **4.1. Characterization of Domestic Wastewater in Turkey and Comparison with Different Countries**

During the entire experiments, real domestic wastewater was used without any nutrient addition. The basic composition of domestic wastewater used during the experiments was presented and discussed in Chapter 2. The used domestic wastewater can be categorized as a wastewater of medium strength (Tchobanoglous and Burton, 1991). The strength of domestic wastewater used exhibited variations due to the consumption of water. Sewage characteristics of this study in different cities for different countries and continents are presented in Table 4.1. The influent domestic wastewater values of this study can be considered as “typical domestic wastewater” for Turkey (Orhon et al., 1997).

The tCOD and sCOD values in Ankara-Turkey were considerably lower than Amman, Egypt, refugee camps of Palestine and Brazil while presented similarities with Istanbul and Netherlands. The tVFA-COD values were given for Palestine, Egypt and Netherlands were higher than that of the average of the tVFA-COD value of this study. The  $\text{NH}_4\text{-N}$  and TKN values in Ankara (in this study) were lower than Palestine, Jordan and Netherlands and more or less the same with

Brazil, Istanbul and Egypt. The total-P and PO<sub>4</sub>-P values of domestic wastewater of this study were about the same with Netherlands but higher than that Istanbul (more than two times). The average protein-COD of domestic wastewater used in this study was lower than Palestine and Jordan like tCOD and sCOD values of these countries in Table 4.1. The VSS/TSS ratio gives an idea about the biodegradability of wastewater (high VSS/TSS ratio indicates high biodegradability Mahmoud, 2002). The VSS/TSS ratio of domestic wastewater showed a great variation (48%- 94%) depending on the wastewater characteristics.

Table 4.1. Domestic Wastewater characteristics of this study and sewage characteristics of different cities in different countries and continents

Parameter (mg/L)	<sup>1</sup> Turkey Ankara	<sup>2</sup> Pales- tine	<sup>3</sup> Jordan Amman	<sup>4</sup> Egypt Rural	<sup>5</sup> Turkey Istanbul	<sup>6</sup> Nether- lands	<sup>7</sup> Brazil	<sup>8</sup> Colum- bia
tCOD	486	1586	1183	824.9	410	528	727	267
sCOD	226	393	608	270.2	140	225	-	112
tVFA- COD	39	160	104-177	-	-	55	-	-
NH <sub>4</sub> - N	38	80	80	26	30	48	34	17
TKN	41	104	109	33.8	43	70	44	24
Total-P	21	13	-	8.9	7.2	18	11	1.3
PO <sub>4</sub> -P	10	12.9	-	3.87	4.5	14	8	-
Protein- COD	155	224	272	-	-	-	-	-
TSS	213	736	420	310	210	-	492	215
VSS	146	617	330	277	145	-	252	108
VSS/TSS ratio	70	84	79	89	70	-	51	50

1, this study; 2, Mahmoud (2002); 3, Kersens (2001); 4, Tawfik (1998); 5, Orhon et al. (1997); 6, Elmitwalli (2000); 7&8, Haandel and Lettinga (1994). All parameters are in mg/L except: VSS/TSS ratio.

The average VSS/TSS ratio, presented in Table 4.1, is typical for Turkey (Orhon et al., 1997) and is high when comparing with the data obtained from Brazil and Colombia. In this study, the VSS/TSS ratio is low compared to Palestine, Jordan and Egypt.

The results of tVFA/tCOD, tVFA/sCOD and sCOD/tCOD ratios are presented in Table 4.2. The results show that there are considerably differences between summer and winter season for the aforementioned parameters in domestic wastewater used in this study.

It is observed that around 9% and 40% of dissolved COD are in the VFA form, in winter and summer time, respectively. The results also show that around 49% of the total COD are present in hydrolyzed form in winter and 40% of tCOD is hydrolyzed in summer. The acidified fraction is around 4% and 16% in winter and summer time, respectively.

Table 4.2. Percentages of hydrolysis and acidification of total COD and acidification of dissolved COD of domestic wastewater of the present study

	Parameter (dimensionless)	Winter	Summer
Acidified fraction	tVFA/tCOD ratio	4 %	16 %
Acidified fraction of dissolved COD	tVFA/sCOD ratio	9 %	40 %
Hydrolysed fraction	sCOD/tCOD ratio	49 %	40 %

The Acidified fractions (tVFA/tCOD ratios) were determined as 10% in Palestine and 9% in the Netherlands (Mahmoud, 2002; Elmitwalli, 2000). The acidified

fractions (tVFA/tCOD ratios) of the present study were 4% in winter and 16% in summer with a yearly average of 12%. Although the yearly average acidified fraction (tVFA/tCOD ratio) of domestic wastewater in this study were very close to the given values for Palestine and Netherlands, the difference between summer and winter time was very high for this study. The seasonal difference also is high for the acidified part of dissolved fraction (tVFA/sCOD ratio). The aforementioned ratio is 9% in winter and 40% in summer with a yearly average of 33% in this study. The higher tVFA/COD ratio is an indicator of the higher acid production namely VFA's. It can be concluded that the end products in hydrolysis step is converted to VFA's. The tVFA is considerably higher in summer compared to winter. The temperatures of sewage mixture is observed as 12-14°C in winter and 23-25°C in summer in the present study. The variation in acidification percentages can be explained by the difference in sewage mixture temperatures during summer and winter periods.

In this study, the hydrolyzed fraction (sCOD/tCOD ratio) was observed as 49% and 40% for winter and summer time, respectively. The hydrolyzed fractions were given between 25% and 39% for the cities of Palestine (Mahmoud, 2002) and the results of the present study seemed compatible with the results given in the literature. The hydrolyzed fraction converted to stable end products before collection and experimentation of domestic wastewater in the present study. The reason of decrease in sCOD for influent domestic wastewater (average 260 mg/L for winter and 146 mg/L for summer) and tCOD (average 536 mg/L for winter and 368 mg/L for summer) in summer can be explained with higher biodegradation of organic compounds before collection and during experimentation in summer.

The BOD<sub>5</sub>/tCOD ratio indicates the percentage of the tCOD in raw sewage, which is potentially biodegradable under aerobic conditions. The BOD<sub>5</sub>/tCOD ratios in domestic wastewater are illustrated in Figure 4.1. It was observed that the BOD<sub>5</sub>/tCOD ratio varied between 0.25 and 0.60 (dimensionless) during this study. The BOD<sub>5</sub>/tCOD ratio is one important factor for efficient performance of the anaerobic system treating complex wastewaters. A wastewater with a BOD<sub>5</sub>/tCOD ratio higher than 0.5 was considered as highly biodegradable under anaerobic treatment at mesophilic conditions (40°C) (Mohan et al., 2005).

The treatability of the COD in an influent depends to wastewater type. However, in this study, it was observed a low BOD<sub>5</sub>/tCOD ratio was less than 0.4. In the present study, high variations in BOD<sub>5</sub>/tCOD ratio changing from 0.25 to 0.60 (dimensionless) in domestic wastewater were observed. Seasonal variations in rainfall caused to vary of BOD<sub>5</sub>/tCOD ratios in the influent of domestic wastewater.

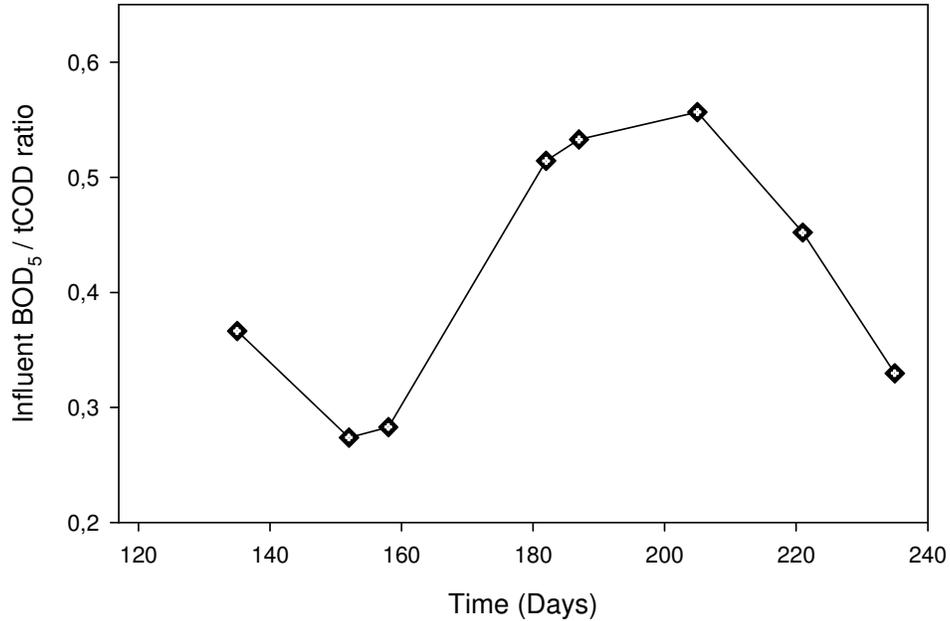


Figure 4.1. The ratio of BOD<sub>5</sub>/ tCOD of influent domestic wastewater

#### 4.2. Startup and Operation of Acidogenic UAF and UASB Reactors (Set 1)

To steady-state conditions for hydraulics and removal efficiencies were reached in about 4 months. The steady state conditions is defined with tVFA production in the effluent and pH level in the reactor with changes less than 20% and 5%, respectively in two weeks. The mean HRT during the start up was kept at  $24 \pm 1.6$  h. Start-up was completed, when the pH values ranged less than  $\pm 5\%$  for pH= 7.77 and 7.93 and tVFA production at a concentration of 55 and 36 mg/L in the effluent with a change less than 20%. After the start-up, the HRT was lowered stepwise from the initial 24 h to 8 h and finally to 2 h. The experimental set-up and operational conditions are given in Table 3.6 (Chapter 3.4).

The operational period in Set 1 can be described in three periods; Period 1, Period 2 and Period 3 as hydraulic retention times are equal to 24 hours, 8 hours and 2 hours, respectively.

#### **4.2.1. Effects of HRT and OLR on the effluent tCOD, sCOD and removal efficiencies in UAF and UASB Reactors**

The OLR was very low and ranged between 90 mg sCOD/L.day and 130 mg sCOD/L.day in Period 1 for UAF and UASB reactors in Set 1. The Influent sCOD and tCOD values ranged between 173-250 mg/L and 247-610 mg/L, respectively, in Period 1 for UAF and UASB reactors. Influent, effluent concentrations and removal efficiencies for sCOD and tCOD are illustrated in Figure 4.2 for UAF Reactor.

The removal efficiencies varied between 64 and 71% with an average of 66% for tCOD in Period 1 (24 h HRT). The average values in effluent of the UAF reactor were measured as 77 mg/L and 152 mg/L for sCOD and tCOD, respectively. In UAF Reactor, the effluent COD values ranged from 53 to 187 mg/L with an average of 101 mg/L for sCOD and from 125 to 210 mg/L with an average of 171 mg/L for tCOD in Period 2 (8 h HRT). The average reduction for tCOD was obtained as 48% in Period 2.

The total COD in domestic wastewater consists from readily degradable-soluble COD, inert COD and slowly degradable COD. The literature studies showed that the tCOD converted to sCOD in the hydrolysis step of acidification reactor for the treatment of domestic wastewaters. The sCOD is converted to volatile fatty acids such as acetic acid, propionic acid, and butyric acid, carbon dioxide and hydrogen.

Acetic acid is an excellent substrate for the next methanogenic step. sCOD/ tCOD ratio should be increased in the acidification reactors since the tCOD converted to sCOD. Contrarily, in this study the sCOD/ tCOD ratio decreased since a part of sCOD (based on VFA) converted to methane. The decreases in sCOD could be attributed to activity of the methanogens although the methane gas could not be measured in step 1.

In acidification, the sCOD concentrations increased while the tCOD concentrations does not change significantly (Wang et al., 2002). It is already stated in literature that there is minimal reduction of tCOD in the acid fermentation stage since the complex compounds such as short-chain fatty acids, alcohols produced, and new bacteria cells exert an oxygen demand (Speece, 1996). In general, it was expected rises in effluent sCOD due to hydrolysis of organic matter (Wang et al., 2002; Guerrero et al., 1999). On the other hand the VFA concentrations increased since a part of sCOD consisting from the VFA. Since a certain fraction of wastewater used in this study was inherently acidified form before collection and operation in the UAF reactor, the sCOD/tCOD fraction of this wastewater is high. Therefore it can be said that the wastewater is hydrolyzed before used in the UAF reactor. In the other words, in the influent domestic wastewater, a certain part of tCOD was in solubilized and acidified form. In this thesis, Set 1 experiments were performed in wintertime and it was observed that about 49% of influent domestic wastewater has already been hydrolyzed (sCOD/tCOD=49%, Table 4.2, Section 4.1). The ratio of the acidified fraction to the hydrolyzed soluble COD was determined as 9% (tVFA/sCOD fraction, Table 4.2) for winter period. In this study the sCOD does not increase and the removals were observed for tCOD in the UAF reactor. The declining in sCOD, tCOD and VFA could be acceptable since the acidogenic reactors were not completely as in acidification phase (Yılmaz, 2007). Therefore, as time passing

some methanogens grow and started to consume VFA in acidification phase. The degradation of acidified portion of sCOD to end products would be possible at short HRTs.

By decreasing HRT from 24 h to 8 h, it was observed a certain decrease in tCOD reductions in UAF reactor. In an acidogenic reactor, it was not expected any reduction in tCOD concentration. The characteristics of domestic wastewater used in this study showed that about half of the tCOD was hydrolyzed and 4% of the tCOD was acidified before operation (Table 4.2). As reported by van Haandel and Lettinga, (1994) it is not possible to inhibit the methanogenic bacteria by only decreasing HRT because the environment for methanification was proper since the pH ranged between 7.2 and 7.8 for HRTs of 8 h and 24 h under mesophilic temperatures in UAF reactor. In this study, in 8 h and 24 h HRTs, most of the COD removals can be explained with methanogens which was enhanced in UAF reactor.

Period 3 was initiated with operation of UAF reactor at an HRT of 2 h. The sCOD reduction percentage was at a maximum of 34% with an average of 11% in the UAF reactor and some increases were observed in Period 3 (HRT=2 h). The effluent tVFA concentration reached about to 145 mg/L as Hac at that HRT. This suggests that about 28% of influent tCOD is found as tVFA in the effluent. The results of this study with effluent tVFA of 20% of total influent COD were found compatible with an effluent tVFA value of 110 mg/L at 2.2 h HRT (Ligero et al., 2001). They studied the pretreatment of domestic wastewater by a hydrolytic upflow digester for sedimentation and hydrolysis of suspended solids and for acidification of solubilized substances. Previous studies that could be used as a reference to compare the results of this study are scarce. Gonçalves et. al. (1994) obtained optimum acidification results at an HRT of 2.8 h and at a temperature of

20°C with a fermentation rate of 0.17 mg VFA-COD/mg tCOD for domestic wastewater. Generation rate of acids increased and small increases were observed in effluent sCOD as HRT decreased to 2 h in the present study.

The tCOD reduction percentages were between 29% and 67% with an average of 40% in Period 3. The removal of a certain part of tCOD can be explained with the conversion of VFAs to the end products by methanogenic bacteria whereas VFAs existing in influent domestic wastewater. Methane percentage of biogas (17%) proved that there were methanogenic bacteria and some methanification was happened in Period 3 in UAF reactor.

A sample calculation is given in Appendix D for the removals of COD components in Period 3.

The sCOD and tCOD variations in influent and effluent of UASB Reactor were presented in Figure 4.3. It was observed that the removal efficiencies ranged between 66% and 88% with an average of 81% for tCOD in UASB Reactor at Period 1. The average effluent values of UASB Reactors were determined as 57 mg/L and 72 mg/L for sCOD and tCOD Reactors, respectively for 24 h HRT.

In UASB Reactor, the effluent COD values ranged from 57 to 68 mg/L with an average of 63 mg/L for sCOD and from 114 to 208 mg/L with an average of 149 mg/L for tCOD in Period 2. The average reduction was 65% for tCOD, respectively.

A certain part of granular sludge in UASB reactor was washed out on day 223. In the evaluation of the treatment efficiencies, the values obtained between days 192 and 223 are taken into consideration in Period 3, since the tCOD values (tCOD

removal decreased from 67% to 28%) of days 223-237 indicated the effect of wash out of granules on the treatment efficiency.

The averages of influent sCOD and tCOD are 295 mg/L and 509 mg/L, respectively for the days 192 and 237 in the UASB Reactor. The averages of effluent sCOD values were 158 mg/L and 179 mg/L for the days 192-223 and 223-237, respectively in UASB Reactor.

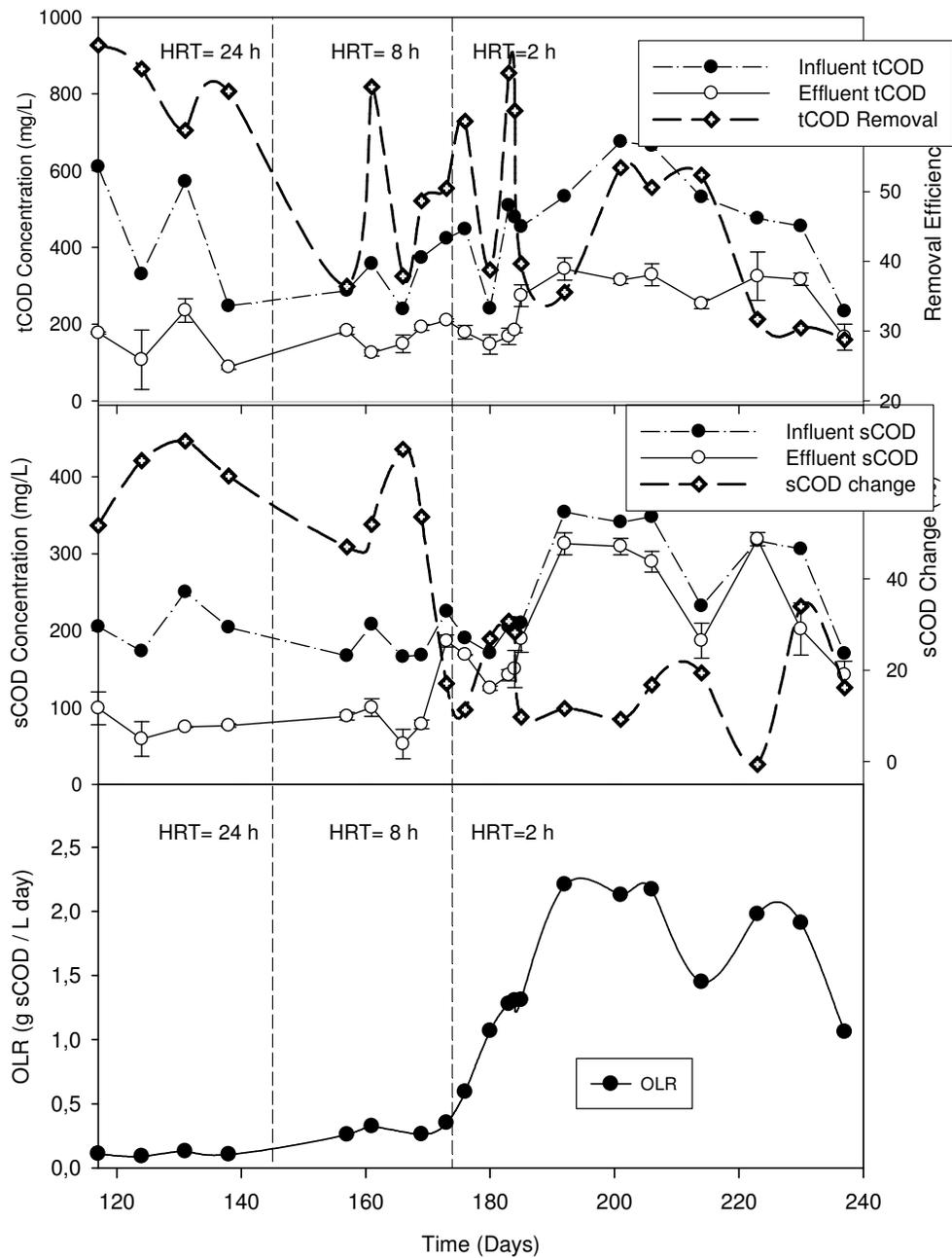


Figure 4.2. Effect of HRT and OLR on sCOD, tCOD effluents, sCOD changes and tCOD removal efficiencies versus operation period for acidogenic UAF reactor.

The averages of effluent tCOD were 253 mg/L and 248 mg/L for days 192-223 and 223-237, respectively, in the UASB reactor. The tCOD treatment efficiency decreased from 56% to 28% in Period 3 at 2 h HRT . The reason of such a decrease in the tCOD efficiency can be explained by the loss of granules from the upper part of the UASB reactor. Since the average tVFA production was low (87 mg/L as Hac, Section 4.2.5) and the methane percentage of total gas was 52% it can be concluded that UASB reactor operated in methanogenic phase in Period 3 at 2 h HRT.

In the UAF Reactor, the data for days 192-237 is evaluated in Period 3. The influent wastewater COD ranged between 170 and 354 mg/L with an average of  $295 \pm 125$  mg/L and between 233 and 675 mg/L with an average of  $510 \pm 277$  mg/L for sCOD and tCOD, respectively.

The removal efficiencies at the studied HRTs for tCOD are shown in Table 4.3 for the UAF and the UASB reactors. It was observed a decrease in tCOD treatment efficiencies with the decrease in HRT and increase in OLR in both reactors. It was observed increases in effluent sCOD concentrations with the decrease in HRT. This could be attributed to hydrolytic organisms degrading the soluble organic compounds through hydrolysis phase and to the acidogenic bacteria has not enough time to degrade the soluble organics at lower HRTs. The removals were slightly lower in 8 h HRT than 24 h HRT in the UAF and the UASB reactors for tCOD efficiencies. The increases in effluent concentrations for sCOD in the UAF reactor were noticeable with the decrease in HRT from 8 h to 2 h. The effluent sCOD concentrations of the UAF reactor were considerably high compared to the UASB reactor in 2 h HRT.

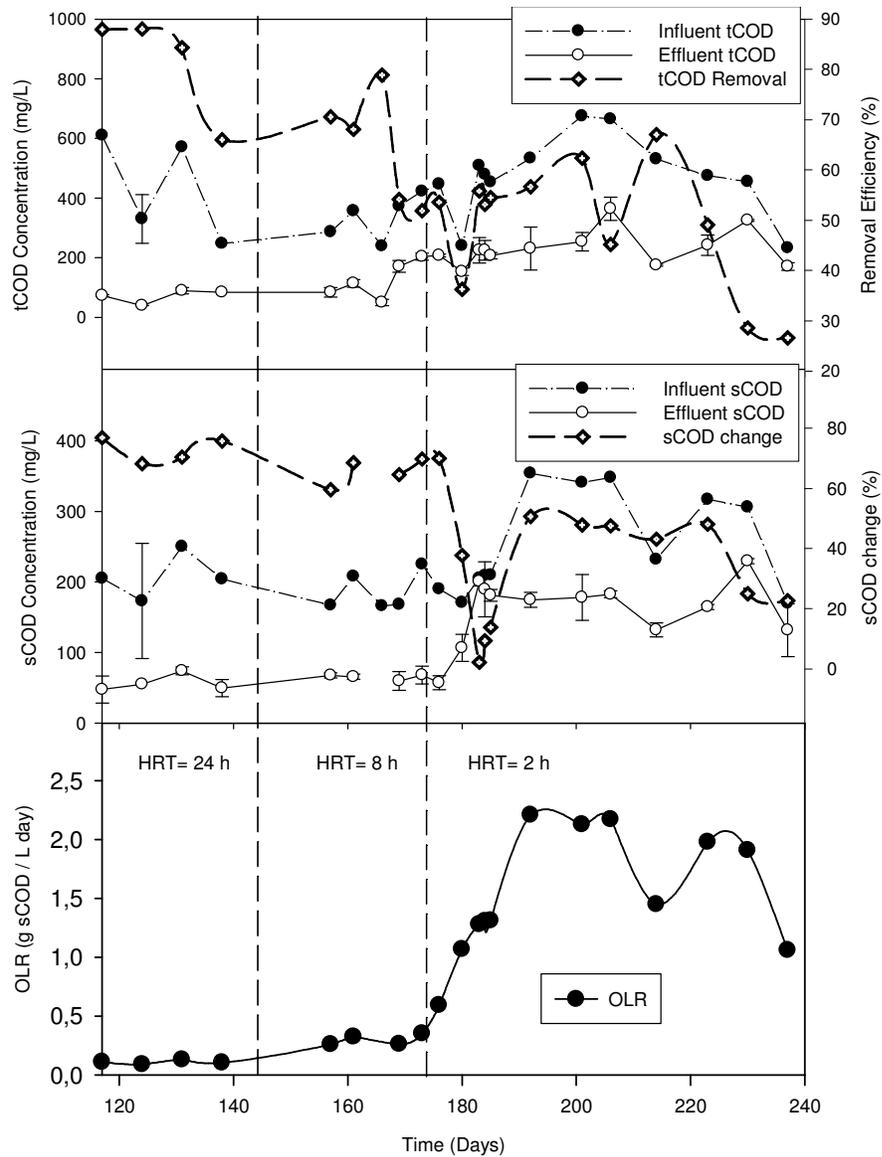


Figure 4.3. Effect of HRT and OLR on sCOD, tCOD effluents, sCOD changes and tCOD removal efficiencies versus operation period for acidogenic UASB reactor.

Table 4.3. The COD removal efficiencies of acidogenic UAF and UASB Reactors versus HRT (OLR)

Period	Operating parameters				sCOD		tCOD removal efficiency (%)	
	HRT (h)	OLR (gCOD/L day)	Influent sCOD (mg/L)	Influent tCOD (mg/L)	Effluent UAF	Effluent UASB	UAF	UASB
	1	24	0.09-0.13	173-250	247-610	59-77	48-74	64-71
2	8	0.26-0.59	166-225	239-423	53-187	57-68	36-65	54-79
3	2	1.06-2.21	171-348	233-675	125-309	87-227	29-67	36-67

In Period 3, the average changes for sCOD were -11% and -48% for UAF and UASB reactors, respectively (- sign shows reduction). The ANOVA test statistics performed for sCOD changes of UAF and UASB reactors in Period 3 (HRT=2 h) indicated that the sCOD changes of the UASB reactor were significantly higher than the UAF reactor ( $F(1, 8) = 95.97$ ,  $p \leq 0.001$ , Table B.1, Appendix B). The sCOD changes of UAF and UASB reactors were statistically different in Period 3. While the average treatment efficiencies for tCOD were 16% higher in UASB reactor than UAF reactor. The ANOVA test was applied for tCOD removal efficiencies calculated in UAF and UASB reactors at Period 3 (HRT=2 h). The ANOVA test statistics indicated that the tCOD removal efficiencies of the UASB reactor were significantly higher than the UAF reactor ( $F(1, 8) = 41.42$ ,  $p < 0.001$ , Table B.2, Appendix B) in Period 3 (HRT=2 h). The tCOD removals between UAF and UASB reactors were statistically different in Period 3. By decreasing the HRT from 8 h to 2 h, the average tCOD removal efficiency decreased from 53% to 40% in the UAF reactor while it decreased from 65% to 56% in the UASB reactor in the decrease of the HRT from 8 h to 2 h.

In this study inherent acidification and hydrolysis were observed in influent domestic wastewater. The decreases in sCOD and tCOD could be acceptable since

the acidogenic reactors were not completely in acidification phase. As time passing some methanogens grew and started to consume the VFA produced. On the other hand as HRT decreased, the effluent sCOD concentrations in both of the reactors increased and tCOD removals decreased. The VFA to sCOD ratio was higher in UAF (0.54) reactor than UASB reactor (0.32) as given in Section 4.2.7 at 2 h HRT. This indicated a better acidification was provided in UAF reactor.

#### **4.2.2. Effects of Organic Loading Rate (OLR) and Hydraulic Retention Time (HRT) on the variations of sCOD change /VSS ratio**

Since the soluble organic compounds in domestic wastewater was used by the anaerobic biomass, it is important to compare the soluble COD changes by the microorganisms in comparison to the available biomass in the reactors. The ratio of sCOD changes to VSS content was evaluated for UAF and UASB reactors (i.e. sCOD change (%) / g VSS). Figure 4.4 shows the effects of OLR and HRT on the sCOD change /VSS ratio in UAF and UASB reactors. It is observed that, UAF reactor had an average -7.23 % sCOD change/ g VSS in low organic loading rates (90-318 mg COD/L. day) in Period 1 (See Figure 4.4). However, the sCOD change (%) /g VSS ratio was -4.48 in UASB reactor, indicating that UASB reactor lower sCOD (%) changes /g VSS ratio than that UAF reactor in Period 1.

In the comparison of sCOD change (%) / g VSS, the UAF reactor had 38% higher reduction (%) / g VSS than that of UASB reactor at organic loadings of 260-350 mg sCOD/L.day in Period 2. The differences between sCOD changes /VSS ratios in UAF and UASB reactors were about the same for Period 1 and 2. In the UAF Reactor, the ratio (% sCOD change / g VSS) was between -5.4 and -8.1, while in UASB Reactor this ratio were between -3.7 and -4.7 in Period 1 and 2. It was

observed that this ratio ranged between 0 and -3 and between -1.4 and -3.4 for UAF and UASB Reactors, respectively, in Period 3. It can be concluded that the higher VSS content of UASB reactor resulted in a more uniform sCOD changes (%) / g VSS ratio. This ratio indicated a higher decrease in sCOD changes (%) / g VSS in UAF reactor with the decrease of HRT from 24 h to 8 h and 2 h. It was observed that the sCOD changes (%) / g VSS ratio decreased when the HRT decreased in both the UAF and UASB reactors. The Bivaried Correlation test was conducted to investigate whether there is a significant relationship between HRT and sCOD change (%) / g VSS ratios in UAF and UASB reactor scores. As the HRT decreased from 24 h to 8 h and 2 h , sCOD change (%) / g VSS ratio decreases were significant in UAF and UASB reactors ( $r(18)=0.795$ ,  $p<0.001$  and  $r(18)=0.686$ ,  $p=0.001$ , Table B.4 and Table B.5, Appendix B). The statistical correlation showed that as the HRT decreased, the sCOD changes (%) / g VSS ratios decreased in both reactors.

With the decrease in HRT, the OLR increased. It was observed that the UAF reactor displayed better sCOD reduction (%) / g VSS performances when the OLR was lower than 1.1 g COD / L.day, It was observed increases in sCOD changes (%) / g VSS ratio with the increase in OLR in the UAF reactor. The Bivaried correlation test was conducted to understand whether there is a significant relationship between increase in OLR and increase in sCOD change (%) / g VSS of UAF reactor. The correlation was statistically significant in the UAF reactor ( $r(18)=-0.843$ ,  $p<0.001$ , Table B.6, Appendix B). There was a statistical relation between increase in OLR and sCOD change (%) / g VSS in UAF reactor. As the OLR increased the sCOD change (%) / g VSS ratio increased. However it was not observed a statistical correlation between sCOD change (%) / g VSS ratio and OLR in the UASB reactor ( $r(18)=-0.391$ ,  $p=0.089>0.05$ ), Table B.7, Appendix B). In UASB there was no statistical correlation between the increases in OLR and the

sCOD changes (%)/gVSS ratio. The amount of VSS is very important in the sCOD change /VSS ratio. A decrease in VSS in UASB Reactor (washout at day 223), the sCOD reduction decreased from 48% to 24%). The results showed that decreases in VSS in the UASB reactor causes lowering of % sCOD decrease / g VSS ratio. The ANOVA statistical Analysis test was performed to understand the statistical significance of sCOD changes (%)/gVSS ratio in UASB and UAF reactors in Period 3 (HRT=2 h). The ANOVA test results indicated a significant statistical difference in sCOD changes (%)/gVSS ratio between UAF and UASB reactors in Period 3 ( $F(1, 12) = 6.69, p = 0.024$ , Table B.8, Appendix B). In Period 3, the sCOD change (%)/gVSS ratios in UAF and UASB reactors were statistically different. The sCOD reduction (%) / g VSS ratio in UASB reactor is higher than in UAF reactor at OLR higher than 1.1 g COD/ L day.

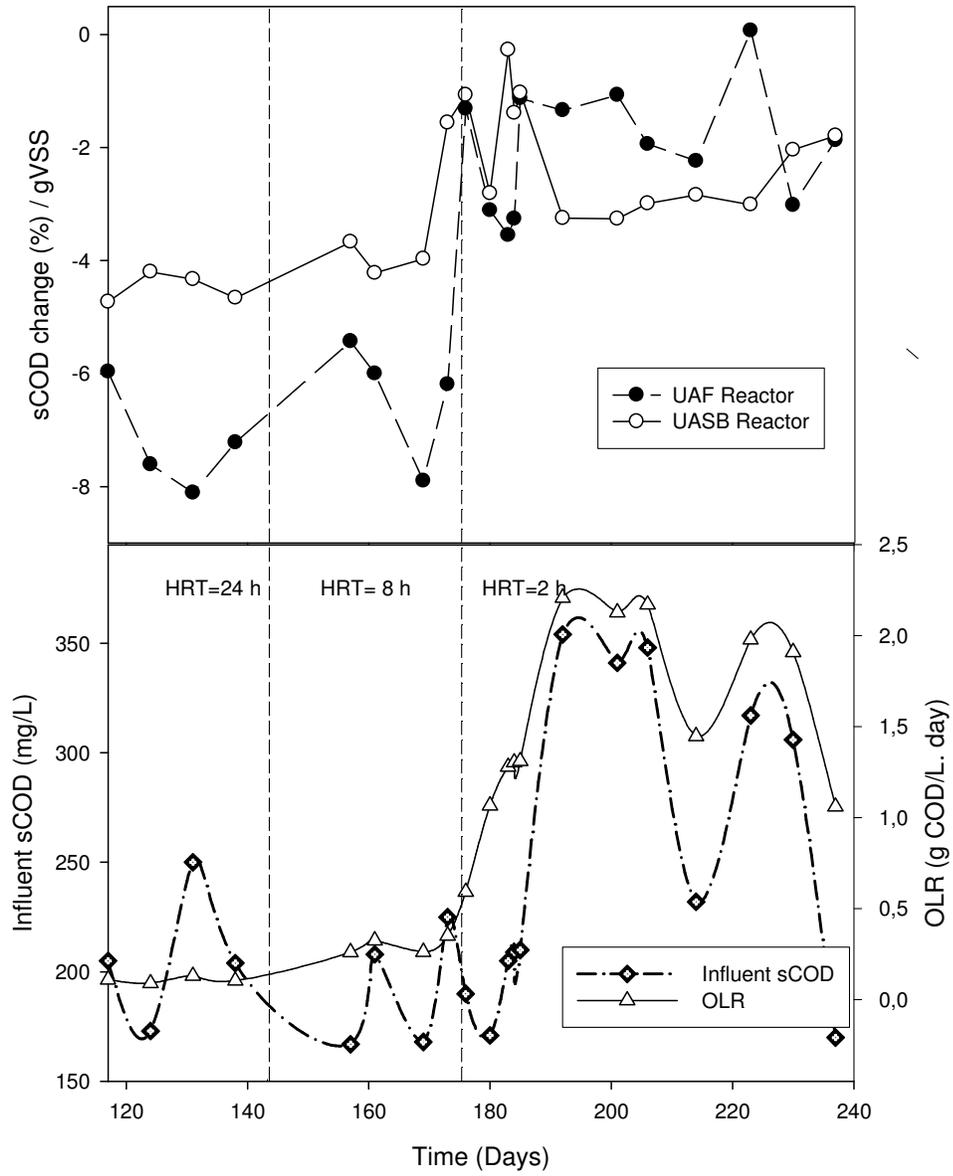


Figure 4.4. Effects of OLR/HRT on the sCOD changes/VSS ratio in UAF and UASB reactor versus time.

### **4.2.3. Effects of HRT and OLR on the variation of pH and ORP in UAF and UASB Reactors**

The HRT of the UAF and UASB reactors were adjusted to 24 hour at the first period of the acidification study. The reactors were operated with an HRT of 24 hours until day 145. The variations of HRT and applied organic loading rates versus time are illustrated in Figure 4.5.a. The variations of pH with time were illustrated depending on the changes in HRT/OLR in the same figure. The time is plotted in x-axis in this Figure and the HRT/OLR applied to the reactors are shown as y-axis in Figure 4.5.a. The effluent pH values are shown in vertical axis in Figure 4.5.b.

The pH measurements were done every day. The pH values in the UAF Reactor represented lower pH values than that UASB Reactor. The pH values were ranged between 7.57 to 7.95 and 7.62 to 8.25 and averaged as 7.77 and 7.93 for UAF and UASB reactors, respectively (See Figure 4.5.b) in Period 1 (HRT=24h ) of this study. Although the final pH values depend mainly on the nature of the wastewaters (Alexiou et al., 1994), the optimum pH for methanogens is within the range 7.5- 8.5 (Hobson and Wheatly, 1993). The effluent pH values of the present study at 24 h HRT showed similarities with the results of Hobson and Wheatly (1993).

In the transient period (days between 145-151), HRT was gradually decreased from 18 h to 12 h and 8 h in Period 2 (days 152-173) the OLRs of the reactors were increased from 0.09- 0.13 g COD/L day to 0.261- 0.324 g COD/L day by decreasing the HRT to 8 h. It is important to note that the strength of influent wastewater showed variations in COD day by day and decreases in HRT resulting in a detectable increase in OLR.

The reactors were operated in steady state conditions by considering the change in pH and VFA values in a period of four days after the start-up period at 8 h HRT. In steady state conditions, the pH values were ranged from 7.21 to 7.56 with an average of 7.32 and from 7.38 to 7.70 with an average of 7.59 for UAF and UASB reactors, respectively.

Although a certain decrease in pH values was observed (average pH, from 7.77 to 7.32 in UAF reactor and from 7.93 to 7.38 in UASB reactor) with the decrease in HRT from 24 h to 8 h, the reactors were considered to still the optimum methanogenic pH range as reported by Hobson and Wheatly, 1993 (See Figure 4.5.b).

The reactors were experimented for 2 hours HRT during days 178 and 237. On day 186, the stopper of the UASB Reactor was flushed out and the half of the granules floated. The granules changed place with water and some granules contacted with air at the top since it was observed some gas pockets inside UASB Reactor. It was added one more spiral cable besides the existing one to prevent the collection of gas pockets at the upper part of the UASB Reactor.

In 24 h HRT, the ORP values were observed around -200 mV and -300 mV for UAF and UASB Reactors, respectively. The optimum range of ORP is known as -60 mV to -220 mV, for biological denitrification and it is the almost the same as for acidogenesis (Noike et al. 2004). Decreasing of HRT to 8 hours resulted with positive ORP values for a period of 7 days and the reactors are considered unstable for anaerobic conditions. After that period, the ORP values of reactors ranged around -150 mV and -250 mV for UAF and UASB reactors, respectively. Negative ORP values about -300 mV indicates the strict anaerobic conditions (Gerardi M.H., 2003).

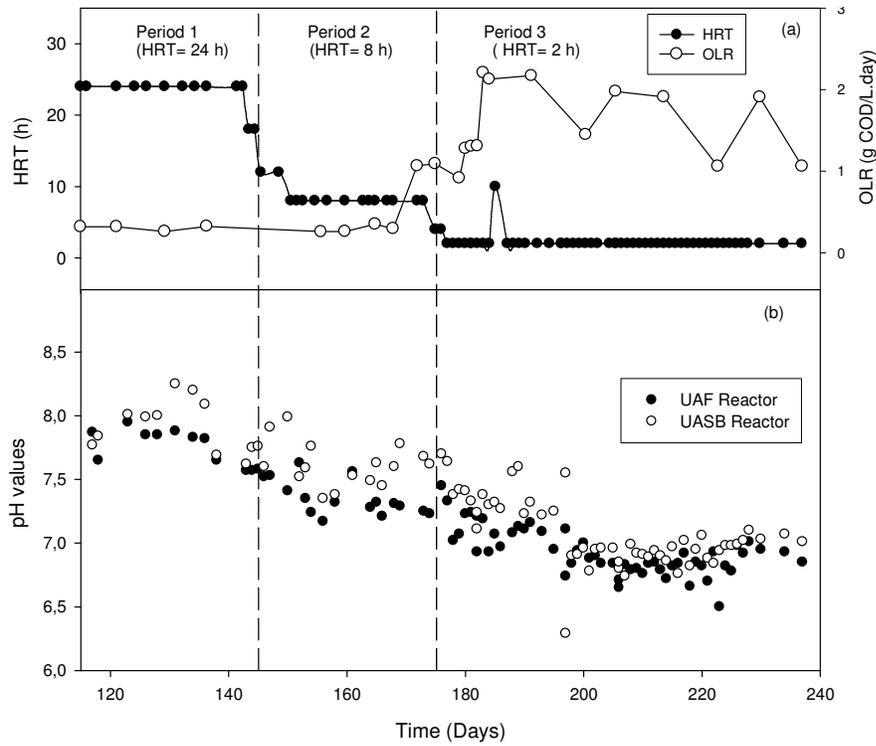


Figure 4.5. Variatons of pH of acidogenic UAF and UASB Reactors at applied OLR's and HRT's.

The dissolved oxygen of the influent wastewater was measured as 3 mg/L at the collection point of domestic wastewater and 2.5 mg/L at 6 °C in the refrigerator and it is planned to provide anoxic/anaerobic conditions for the treatment of domestic wastewater. The reactors were flushed with N<sub>2</sub> gas for half an hour to ensure the anaerobic conditions in day 197. After the date of flushing reactors with N<sub>2</sub>, the influent wastewater also is flushed with nitrogen gas for 5 minutes to ensure the anaerobic conditions from day 197 to 207.

After day 207, sodium thioglucollate was added to the influent wastewater to reduce the redox potential and sustain the anaerobic conditions. The addition of sodium thioglucollate was proposed between (w/w) 0.01-0.2% for anaerobic

conditions (Sponza and Işık, 2004). In the case of addition of sodium thioglucollate, it was experimentally monitored that in order to have a dissolved oxygen (DO) concentration of 0.5-1 mg/L it should be added 0.01 g/L sodium thioglucollate to the influent wastewater. The reason of aiming 0.5-1 mg/L DO is for maintaining the anoxic conditions since acidogenesis is occurred efficiently as reported by Yerushalmi et al. (2001) and Ergüder (2005). On day 223, the flows of the reactors were increased by mistaken and a certain amount of granules from the UASB Reactor washed out while there was no loss of the sludge in UAF Reactor. The effects of VSS losses in UASB reactor for days 223-237 was explained in Section 4.3.1.

In Period 3 (HRT= 2 h), The OLR of the reactors ranged between 1.061 and 2.171 g sCOD/L day. The reason of the variation in OLR is because of the variation of the strength of influent domestic wastewater although the flow rate was not changed.

The pH values ranged between 6.50 and 7.01 with an average of 6.83 and ranged between 6.80 and 7.10 with an average of 6.93 in UAF and UASB reactors, respectively. It was observed that a certain decrease in pH values with the decrease in HRT from 24 h to 8 h and 2 hours in both reactors (See Figure 4.5.b). It was known that pH may vary during acidogenesis, which causes the system tended to buffer itself towards a pH value in the range of 5-7 if no control was carried out (Breure and Van Andel ,1984 ; Bull et. al., 1984 and Dinapoulou et al., 1988). On the other hand the final pH value depended on the nature of the wastewaters (Alexiou et al., 1994). As an example, in this part of this study, the influent domestic wastewater values were between 7.57 and 7.79, 146 and 235 mg/L as CaCO<sub>3</sub>, 27 and 253 mg/L, 124 and 354 mg/L for pH, alkalinity, VSS and sCOD, respectively. In this study, it was observed that the effluent pH of domestic

wastewater ranged between 6.5 and 7.1 in Period 3. This pH range provides available environment for acidogenesis of domestic wastewater during anaerobic treatment (See Figure 4.5.b).

Almost in all of the results in 2 h HRT, the pH values were lower in UAF Reactor than that of UASB Reactor (See Figure 4.5.b). By decreasing the HRT from 24 h to 2 h, it was obtained a decrease in average pH. The average pH decreased from 7.77 to 6.83 in UAF reactor for aforementioned HRTs.

#### **4.2.4. Effects of HRT and OLR on the effluent tVFA in UAF and UASB Reactor**

The average effluent tVFA values were 55 and 37 mg/L as Hac for UAF and UASB Reactors, respectively in Period 1 at an HRT of 24 h. The average effluent sCOD values were 83 mg/L and 70 mg/L while tVFA values were 66% and 53% of sCOD in UAF and UASB Reactors, respectively. The COD equivalents of each volatile acids for the conversion were taken as follows: acetic acid, 1.066; propionic acid, 1.512; butyric acid, 1.816; valeric, 2.036; caproic acid, 2.204 (Demirel and Yenigun, 2004). It can be concluded that more than half of the COD which was in soluble form was acidified in both of the reactors (see Figures 4.2 and 4.3).

The tVFA mostly composed of acetic acid indicating the presence of acetogens which convert the acetic acid to methane gas in methanogenic conditions (Gujer and Zehnder, 1983). In Table 4.4, it can be seen that the average fraction of acetic acid (Hac) in the UAF reactor were 21%, 18% and 12% at HRT's 24 h, 8 h and 2 h, respectively. In the UASB reactor, the average fraction of acetic acid was 50%,

28% and 51% at HRT's 24 h, 8 h and 2 h, respectively. In this reactor, acetic acid was observed mostly while propionic acid and butyric acid was significantly higher in UAF reactor. The fraction of acetic acid was higher in the UASB reactor than the UAF reactor in all the studied HRT's. The fraction of acetic acid concentration in the UAF reactor was lowest in 2 h HRT compared to the other studied HRTs. This can be explained with the low acetic acid production via acetogens at low HRTs. On the other hand the UAF reactor had more acidogenic/acetogenic conditions at 2 h HRT than the 8 h HRT and 24 h HRT. Acetic acid is the major intermediate in the bioconversion of organic matter to methane and CO<sub>2</sub>. About 70% of total methane produced in anaerobic digestion originates from acetic acid (Gujer and Zehnder, 1983). Propionic and butyric acid are intermediary products and an additional step (conversion to acetic acid, hydrogen or CO<sub>2</sub>) is required for methanification. Therefore higher acetic acid content indicates the higher methane production and the methanification phase in the UASB reactor.

The effluent tVFA values of reactors are presented in Figure 4.6. It was not determined a significant change in the effluent tVFA values with decrease in HRT from 24 h to 8 h both in the UAF and the UASB Reactors. The average effluent tVFA values of reactors were 60 and 36 mg/L (as Hac) for the UAF and the UASB reactors, respectively at HRTs of 8 h.

The calculations for VFA measurements are done for the days 197 and 237 in both reactors in Period 3. The tVFA concentrations ranged between 90 and 166 mg/ L (as Hac) with an average of 145 mg/L (as Hac) in the effluent of UAF reactor in 2 h HRT. The same parameter ranged between 69 and 101 mg/L (as Hac) with an average of 87 mg/L (as Hac) in the effluent of the UASB reactor in 2 h HRT. The summary of the pH and tVFA values of the UAF and the UASB

reactors at different HRT's and OLRs is given in Table 4.5. It was observed an increase in tVFA for the UAF reactor compared to the UASB reactor. The tVFA concentration increased 2.4 times in UAF reactor by decreasing HRT from 8 h to 2 h. The tVFA was also 1.7 times higher in the UAF reactor than that of the UASB reactor in Period 3.

Table 4.4. Average of main acid types and fractions at studied HRTs for UAF and UASB reactors

UAF Reactor	Hac (mg/L)	HPr (mg/L as Hac)	Butyric (mg/L as Hac)	Other (mg/L as Hac)	Hac (%)	HPr (%)	Buty-ric (%)	Other (%)
24 h	14.56±2.23	12.39 ± 2.74	31.59±11.51	9.22±3.81	21	17	50	12
8 h	14.14±2.29	14.47 ± 3.80	37.06 ± 7.56	9.89 ± 5.78	18	18	53	11
2 h	20.19±5.01	43.46 ± 20.22	74.60±17.98	12.32±6.79	12	29	52	7
UASB Reactor	Hac (mg/L)	HPr (mg/L as Hac)	Butyric (mg/L as Hac)	Other (mg/L as Hac)	Hac (%)	HPr (%)	Buty-ric (%)	Other (%)
24 h	31.78±7.01	9.11±1.73	19.11±5.22	7.11 ±5.70	50	12	29	9
8 h	11.33±2.92	7.08±0.79	17.58 ± 6.04	5.50 ±2.70	28	16	45	11
2 h	43.50±13.74	19.39±5.69	17.66 ± 3.30	8.09 ±4.01	51	22	19	8

The pH values displayed an opposite trend and decreased (from 7.32 to 6.83) with decreasing HRT (from 8 h to 2 h) in UAF reactor. The VFA production was highest in UAF Reactor in 2 h HRT and it was determined as 58 % of the effluent sCOD.

The reason of considerably higher tVFA values in UAF Reactor than that UASB Reactor can be explained with the conversion of high amounts of VFAs to methane gas in UASB reactor.

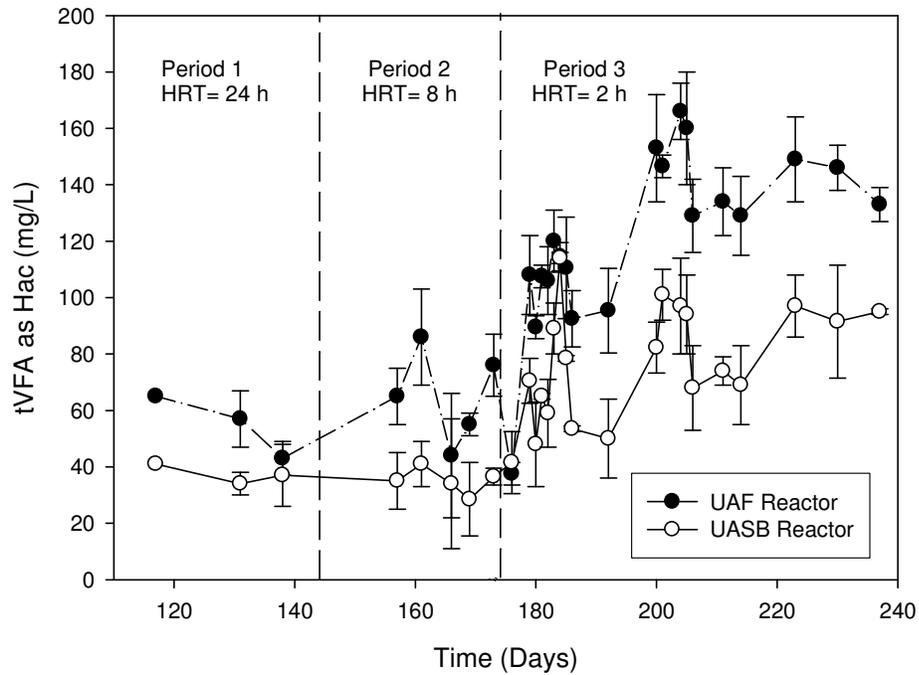


Figure 4.6. Total VFA variations in the effluent of the reactors during operation versus HRT.

Table 4.5. The comparison of effluent VFA and pH values of UAF and UASB Reactors

HRT (hour)	OLR (g sCOD/ L.day)	UAF Reactor		UASB Reactor	
		pH	VFA (mg/L as Hac)	pH	UASB Reactor VFA (mg/L as Hac)
24	0.09- 0.13	7.6- 8 (7.77)*	43- 65 (55)	7,6-8.3 (7.93)	34- 41 (37)
8	0.261-0.364	7.2-7.6 (7.32)	38- 86 (60)	7.4-7.7 (7.59)	29-41 (36)
2	1.1-2.2	6.5-7.0 (6.83)	90- 166 (145)	6.8-7.1 (6.93)	69-101 (87)

\*The values inside paranthesis display average values

#### 4.2.5. Effects of HRT/ OLR on the variation of Alkalinity in Set 1

The alkalinity of a wastewater is important because it is a measure the capacity to neutralize the acids while buffering the pH in the presence of acids (Sawyer et al., 1994). The HRT, alkalinity and applied organic loading rates versus time are illustrated in Figure 4.7. The variations of alkalinity with time are illustrated depending on the changes in HRT/OLR in the same Figure. The resulting influent and effluent alkalinities are shown in vertical axis of Figure 4.7.b, respectively.

Influent domestic wastewater alkalinities were ranged between 184 and 221 mg/L as CaCO<sub>3</sub> with an average of 208 mg/L as CaCO<sub>3</sub> (Figure 4.7). The effluent alkalinities ranged between 70 and 85 mg/L as CaCO<sub>3</sub> and between 92 and 128 mg/L as CaCO<sub>3</sub> with averages of 82 mg/L as CaCO<sub>3</sub> and 109 mg/L as CaCO<sub>3</sub> for UAF and UASB Reactors, respectively. Tchobanoglous and Burton (1991) tabulated the typical alkalinities as 50, 100 and 200 mg/L for weak, medium and high alkalinity untreated domestic wastewater and in general, the domestic wastewater used in this study was observed at high range. When the effluent alkalinities and pH values (See Figure 4.5, Subsection 4.2.4) of the UAF and UASB reactors are considered, the UAF reactor effluent alkalinities and pH values were slightly lower than the UASB reactor at 24 h HRT.

The alkalinities of UAF reactor were also lower than that UASB reactor in Period 2 (HRT=8 h) like Period 1 (HRT=24 h). The average effluent alkalinities were 98 mg/L (as CaCO<sub>3</sub>) and 111 mg/L (as CaCO<sub>3</sub>) for UAF and UASB Reactors, respectively, in Period 2. It was not observed a significant change in alkalinity values between Period 1 and Period 2.

In Period 3, the effluents of the reactors showed that the average alkalinity values were 106 and 120 mg/L (as CaCO<sub>3</sub>), for UAF and UASB reactors, respectively (see Figure 4.7). The alkalinity measurements indicated that there was a reduction in effluent alkalinity values but it was not determined a certain decrease with decreases in HRTs in both reactors.

On the other hand, NH<sub>4</sub>-N produced from the proteins solubilized in the mixture of granules which are on the support material in UAF reactor. The NH<sub>4</sub>-N is in soluble form in this reactor and causes decreases in pH and this indicates acidification of the UAF reactor at lower pH values together with high VFA production as reported by Speece, (1996). However, the configuration of UASB reactor (hydrolysis and acidification in the lower part of the reactor and methanification in the upper part of the reactor) stimulate the alkalinity production in the upper phase resulting in methane production in UASB reactor (Speece, 1996).

With the aforementioned knowledges it can be concluded that the alkalinity in UASB reactor is higher than UAF reactor.

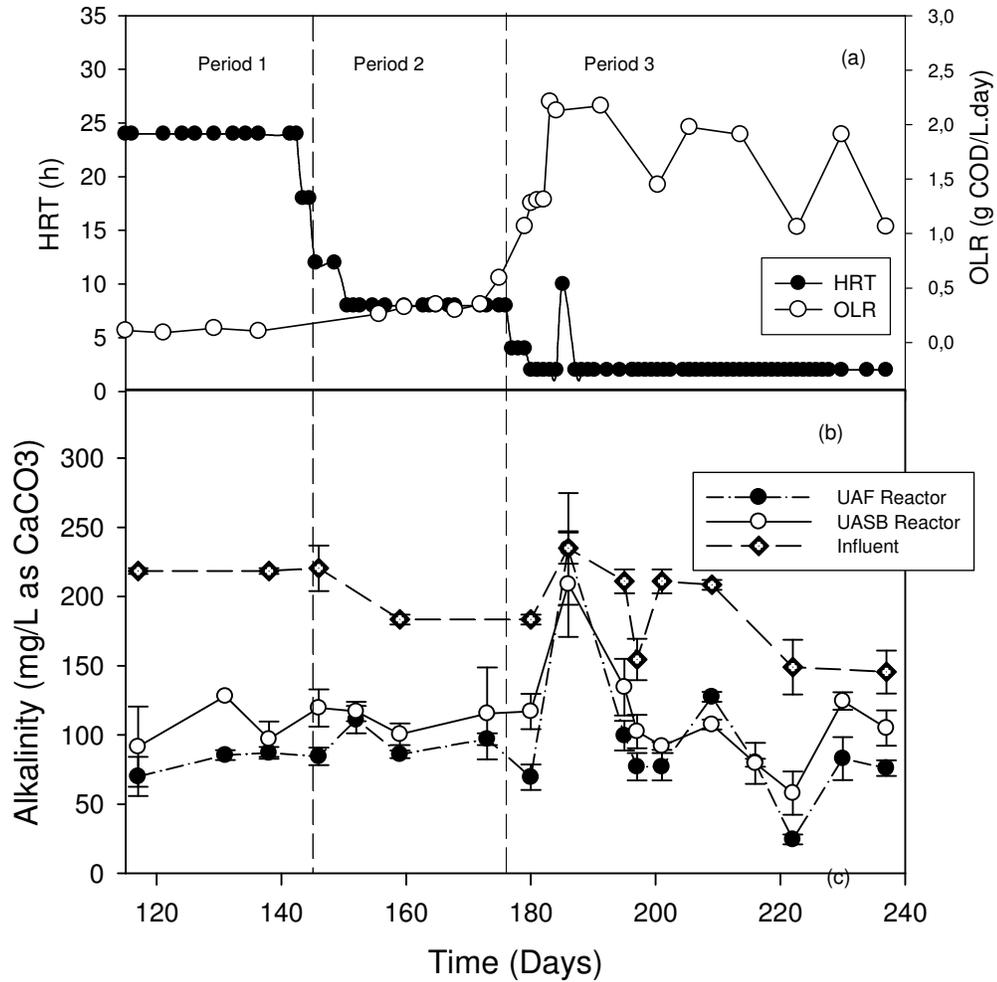


Figure 4.7. Variations of alkalinity of acidogenic UAF and UASB Reactors at applied OLR's and HRT's versus operation time.

#### 4.2.6. tVFA/ sCOD ratio in Set 1

The VFA/sCOD ratios in UAF and UASB reactors at different HRT's are calculated to check if hydrolysis was rate limiting in anaerobic treatment. One

way of defining the degree of success of acidogenesis is the measurement of tVFA/sCOD ratio. This ratio represents the amount of solubilized matter which has been converted to tVFA. High tVFA/sCOD ratios (such as 0.8 or more) indicated that hydrolysis could be rate limiting step (Maharaj, 1999). The studies performed on municipal/industrial wastewater by Maharaj and Elefsiniotis (2001) indicated high tVFA/sCOD ratios such as 0.8 to 1.0. The reactor samples aforementioned were accepted as rate limiting in anaerobic treatment. In the present study, the averages of tVFA/sCOD ratios at studied HRT's are tabulated in Table 4.6 to see if hydrolysis was rate limiting in the anaerobic treatment. The tVFA is calculated as acetic acid equivalents of produced acids. It was observed that the highest tVFA/sCOD ratios are 0.54 and 0.32 for UAF and UASB reactors, respectively in 2 h HRT. The tVFA/sCOD ratios were less than 0.8 and the hydrolysis step can be considered as not rate limiting both in UAF and UASB reactors for anaerobic treatment.

Table 4.6. tVFA/sCOD ratios of UAF and UASB Reactors

HRT (hour)	tVFA/sCOD ratio (dimensionless)	
	UAF Reactor	UASB Reactor
24	0.26	0.18
8	0.31	0.19
2	0.54	0.32

#### 4.2.7. Effect of HRT on the Acidification Degree (Set 1)

The degree of acidification was used to compare the acidification efficiencies of the reactors as an important parameter in the present study. The degree of acidification was defined as the ratio of the COD equivalent of acidification products (VFA) in the wastewater to the total COD (Dinopoulou et al., 1988; Yu and Fang, 2000, Fang et al., 2001). The following formula was used to express the degree of acidification in the present study:

Degree of acidification (%)=COD-VFA/ tCOD (dimensionless)...Equation No 1.

Whereas;

COD-VFA= COD equivalent of VFA (mg/L),

tCOD= Influent total COD (mg/L).

The COD equivalent of each volatile acids were taken as follows for the conversion of related VFA to VFA-COD: acetic acid, 1.066; propionic acid, 1.512; butyric acid, 1.816; valeric acid, 2.036; caproic acid, 2.204 (Demirel and Yenigün, 2004). Table 4.7 shows the acidification degree based on acetic acid in UAF and UASB reactors at different HRTs.

Table 4.7. Degree of acidification of UAF and UASB Reactors

HRT (hour)	Degree of acidification (%)= COD-VFA/tCOD (dimensionless)	
	UAF Reactor	UASB Reactor
24	13	8
8	19	11
2	31	18

The degree of acidification is calculated as 13%, 19% and 31% for HRTs of 24 h, 8 h and 2 h, respectively, in UAF Reactor. The calculated acidification degrees were 8%, 11% and 18% for HRTs of 24h, 8 h and 2 h respectively, in UASB Reactor. The degree of acidification increased with the decrease in HRT in both of the reactors. Degree of acidification is an important parameter to choose the better conditions for acidification phase. It can be concluded that UAF reactor worked better (based on acidification degree) in Period 3 compared to UASB Reactor in Period 3 at a HRT of 2 h. The tVFA produced in UAF reactor is higher than in UASB reactor.

Acidification is strongly dependent on the complexity of the pollutants in wastewater. Cohen et al. (1979) and Zoetemeyer et al. (1982) investigated that a waste containing over 70% of glucose, starch, and other easily degradable carbohydrates and they found that the acidification was possible at HRTs less than 12 h. Only 30-60% of beef extract could be acidified at 6-17 h of HRT (Dinopoulou et al. 1988) and 40% of gelatin at 5 h (Breure et al. 1984). The optimum HRT is experimented as 12 h for simulated dairy wastewater with an acidification of 54.3 % (Fang and Yu, 2001). The acidification efficiency was 23 % in a fish meal factory at an HRT of 24 h (Guerrero et al., 1999).

In the present study, in low strength wastewater treatment (domestic wastewater), it was observed 31% acidification at an HRT of 2 h in UAF reactor in Period 3. It is not possible to compare the acidification degree of the present study since there is not any acidification study performed with domestic wastewater. The degree of acidification resulted as 31% in UAF reactor at 2 h HRT. This range of acidification shows similarities with different types of wastes as aforementioned. In the present study the highest acidification degree was obtained as 31% in the UAF reactor at 2 h HRT compared to the UASB reactor and the other studied

HRTs. Therefore it can be concluded that the UAF reactor at 2 h HRT exhibited an optimum acidification degree for domestic wastewater in the present study.

#### **4.2.8. Effects of HRT on VSS and TSS concentrations and TSS reductions**

The anaerobic degradation process could be separated into two main phases as acid fermentation and methanification. Acid fermentation phase progresses with hydrolysis and solubilization of substrate, because the particular (inert, refractory) organic matter undergoes liquefaction by the extracellular enzymes before being taken up by the microorganisms. The extent of organic substrate solubilization can be viewed from the destruction of the suspended solids reduction (Banarjee et al., 1999).

The influent, effluent concentrations and removals for TSS of UAF and UASB Reactors under steady state conditions are given in Appendix C Table C1. In this table the HRTs of 24 h, 8 h and 2 h show the Period 1, 2 Period 3, respectively in Set 1. The influent VSS and TSS concentrations were between 100 mg/L and 253 mg/L for VSS and 138 mg/L and 367 mg/L for TSS. The average TSS reduction percentages are calculated as 95% and 96% for UAF and UASB Reactors, respectively, in Period 1 (See Appendix C Table C1 and Figure 4.8).

It was not observed any change in the reduction of TSS and VSS values by decreasing the HRT from 24 h to 8 h (Figure 4.8). Maharaj et al (2001) stated that organic matter must first be hydrolyzed or liquified before being assimilated by bacteria, resulting in a reduction in volatile suspended solids (VSS) concentration. The difference between VSS reductions experienced at HRT's of 24 h and 8 h was not significant. This indicated that percent VSS reduction is independent of HRT

within the range of 24 h and 8 h in the UAF and UASB reactors, respectively. The statistical analysis performed whether HRT decrease from 24 h to 8 h affected the VSS reductions for UAF and UASB reactors (For the UAF reactor,  $F(1,4) = 1.298$ ,  $p > 0.05$ , non-significant; for the UASB reactor,  $F(1,4) = 0.153$ ,  $p > 0.05$ , non significant (Appendix, Tables B16- B17).

The percent TSS removal values were similar to those obtained for VSS throughout Periods 1 and 2. The effluent VSS concentration ranged between 0 and 18 mg/L with an average of 6 mg/l both UAF reactor and UASB reactors. The effluent VSS concentrations were similar for UASB and UAF reactor for Period 1 and 2.

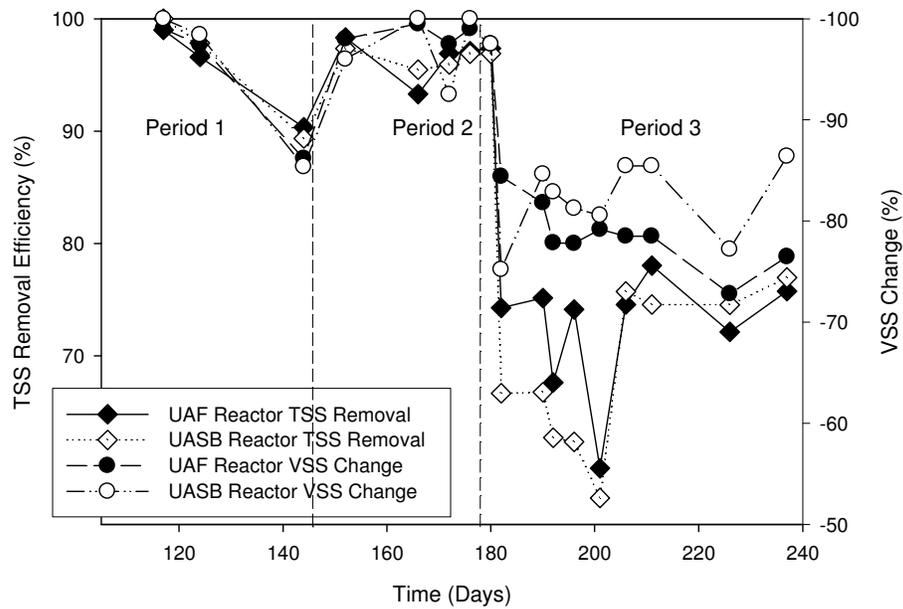


Figure 4.8. Effect of HRT on VSS and TSS Removal in UAF and UASB Reactor.

The average removal were obtained as 72% for TSS in the UAF reactor, and 70% for TSS in the UASB Reactor and average VSS reductions were 77% in the UAF reactor and 83% in the UASB reactor in Period 3. The multiple regression statistics were performed to check whether there was a correlation between VSS reductions, sCOD reductions, tCOD removal and tVFA production in UAF reactor. The multiple regression analysis showed that there was a strong correlation between VSS reductions, sCOD reductions, tCOD removals and tVFA production ( $R^2=0.96$ , adjusted  $R^2=0.93$ , Table B 23, Appendix B). The ANOVA test statistics indicated that the differences between VSS reductions, sCOD reductions and tCOD and tVFA production are significant ( $F(5,6)=29.45$ ,  $p=0.001$ , Table B 23, Appendix B). By decreasing HRT from 8 h to 2 h, the VSS reductions decreased with decreasing of sCOD reductions and tCOD removals while the VFA production increased in the UAF reactor. Since the influent domestic wastewater was inherently acidified and pre hydrolysed, the effluent VFA concentrations increased. The produced VFA was not sufficiently converted to methane. The methane percentage of the total gas was recorded as 17% in UAF reactor.

The multiple regression analysis was performed to check whether a correlation between decrease in VSS reductions and TSS removals sCOD reductions, tCOD removals and tVFA production with the decrease in HRT from 8 h to 2 h in UASB reactor. It was observed that that there was a strong linear correlation between VSS reductions, TSS removals, sCOD reductions, tCOD removals and tVFA production ( $R^2=0.997$ , adjusted  $R^2=0.871$ , Table B 24, Appendix B). The differences were significant between VSS reductions and tVFA production ( $p=0.009$ ) in the UASB reactor. The results showed that the VSS reductions, TSS removals, sCOD reductions and tCOD removals decreased with the decrease in

HRT from 8 h to 2 h. However, VFA production increased as VSS reductions decreased with the HRT decrease from 8 h to 2 h. Banerjee et al (1999) obtained 34-77% VSS reductions in the acidogenesis of the potato-processing wastewater. The VSS reductions of the present study at 2 h HRT were compatible with the results in the literature. The TSS removals were about the same in UAF and UASB reactors but the VSS reductions were 6% higher in UASB reactor in Period 3. The Anova statistics showed that there is a significant difference between VSS reductions in UAF and UASB reactors at Period 3 ( $F(1, 14)=13.33$ ,  $p=0.003<0.05$ ). The VSS reductions in UASB reactor were significantly higher than in UAF reactor. The reason of high VSS reduction in the effluent of UASB reactor can be explained with the methanogenic step which is better provided in UASB reactor since the sCOD reduction and methane percentage are 48% and 52% in UASB reactor while the same parameters are 11% and 17% in UAF reactor, respectively (see sections 4.2.1 and 4.2.10). In this phase, methanogenic archaea converts methane since the sludge (VSS) generated is converted to gas products (to the VFA) (Speece, 1996).

When raw domestic wastewater was directly fed to the anaerobic reactors, a drop in biomass methanogenic activity and biomass floating phenomena appeared, and the process became unstable and inefficient (Uemura and Harada, 2000). These problems were minimized when presettled domestic sewage (Elmitwalli et al., 2002) is used as influent, and also in the cases of total or partially acidified wastewater or mainly soluble wastewater (Rebac et al., 1999). Therefore, several authors indicate that removal of SS is needed prior to the anaerobic treatment in anaerobic reactors (Zeeman and Lettinga, 1999; Elmitwalli et al., 2002). In the anaerobic treatment of domestic wastewater, generally the removal of suspended solids occurred with hydrolysis and acidification in the acidogenic phase (Alvarez, 2008).

In this study an inherent acidification and hydrolysis occurred in the influent wastewater before feeding the UAF reactor. Since this reactor contains a partial methanogenic activity, the VSS concentrations decreased through acidification. Reductions of VSS is resulted in an improved removal of dissolved and colloidal solids in the methanogenesis step.

High amounts of TSS removals obtained in UAF reactor (72%) and UASB reactor (70%) showed that most of suspended solids removed by hydrolysis and partly methanified the TSS in those reactors. The influent domestic wastewater was mostly hydrolyzed (49% in winter time) as explained in Section 4.1.

The VSS reductions was higher in the UASB reactor (83%) than the UAF reactor (77%) which proves that methanification was higher in the UASB reactor. Alvarez et al. (2003) studied hydrolysis and acidification of domestic wastewater in a hydrolytic upflow sludge bed reactor in 2.9 h HRT. Alvarez et al. (2003) presented removals as 81-88% for suspended solids and 46-59% for tCOD and the VFA fraction of tCOD with respect to effluent tCOD ranged from 27% to 43%. The results of the present study was compatible with the findings of that study.

#### **4.2.9. Effects of HRT on Total Volatile Solids (TVS) and Total Solids (TS) reduction in UAF and UASB reactor**

Total Volatile Solids reduction is another important parameter, which also shows if hydrolysis step prior to acidification process is properly completed. In acid fermentation phase, higher TVS reduction means better hydrolysis. It was observed that the average TVS reductions were 68 and 74% for UAF and UASB

reactors, respectively (See Figure 4.9). In this study, these reductions are typical for high rate anaerobic reactors with HRT's of 24 hour Chen and Shiyu (1996). The same researchers found 70% and 71% TVS reductions were observed in UAF and UASB reactors, respectively.

The average TVS values were measured as 222 mg/L and 78 mg/L for influent and effluent wastewaters in UAF Reactor and average TVS reduction was 63 % in Period 2 at a HRT of 8 h. The decrease in TVS for UAF Reactor was only 5% with reducing the HRT from 24 h to 8 h. This can be considered as an insignificant reduction in hydrolysis of wastewater. The results obtained for TVS values for the UASB Reactor was very similar to the UAF Reactor with average effluent values of 77 mg/L and a reduction of 64 %. In Period 3, the volatile solids of the influent wastewater averaged 26% of total solids at a HRT of 2 h. The average TS reductions were about the same for UAF and UASB reactors (16%). The Anova test statistics indicated that the difference between TS reductions in UAF and UASB reactors was not-significant in Period 3 ( $F(1,18)=0.007$ ,  $p=0.937$ , Table B.10, Appendix B). The TS concentrations in UAF and UASB reactors were not statistically different in Period 3. The TVS reductions were at most 48% with an average of 30% for UAF reactor and ranged from 20-75% with an average of 47% for UASB reactor. The TVS reduction is higher in UASB reactor compared to UAF reactor. The Anova test statistics indicated that there is a significant difference between TVS reductions in UAF and UASB reactors in Period 3 ( $F(1, 18)=4.85$ ,  $p=0.041$ , Table B.11, Appendix B). In Period 3, the TVS reductions were significantly different in UAF and UASB reactors. The TVS reduction in UASB reactor is high compared to UAF reactor. The total solids content of domestic wastewater was about 1 g/L and it can be categorized as low solid content as reported by Tchobanoglous and Burton (1991). Because of low

solid content of influent domestic wastewater and a certain ratio of solids are volatile in the influent (30%), the hydrolysis step is not rate limiting.

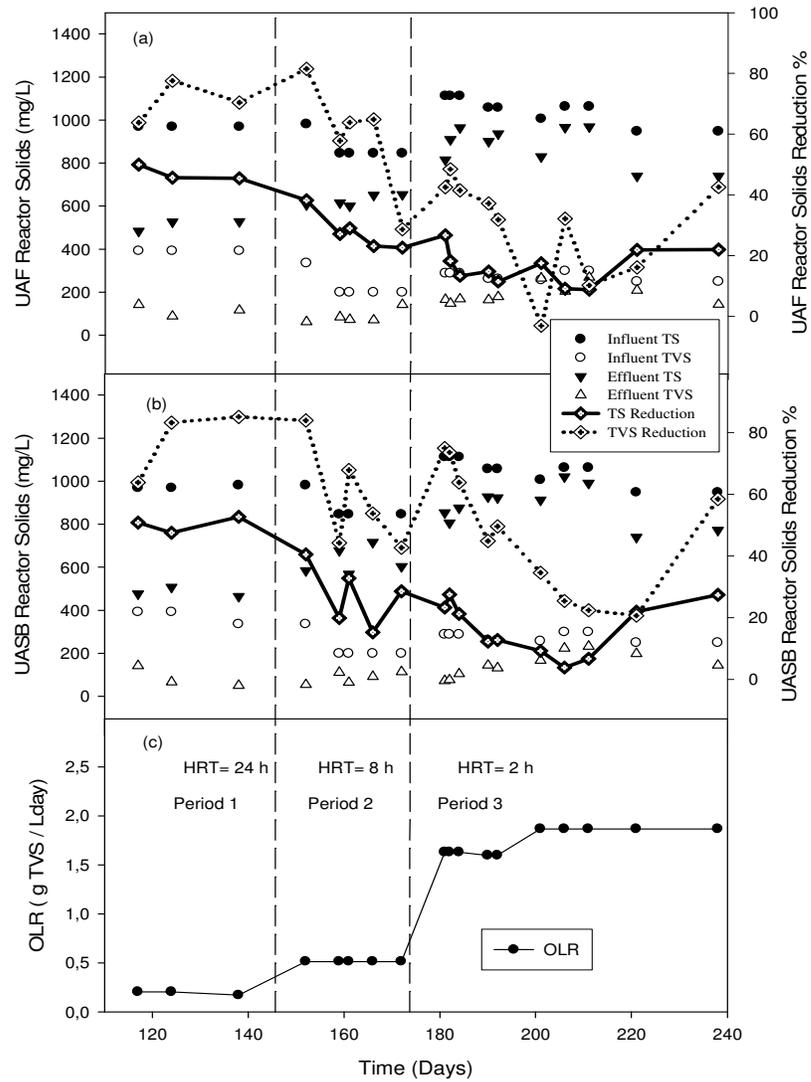


Figure 4.9. Influent, effluent concentration and reduction percentages for TS and TVS in UAF Reactor (a), UASB Reactor (b) and (c) indicates the variation of OLR with time.

#### 4.2.10. Gas Production and Variations of Gas Composition

The gas samples have not been collected and analyzed properly before day 201. Gas production and analysis were performed in Period 3 (HRT= 2 h). The gas production results between days 201 and 237 are shown in Figure 4.10. After day 207, sodium thioglucollate addition was done to provide the anaerobic conditions in the influent wastewater since the dissolved oxygen was 3 mg/L (see subsection 4.2.4). Gas analysis revealed that the biogas contained nitrogen, methane and carbon dioxide. On day 197, the reactors were flushed with N<sub>2</sub> gas and influent domestic wastewater was also flushed with N<sub>2</sub> gas to ensure the anaerobic conditions during days 197 and 207. After day 207, sodium thioglucollate addition was done to influent wastewater to provide anaerobic conditions as previously mentioned (see Section 4.2.3).

Daily gas compositions and productions are given in Figure 4.10 for the days that gas analyses were performed in Period 3 (HRT=2 h). Daily gas productions indicated variations. The variations in gas production rates of the UAF were higher than the UASB and ranged from 240 mL to 700 mL in UAF reactor. Gas pockets appeared in the filter, they became larger and reached a limiting size when they would break loose and rise through the filter. The daily gas composition exhibited great variations in UAF reactor (See Figure 4.10) because of irregular gas pockets in the filter. Kobayashi et al. (1983) explained that actual gas production would occur at a much more even rate than occur in “spurts”. It also appeared gas pockets inside the granules of the UASB reactor while the gas pockets were raised more uniformly through the granules. It was observed that the daily gas collection of the UASB reactor was more uniform than the UAF reactor.

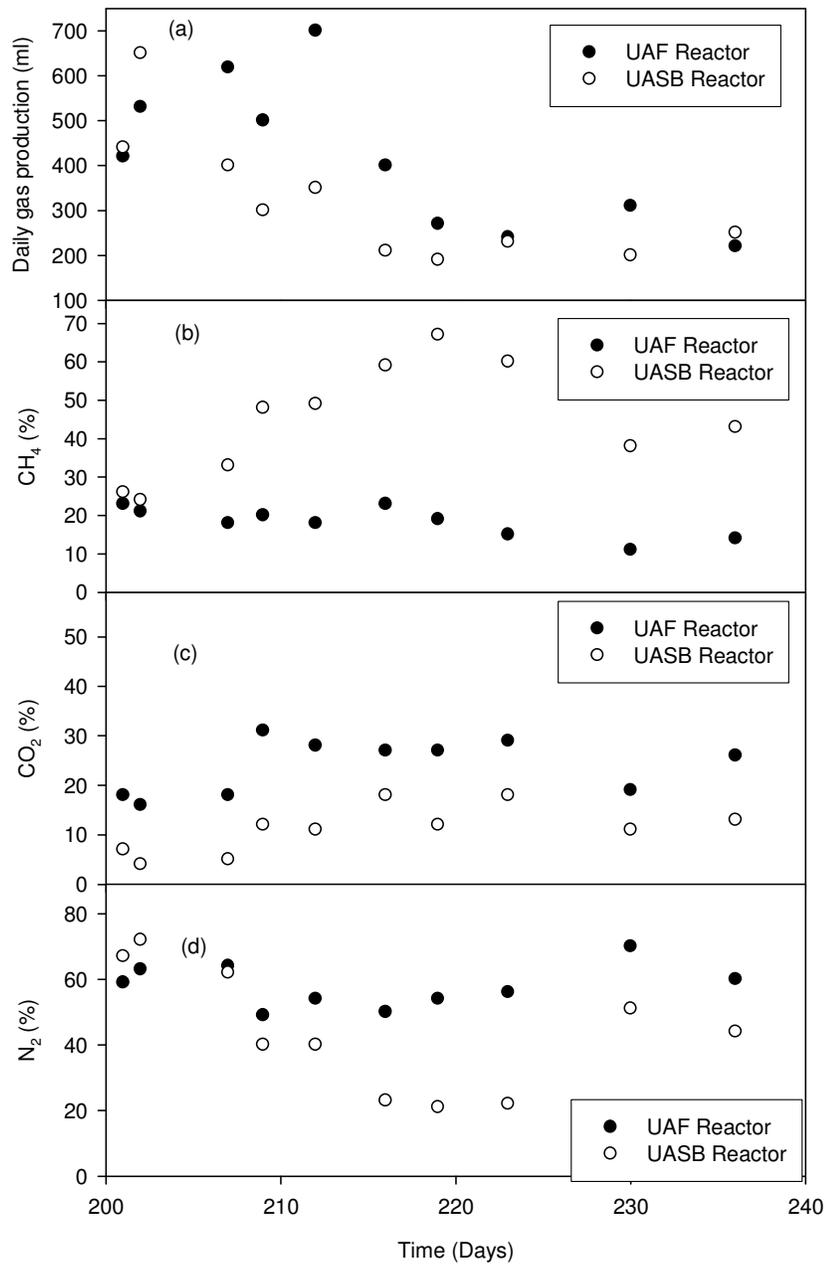


Figure 4.10. Daily Gas Production (a), Percentage of Methane (b), Percentage of Carbondioxide (c) and Percentage of Nitrogen Gas (d) in Period 3 (2 h HRT).

It was observed higher gas quantities with higher percentages of nitrogen gas production during days 197-207 than days 207-237. This can be explained by the saturated nitrogen gas in influent wastewater. This converted to nitrogen gas in that period. The decrease in the percentage of nitrogen gas content was observed when the influent wastewater was not saturated with nitrogen gas (10% and 37% in UAF and UASB reactors), respectively. Without taking the saturation of nitrogen gas in the influent in to consideration, the data of the days 207-237 are used to compare the reactor performances.

During days 207 and 215, the daily gas production in UAF reactor was recorded as 600 mL. The methane percentage of this reactor varied between 15-20% while the CO<sub>2</sub> content and N<sub>2</sub> content of the total gas were between 18% and 25% and 50 and 60%, respectively. The effluent alkalinity of UAF reactor was measured as 108 mg/L as CaCO<sub>3</sub>. The average total alkalinity concentration in influent UAF reactor was 209 mg/L as CaCO<sub>3</sub>. The effluent pH and tVFA concentration were recorded as 6.85 and 132 mg/L as Hac. The average initial tVFA in influent wastewater was 20 mg/L as Hac. The result of this study showed that the VFA concentration increased in effluent. The alkalinity consumed by a small amount of methanogens since the methane percentages were between 15-20%. The CO<sub>2</sub> percent of total gas was recorded at a high level. Similarly, the N<sub>2</sub> percentage of total gas was high. This shows that UAF reactor exhibits acidogenic properties. The consumption of alkalinity indicates that a small amount of VFA was converted to small amount of methane gas.

On day between 230 and 237, the daily gas production reduced to 200-300 mL in UAF reactor at 2 h HRT in the UAF reactor. The methane percentage of the UAF reactor between aforementioned days was recorded as 10-16%. The CO<sub>2</sub> and N<sub>2</sub> content of the total gas were recorded between 20-28% and 60-80%, respectively.

It was observed that daily gas production decreased. Furthermore, the CO<sub>2</sub> percentage of daily gas remained similar as with days 207 and 215. Conversely, the methane percentage decreased. The influent and effluent alkalinities were 145 and 80 mg/L, respectively. This shows that alkalinity was used by part of methanogens since the methane percentage was above 9-10% and CO<sub>2</sub> content is high. The methane percentage on days 230 and 237 is lower than days between 207 and 215. The influent and effluent tVFA concentrations were 20 and 140 mg/L as Hac, respectively. This showed the partly acidification of UAF reactor at a HRT of 2 h.

In the UASB reactor, the daily gas production was recorded as 280 and 350 mL for days 207 and 215. The methane percentage of the UASB reactor varied between 40 and 50% while the CO<sub>2</sub> and N<sub>2</sub> content of the total gas were between 9-12% and 40-65%, respectively. It was observed that the methane percentage is high and CO<sub>2</sub> is low compared to the UAF reactor. The alkalinity of UASB reactor was measured as 108 mg/L in the effluent of UASB reactor while in the influent of UASB reactor was 209 mg/L. The influent and effluent tVFA concentrations were 20 and 93 mg/L as Hac, respectively. The high methane percentage in UASB reactor could be attributed to the conversion of VFA to methane gas while the alkalinity was consumed by methanogens at a pH of 7.0.

On days between 230 and 237, the daily gas production was low (225 mL). The methane percentage was between 40 and 60% while the CO<sub>2</sub> percentage was low (11%). The tVFA concentration in influent and effluent of UASB reactor were 20 and 93 mg/L as Hac. The alkalinity decreased from 145 to 115 mg/L as CaCO<sub>3</sub> in the effluent of UASB reactor. The recorded methane percentage of total gas was 40-60%. The decrease in alkalinity and less production of tVFA compared to days between 207 and 215 indicates that the methanogens converted the VFA to

methane by using the alkalinity at a pH of 7.04. This data showed that the UASB reactor is operated under methanogenic conditions during Period 3.

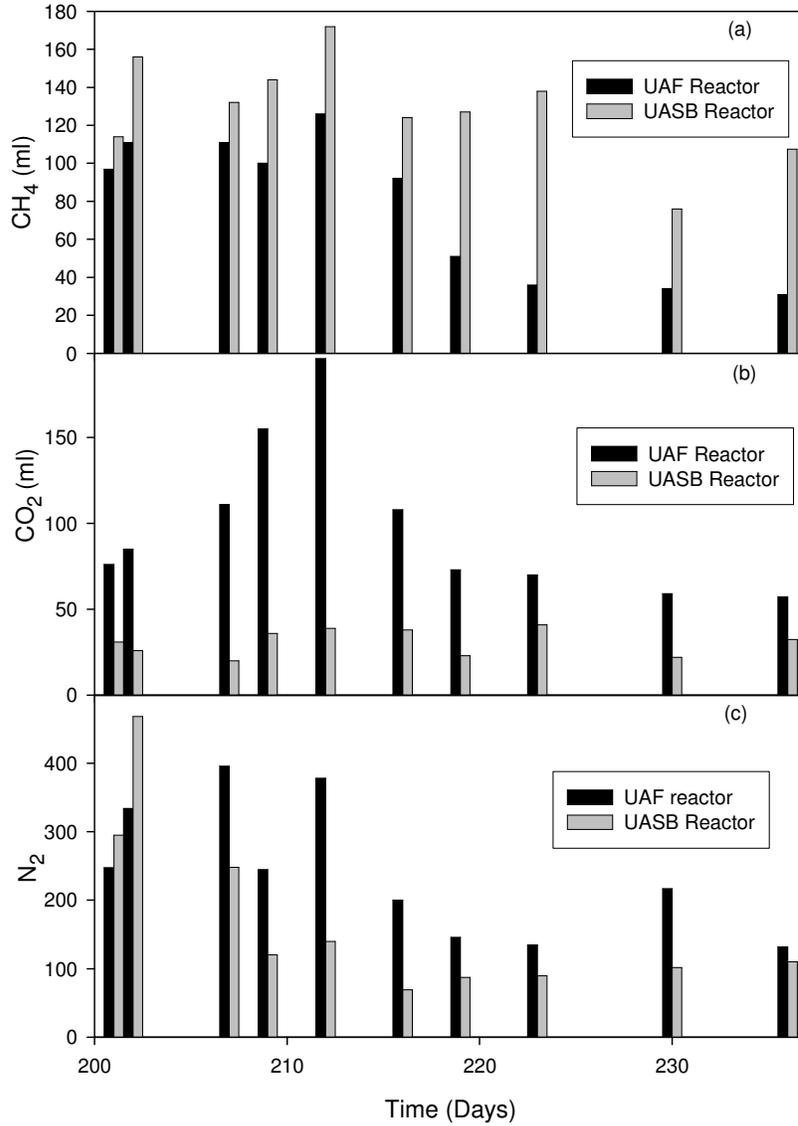


Figure 4.11. Effect of HRT on CH<sub>4</sub>, CO<sub>2</sub> and N<sub>2</sub> production in UAF and UASB reactors at 2 h HRT.

The percentage of methane content was ranged between 11% and 23% with an average of 17% and ranged between 43 and 67% with an average of 52% in UAF and UASB reactors, respectively at Period 3. The high methane percentage of UASB reactor compared to UAF reactor, can be explained with low effluent tVFA concentration, low effluent sCOD concentration and high pH values (6.80-7.42). The sCOD and tVFA concentration in the effluent of UAF reactor was higher than that UASB reactor while the pH was lower in UAF reactor than that UASB reactor. The carbon dioxide percentage of UAF was higher than UASB as opposite to methane fractions. The percentage of carbon dioxide ranged between 19% and 33% with an average of 27% and ranged between 11% and 18% with an average of 14% in UAF and UASB Reactors, respectively.

The daily average quantity of methane collected was observed as 67 mL and 127 mL for UAF and UASB reactors, respectively at a HRT of 2 h at Period 3 (Figure 4.11). There is a difference between the expected methane and the experimentally measured methane. This can be explained by the soluble methane in the effluent. Furthermore, losses in the quantity of methane were greater than the gas collected in many cases as mentioned by Kobayashi et al. (1983). The average daily collected carbon dioxide was determined as 103 mL and 33 mL for UAF and UASB reactors, respectively. Carbon dioxide is one of the by-products of acidification. By considering higher carbon dioxide content and lower methane content in the total gas in UAF reactor, it can be concluded that UAF reactor has more acidifying conditions than that UASB reactor. In Period 3, the other parameters such as pH, tVFA, sCOD and VSS reductions supported that acidification was better provided in UAF reactor than that UASB reactor: The average pH values were higher in UAF reactor (6.83) than that UASB reactor (6.93). The average tVFA concentration was higher in UAF reactor (145 mg/L)

than UASB reactor (87 mg/L) and the sCOD reduction was higher in UASB reactor (48%) than that UAF reactor (11%).

#### **4.2.11. Effect of HRT on the NH<sub>4</sub>-N, Proteins, PO<sub>4</sub>-P and P variations in UAF and UASB Reactors**

The influent, effluent concentrations for NH<sub>4</sub>-N are shown in Figure 4.12. The effluent NH<sub>4</sub>-N was mostly higher than that influent values in UAF and UASB reactors because of increases in NH<sub>4</sub>-N concentration. It was not observed a constant change in NH<sub>4</sub>-N releases with the change in HRT. The reason of the increase of NH<sub>4</sub>-N can be explained by the anaerobic bioconversion of proteins to amino acids and then to ammonia as reported by Demirer and Chen (2005); Martin et al. (2003); Cheng and Liu, (2002), Kobayashi et al. (1983).

Degradation of proteins (deamination) under anaerobic conditions resulted in generation of ammonia as mentioned above. It can be summarized that the protein degradations was higher at longer HRTs since the ammonia nitrogen production was higher. In the present study, it was observed an increase in effluent ammonia concentrations. These results are consistent with the results found in literature (Demirer and Chen, 2005).

The influent protein concentrations were between 120 mg/ L and 140 mg/ L during Set 1 and the effluent protein values were increased about from 30 mg/L to 60 mg/L with the decrease in HRT (see Figure 4.13). The average protein reduction percentages versus HRT are shown in Figure 4.14. It is observed that the protein removal efficiency decreased from 72% to 69% a decrease of 72% to

69% and 48% with the decrease in HRT from 24 h to 8 h and 2 h, respectively, in the UAF reactor.

The reduction percentages in proteins were observed as 75%, 67% and 54% at HRT's of 24 h, 8 h and 2 h, respectively in the UASB reactor. The reduction in percent protein removal was higher at HRTs of 8 h and 24 h. However the protein removal efficiencies were lower at HRT of 2 h in both reactors.

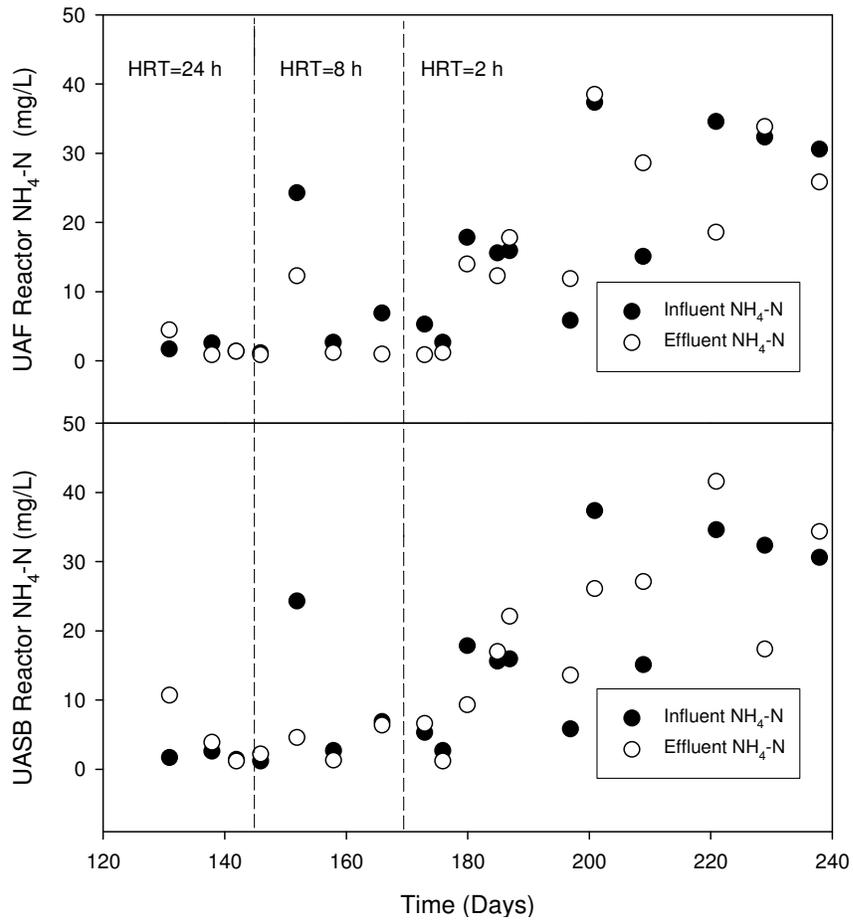


Figure 4.12. Influent and effluent concentrations of NH<sub>4</sub>-N in UAF Reactor and UASB Reactor.

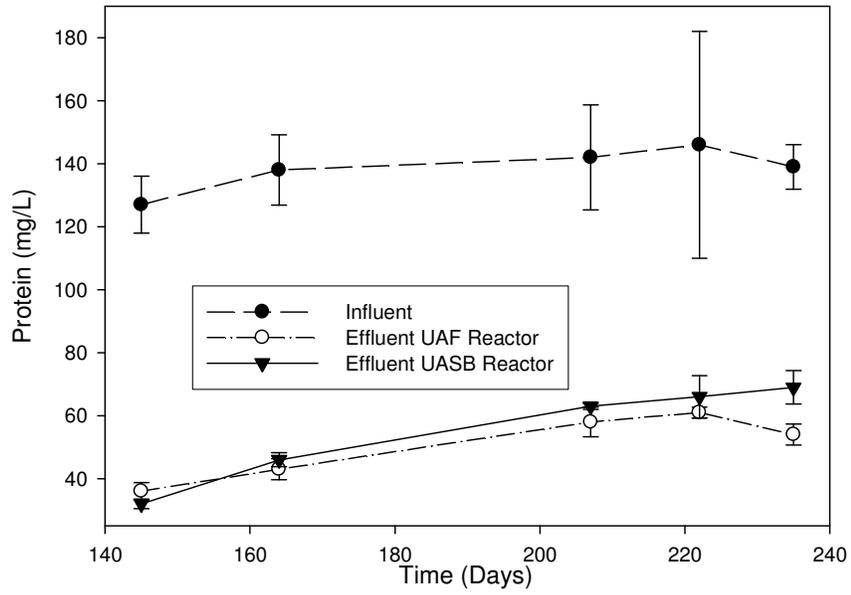


Figure 4.13. Proteins in the influent and effluent UAF and UASB reactors.

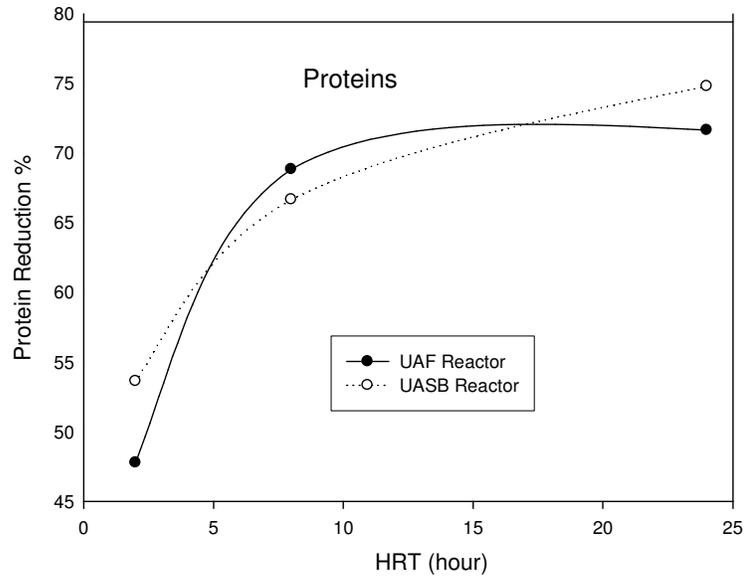


Figure 4.14. Reduction of proteins in UAF and UASB reactors.

Influent and effluent phosphate and phosphorous concentrations for UAF and UASB reactors are displayed in Figure 4.15. The phosphate reductions were averaged as 22%, 14% and 11% in 24 h, 8 h and 2 h HRT respectively, for UAF reactor. The averaged phosphate reductions were 14%, 6% and 6% in 24 h, 8 h and 2 h HRT, respectively for UASB reactor. The phosphate reductions were higher in UAF reactor and it decreased about to the half of their values with decrease in HRT from 24 h to 8 h. The phosphate reduction became constant with the decrease in HRT from 8 h to 2 h. The influent and effluent phosphorous were determined only for 2 h HRT and average reductions determined as 13% and 16% for UAF and UASB reactors, respectively. The low average reductions observed for phosphate and phosphorous are expected since anaerobic digesters are known to reduce negligible amounts of nutrients as mentioned by Lusk (1998) and Demirer and Chen (2005). The UAF and UASB reactors at these HRTs (2 h-24 h) could not be proposed for the anaerobic treatment of phosphate and phosphorous in domestic wastewater treatment.

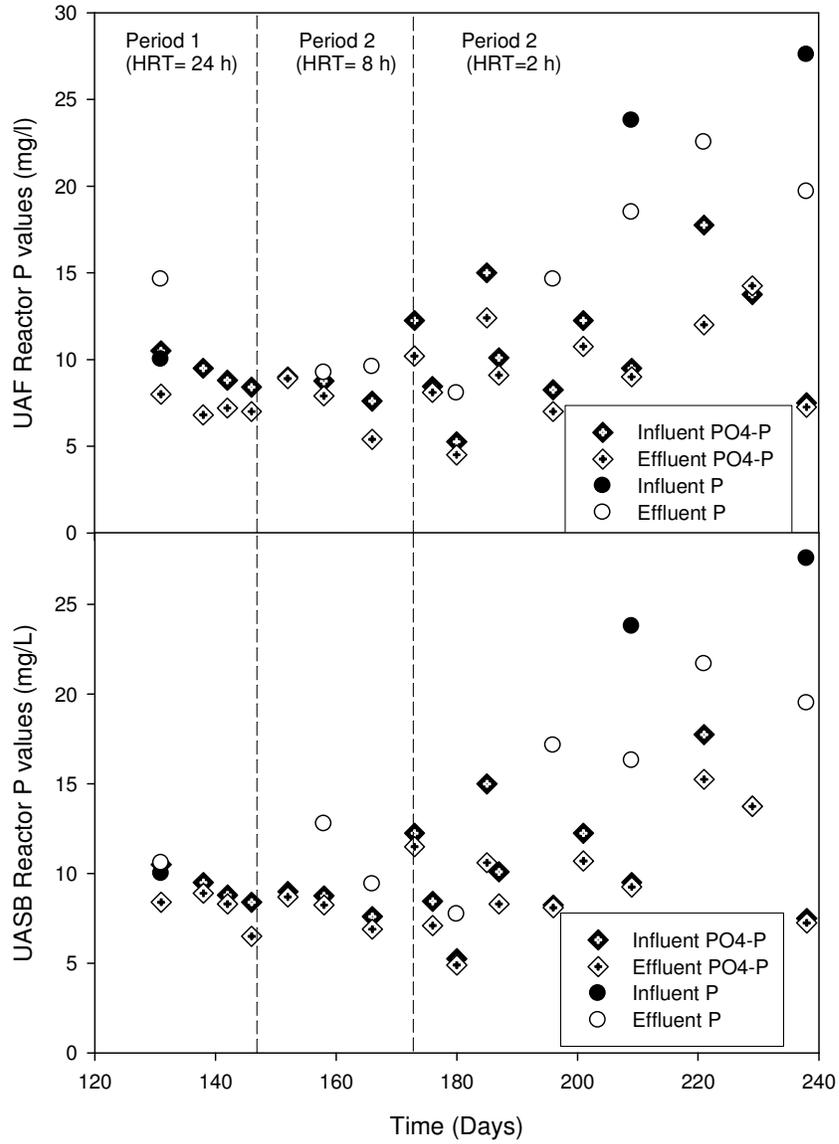


Figure 4.15. Influent and effluent phosphate and phosphorous concentrations versus operational period.

#### 4.2.12. Results of Acidification in UAF and UASB Reactors

The main findings of the acidogenic reactor studies were summarized below:

- The pH of UAF and UASB reactors decreased with the decrease in HRT from 24 h to 8 h and 2 h in the treatment of domestic wastewater. The lowest pH measurements were obtained around 6.5- 7.01 in UAF and 6.8- 7.1 in UASB at a 2 h HRT. By decreasing the HRT from 24 h to 2 h, it was obtained a decrease in average pH from 7.77 to 6.83 in UAF reactor. The pH of reactors decreased with the production of volatile acids. In UAF reactor, the average VFA value was 55 mg/L (as Hac) in Period 1 (HRT= 24 h) and increased to 145 mg/L (as Hac) in Period 3 (HRT=2 h). The reason of this could be explained by high degree of VFA production with low degree of methanification in UAF reactor compared to UASB reactor in Period 3. In the UASB reactor, the average VFA concentration was 37 mg/L (as Hac) in Period 1 and increased to 87 mg/L (as Hac) in Period 3. The tVFA concentration was 1.7 times higher in UAF reactor than that of UASB reactor in Period 3.
- The tCOD treatment efficiencies were 36-71% and 54-88% for UAF and UASB reactors, respectively in Periods 1 and 2. The sCOD decreases were 47-70% and 60-77% for UAF and UASB reactors, respectively in Periods 1 and 2. The average tCOD removals decreased to 45% and 56% in UAF reactor and UASB reactor, respectively in Period 3 while sCOD average decreases were 11% and 48% for UAF reactors and UASB reactors, respectively. It was observed that the sCOD concentrations in effluent are higher than influent sCOD or very low decreases in sCOD in UAF reactor was observed in Period 3. This can be explained with the hydrolysis of

slowly degradable organics in the reactor since the content of organic compounds in domestic wastewater consist from the slowly degradable organics, soluble degradable organics and from the inert organics which is not utilized by the bacteria (Orhon et al., 1997). The soluble organic compounds utilized by the microorganisms while the slowly degradable organics are degraded to soluble organics by means of hydrolytic organisms. Then these soluble compounds are further degraded by acidogenic bacteria in the acidogenesis phase. By considering the effluent tVFA/sCOD ratios (0.54 in UAF and 0.32 in UASB reactors) it was observed that the hydrolysis step was not rate limiting in both reactors in Period 3.

- Lower pH values, higher tVFA effluent values and lower tCOD treatment efficiencies in UAF reactor were obtained compared to UASB reactor. This can be considered as an indication of acidification step with high tVFA concentration in UAF reactor than that UASB Reactor in Period 3.
- The TSS removals in effluent were about the same in UAF and UASB reactors but the VSS reductions were 6% higher (statistically significant) in UASB reactor than that UAF reactor in Period 3. The reason of high VSS reduction in the UASB reactor can be explained by the methanogenic step which was better provided in UASB reactor since the main part of the sludge in this reactor converted to methane successively (average methane percentage of biogas was 17% in UAF reactor and 52% in UASB reactor).
- The methane producing bacteria consume the volatile acids and generated alkalinity. In Period 3, the average alkalinity concentrations in effluents were 106 and 120 mg/L (as CaCO<sub>3</sub>) for UAF and UASB reactors,

respectively. Although the influent alkalinities in both reactors were higher than the effluent alkalinities, the consumption of volatile acids was lower in UAF reactor, resulting with lower alkalinities and low methane gas production. The VFA produced could not be converted to methane, immediately since the buffering capacity (alkalinity) is low in UAF reactor. The tVFA composed from 12% acetic acid, 29% propionic acid and 52% butyric acid in UAF at 2 h HRT. All remaining acid VFAs were 7% in UAF reactor at that HRT. The composition of tVFA in UASB reactor was 51% acetic acid, 22% propionic acid and 19% butyric acid. The composition of other acid types was 8% in the UASB reactor in 2 h HRT.

- Methane, besides carbon dioxide, hydrogen, hydrogen sulfur and nitrogen, are the main product of methanification and carbon dioxide is one of the by-products of acidification. It was obtained higher carbon dioxide content and lower methane content in UAF reactor compared to UASB reactor in Period 3 (HRT=2 h). Therefore it can be concluded that UAF reactor had more acidifying conditions than UASB reactor.
- In low strength wastewater such as domestic wastewater treatment, it was obtained an average acidification degree of 31% at an HRT of 2 h in UAF reactor in Period 3. This value was the highest average acidification degree when compared with the UASB reactor and with the other studied HRTs (average acidification degree was 18% in UASB reactor at an HRT of 2 h; average acidification degree was observed as 19% in UAF reactor at an HRT of 8 h). The average degree of acidification showed similarities with various types of wastes reported by Guerrero et al.(1999) for fish meal factory wastewater and Fang and Yu (2001) for dairy wastewater. In their

studies, 23% and 54.3% acidification was observed for fish meal factory wastewater and dairy wastewater, respectively.

As a result of considered parameters given aforementioned, the UAF reactor was selected as the acidification reactor in two-phase (UAF and UASB) reactor system in the treatment of low strength domestic wastewater at a HRT of 2 h.

#### **4.3. Start-up and Operation of Two-Phase UAF and UASB and One-phase UASB Systems in Set 2**

The aim of the second continuous reactor operation was to determine and to compare the performances of two-phase UAF and UASB system and one-phase UASB system. The UAF reactor was selected as the acidification reactor at a HRT of 2 h. The acidification reactor, UAF, was followed by a UASB reactor in two-phase system. In one-phase system, a methanogenic UASB reactor was operated. The experimental set-up and operational conditions are given in Figure 3.2 and Table 3.8 (Chapter 3.4).

The UAF reactor was operated at a HRT of 2 h during the entire study (till the day 309). The UASB reactors were started to operate in July 2, 2007 (on day 237). To steady state conditions for both UASB reactors were reached on day 259. The time elongation for steady state conditions in UASB reactors for about 20 days was mostly because of the lack of flow adjustments, clogging of pipes and malfunctioning of pumps. The HRT's of the UAF and the UASB reactors throughout operation period are given in Figure 4.16. The HRT's were 2 h and 8 h for the UAF and for the 2<sup>nd</sup> Reactor (UASB following UAF) of the two-phase

reactors, respectively at the start-up and at the 1<sup>st</sup> period. The HRT of the one-phase reactor was 10 h for the same operation period.

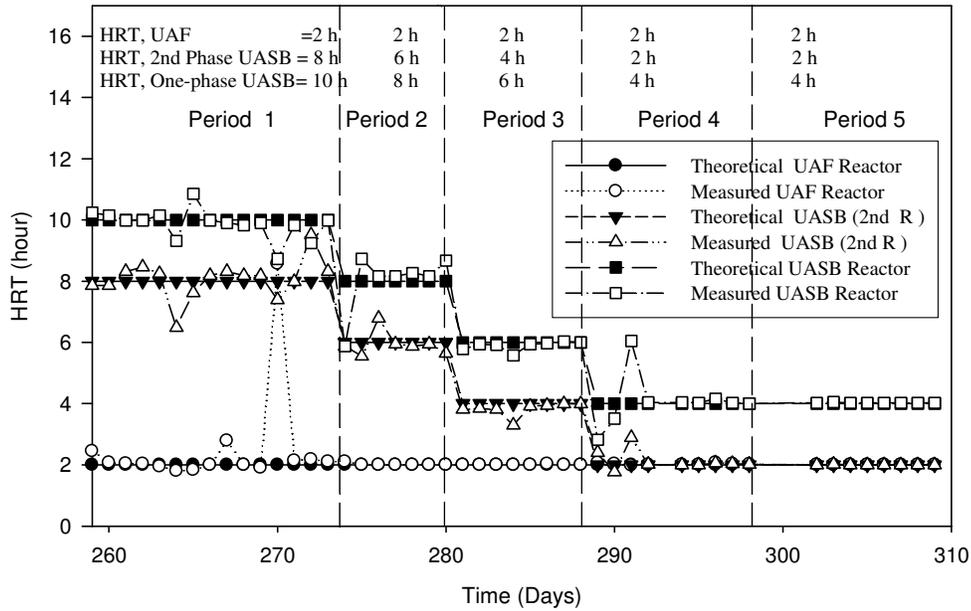


Figure 4.16. Operation of One-phase (UASB) and Two-phase(UAF and UASB) reactor systems.

The HRT of the 2<sup>nd</sup> reactor decreased stepwise from 8 h to 6 h, from 6 h to 4 h and from 4 h to 2 h and the HRT of UASB reactor (one-phase reactor) decreased from 10 h to 8 h, 6 h and 4 h with 7 to 10 days of period of time in each Period. The Period 1 (days 237-273), Period 2 (days 274-280), Period 3 (days 281-288) and Period 4 (days 289-295) were performed in mesophilic temperatures (at 35±2°C) and Period 5 (days 296-308) performed in psychrophilic temperatures (20±2°C).

#### **4.3.1 Effects of HRT/OLR on the Effluent tCOD, sCOD and Removal Efficiencies in Two-Phase UAF and UASB System and One-Phase UASB System**

In the summer time the strength of influent domestic wastewater varied in a large extent. The OLR of the UAF reactor ranged between 0.74 g sCOD/L.d and 3.78 g sCOD/L.d during the studied periods for the acidification reactor in two-phase (UAF and UASB) system. The flow rate is not adjusted, it was kept constant. The influences of HRT-OLR on the variations of sCOD and tCOD concentrations in the influents of domestic wastewater are presented in Figures 4.18-b, 4.19-b and 4.20-b. The influent and effluent sCOD and tCOD values and treatment efficiencies for tCOD are given in Table 4.8. In this Table, the HRTs of the UAF reactor was 2 h for Periods 1-4 at  $35\pm 2^\circ\text{C}$  and 2 h for Period 5 at  $20^\circ\text{C}$ . The HRTs of the 2<sup>nd</sup> reactor were 8 h, 6 h, 4 h and 2 h for Periods 1, 2, 3 and 4, respectively, at  $35\pm 2^\circ\text{C}$ . The HRT and temperature was 2 h and  $20\pm 2^\circ\text{C}$  for Period 5 in the 2<sup>nd</sup> reactor. The HRTs of the UASB reactor were 10 h, 8 h, 6 h and 4 h for Periods 1, 2, 3 and 4, respectively, at  $35\pm 2^\circ\text{C}$ . The HRT and temperature was 4 h and  $20\pm 2^\circ\text{C}$  for Period 5 in the UASB reactor. The influent sCOD and tCOD values ranged between 44-194 mg/L and 184-793 mg/L, respectively, during the experimented periods. The effluent sCOD and tCOD values and treatment efficiencies for tCOD and changes for sCOD were tabulated in Table 4.8.

The UAF reactor influent and effluent values and changes for sCOD and removal efficiencies for tCOD are illustrated in Figures 4.18 and given in Table 4.8. In Periods 1-4 (HRT= 2h, T= $35\pm 2^\circ\text{C}$ ), the average changes varied between +16% and -52% for sCOD. The removal efficiencies varied between 28% and 70% for tCOD. The tCOD removal efficiencies were more than 60% for influent tCOD values higher than 300 mg/L. The tCOD removals were 71%, 63% and 63% for

influent tCOD values of 466, 793 and 332 mg/L during Periods 1, 2 and 4, respectively at  $35\pm 2^\circ\text{C}$ . In Period 3, the influent tCOD was 184 mg/L and the tCOD removal efficiency decreased to 28%. It can be said that for lower tCOD values (less than 200 mg/L), the tCOD removal efficiency decreased from 61% to 28% in UAF reactor at a HRT of 2 h. The tCOD removal efficiencies of the present study showed similarities with the results of Kato et al (1997). As the influent COD concentration decreased below 200 mg/L, the COD removal efficiency decreased less than 37% at 2.6 h HRT in a UASB reactor.

In Period 5, the tCOD removal efficiency decreased from 63% to 44% at an influent tCOD value of about 300 mg/L in the UAF reactor. The reason of such a decrease can be explained with the decrease in temperature from  $35\pm 2^\circ\text{C}$  to  $20\pm 2^\circ\text{C}$ . Temperature decreases from mesophilic range to psychrophilic range resulted decreases in removal efficiencies (about 19%) in tCOD in the UAF reactor.

It was observed increases in effluent sCOD values when the influent tCOD values were relatively high on days 274-280 (Table 4.8, Period 2). On the other hand, the influent sCOD values were very low (Table 4.8, Period 4) in the UAF reactor. The reason of increases in effluent sCOD values can be explained by the high rate conversion of tCOD to sCOD and does not have enough time for the removal of sCOD. The purpose of chosen HRT as 2 hours in the UAF reactor was to provide the phase separation and the acidification in the UAF reactor and the methanification in the 2<sup>nd</sup> reactor.

The sCOD and tCOD variations in the influent and effluent of the 2<sup>nd</sup> phase reactor, the removal efficiencies for the 2<sup>nd</sup> phase UASB reactor and the two-phase system (UAF+ 2<sup>nd</sup> phase UASB reactor) removal efficiencies were

presented in Figure 4.18. The HRT of the 2<sup>nd</sup> phase reactor decreased from 8 h to 4 h but the OLR of the 2<sup>nd</sup> reactor did not increase at a similar rate because of the decreases in the effluent sCOD of the UAF (2<sup>nd</sup> phase reactor placed after the UAF reactor). The OLR of the 2<sup>nd</sup> phase reactor fluctuated from 0.35 g sCOD/L.d to 1.22 g sCOD/L.d. It was observed tCOD removal efficiencies and sCOD reductions of 51-65% and 52-61%, respectively at mesophilic (35±2°C) temperatures in the 2<sup>nd</sup> reactor. Lettinga et al. (1993) reported that tCOD removal efficiencies were 55-75% at a HRT, temperature and influent tCOD values of 5-6 h, 24-26°C and 200- 300 mg/L, respectively. The influent tCOD values of the 2<sup>nd</sup> phase reactor were accepted as the effluent values of the UAF reactor and the calculations were done accordingly. Although a part of tCOD was removed in the UAF reactor, the tCOD removal efficiency results of the 2<sup>nd</sup> phase reactor are compatible with the results given in the above literature.

In the 2<sup>nd</sup> phase reactor, the tCOD treatment efficiency was 8% higher in 20°C than that 35°C (average tCOD treatment efficiency of the 2<sup>nd</sup> phase reactor at mesophilic temperatures and psychrophilic temperatures were 60% and 68%) (Table 4.8). It is important to note that an increase in tCOD treatment efficiency (to 70%) of the UAF reactor resulted a decrease in tCOD treatment efficiency (to 51%) of the 2<sup>nd</sup> phase reactor (Table 4.8, Period 1). On the other hand a decrease in tCOD treatment efficiency (to 44%) of the UAF reactor resulted an increase in tCOD treatment efficiency (to 68%) of the 2<sup>nd</sup> phase reactor (Table 4.8, Period 5). It can be concluded that the 2<sup>nd</sup> phase reactor balanced the removal efficiencies. Higher removal efficiencies (65%) in the 2<sup>nd</sup> phase reactor were obtained when the removal efficiencies were lower (28%) in the UAF reactor. Furthermore, lower tCOD removal efficiencies were observed (51%) in the 2<sup>nd</sup> phase reactor when the removal efficiencies were higher (70%) in the UAF reactor. The effluent tCOD values were between 97 mg/l and 39 mg/L in the 2<sup>nd</sup> phase reactor.

The effects of higher influent tCOD values (273 mg/L) were resulted with relatively higher effluent tCOD values (97 mg/L) in UAF reactor. The lower tCOD influent values (97 mg/L) were resulted relatively lower effluent tCOD values (39 mg/L) in the 2<sup>nd</sup> phase reactor.

Any reduction of sCOD in the 2<sup>nd</sup> phase reactor was not observed with the decrease of the HRT from 8 h (Period 1) to 6 h (Period 2), from 6 h (Period 2) to 4 h (Period 3) and from 4 h (Period 3) to 2 h. The average sCOD reductions of the 2<sup>nd</sup> phase reactor was 55% at 35±2°C. The average sCOD reduction decreased to 44% with the decrease of the ambient temperature to 20±2°C. This increase in the effluent sCOD at psychrophilic temperatures can be explained by the VFA form of some of sCOD and methanogenesis step was not enhanced as it was in mesophilic temperatures.

The sCOD changes and tCOD treatment efficiencies of the two-phase UAF and UASB system are shown with red lines in Figure 4.18. The tCOD removal efficiencies of the two-phase UAF and UASB system were between 74% and 88% with an average of 84% at mesophilic temperatures. It was not observed any decreases in tCOD treatment efficiencies with decreases in total HRT from 10 h to 4 at 35±2°C in the two-phase UAF and UASB system. The tCOD treatment efficiency in Period 3 decreased from 88% to 74% and this decrease in removal efficiency could be explained as the result of the low influent tCOD value (184 mg/L). Although the removal efficiency decreased from 88% to 74%, the effluent tCOD value was 48 mg/L, and it was being in the range to be acceptable by the receiving waters according to Turkish Water and Wastewater Regulation (SKKY, 2005).

Table 4.8. The average influent and effluent sCOD and tCOD values of UAF reactor, 2<sup>nd</sup> reactor and UASB reactor

Days	Influent sCOD (mg/L)	Effluent sCOD (mg/L)			sCOD Change (%)*				
		UAF Reactor	2 <sup>nd</sup> phase Reactor	UASB Reactor	UAF Reactor	2 <sup>nd</sup> phase Reactor	UAF+ 2 <sup>nd</sup> phase Reactor		
		UAF Reactor	2 <sup>nd</sup> phase Reactor	UASB Reactor	UAF Reactor	2 <sup>nd</sup> phase Reactor	UASB Reactor		
Period 1	259-273	194±50	93±8	45±12	41±8	-52	-52	-77	-79
Period 2	274-280	113±6	127±4	49±1	50±2	+13	-61	-56	-55
Period 3	281-288	89±3	58±6	29±4	29±3	-36	-50	-68	-67
Period 4	289-295	44±3	51±4	23±4	33±3	+16	-55	-48	-26
Period 5	296-308	65±4	64±2	35±3	48±5	-1	-44	-44	-25
Days	Influent tCOD (mg/L)	Effluent tCOD (mg/L)			tCOD Treatment Efficiency (%)				
		UAF Reactor	2 <sup>nd</sup> phase Reactor	UASB Reactor	UAF Reactor	2 <sup>nd</sup> phase Reactor	UAF+ 2 <sup>nd</sup> phase Reactor		
Period 1	259-273	466±19	136±21	67±8	84±10	70	51	86	87
Period 2	274-280	793±27	273±18	97±12	91±18	61	65	88	87
Period 3	281-288	184±35	135±7	48±2	55±26	28	65	74	74
Period 4	289-295	332±17	97±7	39±4	76±11	63	60	88	73
Period 5	296-308	310±23	166±12	53±4	105±12	44	68	83	64

\*(-) sign shows reduction, (+) sign shows increase.

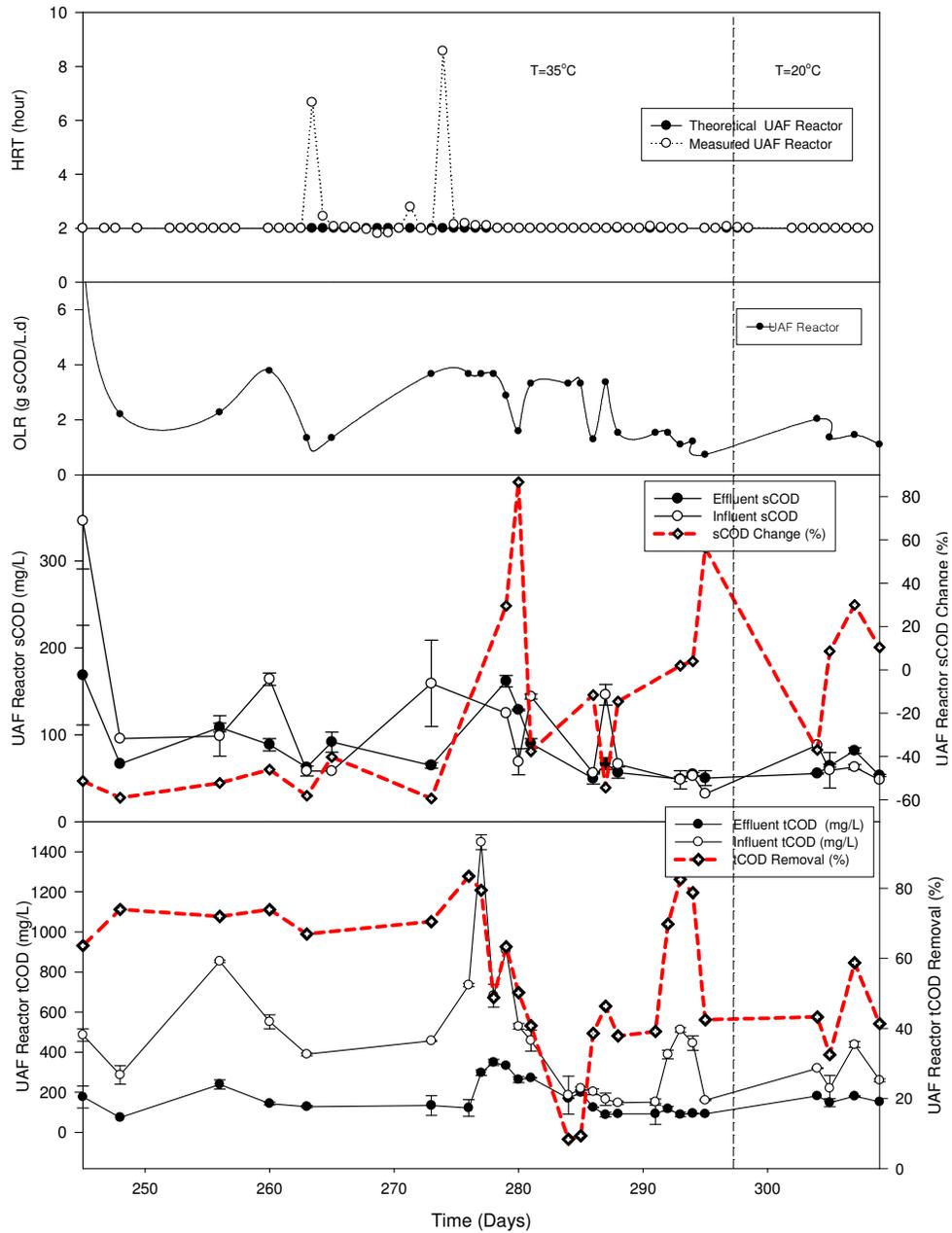


Figure 4.17. Effect of OLR/HRT on sCOD, tCOD effluents, sCOD changes and tCOD removal efficiencies versus operational period for acidogenic UAF reactor ( $T=35\pm 2^{\circ}\text{C}$  in days 237-295 and  $T=20\pm 2^{\circ}\text{C}$  in days 295-309).

In Period 5, the total HRT of the two-phase UAF and UASB system was 4 h and the temperature was  $20\pm 2^{\circ}\text{C}$ . It was observed similar tCOD removal efficiencies (average 83%) in Period 5 at psychrophilic temperature like at mesophilic temperatures. The ANOVA test statistics showed that the difference in tCOD removals between mesophilic and psychrophilic temperatures was non-significant in the two-phase UAF and UASB system ( $F(1,7)=0.25$ ,  $p=0.63$ , Table B.12, Appendix B). It can be concluded that it was not observed any negative effect of decreases in temperature on the total COD removal efficiencies of the two phase UAF and UASB system. One reason of that might be explained with the influent domestic wastewater characteristics. The average hydrolyzed fractions (sCOD/tCOD ratio) of influent domestic wastewater were 13% and 21% for Period 4 and 5, respectively, at 35 and  $20^{\circ}\text{C}$ . It can be clearly seen that the average hydrolyzed fraction of influent domestic wastewater was higher at  $20^{\circ}\text{C}$ . When the influent domestic wastewater was collected from the collection point, it was better hydrolysed when it was operated at  $20^{\circ}\text{C}$  than that at  $35^{\circ}\text{C}$ . Therefore the expected decreases in total COD removal efficiency was not observed at  $20^{\circ}\text{C}$ . Another reason for that can be explained with the specific activity of sludge improved under psychrophilic conditions, which indicated that there was growth and enrichment of methanogens and acetogens in the anaerobic system (Lettinga et al. (2001). Wiegel (1990) indicated that microorganisms are classified into 'temperature classes' on the basis of the temperature span in which the species are able to grow and metabolize. The overlapping growth temperature ranges indicated that there was not a clear boundary between these classic groups of psychrophilic, mesophilic and thermophilic microorganisms (Wiegel, 1990). Whereas the bacterial growth rates of methanogenic thermophiles and mesophiles from anaerobic reactors are well determined. Under psychrophilic conditions, chemical and biological reactions proceed much slower than under mesophilic conditions. Most reactions in the biodegradation of organic matter needs more

energy to proceed at low temperatures. However some reactions, such as hydrogenotrophic methane production and acetate formation from hydrogen and bicarbonate require less energy (Lettinga et al., 2001). In general, lowering the operational temperature leads to a decrease in the maximum specific growth rates but it might also lead to an increased net biomass yield (g biomass/ g substrate converted) of methanogenic population or acidogenic sludge (Lin et al., 1987; Berg, 1977;Lier, 1997).

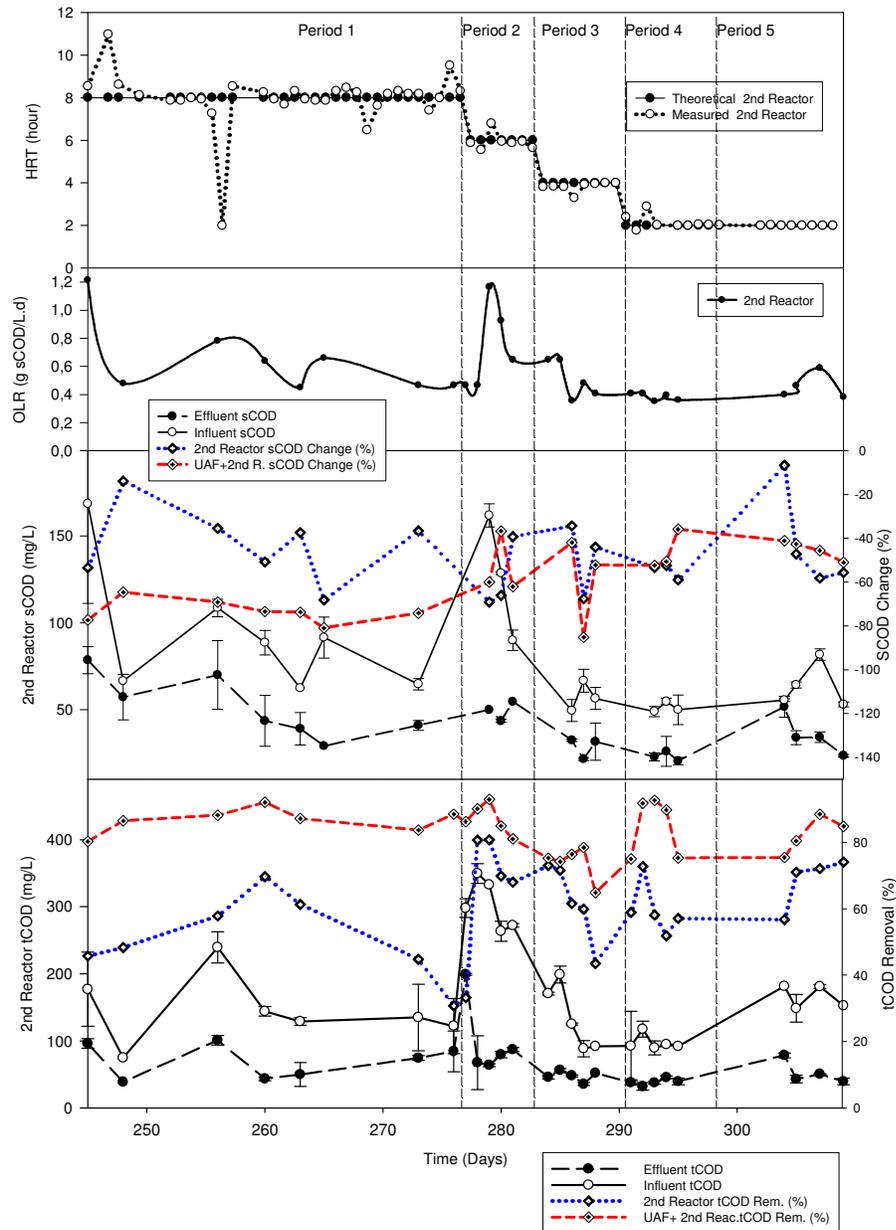


Figure 4.18. Effect of OLR/HRT on sCOD, tCOD effluents, changes for sCOD and removal efficiencies for tCOD versus operational period for 2<sup>nd</sup> phase reactor and two-phase UAF and UASB system ( $T=35\pm 2^{\circ}\text{C}$  in days 237-295 and  $T=20\pm 2^{\circ}\text{C}$  in days 295-309).

In two-phase UAF and UASB system an acidified wastewater was introduced to the methanogenic reactor in this study. When imposing non-acidified wastewater to the treatment system, using a methanogenic reactor, under psychrophilic conditions, the formation of a layer of acidifying sludge around the granules is a serious problem. This layer formation can lead to gas entrapment inside the granules, which initiates flotation of these granules (Rebac, 1998). In the present study, it was not faced with such a problem because acidification was provided in the UAF reactor and resulted with no flotation of granules. Therefore, acidification reactor (UAF) enhanced the efficiency of the methanogenic reactor (UASB) in the two-phase system in this study.

It was observed a decrease in sCOD reductions of the two-phase UAF and UASB system with the decrease in HRT. By not taking into consideration the Period 3 (influent tCOD value was very low), the sCOD reductions were 77%, 56% and 48% at total HRTs of 10 h, 8 h and 4 h, respectively in the two-phase UAF and UASB system at  $35\pm 2^{\circ}\text{C}$  (Table 4.8). It was observed 4% decreases in sCOD reductions with decrease in temperature from  $35\pm 2^{\circ}\text{C}$  to  $20\pm 2^{\circ}\text{C}$  with an average sCOD reduction of 44% in the two-phase UAF and UASB system. The ANOVA and multiple regression analysis showed that a strong correlation between sCOD reduction and temperature was observed ( $F(1,6)=25,81$ ,  $R^2=0.84 - 0.843$ ,  $p=0.0038$  (Appendix B, Table B19) ). Decreases in temperature caused decreases in sCOD reductions in the two-phase UAF and UASB reactor system. The reason of decrease in sCOD reductions with the decrease in two-phase UAF and UASB system might be explained as the high rate of the conversion of tCOD to sCOD. The sCOD in VFA form did not enhance the methanification in 4 h HRT as it was in 10 h HRT.

The tCOD and sCOD variations in influent and effluent of one phase UASB system with respect to HRT and OLR were presented in Figure 4.20. The OLR of the one-phase UASB reactor ranged between 0.42 g sCOD/L.d and 1.60 g sCOD/L.d during the five periods. Although the HRT of the one-phase UASB system decreased from 10 h to 4 h step by step, the OLR of reactor did not increase, respectively, because of the variations in influent COD concentrations. The tCOD treatment efficiency was 87% at HRTs of 10 h (Period 1) and 8 h (Period 2). Then it decreased to 74% and 73% at HRTs of 6h (Period 3) and 4 h (Period 4), respectively (Table 4.8). The reason of decrease of removal efficiency to 74% at 6 h HRT may be explained with low influent tCOD values. The results were compatible with the results of Lettinga et al (1993) in which 55-75% tCOD removals obtained at a HRT of 5-6 h with influent tCOD values of 200-300 mg/L. In 4 h HRT (Period 4), the influent tCOD was in medium strength and it was observed a decrease in removal efficiency about 16% (compared to Periods 1 and 2) at 35±°C. When the results of this work are compared with the literature, it is possible to conclude that the tCOD removal efficiencies are higher than the results attained by Castillo et al (1997). Castillo et al (1997) reported that the overall efficiency of the one-phase UASB system removing tCOD doubled when the HRT increased from 3 h (35%) to 7.5 h (tCOD removal efficiency= 70%). In the present study, removal efficiencies were much better at 4 h (73%) and 8 h (87%) than the results published by Castillo et al (1997). The reason of higher removal efficiencies at the present study can be explained with the experiments performed at higher temperatures (at 35°C) than the experiments performed at low (20°C) temperatures by Castillo et al (1997).

The results of the present study (73% removal at an HRT of 4 h with an influent tCOD of 332 mg/L) are comparable with the results by Barbosa and Sant'Anna (1989). They reported that 74% tCOD removal efficiency was obtained for an

average influent tCOD value of 627 mg/L at an HRT of 4 h and at a temperature of 19-28°C.

The tCOD removal efficiency decreased from 73% to 64% (12%) by decreasing the reactor temperature from 35°C (Period 4) to 20°C (Period 5). The ANOVA test statistics indicated that the difference between tCOD removals in mesophilic and psychrophilic temperatures was significant in one-phase reactor ( $F(1,7)= 11.40$ ,  $p=0.012$ , Table B.13, Appendix B). The tCOD removal efficiencies were reported as 60% and 65%, respectively by Garcia et al (1998) and Kalogo et al (2001) at an HRT of 4 h and temperature of 10-26°C. The one-phase UASB system removal efficiencies of the present study at 4 h HRT are compatible with the literature results.

In the present study, 12% decrease in tCOD removal efficiency can be accepted as a prospective result by decreasing the temperature range from mesophilic (35°C) to psychrophilic conditions (20°C). Comparing the summer and winter results measured in the UASB reactor studied by Castillo et al (1997), it could be concluded that the effect of temperature was very noticeable and the removal efficiencies were around 20%- 30% higher in summer than that in winter.

In this study, the average sCOD reductions ranged between 79% and 55% with an average of 67% at HRTs of 10 h- 6 h. Lettinga et al. (1993) reported that 50-60% sCOD reductions were attained at HRTs of 5- 6 h at temperatures of 24-26°C. The reasons of higher sCOD reductions of the present study (average 67%) can be explained by the increases in temperature (35°C) and high rate of tCOD conversion to sCOD.

The average of sCOD reductions at 10 h HRT was 79% and the average sCOD reduction decreased to 26% at 4 h HRT. The average sCOD increase was 53% with the decrease of HRT from 10 h to 4 h in the one-phase UASB system reactor. The average of tCOD removals at 10 h HRT was 87% and decreased to 73% in 4 h HRT. By decreasing the HRT from 10 h to 4 h, the average tCOD removal decreased only 14% but the average sCOD reduction decreased 53%. The higher decrease in sCOD reduction than the tCOD removal at that HRT can be explained as the rate of the removal of sCOD was lower than tCOD and a high percentage of sCOD might be in VFA form at 4 h HRT. The reduction of sCOD of one-phase UASB system did not show a variation by decreasing of temperature from 35°C to 20°C. The removal efficiency of sCOD decreased from 26% to 25% in 4 h HRT as the temperature decreased from 35°C to 20°C. This could be explained with the decreases in the hydrolysis of slowly degradable organics to soluble organics at low temperatures in one-phase reactor (Luostarinen et al., 2007). The regression and ANOVA analysis showed that there is a weak correlation between sCOD and temperature while the differences between sCOD and temperature was not so significant in one-phase UASB system ((F(1,6)=13.233,  $p=0.015$ ,  $R^2=0.726$ ,  $R^2=0.526$ ), Appendix B. Table B 20).

Comparison of one-phase UASB system and the two-phase UAF and UASB system for COD reductions and effluent values:

In the comparison of two sets of reactors, the two-phase UAF and UASB system and one-phase UASB system, the total HRT of UAF and 2<sup>nd</sup> phase UASB system and one-phase UASB system were the same for the specified Periods. The HRTs of the reactors are shown in Figures 4.18, 4.19 and 4.20. The Periods 1-4 and Period 5 were performed at 35±2° C and 20±2°C, respectively.

In Periods 1-3, the tCOD removal efficiencies were about the same for the two-phase UAF and UASB system and the one-phase UASB system (Table 4.8). For the Periods 1 (HRT=10 h) and 2 (HRT=8), the average of tCOD removals was 87% for the two-phase UAF and UASB system and for the one-phase UASB system. The averages of influent tCOD were 466 and 793 mg/L in Period 1 and Period 2, respectively. The tCOD removal efficiencies decreased from 88% to 74% with the decrease in influent tCOD value to 184 mg/L in Period 3. The reason of the decrease in removal efficiency can be explained with the decrease in influent tCOD and not with the decrease in total HRT to 6 h. The removal efficiency of the two-phase UAF and UASB system showed an increase (88%) with the increase in influent tCOD (332 mg/L) in Period 4 (total HRT decreased to 4 h). In Period 4 (HRT=4h), the tCOD removal efficiency in one-phase UASB system was 73% and it was lower (15%) than the removal efficiency in the two-phase UAF and UASB system at that HRT and at 35°C. There was no negative effect to the tCOD removal efficiency of the decrease of HRT to 4 h in the two-phase UAF and UASB system but the decrease in tCOD removal efficiency was noticeable. In one-phase system it was provided hydrolysis, acidification and methanification phases as much as the two-phase system at HRTs 10h to 6 h.

However the decrease in HRT from 6 h to 4 h indicated the importance of the phase separation. In Period 4 (HRT=4 h), the effluent tCOD values were 39 mg/L and 76 mg/L for the two-phase UAF and UASB system and the one-phase UASB system, respectively. The effluent tCOD values were compatible with the Turkish Regulations and reached to the COD criteria given for the acceptance of receiving waters (Table 21, Turkish Water Pollution and Prevention Regulation (SKKY), 2005).

In Period 5 (T=20°C), the tCOD removals were 83% and 64% in the two-phase UAF and UASB system and one-phase UASB system, respectively. The removal efficiency was 23% higher in the two-phase UAF and UASB system than the one phase UASB reactor at 20°C. It was not observed any significant effect of temperature decrease to the tCOD removal efficiency in two-phase system. As the temperature decreased from 35°C to 20°C in the two phase system, the tCOD removal efficiency was not influenced. The decrease in tCOD removal efficiency was considerably high in one-phase UASB reactor compared to two phase (73% and 64% in one phase UASB system and two phase system, respectively). The effluent tCOD values were 53 mg/L and 105 mg/L for the two-phase UAF and UASB system and one-phase UASB system, respectively. In Turkish Regulations the upper limit of a composite effluent sample for COD in domestic wastewater is 90 mg/L in populations more than 100000 and 100 mg/L for populations of 10000- 100000. Therefore, the effluent tCOD of the UAF+2<sup>nd</sup> reactor system effluent was compatible. However, one-phase UASB system effluent was not compatible with the receiving water COD criteria in Turkish Regulations. A phase separation can be advantageous to have the effluent tCOD results below 90 mg/L at HRTs around 4 h and/or at temperatures below 20°C. As a conclusion, the two-phase UAF and UASB system had considerably better tCOD removal efficiencies than one-phase UASB system at 4 h HRT at 35°C and 20°C. This also indicated that the two-phase UAF and UASB system has a better potential for treating domestic wastewater at ambient temperatures and at lower HRTs.

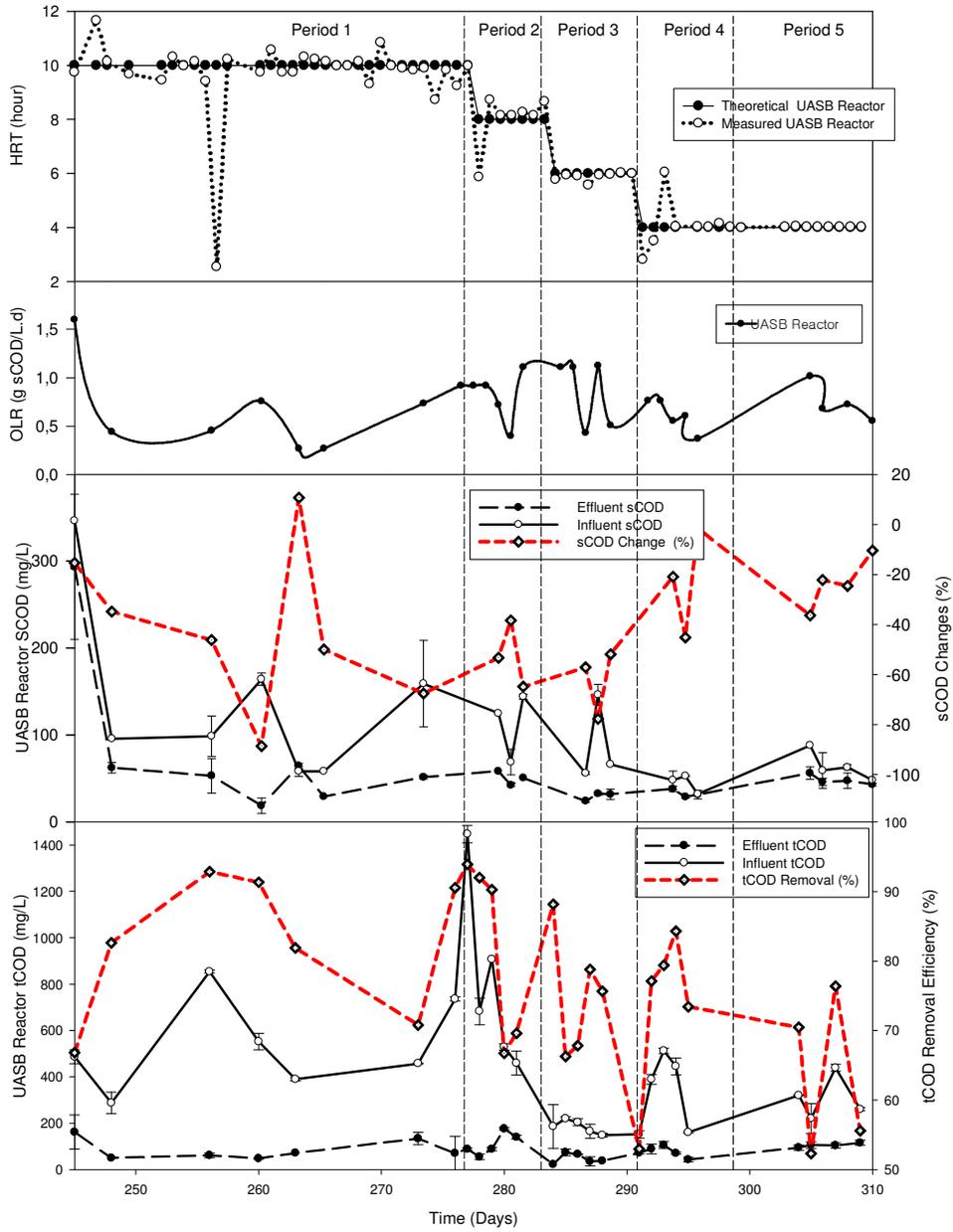


Figure 4.19. Effect of OLR, HRT and temperature on sCOD, tCOD effluents, sCOD changes and tCOD removal efficiencies versus operational period for one-phase UASB system ( $T=35\pm 2^{\circ}\text{C}$  in days 237-295 and  $T=20\pm 2^{\circ}\text{C}$  in days 295-309).

In the past, it had been demonstrated that the two-phase digestion process exhibited better performances than the conventional one-phase digestion process (Ghosh, 1986; Zhang et al., 1991; Cha and Noike, 1997) for different types of wastewaters. The results of the present study for domestic wastewater at a HRT of 4 h verified the results in literature for domestic wastewater treatment. The key feature of the two-phase process was the permission for the enrichment of different bacteria in each digester by independently controlling the operational conditions of the digester (Cha and Noike, 1997).

In the studies performed by El Gohary and Nasr (1999), one-stage and two-stage UASB reactors were experimented at the same HRT (8 h) for comparison. In this study, the average one-stage and two-stage tCOD removals were 77% and 79%, respectively. In the present study, the average tCOD removal results of two-phase system (88%) and one-phase system (87%) performances were better than that the results of El Gohary and Nasr (1999). The tCOD removal results of the two-phase UAF and UASB system of the present study at 4 h HRT (88% tCOD removal) were better than the two-stage UASB reactor (79% tCOD removal) in the study performed by El Gohary and Nasr (1999). It can be said that phase separation resulted better tCOD removals than staged treatment.

A significant difference in sCOD reductions between the two-phase UAF and UASB system and one-phase UASB system was not observed in Period 1 (HRT=10 h), Period 2 (HRT=8 h) and Period 3 (HRT=6h) (for 6 h-10h,  $F(1, 30) = 0,040$ ,  $p > 0.05$ . Table B.18-Appendix B). But in Period 4 (HRT=4 h), the average sCOD reduction of the two-phase UAF and UASB system (48%) was higher than that one-phase UASB system (26%). The decreases in temperature (20°C) did not significantly affect the sCOD reductions and the sCOD reductions became 44%

and 25% for two-phase system study and one-phase system, respectively. It can be concluded that most of the solubilized COD was better removed in the two-phase UAF and UASB system than that one-phase UASB system. In this study the influent was a prehydrolyzed domestic wastewater.

#### **4.3.2 The Variations of BOD<sub>5</sub> in Influent, Effluent Samples and BOD<sub>5</sub> Removal Efficiencies in One-Phase and Two-Phase Systems**

Influent, effluent concentrations and removal efficiencies for BOD<sub>5</sub> are given in Table C 2 (Appendix C) and illustrated in Figure 4.20 for UAF reactor, 2<sup>nd</sup> phase UASB reactor, two-phase UAF and UASB system and one-phase UASB system. The variations in the strength of influent were very high and the influent BOD<sub>5</sub> values ranged between 65±25 mg/L and 200±10 mg/L during the days 274-309.

During the Periods 1-4, the UAF reactor was experimented at a HRT and temperature of 2 h and 35°C, respectively. The reason of variations in effluent values (Figure 4.20.b) can be explained with the variations in influent BOD<sub>5</sub> concentrations. For example, for an influent BOD<sub>5</sub> concentration of about 185 mg/L, the effluent BOD<sub>5</sub> concentrations were between 76 mg/L and 95 mg/L and for a much lower influent BOD<sub>5</sub> concentration of 65 mg/L, the effluent BOD<sub>5</sub> was 32 mg/L (Table 4.11). Although the effluent BOD<sub>5</sub> concentrations showed peaks depending on the influent BOD<sub>5</sub> concentrations, the BOD<sub>5</sub> removal curve for Periods 1-4 was almost flat in the UAF reactor (Figure 4.20.b) and ranged between 49% and 59% (Table 4.11) at 35°C. Although the ambient temperature of the UAF reactor decreased to 20°C in Period 5, the BOD<sub>5</sub> removal efficiency increased to 73% (influent BOD<sub>5</sub> concentration increased to 200 mg/L). With the decrease in influent BOD<sub>5</sub> concentration (135 mg/L) in Period 5 (the last

experiment), the efficiency again decreased to 53% at 20°C. It can be concluded that BOD<sub>5</sub> removals in the UAF reactor were not affected by temperature decrease (from 35°C to 20°C). The removal efficiency was mostly affected by the BOD<sub>5</sub>

concentration of influent in UAF reactor. Similar to tCOD removal the hydrolyzed fraction of influent domestic wastewater was higher when the sample was collected as the reactor was operated at 20°C. Recent studies showed that methanogenic activity under psychrophilic conditions could be carried out and the methanogens are able to grow at low temperatures (Rebac, 1998).

The effluent BOD<sub>5</sub> values varied between 20 mg/L and 25 mg/L and it was not observed any effect for decreases in HRTs and in temperatures from 8 h to 2 h and from 35°C to 20°C in the 2<sup>nd</sup> reactor, respectively. The effluent BOD<sub>5</sub> values for the investigated HRTs can be seen almost as a flat line in Figure 4.20.c. In the 2<sup>nd</sup> phase UASB reactor (effluent of two-phase system), the effluent BOD<sub>5</sub> values were below 25 mg/L in all reactors which satisfied the Turkish Regulations (In composite samples, the acceptable limits for BOD<sub>5</sub> varied between 35 and 50 mg/L for different receiving waters). The effluents of the two-phase system also satisfied the discharge standards of European Union Discharge Standards set as 25 mg/L (EU, 1991). It can be concluded that the two-phase UAF and UASB system satisfied the Turkish and EU Discharge Standards at HRT ranges between 10 h (2h+8h) and 4 h (2 h+2 h) at 35°C and 20°C, respectively.

The effluent BOD<sub>5</sub> values of the UAF reactor were considered as the influent in the calculations of the treatment efficiencies of the 2<sup>nd</sup> phase reactor. The BOD<sub>5</sub> removal efficiencies were between 64% and 74% for the influent BOD<sub>5</sub> values of 55 mg/L and 95 mg/L in the 2<sup>nd</sup> phase reactor. For an influent BOD<sub>5</sub> value of 32 mg/L, the removal efficiency decreased from 64% to 25%.

In the present study, for the average influent BOD<sub>5</sub> values of 185 mg/L, the removal efficiencies were 86% and 87% in Period 1 and Period 3, respectively at 35°C. The BOD<sub>5</sub> treatment efficiency decreased to 63% for influent BOD<sub>5</sub> value of 65 mg/L in Period 4. It was observed that a decrease in the strength of influent domestic wastewater (below 100 mg/L) resulting with a decrease in BOD<sub>5</sub> removal efficiency of the two-phase UAF and UASB system below 65%. The BOD<sub>5</sub> performance of two-phase UAF and UASB system surpassed the expectation at Period 5 (HRT=4 h, T=20°C). In temperatures of 24-26°C, the BOD<sub>5</sub> removal was between 69 and 83% for an influent value of about 153 mg BOD<sub>5</sub>/L, at a HRT of 12 h in an Anaerobic Filter (AF)+UASB pilot plant study (Joshi et al., 1987). It was observed that the BOD<sub>5</sub> treatment efficiencies were between 85% and 90% for the influent BOD<sub>5</sub> values of 135 mg/L and 200 mg/L at 20°C in Period 5. As a result of two-phase system, it can be stated that the removal efficiencies were mostly depended on the influent BOD<sub>5</sub> concentrations compared to HRT and presented small variations in total HRTs of 4-10 h.

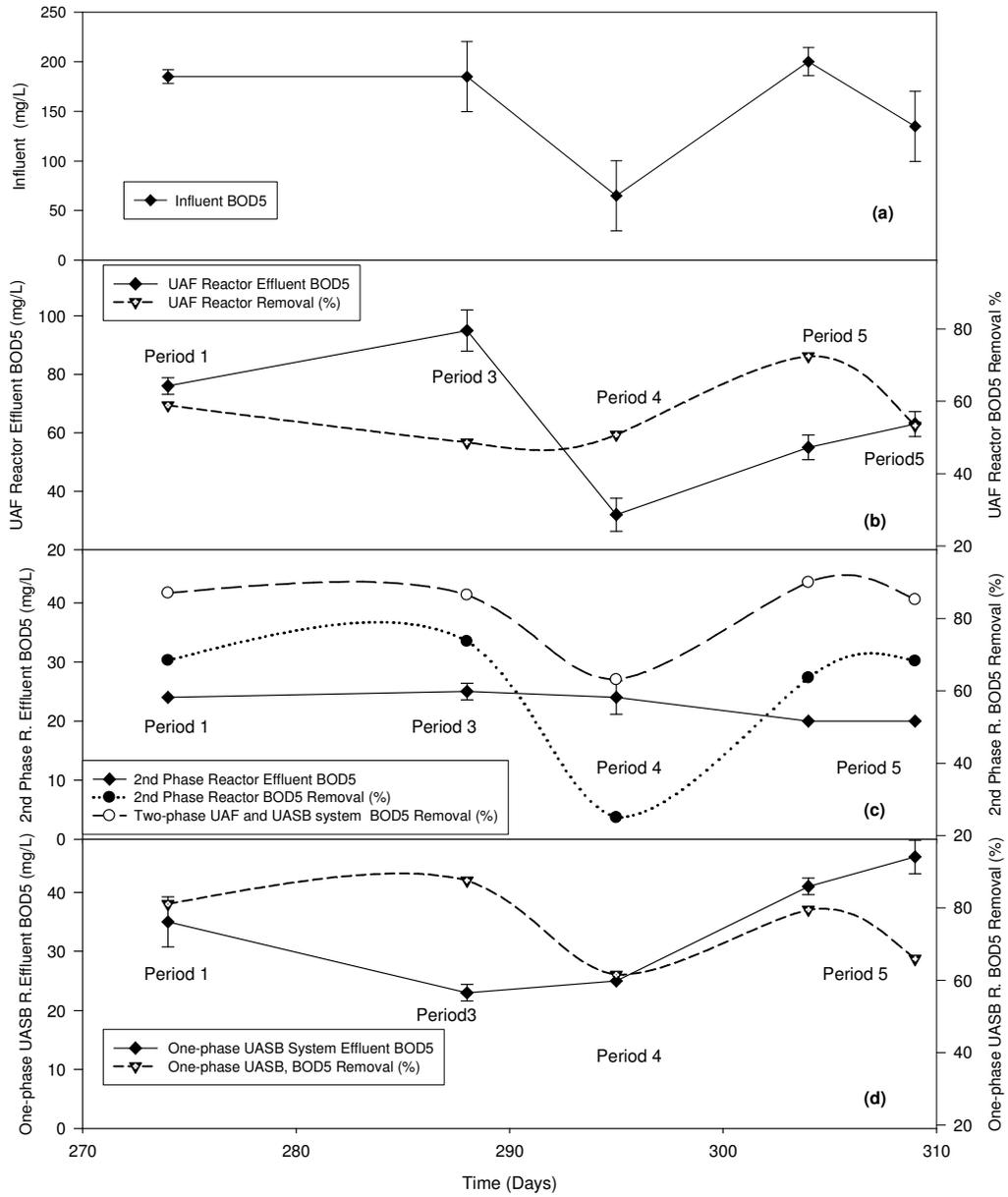


Figure 4.20. Influent and effluent BOD<sub>5</sub> values and BOD<sub>5</sub> removal efficiencies in UAF, two-phase UAF and UASB system and one-phase UASB system.

In one-phase reactor, the hydraulic retention times of UASB reactor were 10 h, 6 h and 4 h in Period 1, Period 3 and Period 4, respectively at 35°C. The Period 5 was performed at a HRT of 4 h and a temperature of 20°C. In Periods 1, 3 and 4, the effluent BOD<sub>5</sub> values of UASB reactor were 23-35 mg/L which are below the effluent discharge standards of Turkey and set as 35 mg/L for 24 h composite samples of treated domestic wastewaters (SKKY, 2005). However, in Period 5, the average effluent BOD<sub>5</sub> values were between 41-46 mg/L with an average of 44 mg/L, which is above from the Turkish discharge standards. For the influent BOD<sub>5</sub> values of 100 to 250 mg/L, the effluent BOD<sub>5</sub> values were between 40 and 80 mg/L at a temperature range of 16-23°C (Vieira, 1988; Vieira and Garcia, 1992). Results of Lettinga et al (1993) demonstrated that UASB systems were effective in removing and stabilizing the organic pollutants at HRTs as low as 4 h, i.e. a BOD<sub>5</sub> reduction up to 65-80% could be achieved at 24-26°C. It was observed that, the BOD<sub>5</sub> reductions of the present study were between 66% and 80% at a HRT and temperature of 4 h and 20°C, respectively in UASB reactor. The results of the present study are in agreement with the literature results mentioned above (Vieira, 1988; Vieira and Garcia, 1992; Lettinga et al., 1993).

In the comparison of BOD<sub>5</sub> effluent values and removal efficiencies of two-phase UAF and UASB system and one-phase UASB system, it was observed that;

The BOD<sub>5</sub> removal efficiencies at Period 1 (HRT=10 h) and at Period 3 (HRT=6 h) were above 80% for both of the reactor systems at 35°C. At Period 4 (HRT=4 h) the BOD<sub>5</sub> removal efficiencies decreased from 87% to 63% and from 88% to 62% for the two-phase system and one-phase system, respectively at 35°C. In Periods 1-4, the effluent BOD<sub>5</sub> values were 24-25 mg/L and 23-35 mg/L for two-phase system and one-phase system, respectively. The effluents of both sets of

reactors were below the BOD<sub>5</sub> discharge standards (35 mg/L) of Turkey (SKKY, 2005).

In Period 5 (HRT=4 h and T=20°C), the averages of BOD<sub>5</sub> removals were 88% and 73% for two-phase system and one-phase system, respectively. In Period 5, the average BOD<sub>5</sub> effluent values were 20 mg/L in two-phase system and it easily satisfied the Turkish Regulations set as 35 mg/L (SKKY, 2005) and European Union (EU) Discharge Standards set as 25 mg/L (EU, 1991). In one-phase reactor the effluent BOD<sub>5</sub> values were between 41-46 mg/L and they were above the limits set by SKKY (2005) of Turkish Regulations and EU Discharge Standards (EU, 1991). As a conclusion, the effect of two-phase system was important in the Turkish and EU effluent standards whereas it was not possible to satisfy these standards by one-phase UASB reactor at 20°C for BOD<sub>5</sub> parameter.

For providing an effluent with BOD<sub>5</sub> lower than 35 mg/L, stabilization ponds could be operated at hydraulic retention times of 15 to 20 days (CETESB, 1989). In case of limited land availability, aerobic treatment systems like the conventional activated sludge process, extended aeration or oxidation ditches may be used. Although they are expensive and require the use of electromechanical devices for aeration and mixing, aerobic processes may provide an effluent of about 20 mg BOD<sub>5</sub>/L (Vieira and Garcia, 1992). But in case of conventional activated sludge process, sludge stabilization must also be provided. For satisfying related Turkish Regulations and EU Discharge Standards, two-phase system can be a feasible alternative by providing effluent BOD<sub>5</sub> values less than 35 mg/L.

### 4.3.3 Effect of HRT on the Variation of pH in Two-phase and One-phase Systems

The pH variations with the applied HRT and organic loading rates are illustrated in Figure 4.21. Although the HRT of 2<sup>nd</sup> Reactor (UASB reactor following UAF reactor), and the UASB (one-phase reactor) decreased stepwise during the Periods 1 to 4, the organic loading rates did not show increases at similar rates because of the variations and decreases in influent sCOD values (Figure 22-b). The HRT of the UAF reactor was 2 h during this study. The average pH values did not show a significant variation and ranged between 7.05 and 7.17 during the studied periods. Small variations in pH values of UAF reactor were observed with the change in average influent pH values of domestic wastewater. As an example of that, the averages of influent pH values were 7.73 and 7.40 and the average effluent pH values were 7.17 and 7.05, for Period 1 and Period 2 respectively. Although the decrease in influent pH values was 0.33 from Period 1 to Period 2, the decrease in the effluent pH values was only 0.12 in UAF reactor. The effluent pH values were between 6.87 and 7.25 with an average of 7.10 while the average of influent pH values was 7.71. It was not observed a variation in the effluent pH values of UAF reactor with the change in temperature from 35°C to 20°C during this study. Kalogo et al. (2001) found that no excessive acidification determined in UAF reactors because pH never dropped below 6.8. In the present study since the pH did not decrease below 6.87, it can be concluded that no excessive acidification occurred in UAF reactor.

In the 2<sup>nd</sup> reactor, the effluent pH values were between 7.23 and 8.06 (Figure 18-a) and the average effluent pH values were 7.71, 7.56, 7.73, 7.89 and 7.71 for Periods 1, 2, 3, 4 and 5, respectively. It was observed a certain decrease in pH values from 7.74 to 7.56 with the decrease in effluent pH values of the UAF

reactor (from 7.17 to 7.05). The optimum pH level for methanogens is within the range 7.5- 8.5 (Hobson and Wheatly, 1993) and the effluent pH of the 2<sup>nd</sup> phase reactor averaged between 7.56 and 7.89 for the studied five different HRTs and two different temperatures. Therefore, the 2<sup>nd</sup> phase UASB reactor can be considered in optimum methanogenic range at the two-phase system whereas . average methane percentages, and sCOD reductions were observed as 38% and 55%, respectively, for a very low influent average sCOD concentration of 83 mg/L (Table 4.8 and Table C 5).

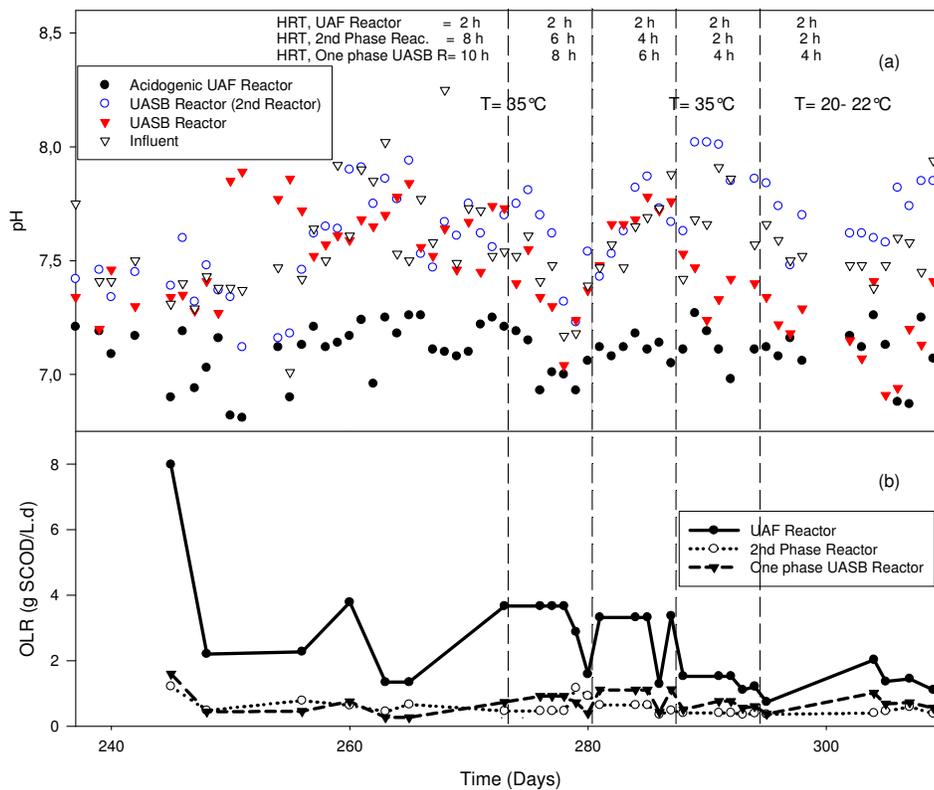


Figure 4.21. Variations of pH in the two-phase UAF and UASB system and the one-phase reactor (a) at applied OLR's (b) and HRT's.

In one-phase UASB system, the effluent pH values were between 6.91 and 7.84 (Figure 22-a). It was observed average effluent pH values of 7.64, 7.36 and 7.66 for HRTs of 10 h, 8 h and 6 h, respectively. In Period 2, the decrease in effluent pH can be considered as the result of relatively lower influent pH values during Period 2. Although the average influent pH value in Period 4 (HRT=4 h) was high (pH=7.71), the average effluent pH value decreased to 7.33. The reason of this could be explained by the one-phase UASB reactor having lower pH values than optimum methanogenic pH values. This may be explained by acidogenic processes continued at that HRT. In Period 5 (HRT=4 h, psychrophilic temperatures), it was observed that the effluent pH values were between 6.91 and 7.22 with an average of 7.17. The resulting low average effluent pH value at a HRT of 4 h in one phase UASB reactor can show that the methanogenic processes are not properly proceeded whereas COD removals decreased to 25% and methane percentages were 14% at Period 5 (Table 4.8 and Appendix C 5)

#### **4.3.4. Variations of VFA and Alkalinity in Influent and Effluent Samples in One-phase and Two-phase (UAF and UASB) Systems**

The influent and effluent pH, VFA and alkalinity values of the reactors during the one-phase and two-phase systems are presented in Appendix C (Table C 4). It was observed that all reactors were in acceptable ranges in terms of effluent VFA and alkalinity contents. The assessment was made considering the optimum operational conditions given for anaerobic cultures (below 250 mg/L) for VFA, (Speece, 1996). Typical alkalinities of untreated domestic wastewater in medium strength is 100 mg/L as CaCO<sub>3</sub> and has a range of 50-150 mg/L as CaCO<sub>3</sub> for medium and strong domestic wastewater (Metcalf and Eddy, 1991). The influent and effluent alkalinities of the UAF, the 2<sup>nd</sup> phase reactor and the one-phase

UASB reactor are shown in Figure 4.22. As seen in Appendix C (Table C.4) , in the present study, the influent domestic wastewater alkalinities were between 70 and 126 mg/L as CaCO<sub>3</sub> and they were almost in the medium range given in literature.

In the UAF reactor, it was observed decreases in effluent pH and effluent alkalinities and increases in effluent VFAs in Periods 1, 2, 4 and 5 except the Period 3 as seen in Appendix C, Table C.4 . In Period 3, there was a decrease in tVFA from 55 to 49 mg/L (as Hac) and the influent tVFA was the highest tVFA among the influents during the Periods 1-5. The reason of the decrease in effluent tVFA of the UAF reactor in Period 3 was most probably because of the degradation of wastewater and acidification in holding tank during the collection and storage time. However, if the pH drops below an acceptable range, the biomass will be inhibited and it is unable to convert the VFA to the CH<sub>4</sub> (Speece, 1996). In general, it was observed decreases in pH values and alkalinities but there were available alkalinities in the effluents in all the experimented periods and it was considered that there was enough alkalinity to convert the COD to VFA in the UAF. It was observed decreases in the effluent pH values as explained in Section 4.3.3. But the pH was in acceptable range for anaerobic treatment and there was considerably high VFA production in the UAF. The decreases of the pH in the effluent accompanied with increases in VFAs as given in Appendix C, (Table C4). Kalogo et al. (2001) stated that the drop of pH and concomitant increase of VFA concentration in the effluent, and also production of gas bubbles, are characteristic features of acidogenesis. The pH of the effluent constantly dropped, indicating that acid producing metabolic reactions were occurred. There was, however, no excessive acidification in the UAF reactor because the pH never dropped below 6.8 (Kalogo et al., 2001). Therefore, it can be concluded that the UAF reactor was in proper conditions as an acidogenic reactor.

Speece (1996) stated that if the pH was maintained in a favorable range the biomass would convert the accumulated VFA to CH<sub>4</sub>, regenerating the bicarbonate alkalinity. In the 2<sup>nd</sup> phase reactor, it was observed that effluent alkalinities increased during the Periods 1-5 (in mg/L as CaCO<sub>3</sub> from 81 to 93, from 77 to 111, from 76 to 129, from 70 to 129, from 86 to 125 in Periods 1, 2, 3, 4 and 5, respectively) as seen in Appendix C (Table C4). The increase in the effluent alkalinities of the 2<sup>nd</sup> phase reactor can be explained with the utilization of VFAs to produce CH<sub>4</sub>. Throughout the Periods 1-5, decreases in tVFA and increases in pH can be all interpreted as the indications of the methanification in the effluent samples.

In the one-phase UASB reactor; pH values did not vary between influent and effluent during the Periods 1-3. The average pH was in optimum methanogenic range given as 7.5- 8.5 (Hobson and Wheatly, 1993). In Period 1, there was a decrease in effluent alkalinity whereas no change in average tVFA was obtained. This can be explained by low utilization of alkalinity with methanogens to produce methane from the tVFAs. In Periods 2 and 3, a part of alkalinity utilized for conversion of tVFA to end products. In Periods 4 and 5, the average effluent pH decreased considerably but they were still in methanogenic range. In Period 4, there was no change between the influent and effluent tVFAs but slightly alkalinity production and consumption was determined in Periods 4 and 5, respectively. The reactors can be considered as stable and enough alkalinity was available during the Periods 1-5.

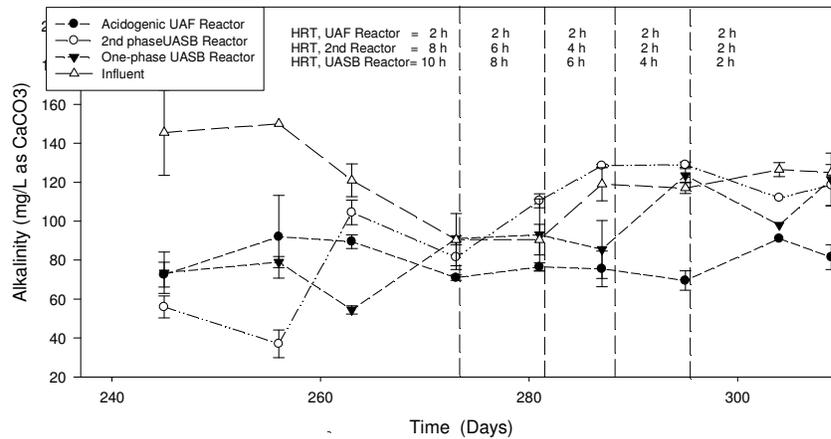


Figure 4.22. Influent and effluent alkalinities of one-phase and two-phase systems.

In the UAF reactor, the average effluent VFA-COD was 51 mg/L (Appendix C, Table C5) while the average effluent sCOD was 82 mg/L (Table 4.8, Section 4.3.1) in 2 h HRT at 35°C. The average effluent acidified fraction (VFA-COD/sCOD ratio) in the UAF was calculated as 61% for that period. The average effluent values of VFA-COD and sCOD were 48 mg/L and 64 mg/L, respectively, at 20°C and in 2 h HRT. The acidified fraction in effluent increased from 61% to 75% at 20°C in the UAF reactor. By decreasing the room temperature from mesophilic (35°C) to psychrophilic (20°C) conditions, the acidified fraction of the UAF reactor increased 14%. Jeyaseelan and Matsuo (1995) explained the reasons of practical impossibility of achieving complete acidification within acid tank as substrate contained different components. Furthermore, the time required for acidification of each of these components was different. It was also reported that complete acidification before entering the methane reactor might cause system failure in anaerobic upflow sludge blanket reactors (Lettinga and Pol, 1991). It was obtained an average acidification of 61% and a maximum acidification of

75% in the UAF reactor. The average effluent tVFAs were between 38 and 54 mg/L (as Hac) (Appendix C, Table C.4), in optimum anaerobic range being below 250 mg/L (as Hac) given by Speece (1996). Therefore, the resulted acidification in the UAF reactor can be considered in the acceptable range.

The VFA-COD increase was between 23% and 245% for the influent VFA-CODs of 12-47 mg/L in 2 h HRT at 35 °C (Appendix C, Table C 4). It was observed removals in effluent VFA-COD (about 11%) in the case of average influent VFA-COD was 59 mg/L. At a room temperature of 20°C, the average increase in VFA-COD was 181% in 2 h HRT (Appendix C, Table C 4).

In the UAF reactor, ratio of average methane production (mL CH<sub>4</sub>/ day) to OLR (g sCOD/ L.day) was 25 mL CH<sub>4</sub>/ (sCOD/L) and 4 mL CH<sub>4</sub>/ (sCOD/L) at 35°C and at 20°C, respectively. Although the UAF reactor was mainly an acidification reactor, it was observed some methanification in the UAF reactor. The decrease in temperature was resulted with a slightly more acidified effluent and less methane production per OLR (g COD/L.day) in the UAF. The objective of this study was mainly to utilize the UAF as an acidification reactor but the existence of relatively high fraction of VFAs in influent wastewater was resulted with partially methanification in the UAF reactor. It is known that phase separation could be achieved by keeping the acidogenic reactor short enough to simulate the growth of acidogenic bacteria and to wash out the methanogenic bacteria (Ghosh and Pohland, 1974; Ghosh et al., 1975). In the present study, it was observed decreases in pH values (range for influent and effluent pH were 7.73-7.40 and 7.17-7.05, respectively), increases in tVFA concentrations (from 11-44 mg/L in influent to 48-54 mg/L in effluent except Period 3) and decreases in alkalinity (from 91-126 mg/L as CaCO<sub>3</sub> in influent to 70-86 mg/L as CaCO<sub>3</sub> in effluent) in UAF reactor. The decrease in pH, increase in tVFA and decreases in alkalinity are

the indicators of acidification. Furthermore, it was observed increases up to 16% in effluent sCOD values which hydrolysis of slowly degradable compounds was enhanced but methanification phase has not been already done. However in Set 2, the increases in tVFA concentrations in the effluent of UAF reactor was not so much as in Set 1. The HRT of the acidogenic reactor (UAF reactor) was adjusted as 2 h for acidogenic phase as explained in Section 4.2. The acidified fraction (tVFA/tCOD ratio) of influent domestic wastewater was considerably higher in Set 1 (summer time) than Set 2. The average acidified fractions were 4% and 16% in winter and in summer, respectively, as explained in Section 4.1 and the influent domestic wastewater was acidified at the point of collection at a high extent.

The influent and effluent tVFAs of the two-phase system and one-phase system are plotted in Figure 4.23. In general, the effluent tVFAs in the UAF reactor were considerably higher than the influent tVFAs and the effluent tVFAs of the 2<sup>nd</sup> phase reactor and the UASB reactor.

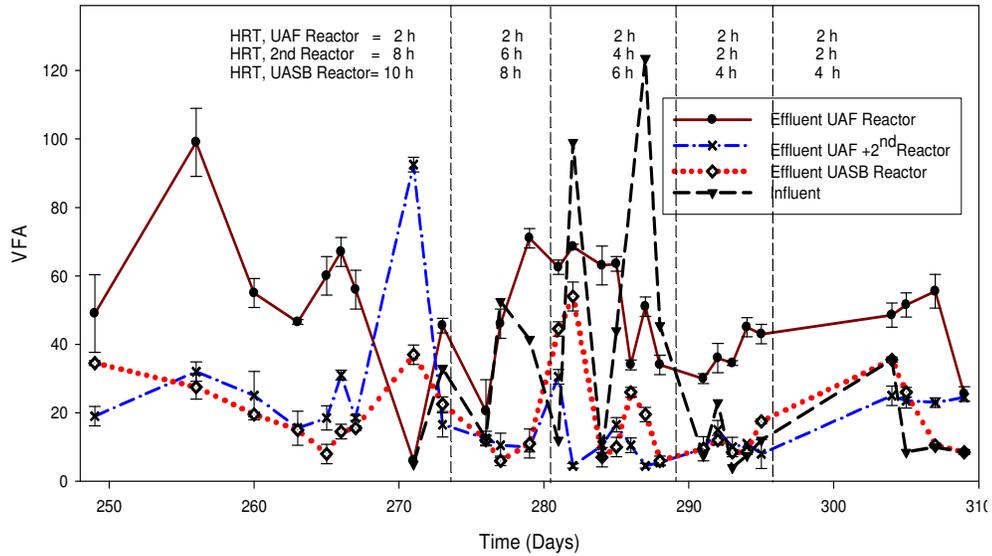


Figure 4.23. Influent and effluent VFAs in the UAF reactor, two-phase UAF and UASB system and one-phase UASB system.

In the 2<sup>nd</sup> phase reactor, the effluent contained a low amount of sCOD amounting to 45, 49, 29, 23 and 36 mg/L of which 33, 15, 11, 12 and 26 mg/L were in the form of VFA at Periods 1, 2, 3, 4 and 5, respectively (Table 4.8 and Table C5-Appendix C). In the average, 53% of the sCOD was in VFA form during the Periods 1-5 in the 2<sup>nd</sup> phase reactor. The average daily methane production increased with the decrease in HRT at 35°C, such as 25 mL CH<sub>4</sub>/day at 8 h HRT, 57 mL CH<sub>4</sub>/day at 6 h HRT, 83 mL CH<sub>4</sub>/day at 4 h HRT and 74 mL CH<sub>4</sub>/day at 2 h HRT (average OLR = 0.53 ± 0.17 g sCOD/L d) in the 2<sup>nd</sup> reactor. The decrease in temperature from 35°C to 20°C did not show a negative effect in the 2<sup>nd</sup> phase reactor and daily methane production increased from 74 mL to 77 mL (the reason can be explained with the decrease in average daily methane production from 58 mL to 5 mL in the UAF reactor by decreasing the room temperature from 35°C to 20°C). In the 2<sup>nd</sup> phase reactor, the ratio of average methane production (mL

$\text{CH}_4/\text{day}$ ) to OLR ( $\text{g sCOD/L.day}$ ) was calculated as 43, 82, 156, 194 and 166  $\text{mL CH}_4/(\text{sCOD/L})$  at Periods 1, 2, 3, 4 and 5, respectively. It was observed a rise in the ratio of methane production to OLR with the decrease in HRT from 8 h to 2 h at 35°C. But the ratio of methane production to OLR decreased from 194 to 166  $\text{mL CH}_4/(\text{sCOD/L})$  with the decrease of temperature from 35°C to 20°C.

As a summary, influent and effluent alkalinity, pH and VFA values were in acceptable ranges for anaerobic treatment in the 2<sup>nd</sup> phase reactor. The change of HRT and temperature from 35°C to 20°C was resulted with minor variations in the ratio of VFA-COD to sCOD in the 2<sup>nd</sup> phase reactor. But the decrease in HRT was resulted with increases in the ratio of average methane production ( $\text{mL CH}_4/\text{day}$ ) to OLR ( $\text{g sCOD/L.day}$ ) in the 2<sup>nd</sup> phase reactor.

In the one-phase UASB system, the effluent contained sCOD amounting to 41, 50, 30, 33, and 48 mg/L of which 20, 28, 15, 12 and 21 mg/L were in the form of VFA in Periods 1, 2, 3, 4 and 5, respectively. In the average, 47% of the sCOD was in tVFA form during the Periods 1-5 in one-phase UASB system. The average daily methane production decreased with the decrease in HRT at 35°C, such as, 119  $\text{mL CH}_4/\text{day}$  at 8 h HRT, 57  $\text{mL CH}_4/\text{day}$  at 6 h HRT and 46  $\text{mL CH}_4/\text{day}$  at 4 h HRT (average OLR =  $0.70 \pm 0.20 \text{ g sCOD/L d}$ ). The decrease in temperature from 35°C to 20°C negatively affected the daily methane production and it decreased from 46  $\text{mL/day}$  to 20  $\text{mL/day}$  in the UASB reactor. In the UASB reactor, the ratio of average methane production ( $\text{mL CH}_4/\text{day}$ ) to OLR ( $\text{g sCOD/L.day}$ ) was calculated as 144, 155, 64, 75 and 27  $\text{mL CH}_4/(\text{g sCOD/L})$  at Periods 1, 2, 3, 4 and 5, respectively. It was observed a decrease in the ratio of methane production to OLR with the decrease in HRT from 10 h to 2 h at 35°C. The ratio of methane production to OLR decreased from 75 to 27  $\text{mL CH}_4/(\text{sCOD/L})$  with the decrease of temperature from 35°C to 20°C.

In the comparison of the methanification reactors (2<sup>nd</sup> phase reactor and one-phase UASB), the tVFA fraction of effluent sCOD was higher in the 2<sup>nd</sup> phase reactor (72%) than the one phase UASB reactor (44%) at 20°C. The conversion of tVFA to methane also was better in the 2<sup>nd</sup> reactor than that the UASB reactor at the experimented temperatures and HRTs. It was observed significant decreases in methane production/OLR ratio with the decrease in HRT in the one-phase system. This ratio increased with the decrease in HRT in the 2<sup>nd</sup> phase reactor and this can be explained as the capacity of gas collection of the 2<sup>nd</sup> phase reactor increased at shorter HRTs and higher OLRs and a certain part of gas was collected in the UAF reactor prior to the 2<sup>nd</sup> phase reactor. There is a positive effect of an acidification reactor prior to the 2<sup>nd</sup> phase UASB reactor. The placement of an acidification reactor prior to an UASB reactor resulted better acidification and better conversion of acidified products to methane at lower HRTs.

#### **4.3.5. Gas Production in Two-Phase UAF and UASB and One-phase UASB Systems**

The daily biogas produced in two-phase UAF and UASB system and one-phase UASB system are depicted in Figure 4.24. The two-phase configuration consisted of UAF reactor and 2<sup>nd</sup> phase reactor for acidogenic and for methanogenic phases, respectively. The one-phase configuration (UASB) was run for compare the removal efficiencies of one and two-phase reactor systems.

The HRT of UAF reactor, acidogenic reactor, was adjusted to 2 h during the Set 2 (Period 1-5). The room temperature was 35°C in Periods 1-4 and 20°C in Period 5. The daily gas production of the UAF reactor fluctuated from 0 to 820 mL with

an average of 250 mL at 35°C in 2 h HRT. The reasons of these fluctuations can be explained with the variations in the strength of influent wastewater and irregular gas pockets appeared in the filter as explained in Section 4.2.11. The actual gas production would occur at rate more than spurts and the average daily gases measured tentatively exhibits similar data with theoretical gas data in UAF reactor for a certain period (Kobayashi, 1983). The daily gas productions of the UAF reactor were between 20 mL and 260 mL with an average of 77 mL in Period 5 at 20°C. The average OLR of Periods 1-3 was 2.74 g sCOD/L.d and decreased to 1.22 g sCOD/L.d at Period 4 (T=35°C) and decreased from 1.22 to 1.49 g sCOD/L.d, at Period 5 (T=20°C) in UAF reactor. Decreases in OLR in Period 4 and Period 5 resulted decreases in gas production as seen in Figures 4.25 and 4.27.a.

The temperature was decreased from 35°C to 20°C in Period 5. This affected the performance of the UAF reactor and average biogas production. This gas production decreased from 237 mL/day in Period 4 to 77 mL/day in Period 5. The average OLRs were 1.22 g sCOD/L.d and 1.49 g sCOD/L.d in Periods 4 and 5, respectively. The OLR in Period 5 exhibited similarities with the OLR in Period 4. Therefore, the reason of decrease in average daily gas production can be explained with the decrease in ambient temperature.

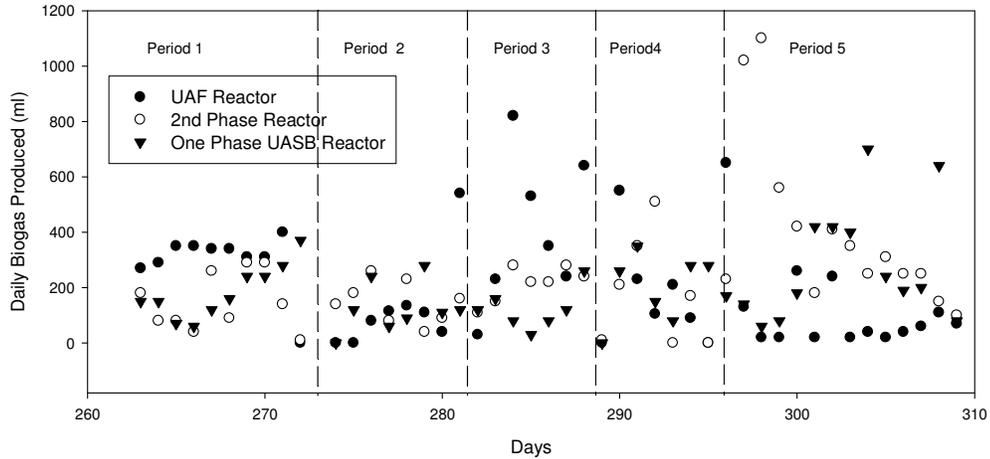


Figure 4.24. Daily Gas Production of UAF, 2<sup>nd</sup> Phase Reactor and One-Phase UASB Reactor.

The averages of biogas produced and the main gas components measured during days 237 and 312 are plotted in Figure 4.25. It was seen that the average carbon dioxide content of the UAF was 22% and the average methane content was 24% in the UAF reactor at 35°C in 2 h HRT. By decreasing the ambient temperature from 35°C to 20°C, the produced total gas decreased by 70% while the methane and CO<sub>2</sub> contents decreased to 14% and 6% respectively. Daily gas production contents of the UAF reactor were plotted in Figure 4.26-a. In this figure, fluctuations in the collected contents of gas volumes can be easily seen. As a result of the decrease in the collected gas volume, contents of methane and CO<sub>2</sub> at temperature 20°C, a sharp decrease in collected volumes of gas fractions are seen in Figure 4.26.a.

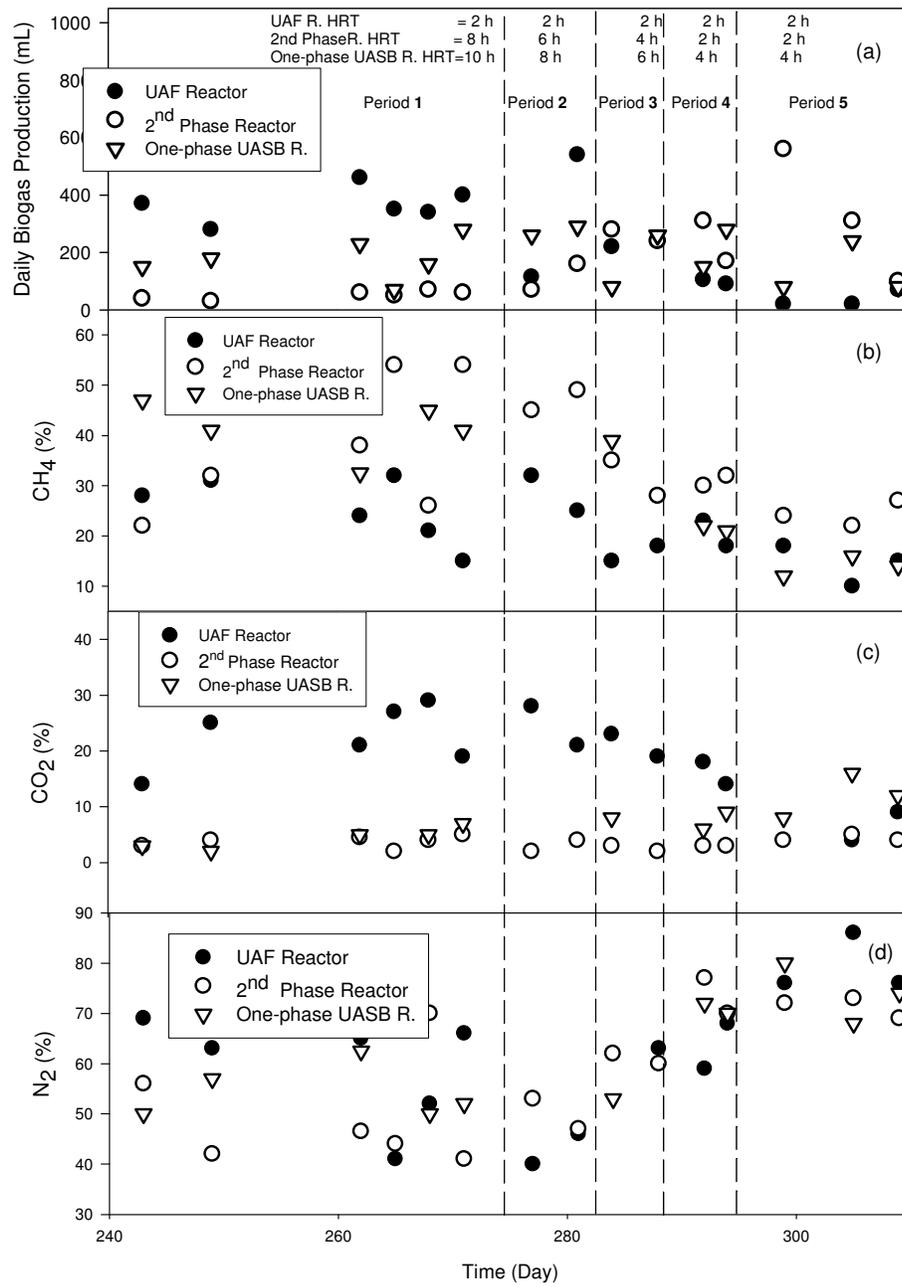


Figure 4.25. Gas production and CH<sub>4</sub>, CO<sub>2</sub> and N<sub>2</sub> percentages in the reactors.

In general, in acidification reactors (i.e. UAF reactor), the methanogenic activity should be retarded. In the present study, a certain amount of methane production was observed in the UAF because the acidification process was provided only by the control of HRT. The most effective separation would be to allow limited time within the acid tank for acidification without having prolonged time for methanification (Jeyaseelan and Matsuo, 1995). On the other hand the HRT should not be short to continue the acidification until the methane tank. It was not possible to inhibit the methanogenic activity completely in the acidogenic UAF reactor since a certain percentage of influent domestic wastewater exhibits acidogenic/acetogenic properties before the collection.

In the 2<sup>nd</sup> phase reactor, the HRTs were 8 h, 6 h, 4 h and 2 h in Periods 1, 2, 3 and 4, respectively at 35°C. In Period 5, the HRT was 2 h and the room temperature was 20°C. The average OLRs were 0.58, 0.70, 0.53, 0.38 and 0.46 g sCOD/L.d in Period 1, Period 2, Period 3, Period 4 and Period 5, respectively. Although the HRT of the 2<sup>nd</sup> phase reactor decreased stepwise from 8 h (Period 1) to 2 h (Period 4/ Period 5), the OLR of the 2<sup>nd</sup> phase reactor did not increase at a certain rate. The influent strength of domestic wastewater of the 2<sup>nd</sup> phase reactor depends on the performance of the UAF reactor (the effluent of the UAF reactor was the influent of the 2<sup>nd</sup> phase reactor).

In the 2<sup>nd</sup> phase reactor, it was observed that there was an increase in daily gas production with the decrease in HRT as seen in Figure 4.24. The averages of daily gas production were calculated as 146 mL/day, 148 mL/day, 214 mL/day, 208 mL/day and 360 mL/day in Periods 1, 2, 3, 4 and 5, respectively. When the contents of the gas were considered, methane percentage decreased from 43% in Period 1 (HRT=8 h) to 31% in Period 4 (HRT=2 h) and from 31% in Period 4 (T=35°C) to 24% in Period 5 (T=20°C). The ANOVA statistics showed the relationships between

decrease in methane percentage and decrease in HRT was not significant in 2<sup>nd</sup> phase reactor at 35°C ( $F(3,9)=0.1397$ ,  $p=0.314 > 0.05$ , Table B.14, Appendix B). As the HRT was decreased, the methane percentage was not changed in the 2<sup>nd</sup> phase reactor. The carbon dioxide content of the 2<sup>nd</sup> phase reactor was between 3% and 4% throughout the Periods 1-5 of the 2<sup>nd</sup> phase reactor.

As a conclusion, it was observed decreases in methane percentages of collected gas with the decrease in HRT and decrease in reactor temperature. However the variation in CO<sub>2</sub> fraction was not significant in the 2<sup>nd</sup> phase reactor since the HRT and temperature variations affected methanification in the 2<sup>nd</sup> phase reactor.

In the UASB reactor, the HRTs were 10 h, 8 h, 6 h and 4 h in Periods 1, 2, 3 and 4, respectively at 35°C. In Period 5, HRT was 4 h while the reactor temperature was 20°C. The average OLRs were 0.49, 0.77, 0.90, 0.61 and 0.74 g sCOD/L.d in Periods 1, 2, 3, 4 and 5, respectively. Although the HRT of the UASB reactor decreased in Period 4 and Period 5, the OLR of the UASB reactor did not increase because of the influent strength of domestic wastewater decreased during Periods 4 and 5. The average daily collected gas volumes in the UASB were 184 mL/day, 128 mL/day, 121 mL/day, 187mL/day and 301 mL/day in Periods 1, 2, 3, 4, and 5, respectively. But when the methane percentages were considered there was a decrease in methane fraction with the decrease in HRT (37%, 44%, 36%, 22% and 14% CH<sub>4</sub>, in Periods 1, 2, 3, 4 and 5, respectively). One-way ANOVA and Post HOC Tests (TUKEY & SCHEFFE) indicated that the relationships between decrease in methane percentage of biogas was significant with the decreases of HRT in one-phase reactor at 35°C ( $F(3,11)=14.252$ ,  $p=0.036$ , Table B.15, Appendix B). As the HRT decreased from 10 h to 8 h to 6 h and 4 h in one phase UASB system, the methane percentages decreased. Although the OLR increased by lowering the HRT stepwise in the UASB reactor, total collected volume of

CH<sub>4</sub> gas decreased as depicted in Figure 4.26-c (daily average CH<sub>4</sub> volume decreased from 71 mL to 46 mL in 10 h HRT to 4 h HRT). The methane percentage of the UASB decreased from 22% to 14% by lowering the room temperature from 35°C to 20°C. Therefore, it was observed a decrease in the total volume of collected methane when the HRT was decreased. The total gas lowered from 46 mL to 20 mL by lowering the temperature from 35°C to 20°C. The effect of temperature in methanogenic activity was considerably high and the methane production decreased by decreasing the room temperature from methanogenic to psychrophilic level. The CO<sub>2</sub> fraction increased from 6% (Period 1) to 8% (Period 4) and 8% (Period 4) to 12% (Period 5) in the UASB reactor. Carbon dioxide is a by-product of acidification phase and the increase in CO<sub>2</sub> fraction with the decrease in HRT could be attributed to comparably higher acidification degree than that methanification step at lower temperatures and at lower HRTs in UASB reactor (Cha and Noike, 1997).

Comparison of gas collection level and gas composition of one-phase UASB system and two-phase UAF and UASB system:

A summary of average daily methane gas production at the presented HRTs and OLR are seen in Table C 6 (Appendix C). In Periods, 1 and 2, OLRs of the 2<sup>nd</sup> phase reactor and the UASB reactor were about the same. However, the methane gas collected is more in the one-phase UASB reactor because a certain amount of methane was collected in the first reactor (UAF) of the two-phase system. Although the OLR of one-phase UASB reactor increased in Periods 3 and 4, CH<sub>4</sub> produced in the one-phase UASB reactor was less than the 2<sup>nd</sup> phase reactor. The characteristics of influent wastewater showed variations in Periods 3 and 4. The total gas and methane production was comparably higher in the 2<sup>nd</sup> phase reactor than that the UAF reactor. In the one-phase UASB reactor, the decrease in HRT (8

h to 4 h) resulted decreases in the methane gas production (OLR increased at that period). Any negative effect in daily methane gas production was not observed by decreasing the HRT from 8 h to 2 h in the 2<sup>nd</sup> phase reactor. Decreases in temperature from 35°C to 20°C affected the UAF and the one-phase UASB reactor. The daily methane productions of the UAF and the one-phase UASB dropped down to about ¼ (from 20 mL/day to 5 mL) and ⅓ (46 mL/day to 20 mL) the methane production at the same HRT, respectively.

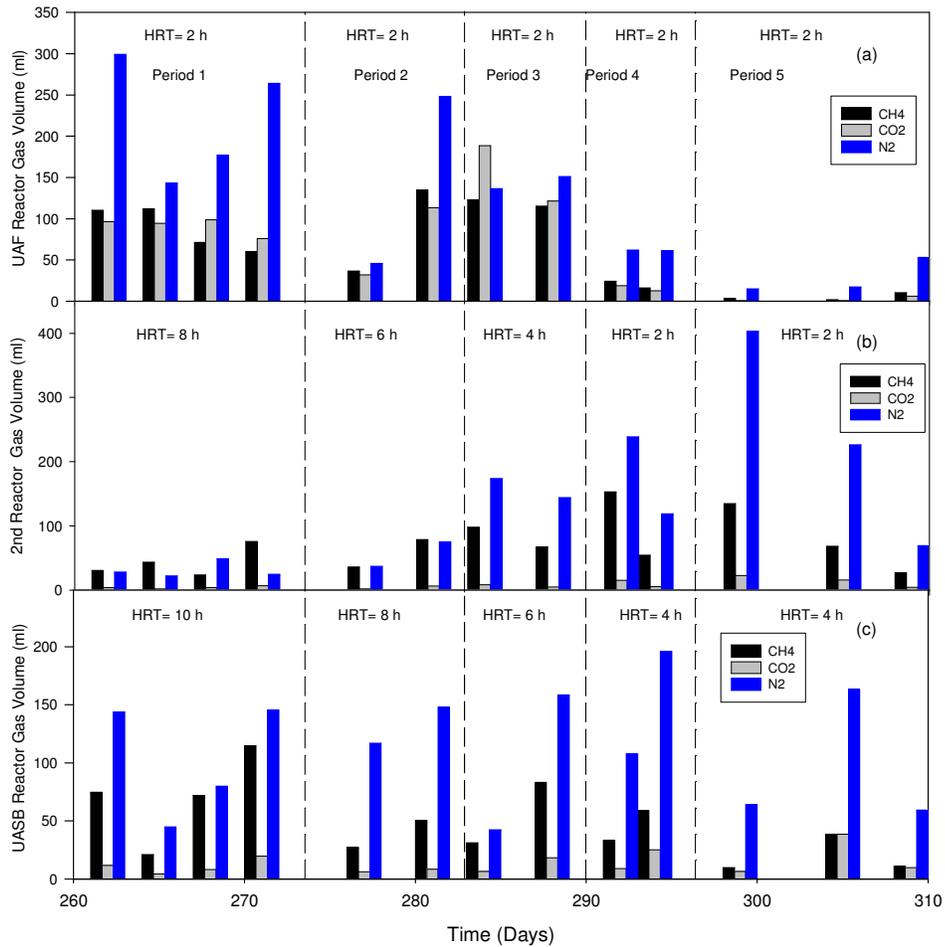


Figure 4.26. Daily gas production contents of UAF reactor, 2<sup>nd</sup> phase reactor and one-phase UASB reactor.

As a summary, decreases in HRT and temperature resulted in insignificant changes in daily methane production in the 2<sup>nd</sup> phase reactor. In the UASB reactor, the increase in the OLR (from 0.49 g sCOD/L.d in 10 h HRT to 0.77 g sCOD/L.d in 8 h HRT) was resulted with increases in methane gas production. But further increases in OLR with decreases in HRT (8 h to 4 h) showed reductions in methane production. The main factor affecting the methane gas production was HRT for the HRTs less than 8 h in one-phase reactor (UASB reactor). The temperature decrease from 35°C to 20°C had a negative effect in gas productions of the UAF reactor and UASB reactor while no effect was observed in gas productions of the 2<sup>nd</sup> phase reactor. The CH<sub>4</sub>, CO<sub>2</sub> and N<sub>2</sub> fractions of the UAF, the 2<sup>nd</sup> phase reactor and the UASB reactor are summarized in Table C 5 (Appendix C)

The CH<sub>4</sub> fraction of the UAF was considerably lower than the 2<sup>nd</sup> phase reactor and the UASB reactor. The CO<sub>2</sub> fractions of the UAF reactor were considerably higher than the 2<sup>nd</sup> phase reactor and the one-phase UASB reactor. The high CO<sub>2</sub> and very low CH<sub>4</sub> fractions are important features of acidogenic reactors as reported by Fang and Yu (2001).

Methane percentages were about the same in Periods 1-4 but the CO<sub>2</sub> content was higher in the UASB reactor than that 2<sup>nd</sup> phase reactor in all HRTs. This can be explained with the acidogenesis/ acetogenesis phases which were in progress in the one-phase UASB reactor during the Periods 1- 4. At 20°C, the methane percentages of the UAF reactor and the one-phase UASB reactor decreased significantly while the methane percentage of the 2<sup>nd</sup> phase reactor showed a slight decrease in the average. The CO<sub>2</sub> content of the UAF reactor decreased about 1/3 at 20°C (7%) of its average CO<sub>2</sub> value measured at 35°C (16%). The major part of the CO<sub>2</sub> formed might have been present in dissolved form in

wastewater for lower temperatures. Singh et al. (1996) observed that the CO<sub>2</sub> content of the gas was very low at lower temperatures and this was due to increase in solubility of CO<sub>2</sub> because of the decrease in temperature. In the present case, the reason of the decrease of CO<sub>2</sub> content at 20°C can be explained with the increase in solubility of CO<sub>2</sub> because of decrease in temperature.

In the study performed by Lettinga et al. (1983), GC analysis performed in methanogenic UASB indicated that the gas contained only 2-4% CO<sub>2</sub>, but the fraction of nitrogen was relatively high dependent on the gas produced. It can be concluded that the higher gas production per unit of volume of sewage treated, the N<sub>2</sub> content is lower and the CH<sub>4</sub> content is higher. The gas fraction results in the 2<sup>nd</sup> phase reactor of the present study were compatible with statement of Lettinga et al. (1983). Lettinga et al. (1983) provided an explanation for the nitrogen in the biogas originated from the dissolved nitrogen in the influent solution; it was stripped from the liquid phase by the methane gas produced in the fermentation process. In one-phase UASB reactor, it was observed a decrease in methane production in Period 4 and especially in Period 5. An increase in N<sub>2</sub> content of biogas can be explained with the less CH<sub>4</sub> production. The increase in N<sub>2</sub> fraction of biogas in the UAF reactor at 20°C can be explained with the reduction in gas production at that period. This result was compatible with the literature results above.

The methane content of average daily produced gas was calculated at STP and divided by COD removed at that period in order to calculate the methane production rate. The conversion rate of COD removed to methane as shown in Table 4.9 were between 0.005 and 0.067 Nm<sup>3</sup>CH<sub>4</sub>/ kg COD removed in the UAF, between 0.158 and 0.233 Nm<sup>3</sup> CH<sub>4</sub>/ kg COD removed in the 2<sup>nd</sup> phase reactor and between 0.029 and 0.199 Nm<sup>3</sup>CH<sub>4</sub>/ kg COD removed in the one-phase UASB

reactor. The reply of why conversion rate of COD removed to methane of the UAF were so low can be explained with the dissolved methane losses in the effluent (Yu and Anderson, 1996). In the study of Gnanadipathy and Polprasert (1993), it was observed that the amount of CH<sub>4</sub> gas produced per kg of COD removed in UASB reactors was the lowest in the reactor which had lowest biomass concentrations in the reactor. Considering the UAF had the lowest amount of biomass in the reactor, the low conversion rate of COD removed to methane is also compatible with the literature.

Table 4.9. Conversion Rate of COD Removed to Methane

	CH <sub>4</sub> conversion rate (N m <sup>3</sup> CH <sub>4</sub> /kg COD removed)		
	UAF	2nd Reactor	UASB
Period 1	0.036	0.217	0.199
Period 2	0.019	0.158	0.079
Period 3	0.067	0.233	0.135
Period 4	0.011	0.202	0.045
Period 5	0.005	0.113	0.029

The conversion rate of COD removed to methane of the methanogenic reactors, 2<sup>nd</sup> phase reactor and one-phase UASB reactor, are comparable with the values presented by other researchers (Yu and Anderson, 1996; Lettinga et al., 1993; Mergeart et al., 1992; Vieira and Garcia, 1992; Collivignarelli, 1990; Schellinkhout and Collazes, 1992; Marango and Campos, 1992). For example, Yu and Anderson (1996) gave conversion rate of COD removed to methane between

0.09-0.12 Nm<sup>3</sup> CH<sub>4</sub>/ kg COD removed. Lettinga et al (1993) indicated that CH<sub>4</sub>-gas production rate was 0.19 Nm<sup>3</sup>/ kg COD removed since the diluted properties of sewage and in sewage wastewaters, generally more than 50% of the methane would leave the reactor with the effluent solution.

However, the conversion rate of COD removed to methane of the 2<sup>nd</sup> phase reactor and the one-phase UASB reactor were smaller than the theoretical value of 0.35 m<sup>3</sup>.CH<sub>4</sub> /kg.COD removed (Metcalf and Eddy, 1991). The probable reasons for the relatively low amount of methane produced compared to COD removal could be attributed to the dissolved CH<sub>4</sub> gas in the effluent overflows and possible gas leakage in the collection system. Other investigators also reported lower CH<sub>4</sub> production rates in the UASB reactors than the theoretical values: Barbosa and Sant'Anna, (1989); Fernandes et al., (1985); Singh et al., (1996); Gnanadipathy and Polprasert (1993). When the results of conversion rate of COD removed to methane were compared, methane recovery per COD removal was considerably high in the 2<sup>nd</sup> phase UASB reactor than that the one-phase UASB reactor.

It was frequently reported that a UASB reactor could perform a good removal of organic matter without an effective production of biogas for organic loadings up to 1.99 g.COD/L.d (Kalogo and Verstrate, 1999; Kalogo et al., 2001). As an economical conclusion, the treatment of low-strength wastewater (such as domestic wastewater); reduces the essential advantage of an anaerobic process as an energy- recovery system. The average amount of methane produced (liters at STP) for COD removed (liters at STP) for COD removed (kg-COD) for whole experimental period was 185 L/kg.COD removed and 98 L/kg.COD removed for the 2<sup>nd</sup> phase reactor and the one-phase UASB reactor, respectively.

### 4.3.6 Comparison of the Specific Methanogenic Activity (SMA) of UASB Reactors

The presence of viable methanogens in the initial, final sludge samples and methane gas production of granules were determined with specific methanogenic activities (SMA) experiments (Speece, 1996). The SMA of sludge samples in this study are shown in Table 4.10. The SMA of sludge samples of the 2<sup>nd</sup> phase reactor and the UASB are compatible with the published results of Lettinga et al (1981), Barbosa and Sant'Anna Jr (1989), Grin et al (1983) and Schellinkhout et al (1985) which were in the range of 0.1-0.25 kg COD-CH<sub>4</sub>/ kg VSS.d.

The SMA of one-staged and two-staged UASB reactors were compared in the treatment of municipal wastewater (El-Ghory, 1999). El-Ghory (1999) reported that the SMA of the first reactor ranged between 0.144 and 0.219 g CH<sub>4</sub>-COD/ g VSS.d, while the SMA of the 2<sup>nd</sup> phase reactor ranged between 0.163 and 0.297 g CH<sub>4</sub> COD/ gVSS.d. The SMA of one-staged reactor was given as 0.21 g CH<sub>4</sub>-COD/ g VSS.d in that study.

Table 4.10. Specific Methanogenic Activity (SMA) of UASB reactors

Reactor	SMA (kg CH <sub>4</sub> -COD/kg VSS.d)
Granular sludge used in UASB reactors at start-up	0.282
Acidogenic UASB (used in acidification system)	0.086
2 <sup>nd</sup> phase reactor ( used in two-phase system)	0.253
UASB (used in one-phase system)	0.222

The one-phase UASB reactor SMA result of the present study (0.222 g CH<sub>4</sub>-COD/ gVSS.d) was very similar to the one-stage UASB reactor SMA results of

El-Ghory (1999) (0.21 g CH<sub>4</sub>-COD/ g VSS.d). The 2<sup>nd</sup> phase UASB reactor SMA result of the present study (0.253 g CH<sub>4</sub>-COD/gVSS.d) was in the range given for the second reactor of the two-stage study (El-Ghory, 1999). The SMA results of the UASB reactors for one-phase and two-phase system were compatible with the results in the literature. The SMA results of the acidogenic UASB were lower than the given literature results. Being the SMA results of the acidogenic UASB lower than those obtained in the 2<sup>nd</sup> phase UASB (methanogenic phase) and one-phase UASB reactor was reasonable and it showed that it was provided better methanogenic conditions at the 2<sup>nd</sup> phase UASB and one-phase UASB reactor than that acidogenic UASB reactor. The SMA of the 2<sup>nd</sup> phase reactor (0.253 kg CH<sub>4</sub>-COD/kgVSS.d) was higher than one-phase UASB reactor (0.222 kg CH<sub>4</sub>-COD/kgVSS.d). It proved that the 2<sup>nd</sup> phase reactor was superior in terms of methanogenesis than the one-phase UASB reactor.

#### **4.3.7. The Variations of Suspended Solids (SS) and SS Removals in One-phase and Two-phase Reactors**

The suspended solid (SS) concentrations in influent, effluent and the removal efficiencies of the reactors at each HRT are presented in Figure 4.27. The influent TSS and VSS concentrations and the HRTs are presented in Figures 4.27.a and 4.27.b, respectively. The influent VSS and TSS values displayed variations and it was observed peaks in VSS and TSS values in Period 2. When the influent tCOD values in Period 2 are compared, it can be seen that the tCOD of the influent was comparably higher than the other studied HRTs.

The average SS values in influent, effluent and removal efficiencies can be seen in Appendix C-Table C7. The influent values of TSS were higher than VSS and the

difference varied between 35-78 mg/L with the exception of the Period 3. In Period 3, the difference between influent TSS and VSS values was 203 mg/L. It could be considered that the biodegradable part of the influent domestic wastewater was lower in Period 3. When the difference between tCOD and sCOD values (Table 4.8 Section 4.3.1) and variations in BOD<sub>5</sub> (Table C2- Appendix C) are considered, it can be seen the ratio of sCOD to tCOD was highest in Period 3 and the influent BOD<sub>5</sub> values were about the same between the Periods 1 and 3. Therefore it was considered that it could be an experimental error in Period 3 and the results were not compared in this study (the results of the Period 3 is shown in Figure 4.27).

In the UAF reactor, the VSS decreases were very close in Periods 1, 2 and 4 (Figure 4.27.c). The average VSS decreases were between 38% and 68% with an average of 49% in Periods 1, 2 and 4 at 35°C in the UAF reactor. It was observed about 21% decreases in VSS reductions with the decrease in ambient temperature (from 35°C to 20°C) in the UAF reactor.

In the effluents of the 2<sup>nd</sup> phase UASB reactor (effluents of two-phase system), it was observed increases in effluent VSS and TSS values with the decreases in HRT from 8 h to 4 h at 35°C (Figure 4.27.d). The effluent VSS values were 24 mg/L, 29 mg/L and 33 mg/L for Period 1 (HRT=8 h), Period 2 (HRT=6 h) and Period 4 (HRT=4 h), respectively at 35°C. The effluent TSS values were 30 mg/L, 38 mg/L and 44 mg/L for Period 1 (HRT=8 h), Period 2 (HRT=6 h) and Period 4 (HRT=4 h), respectively at 35°C. When the effluent VSS and TSS values of Period 4 and 5 were compared the effluent VSS and TSS concentrations slightly increased with the temperature decrease from 35°C to 20°C (HRT=4 h).

The VSS reductions and TSS removal efficiencies of the 2<sup>nd</sup> phase UASB reactor and the two-phase system are presented in Figure 4.27.d. In the 2<sup>nd</sup> phase UASB reactor, the VSS reductions and TSS removals mostly depended on the influent VSS and TSS concentration (effluent of the UAF reactor). When the VSS and TSS influent concentrations of the 2<sup>nd</sup> phase reactor were high, the VSS reductions and TSS removal efficiencies of the 2<sup>nd</sup> phase reactor were high (see Table 4.18).

In the two-phase UAF and UASB system, the average TSS removal efficiencies were 77% and 91% at Periods 1 and 2, respectively. The VSS reductions were 74% and 91% at Periods 1 and 2 in two-phase system. The VSS reductions and TSS removal efficiencies were 83% at Period 4 (HRT=4 h). The decrease in VSS reductions and TSS removal efficiencies with the variation of HRT from 8 h to 4 h was low (about 8% in VSS and in TSS). It was not observed a significant variation in VSS reductions and TSS removal efficiencies as the ambient temperature decreased from 35°C to 20°C in two-phase system. The small variations in TSS removal efficiencies can be attributed to the variations in the influent TSS concentrations.

In one-phase UASB reactor, it was observed a stepwise increase in VSS and TSS effluents with the decreases in HRT from 10 h (Period 1) to 4 h (Period 4) and with the decrease in temperature from 35°C (Period 4) to 20°C (Period 5). The average VSS effluents were 22 mg/L, 56 mg/L, 88 mg/L and 73 mg/L while the average TSS effluents were 44 mg/L, 92 mg/L, 127 mg/L and 103 mg/L for Period 1, Period 2, Period 4 and Period 5, respectively. As reported by Yu and Anderson (1996), the effluent SS concentration increased with the increase in HRT. The reason of the decrease in effluent VSS and TSS values in Period 4 and Period 5 can be explained with the decrease in influent strength (influent TSS

concentration) of wastewater. The average TSS removal efficiencies was and 71%, respectively, for Period 1 (HRT=8 h) and Period 2 (HRT=10 h). The removal efficiency in TSS was 50% at Period 4 (HRT=4h, T=35°C) and decreased to 47% at Period 5 (T=20°C). Schellinkhout et al (1993) reported the results of raw sewage treatment at an HRT of 5.2 h and they obtained 69% TSS removal. Lettinga et al. (1993) reported the TSS treatment efficiencies were 67-81% at HRT=5-6 h and at a temperature of 24-26°C in UASB reactors. When compared the TSS removal efficiencies (average 71% at 8-10 h) of the present study with the given examples by Schellinkhout et al. (1993) and Lettinga et al. (1993) the TSS removals were 69% and 67-81% in the present study. This shows that the present study results were consistent with the literature.

The SS removal efficiencies of UASB reactors were reported as 72%, 70% and 76% by Barbosa and Sant'Anna (1989) Garcia et al (1998) and Kalogo et al (2001), respectively, at a HRT of 4 h and temperature of 10-28°C. For an influent TSS value of 188 mg/L, Vieira (1984) mentioned that the TSS removals were 61% at a HRT of 4 h and at 35°C. When the results of this work are compared to those reported above, it is possible to conclude that the TSS removal efficiencies are lower or comparable with the results performed by other investigators.

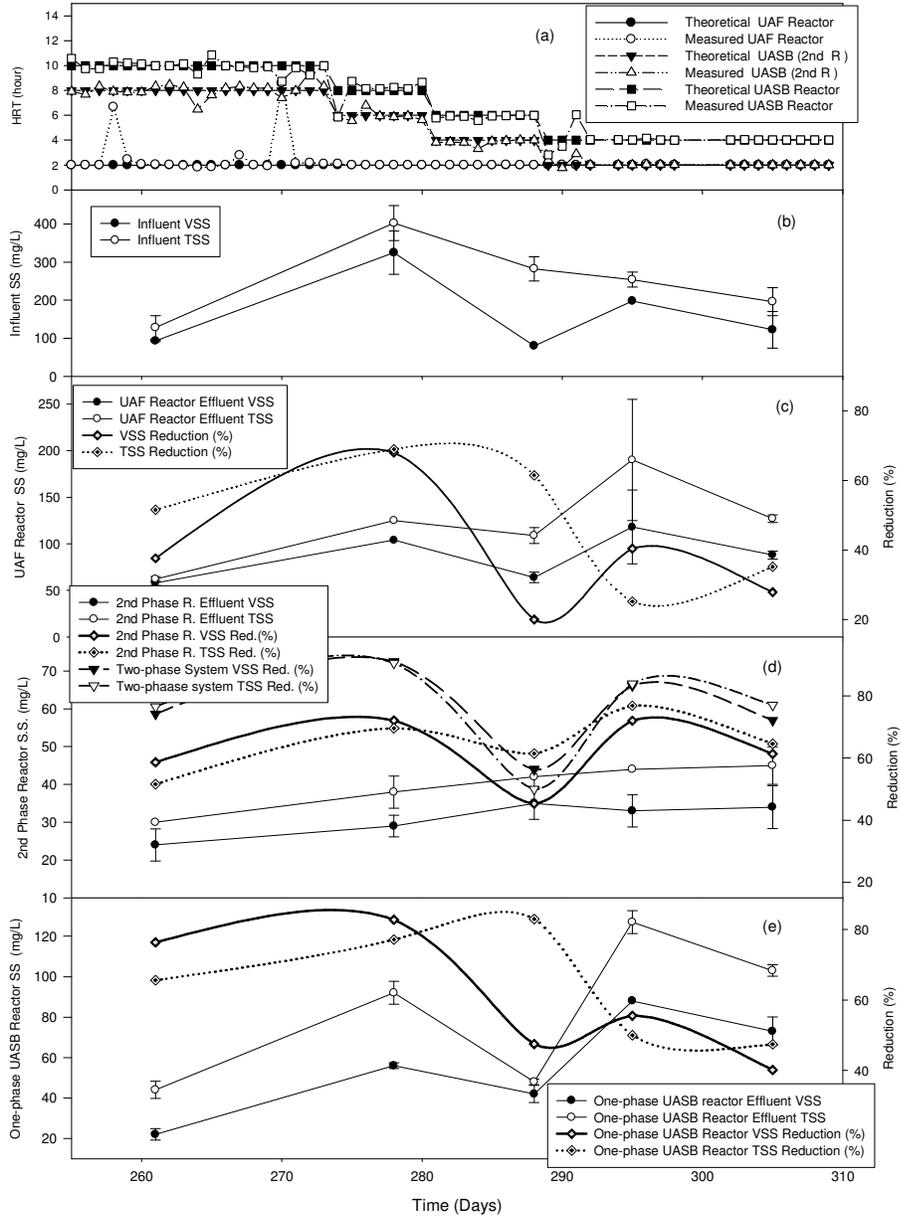


Figure 4.27. Influent, effluent and removal percentages for volatile suspended solids (VSS) and total suspended solids (TSS) at different HRTs of UAF reactor, 2<sup>nd</sup> phase UASB reactor, two-phase system and one-phase system.

Barbosa and Sant'Anna Jr (1989) concluded that the suspended solids in the reactor effluents were low and independent of the usual variations observed in influent suspended solids content (Barbosa and Sant'Anna Jr, 1989). But in this study, the results were different to the data obtained by Barbosa and Sant'Anna Jr (1989). For example, at longer HRTs lower TSS removals were resulted for a lower influent TSS value of about 128 mg/L at 10 h HRT.

The comparison of the reactors are presented in Figures 4.28.b and 4.28.c, for VSS and TSS, respectively at different HRTs (Figure 4.28.a). The VSS reductions of the UAF reactor was considerably lower than the one-phase UASB reactor and two-phase UASB reactor because the HRT of the UAF was fixed to 2 h during the whole study. In VSS reductions, it was obtained higher results in the one-phase system than that two-phase system in Period 1. However, in Periods 2, 4 and 5 the performance of two-phase system was better. The VSS reductions of one-phase system was significantly lower (32%) than two-phase system in Phase 5. Higher TSS removals (77%) for higher influent TSS value of about 403 mg/L was obtained at 8 h HRT in one-phase UASB reactor system was obtained.

In the comparison of two-phase system and one-phase system, it was observed very high TSS removals during the Periods 1-5 as seen in Table 4.18. In Period 4 (HRT=4 h, T= 35°C), and Period 5 (HRT=4 h, T=20°C), the TSS removals were 32% and 30% higher in two-phase system than that one-phase system. It can be concluded that the reductions of VSS and TSS were significantly higher in two-phase system than one-phase system at lower temperatures and HRTs like 4 hours.

During the study, domestic wastewater was directly taken from the collection point of the sewer line and it was not pre-settled. Although there was no pre-

settlement unit, the one-phase and two-phase systems at Period 1 (HRT= 10 h) satisfied the receiving water discharge standards of Turkish Regulation (TSS  $\leq 45$  mg/L, SKKY: 2005) as defined for cities with a population of 84-1000. In Set 2 for total HRT  $\leq 8$ h, the receiving water criteria of Turkish Regulation could not be met in one-phase reactors. However, the two-phase system effluents in the Periods 1-5 satisfied the requirements of above mentioned Turkish Regulations.

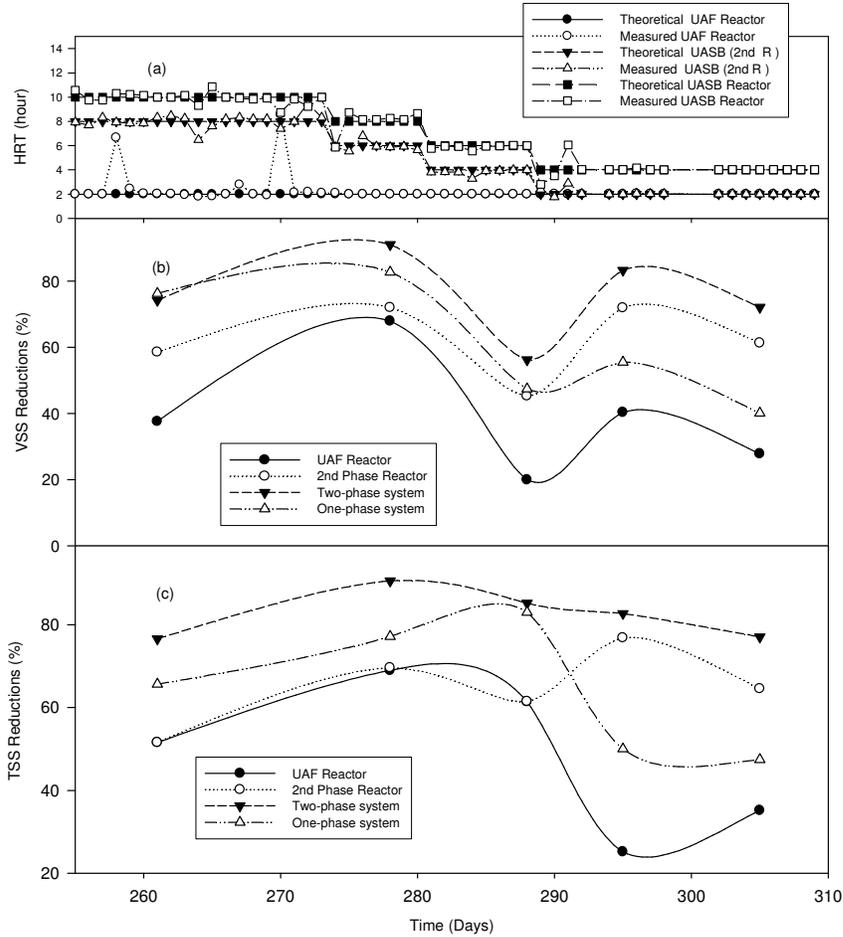


Figure 4.28. Comparison of volatile suspended solids (VSS) and total suspended solids (TSS) reductions at different HRTs in one-phase and two-phase systems.

#### **4.3.8. The Variations of Total Solids and Total Volatile Solids in Two-Phase and One-phase Reactors**

The average Total Volatile Solids (TVS) and Total Solids (TS) values in influent, effluent samples and reductions are presented in Table C 8 (Appendix C). In this Table, the first column gives the studied periods, which were categorized according to HRTs and temperatures applied to the reactors. The HRTs of each reactor are shown in Figures 4.29.a, 4.30.a and 4.31.a for UAF reactor, two-phase system and one-phase system, respectively. The ambient temperatures were 35°C and 20°C in Periods 1-4 and 5, respectively.

The TS and TVS values were between 990-1222 mg/L and 159-239 mg/L in the influent of UAF and the UASB reactors, respectively. Figure 4.29.b and 4-29.c present the TVS and TS concentrations and reductions, respectively in UAF reactor. The TS removal efficiencies in UAF reactor were between 11% and 32% with an average of 20% at a HRT and temperature of 2 h and 35°C, respectively. This removal efficiency decreased to 8% at 20°C. The average of TVS reductions in UAF reactor was 23% at 35°C and decreased to 7% at 20°C. The average decreases in TS and TVS concentrations were calculated as 225 mg/L and 53 mg/L at 35°C in the UAF reactor. The reasons of decrease in TS may be explained with the ending of hydrolysis, acidification and methanification phases resulting in adsorption of soluble solids by microorganisms and entrapment of particular solids in interstitial spaces in the UAF reactor. The average reductions in TS and TVS concentrations in effluent of the UAF reactor were 102 mg/L and 11 mg/L at a temperature of 20°C. It was observed decreases in TS removal efficiencies with the decrease in temperature from 35°C to 20°C. Decreases in TS in low temperatures can be explained with the decrease in methanogenic activity because of decrease in temperature. A better solids removal was obtained at 35°C than at

20°C at 2 h HRT. Therefore it can be concluded that there was some methanogenic activity in the UAF reactor at 35°C.

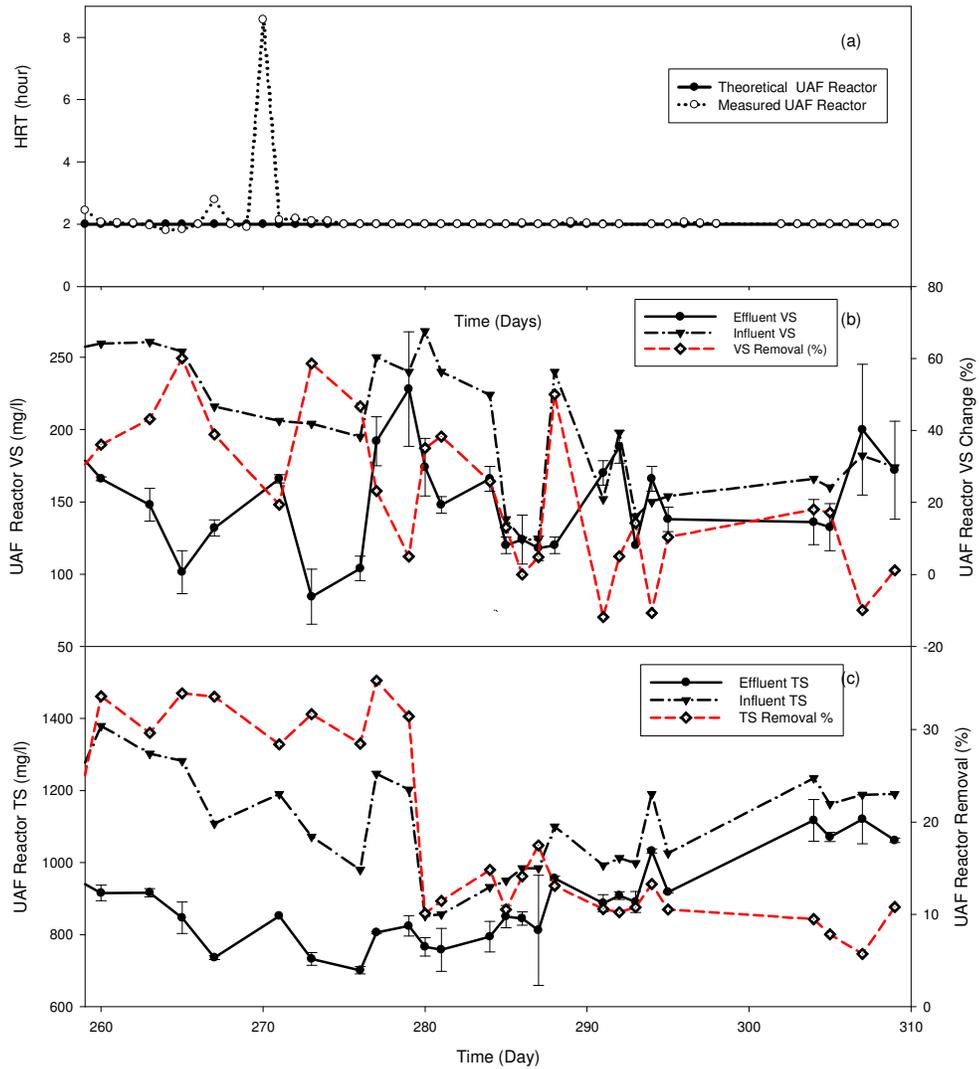


Figure 4.29. Total Volatile solids (TVS) and total solids (TS) concentrations in influent, effluent samples TVS changes and TS removal efficiencies in UAF reactor.

The 2<sup>nd</sup> phase reactor influent, effluent TVS, TS concentrations and TS, TVS removal efficiencies of two-phase system and the 2<sup>nd</sup> phase UASB reactor are presented in Figures 4.30.b and 4.30.c. It was observed a tendency of decrease in TS removal performances in two- phase UAF and UASB reactor system with the decrease in HRT. The TS removals were 33%, 31%, 24% and 20% at HRTs 10 h, 8 h, 6 h and 4 h, respectively at 35°C. The average TS removal efficiency decreased from 20% to 16% with the decrease of temperature from 35°C to 20°C. The small decreases in TS removals could be attributed to the decrease of hydrolysis with decreases in HRT and temperature as mentioned by Lew et al (2004), Man et al (1986), Lettinga et al (1996). Decreases in TVS reductions were observed during the Period 4. The reason of decreases in the TVS reductions on days between 291 and 293 can be explained with the decreases in influent TVS values of raw wastewater.

In two-phase UAF and UASB reactor system, the TVS reductions is compared with the sCOD reductions, VFA production and produced CH<sub>4</sub> percentages at different HRTs at 35°C whether there is a correlation between aforementioned parameters. The multiple regression and ANOVA test statistics were performed between TVS reduction and sCOD reduction, VFA production and CH<sub>4</sub> percentages at decreasing HRTs in two-phase reactor (Appendix B, Table B 21). It was found that there is a strong correlation between sCOD reduction, CH<sub>4</sub> percentage and TVS reduction ( $R^2=0.96$ , adjusted  $R^2=0.84$ ,  $F(3,9)=75.56$ ,  $p=0.029$ ). The correlations between TVS reduction, sCOD reduction, CH<sub>4</sub> percentages and HRT was significant. At longer HRTs, sCOD reductions and CH<sub>4</sub> content of biogas was higher and related to TVS reductions. The TVS reductions decreased with the decreases in HRT in two-phase reactor system. This can be explained with insufficient time for hydrolysis and acidogenesis and methanogenesis for the reduction of TVS at short HRTs.

In one-phase UASB reactor, the regression analysis and ANOVA test statistics showed that there was a strong correlation between TVS reduction and sCOD reduction, VFA production and CH<sub>4</sub> percentages at decreasing HRTs (Appendix B, Table B 22). The regression equation showed that there is a strong correlation between VFA production, sCOD and TVS percent reductions ( $R^2 = 0.95$ , adjusted  $R^2 = 0.81$ ,  $F(3,8) = 45.94$ ,  $p < 0.05$ ). Furthermore, the differences between these parameters are significant.

The one-phase UASB reactor influent, effluent TVS, TS concentrations and TS, TVS reductions are presented in Figures 4.31.b and 4.31.c, respectively. In the UASB reactor, the average TS removals were about 32% at 10 h and 8 h HRT and decreased to 20% and 11% at 6 h and 4 h HRT, respectively at 35°C. A considerable reduction in TS removal efficiencies was observed (66%) with the decrease in HRT from 8 h to 4 h at 35°C. The change in TS removal efficiency (from 11% to 13%) was not significant with the room temperature decrease from 35°C to 20°C at 4 h HRT. Therefore, it can be concluded that the decreases in TS removals could be attributed to the decreases in HRT.

In the comparison of the TS removal efficiencies of the two-phase UAF and UASB system and one-phase UASB system, the TS removals were about the same at the HRTs of 10 h and 8 h in both of the reactors. However, the TS removals of the two-phase system were 4% and 9% higher than that one-phase reactor at HRTs 6 h and 4 h, respectively at 35°C.

As the HRT decreased from 10 h to 4 h, the CH<sub>4</sub> percentage, VFA production, sCOD reductions decreased from 43 to 31%, from 31 to 11 mg/L, from 48 to 38%, from 77 to 48% and from 50 to 27%, respectively, in two-phase UAF and UASB system. As the HRT decreased from 10 to 4 h, the CH<sub>4</sub> percentage, VFA

production and sCOD reduction decreased from 37 to 22%, from 19 to 11 mg/L, from 79% to 27% and 49% to 33% in one-phase UASB system (see Table 4.8 and Table C 3, Table C 5, Table C 8, Appendix C). At low HRTs, there is not enough time for methane production by methanogens and for hydrolysis of organics and VFA production, for removal of sCOD and volatile solids. At lower temperatures, the methane percentage of total gas, VFA production and TVS reductions are lower compared at 35°C in one-phase and two-phase system. This could be explained with low activity in methanogenic bacteria, low hydrolysis and acidification and low TVS reductions. At low temperatures, the sCOD concentrations did not change significantly, this could be explained with lower hydrolysis rate at low temperatures.

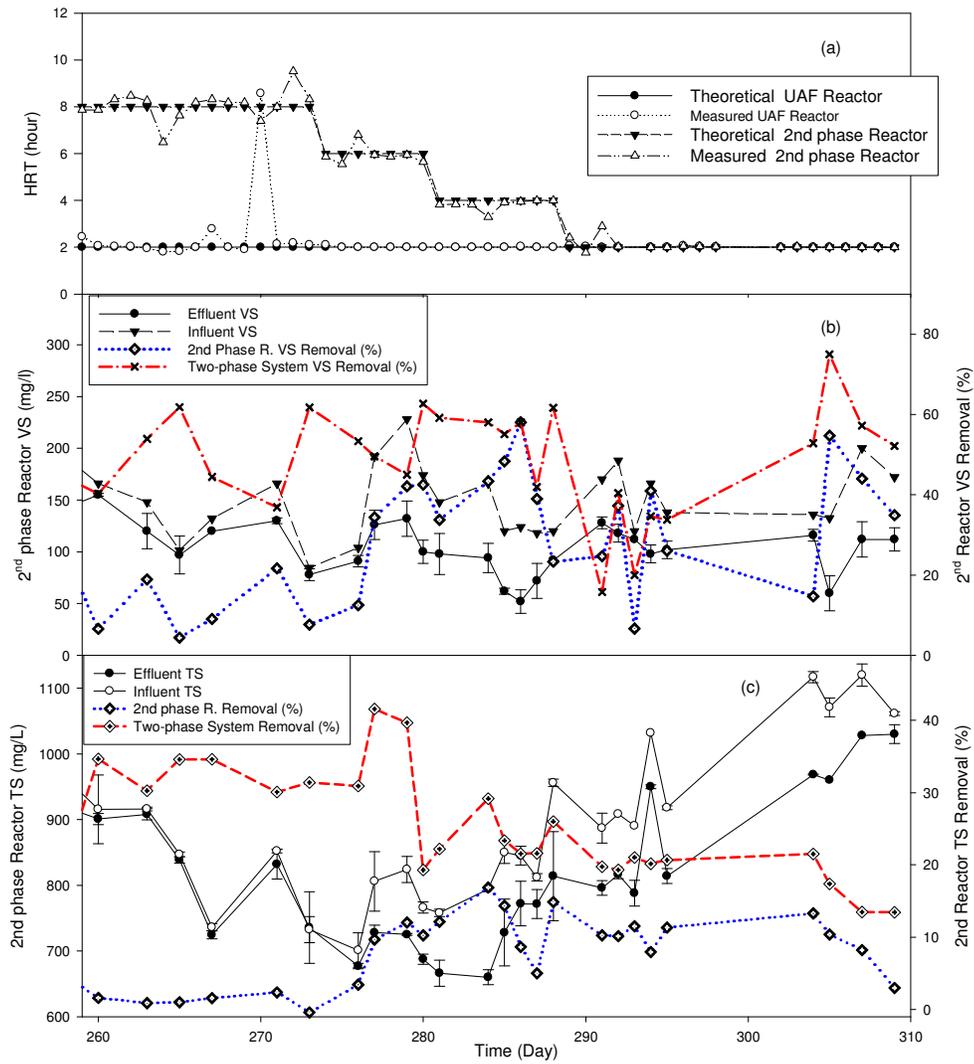


Figure 4.30. Volatile Solids (TVS) and total solids (TS) concentrations in influent, effluent samples and removal efficiencies in 2<sup>nd</sup> phase UASB reactor reactor and two-phase UAF and UASB system versus operation days at studied HRTs.

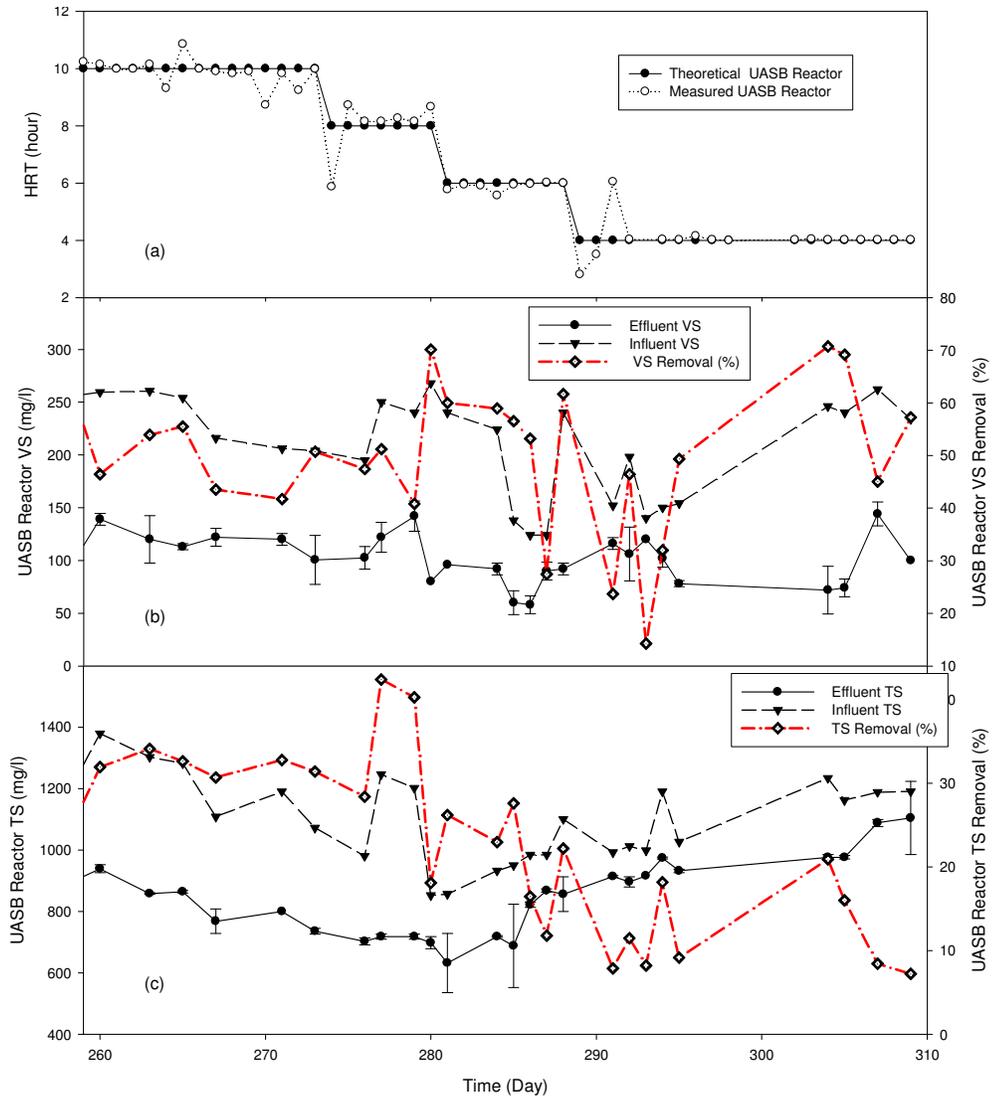


Figure 4.31. Total volatile solids (TVS), Total Solids (TS) concentrations in influent, effluent samples and removal efficiencies in the one-phase UASB reactor.

#### 4.3.9. The Variations of Proteins, NH<sub>4</sub>-N, PO<sub>4</sub>-P and TP Concentrations

The protein measurements were performed to have an understanding about their degradations (deamination) via and generation of ammonia under anaerobic conditions. The influent, effluent and removal percentages of protein for two-phase system and one-phase system are tabulated in Table C 9 (Appendix C). In that table, it was presented the effluent protein concentrations of the 1<sup>st</sup> reactor of the two-phase system (UAF reactor for acidification), 2<sup>nd</sup> phase UASB reactor of the two-phase system and one-phase system, respectively. In Table C 9 (Appendix C), the percent protein removal column of “UAF” showed the removal in the 1<sup>st</sup> reactor of the two-phase system. The “2<sup>nd</sup>” column showed the removal in the 2<sup>nd</sup> phase UASB reactor of the two-phase system. The two-phase system and one-phase system protein removals are presented in UAF+2<sup>nd</sup> reactor and UASB reactor columns, respectively. The influent, effluent concentrations and removals for protein are presented in Figure 4.32.

It was observed that considerable amounts of protein removal were obtained in the UAF reactor, the 2<sup>nd</sup> phase reactor, the two-phase system and the one-phase system. The protein removals were between 44% and 79% in the UAF reactor at 2 h HRT and 35°C temperature (Periods 1-4). The average protein removal of the UAF reactor was 71% at Period 5 (at 2h HRT and 20°C temperature) and it was not affected by the room temperature (decrease in temperature from 35°C to 20°C). At Period 3, the low influent protein concentration (25 mg/L) was resulted with lower protein removals in the UAF reactor. Protein removals in the two-phase UAF and UASB system were between 84% and 96%. The effluent proteins of 2<sup>nd</sup> phase UASB reactor were very low (between 4 mg/L and 11 mg/L). The protein removals of one-phase UASB system were between 76% and 95% and there was no significant influence of HRT on protein removals as decrease from

10 h to 4 h The effluent proteins of one-phase UASB system were low (between 6 mg/L and 14 mg/L). As the room temperature decrease from 35°C to 20°C, the protein removals was not significantly influenced in the two-phase UAF and UASB system and in one-phase UASB system. The overall removal efficiencies for one-phase and two-phase systems were about the same in Periods 1 and 2 (in HRTs 10 h and 8 h). Protein removals were slightly higher in two-phase UAF and UASB system when compared to the one-phase UASB system in Period 3 (HRT=6 h, T= 35°C), Period 4 (HRT=4 h, T=35°C) and Period 5 (HRT= 4 h, T=20°C).

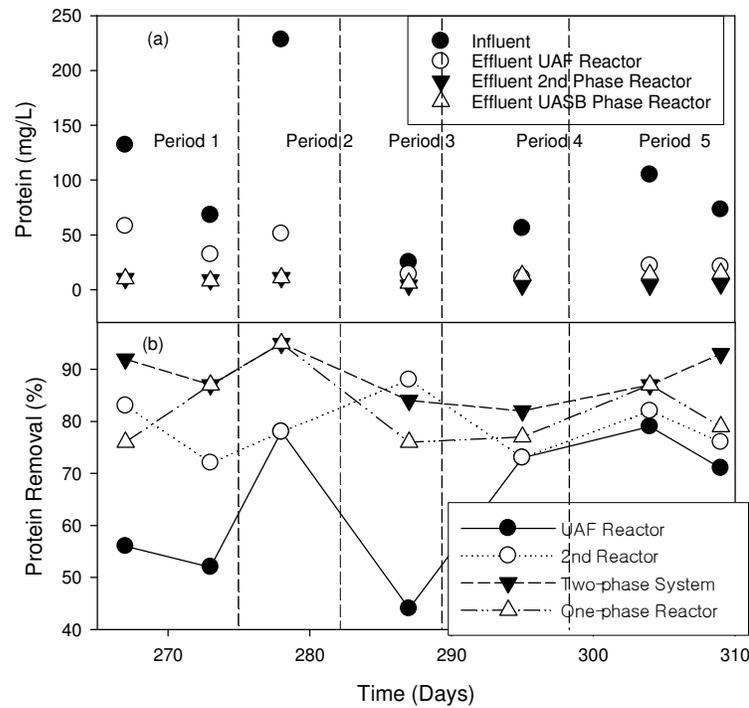


Figure 4.32. (a). Proteins concentrations in the influent and effluent of UAF reactor, two-phase system and one-phase UASB system; (b) Reduction of proteins in UAF reactor, 2<sup>nd</sup> phase UASB reactor, two-phase system and one-phase UASB system.

A typical total nitrogen in domestic wastewater consists of about 60% ammonium nitrogen and 40% organic nitrogen such as protein and amino acids with less than 1% in the form of nitrate and/or nitrite (Barbosa et al., 2003). In the present study, the  $\text{NH}_4\text{-N}$  and TKN the influent  $\text{NH}_4\text{-N/TKN}$  ratios were observed as 0.17, 0.53, 0.72, 0.75 and 0.85 in Period 1, 2, 3,4 and 5, respectively.

The typical  $\text{NH}_4\text{-N/TKN}$  ratios for raw domestic wastewater were given between 0.60 and 0.77 for different cities by Mahmoud (2002). It can be stated that the influent  $\text{NH}_4\text{-N/TKN}$  ratios in Periods 1-5 showed high variations due to influent domestic wastewater concentrations. The influent, effluent concentrations and removal percentages for  $\text{NH}_4\text{-N}$  are given in Table C 10 (Appendix C) and in Figure 4.33. The average releases in  $\text{NH}_4\text{-N}$  concentration in the UAF reactor was 96% in 2 h HRT at 35°C. The reason of the release of  $\text{NH}_4\text{-N}$  concentration in the effluent can be explained by the anaerobic bioconversion of proteins to amino acids and then to ammonia as reported by Demirer and Chen (2005); Martin et al (2003); Cheng and Liu (2002); Kobayashi et al (1983). In Period 3, the protein removal was lowest compared to the other periods and the generation of ammonia was lowest in that period in the UAF reactor (Appendix C- Table C 9 and Table C 10) at 35°C at 2 h HRT. High protein removal and high ammonia generation was observed in the UAF reactor at 2 h HRT and at 35°C. It was observed no change in  $\text{NH}_4\text{-N}$  concentrations at 20°C in the UAF reactor (Period 5). Although it was observed a certain removal of proteins, there was no  $\text{NH}_4\text{-N}$  production in the UAF at 20°C. It can be stated that deamination of proteins can be seen at high temperatures (35°C) in the UAF reactor in 2 h HRT.

In the 2<sup>nd</sup> phase UASB reactor of the two-phase system, it was observed 3-45%  $\text{NH}_4\text{-N}$  decreases during the Periods 1- 4. The HRT of the 2<sup>nd</sup> phase reactor did not affect the  $\text{NH}_4\text{-N}$  decreases at 35°C in the 2<sup>nd</sup> phase reactor. In the

consideration of effluent  $\text{NH}_4\text{-N}$  values of two-phase system, it was seen increases in effluents at the periods when there were considerably high  $\text{NH}_4\text{-N}$  productions (e.i. Period 1 and 2) in the 1<sup>st</sup> reactor (UAF reactor). It was obtained small amounts of  $\text{NH}_4\text{-N}$  decreases (22%-25%) in Periods 3 and 4, respectively in two-phase system at 35°C. In one-phase UASB system, the  $\text{NH}_4\text{-N}$  releases varied between 4% and 67% at 35°C. This can be explained by the degradation of proteins to  $\text{NH}_4\text{-N}$  in anaerobic conditions and by the biological conversion of organic nitrogen into ammonium nitrogen during nitrogeneous organic matter degradation (Barbosa and Sant' Anna Jr, 1989). In Period 5 (T=20°C), it was observed no production of  $\text{NH}_4\text{-N}$ . The reason of this can be explained with high  $\text{NH}_4\text{-N}/\text{TKN}$  ratio (0.85) in the influent. Furthermore, low organic nitrogen content of influent feeding wastewater and temperature decrease from 35°C to 20°C. Typical ratios of organic nitrogen to total nitrogen were calculated as  $0.40\pm 0.20$  and  $0.26\pm 0.03$  from the untreated domestic wastewater data given by Tchobanoglous and Burton, 1991 and Segghezzo et al. 1998, respectively.

In general, it was observed increases in the concentrations of ortho-phosphates in the effluents of UAF reactor, in the two-phase UAF and UASB system and the one-phase UASB system (Table C 10, Figure 4.34). The increase instead of elimination can be attributed to the conversion of organic compound phosphorus to phosphate (Barbosa et al., 2003). On the other hand the  $\text{PO}_4\text{-P}$  released to the reactor since  $\text{PO}_4\text{-P}$  released during anaerobic conditions (Orhon and Artan, 1994). In one-phase reactor, it was observed increases in effluent  $\text{PO}_4\text{-P}$  values in all studied HRTs and temperatures. Although, it was observed increases in effluent  $\text{PO}_4\text{-P}$  values in the UAF reactor, there was some minor decreases in effluent  $\text{PO}_4\text{-P}$  values of the 2nd reactor. It was not possible to propose a relation between HRT and  $\text{PO}_4\text{-P}$  removals in the two-phase system and in one-phase

system. But there was a high degree of  $\text{PO}_4\text{-P}$  production at higher temperatures since  $\text{PO}_4\text{-P}$  released to the environment during anaerobic conditions.

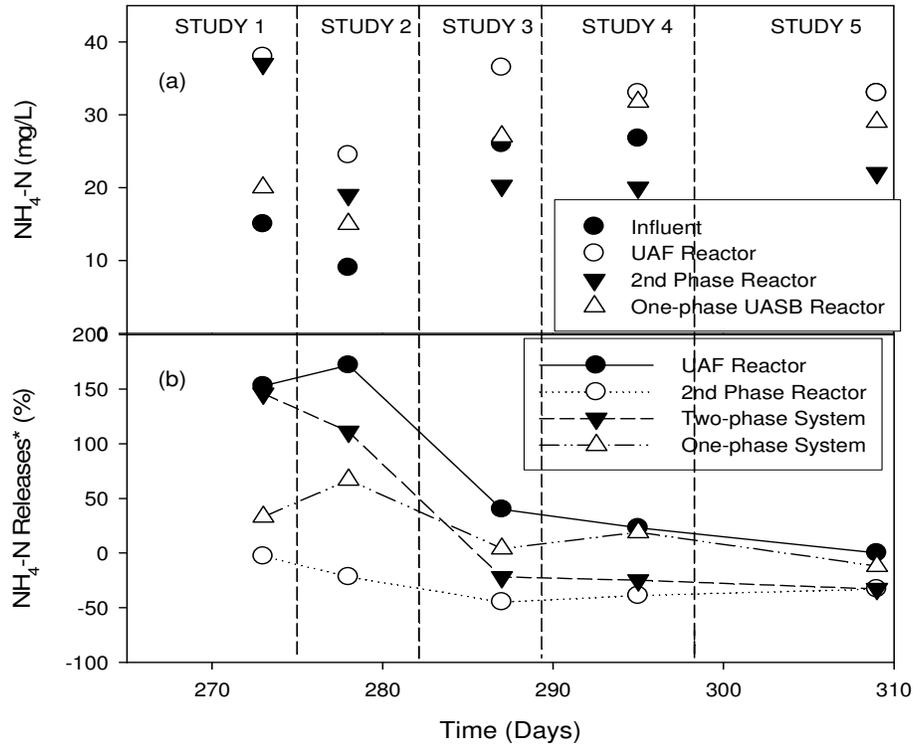


Figure 4.33.  $\text{NH}_4\text{-N}$  concentrations (a) in the UAF reactor, the two-phase UAF and UASB system and the one-phase UASB system; Changes in  $\text{NH}_4\text{-N}$  in UAF reactor, 2<sup>nd</sup> phase UASB reactor, two-phase UAF and UASB system and one-phase UAF and UASB system (b). \* (+) sign shows releases and (-) sign shows decreases.

Phosphorus (TP) removals showed a decrease with the HRT decreases (from 8 h to 4 h) at 35°C in two-phase UAF and UASB system (from 53% to 19%) and in one-phase system (from 40% to 23%) (Figure 4.34.d). In UASB reactor, El-Shafai et al. (2006) presented that the raw sewage phosphorus removals were 20% in warm seasons (at 25-31°C) and 28% in winter time (12.5- 19%) at 6 h HRT, respectively. In the present study, phosphorus removal efficiency was about 38% at 6 h HRT in two-phase reactor system and in one-phase reactor system. Phosphorus removals were given as 49% by Barbosa and Sant Anna Jr (1989). The phosphorus removal of the present study are in agreement with results obtained by Barbosa and Sant Anna Jr (1989) but higher than the results reported by El-Shafai et al (2006). The variations in the treatment efficiencies can be attributed to the variations in the influent characteristics of domestic wastewater. The effluent phosphorus was about the same in two-phase UAF and UASB system and in one-phase UASB system. When the ambient temperature decreased from 35°C to 20°C the effluent phosphorus concentration increased from about 22 mg/L to about 30 mg/L in both set of reactors.

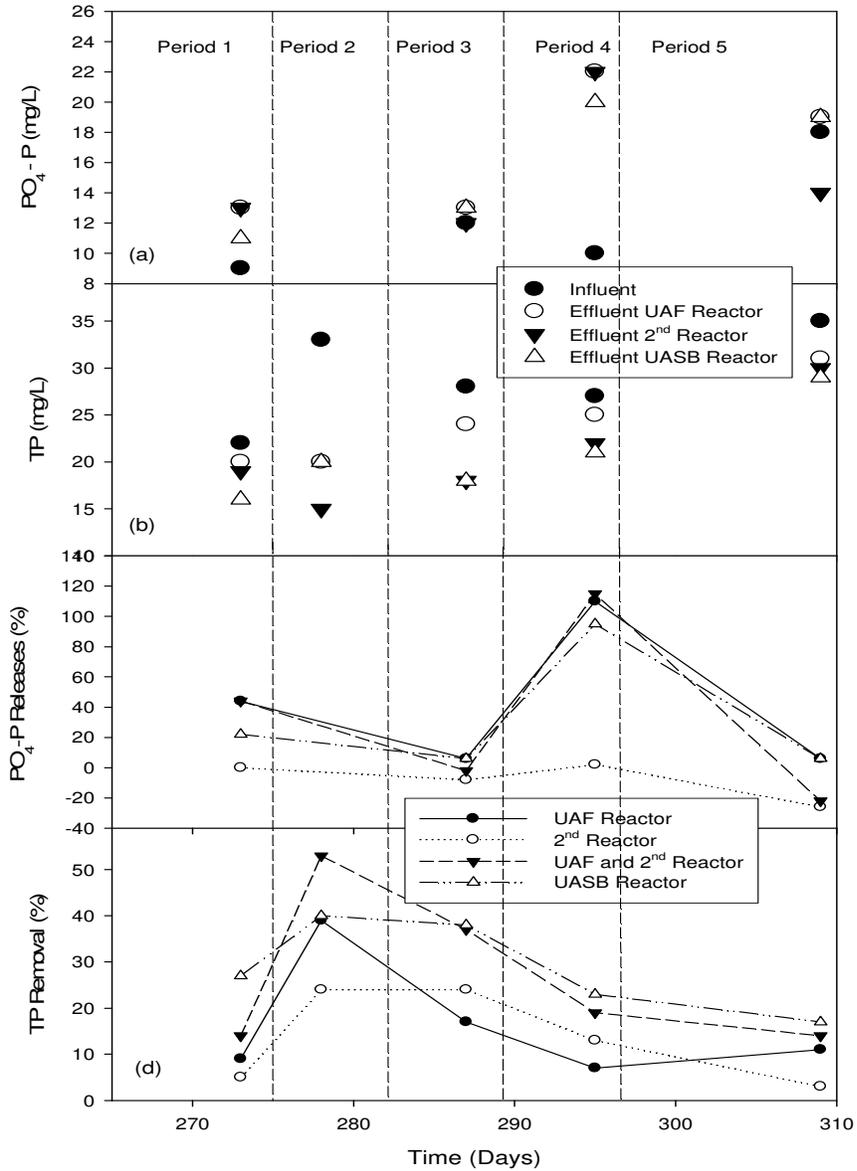


Figure 4.34. PO<sub>4</sub>-P (a) and TP (b) concentrations in the UAF reactor, 2<sup>nd</sup> phase UASB reactor and one-phase UASB reactor; Reduction of PO<sub>4</sub>-P (c) and TP (d) in UAF reactor, 2<sup>nd</sup> phase reactor, two-phase UAF and UASB system and one-phase UASB system. \* (+) sign shows releases and (-) sign shows decreases.

#### **4.3.10. System Feasibility**

The calculations relevant to energy requirement and energy generated for the proposed and compared systems are given in Appendix E. The two-phase UAF and UASB system and one-phase UASB system are compared at 4 h HRT for 35°C and 20°C temperatures.

The effluent results showed that the two-phase UAF and UASB system satisfied the Turkish effluent criteria (SKKY (2005) at 4 h HRT and 20°C. On the other hand, the methane production yield indicated that the amount of produced methane gas would not recover the energy requirement to increase influent temperature from 20°C to 35°C in two-phase system and one-phase system. Therefore, to operate the two-phase system at 20°C can be considered a significant advantage over the other heated anaerobic systems for the treatment of domestic wastewater in dilute character.

There are a variety of systems which can be applied for wastewater treatment. In industrialized countries, the number of alternatives may be more limited due to more stringent effluent quality standards applicable. On the other hand, in developing countries, the number of choices to be compared may be higher. In Turkey, effluent standards display similarities with many of industrialized countries and cost component and operational requirements are very important in the selection of the treatment systems. The main factor in the selection of a treatment system is the cost estimation for the system which satisfy the effluent criteria. In a comparative analysis, the main factors are construction costs such as earthworks, concrete works, process equipment, piping, electrification, instrumentation and operation and maintenance costs such as energy (pumping,

aeration, heating, etc), chemicals (nutrients, sludge conditioning, neutralization, etc.), sludge transport, maintenance and administration costs (von Sperling, 1996).

In general, the critical items are efficiency, reliability, sludge disposal aspects and land requirements for industrialized countries. However, in developing countries, construction costs, sustainability, simplicity and operational costs are mainly considered as major parameters. Although these are important for developing countries they can not be considered at first for industrialized countries (von Sperling, 1996). Typical characteristics of conventional wastewater treatment systems are shown in Table 4.11.

In comparison of activated sludge systems with the UASB systems in terms of land and power requirements, it is reported that these requirements are considerably lower in the UASB systems than the activated sludge systems. The construction costs and quantity of sludge to be handled are much lower in UASB reactor with respect to activated sludge systems. On the other hand, the BOD removal efficiencies are also relatively lower in UASB system than activated sludge systems. However, in a two-phase UAF and UASB system, the effluent criteria for BOD, COD and TSS were satisfied according to SKKY (2005) in this study. In other words, two-phase UAF and UASB system successfully treated domestic wastewater. In fact, the importance of such a two phase system is that effluent criteria can be satisfied by lower land requirements, construction costs with a relatively low sludge to be handled. Additionally, the application of two-phase system in low strength wastewater treatment can provide an extra heat such as  $1.9 \times 10^8$  cal/d and an electric energy with a power 9201 Watt/day (9.2 kW/day) at a HRT of 4 hours and at a temperature of 20 °C. Consequently, a two-phase UAF and UASB system could be recommended for the treatment of domestic wastewater in low strength character.

Table 4.11. Typical characteristics of the conventional wastewater treatment systems (Von Sperling, M. 1996)

TREATMENT SYSTEMS	REMOVAL EFFICIENCY (%)				REQUIREMENTS		CONSTRUCT. COST (US\$/inhab)	TOTAL HYDRAULIC DETENTION TIME (days)	QUANTITY OF SLUDGE TO B HANDLED (m3/inhab.year)
	BOD	N	P	COLI-FORMS	LAND (m2/inhab)	POWER (W/inhab)			
PRELIMINARY TREATMENT	0 - 5	~ 0	~ 0	~ 0	< 0,001	~ 0	2 - 8	-	-
PRIMARY TREATMENT	35 - 40	10 - 25	10 - 20	30 - 40	0,03 - 0,05	~ 0	20 - 30	0,1 - 0,5	0,6 - 1,3
FACULTATIVE POND	75 - 85	30 - 50	20 - 60	60 - 99	2,0 - 5,0	~ 0	10 - 30	15 - 30	-
ANAEROBIC POND-FACULTATIVE POND	75 - 90	30 - 50	20 - 60	60 - 99,9	1,5 - 3,5	~ 0	10 - 25	12 - 24	-
FACULTATIVE AERATED LAGOON	75 - 90	30 - 50	20 - 60	60-96	0,25 - 0,5	1,0 - 1,7	10 - 25	3 - 9	-
COMPLET.MIXED AERAT.-SEDIMENT.POND.	75 - 90	30 - 50	20 - 60	60-99	0,2 - 0,5	1,0 - 1,7	10 - 25	4 - 9	-
CONVENTIONAL ACTIVATED SLUDGE	85 - 93	30 - 40 (a)	30 - 45 (a)	60 - 90	0,2 - 0,3	1,5 - 2,8	60 - 120	0,4 - 0,6	1,1 - 1,5
EXTENDED AERATION (CONTINUOUS FLOW)	93 - 98	15 - 30 (a)	10 - 20 (a)	65 - 90	0,25 - 0,35	2,5 - 4,0	40 - 80	0,8 - 1,2	0,7 - 1,2
SEQUENCING BATCH REACTOR	85 - 95	30 - 40 (a)	30 - 45 (a)	60 - 90	0,2 - 0,3	1,5 - 4,0	50 - 80	0,4 - 1,2	0,7 - 1,5
LOW RATE TRICKLING FILTER	85 - 93	30 - 40 (a)	30 - 45 (a)	60 - 90	0,5 - 0,7	0,2 - 0,6	50 - 90	NA	0,40 - 0,6
HIGH RATE TRICKLING FILTER	80 - 90	30 - 40 (a)	30 - 45 (a)	60 - 90	0,3 - 0,45	0,5 - 1,0	40 - 70	NA	1,1 - 1,5
UPFLOW ANAREOBIC SLUDGE BLANKET	60 - 80	10 - 2 5	10 - 20	60 - 90	0,05 - 0,10	~ 0	20 - 40	0,3 - 0,5	0,07 - 0,1
SEPTIC TANK-ANAEROBIC FILTER	70 - 90	10 - 25	10 - 20	60 - 90	0,2 - 0,4	~ 0	30 - 80	1,0 - 2,0	0,07 - 0,1
SLOW RATE INFILTRATION	94 - 99	65 - 9 5	75 - 99	> 99	10 - 50	~ 0	10 - 20	NA	-
RAPID INFILTRATION	86 - 98	10 - 80	30 - 99	> 99	1 - 6	~ 0	5 - 15	NA	-
SUBSURFACE INFILTRATION	90 - 98	10 - 40	85 - 95	> 99	1 - 5	~ 0	5 - 15	NA	-
OWERLAND FLOW	85 - 95	10 - 80	20 - 50	90 - > 99	1 - 6	~ 0	5 - 15	NA	-

References: Arceivala (1981), Metcalf & Eddy (1991), Prioli et al (1993), Qasim (1985), USEPA (1979, 1981, 1992), Vieira (1993), von Sperling (1994), information from others and author's experience

Notes:

NA: not applicable

Energy requirements do not include the eventual raw sewage pumping

(a) An additional nutrient removal can be obtained through modifications in the process

## CHAPTER 5

### CONCLUSIONS

This study investigated the possible advantages of the utilization of two-phase anaerobic digestion for the treatment of domestic wastewater in Turkey, for the first time. The two-phase reactor has been demonstrated to be a good choice for the anaerobic treatment of the domestic wastewater at low temperatures (20°C) and low hydraulic retention time (HRTs) (e.i. 4 h).

In the first part of the study, the acidification performances of domestic wastewater were investigated by upflow anaerobic filter (UAF) and upflow anaerobic sludge blanket (UASB) reactors. The performances of one-phase UASB system and two-phase UAF and UASB system were evaluated and compared at various (HRTs) for the treatment of domestic wastewater in the second part of the study.

Based on the results of this study, the following conclusions could be made;

- Degree of acidification was higher in the UAF reactor than the UASB reactor. The optimum acidification conditions leading to highest degree of acidification (31%) was obtained in the UAF reactor at 2 hour HRT. The

placement of an acidification reactor (UAF) prior to an UASB reactor resulted better acidification and better conversion of acidified products to methane at low HRTs (4 h) through the treatment of domestic wastewater.

- The tCOD treatment efficiencies (between 74% and 88%) were about the same for both one-phase and two-phase systems at HRT's between 6 h and 10 h. The tCOD treatment in two-phase UAF and UASB system was 15% higher than the one-phase system and the effluent tCOD values of the two-phase UAF and UASB system were considerably lower than the one-phase UASB system at 4 h HRT. At lower HRTs, the average tCOD removal of two-phase UAF and UASB system was better than that one-phase UASB system. The decreases in temperature did not significantly affect the tCOD removal efficiencies in two-phase UAF and UASB system. The tCOD removal efficiencies became 83% and 64% for two-phase UAF and UASB system and one-phase UASB system, respectively, at 20°C. It can be said that tCOD removals were better in two-phase UAF and UASB system than that one-phase UASB system.
- The sCOD reductions in acidified UAF reactor could be explained with the partial methanification of inherently acidified influent wastewater. This cause decreases in tCOD concentrations in the effluent. The VSS reductions in acidified UAF reactor are due to hydrolysis and acidification of inherently acidified influent domestic wastewater ending with methanogenesis.
- The effluent BOD<sub>5</sub> values of both two-phase UAF and UASB system and one-phase system satisfied the limits given by Turkish Water and Wastewater Regulation at HRTs of 4-10 h and at 35°C.

- The effluent tVFA concentration was higher in acidified-UAF reactor than that the UASB reactor at 2 h HRT. The UAF reactor had more acidifying conditions than that UASB reactor. Lower pH values were obtained in UAF reactor compared to UASB reactor at an HRT of 2 h in one-phase reactor system.
- The tVFA composed from 12% acetic acid, 29% propionic acid and 52% butyric acid in UAF at 2 h HRT through acidification study. The composition of tVFA in UASB reactor was 51% acetic acid, 22% propionic acid and 19% butyric acid.
- The highest tVFA/sCOD ratios are 0.54 and 0.32 in UAF and UASB reactors, respectively, in 2 h HRT.
- Decreases in HRT and temperature from 10 h to 4 h and from 35°C to 20°C did not show a negative effect in the total methane gas production in the two-phase UAF and UASB system.
- The conversion rates of COD removed to methane were between 0.005 and 0.067 Nm<sup>3</sup>CH<sub>4</sub>/kg COD removed and between 0.158 and 0.233 Nm<sup>3</sup> CH<sub>4</sub>/kg COD removed in the UAF reactor and in the 2<sup>nd</sup> phase UASB reactor of the two-phase system, respectively. The conversion rate of COD removed to methane varied between 0.029 and 0.199 Nm<sup>3</sup> CH<sub>4</sub>/kg COD removed in the one-phase UASB reactor.

- The total suspended solids (TSS) removals were significantly higher in two-phase UAF and UASB system than that one-phase UASB system at lower temperatures.
- The SMA of the 2<sup>nd</sup> phase UASB reactor (0.253 g CH<sub>4</sub>-COD/kgVSS.d) was higher than one-phase UASB reactor (0.222 g CH<sub>4</sub>-COD/kgVSS.d).
- Proteins degraded to corresponding amino acids ending in ammonia production. A small amount of ammonia was removed under anaerobic conditions.
- In the operation of two-phase UAF and UASB system, an extra energy (e.i. 9.2 kW/day from treatment of a 1 MLD domestic wastewater) could be available provided that the treatment is accomplished at 20°C (without heating the system, the related effluent criteria satisfied).

## CHAPTER 6

### FUTURE WORK

The application of two-phase high rate reactor systems on the direct treatment of raw domestic wastewater using anaerobic technology would be an important progress. The system is simple in terms of operation and maintenance, and therefore could be suitable for rural communities.

- There are lots of touristic villages and hotels at the Southern and Western coasts of Turkey and the seasonal temperature with the volumetric loadings of domestic wastewater vary significantly from summer to winter. It is important to prove the applicability of two-phase systems to the environmental and physical conditions in Turkey. The effect of temperature to the reactor performance is very important. The two-phase UAF and UASB system satisfied the Turkish and EU receiving water standards for domestic water at 20°C. The effect of lower temperatures on the treatment of carbonaceous compounds in sewage wastewater would give a better understanding about the applicability of the two-phase system in the anaerobic treatment of domestic wastewaters in Turkey. The feasibility of the two-phase technology should be investigated at lower temperatures, e.g. 5-20°C for their application in colder regions. More researches relevant to anaerobic degradation of domestic wastewater

should be conducted to widen the applicability of two-phase system at different operational conditions for the treatment of domestic wastewaters.

- The performance of the two-phase system for the treatment of domestic wastewaters under the environmental conditions in Turkey still needs to be demonstrated along relevant research projects at pilot scale. The septic tanks and UASB-septic tanks are used for single households, in isolated locations like farms and recreational facilities not connected to the centralized sewerage system (Zeeman, 1997). The two-phase system can be applied for this purpose to satisfy the effluent receiving water criteria of Turkey. Pilot-scale studies can be applied to understand the usage of two-phase UAF and UASB system in the treatment of domestic wastewater. Pilot studies and some applications on-site could be useful to have a better idea about the treatment of domestic wastewater by two-phase reactors in ambient conditions.
- Economical, financial and feasibility studies to be carried out by the researchers might be necessary to compare the one-phase and two-phase systems for the treatment of domestic wastewaters in Turkey.

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## APPENDIX A

### CALIBRATION CURVES FOR PROTEIN MEASUREMENT

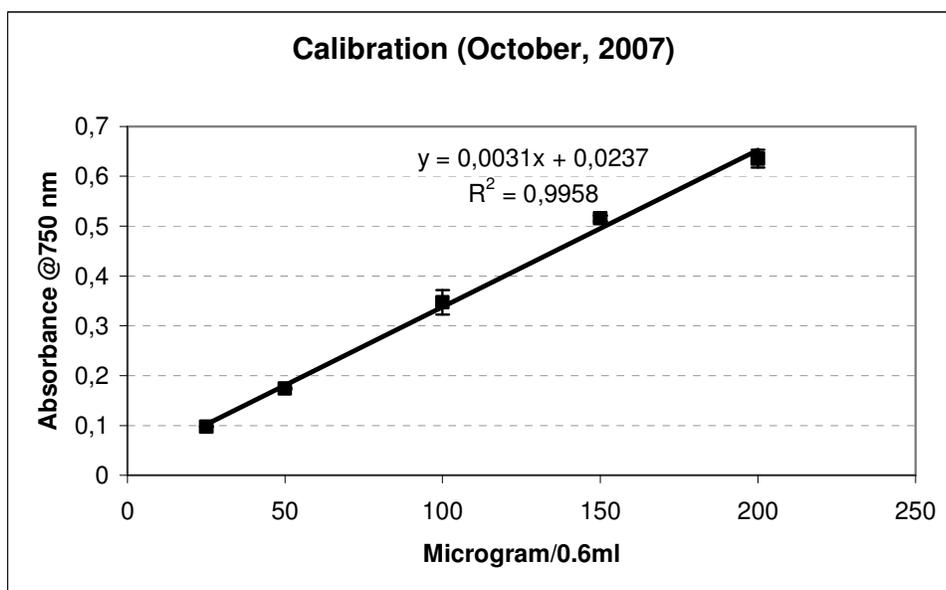


Figure A. 1. Calibration curve for protein measurement in October 2007.

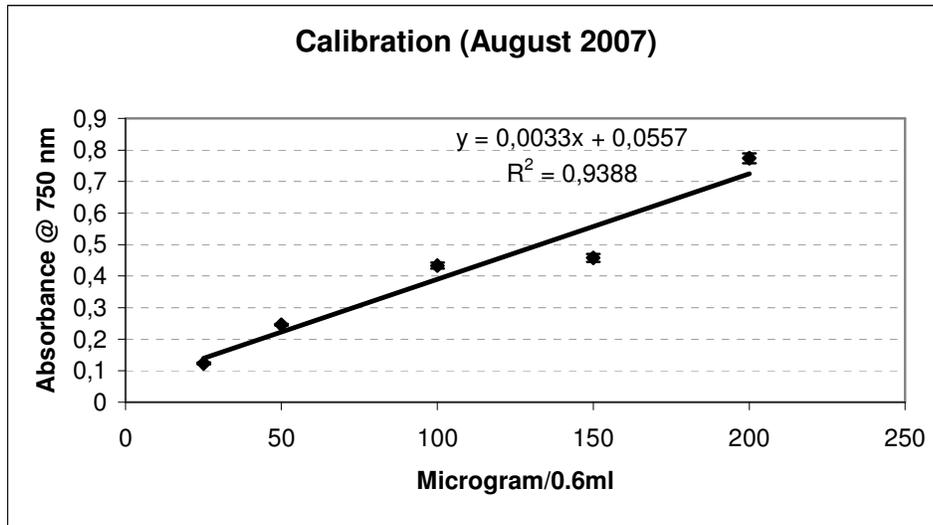


Figure A. 2. Calibration curve for protein measurement in August 2007.

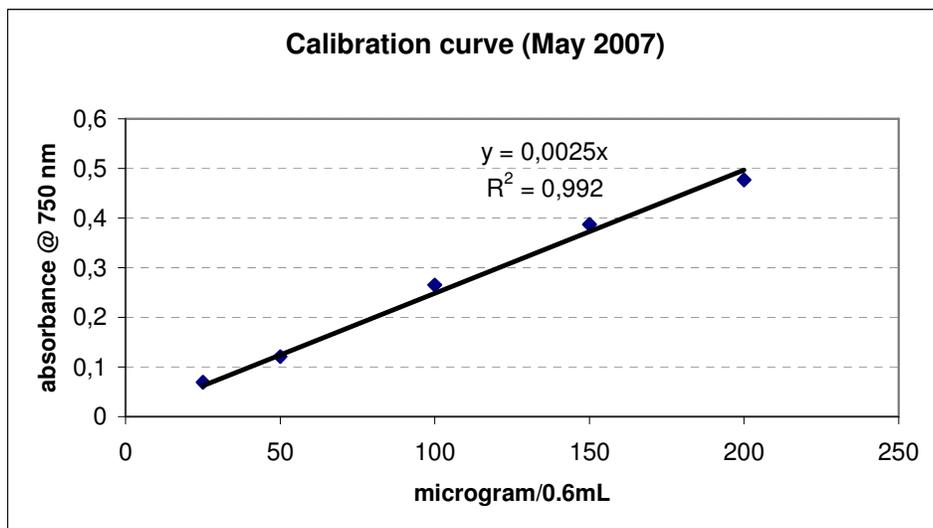


Figure A. 3. Calibration curve for protein measurement in May 2007.

## APPENDIX B

### THE RESULTS OF ONE-WAY ANOVA TESTS AND CORRELATION TESTS

Table B.1. One-way Anova analysis to evaluate the relationship for sCOD changes in the UAF and UASB reactors in Phase 3 (HRT=2 h) for the acidogenic system (for days 176-223).  $F(1,8) = 95.97$ ,  $p \leq 0.001$

sCOD	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
UAF Reactor	5	11.28	7.79	3.483	1.61	20.950	-0.63	19.40
UASB Reactor	5	47.46	2.74	1.227	44.05	50.863	43.10	50.71
Total	10	29.37	19.85	6.276	15.17	43.564	-.63	50.71
ANOVA		Sum of Squares		df	Mean Square	F	Sig.	
SCORES								
Between Groups		3271.757		1	3271.757	95.969	.000	
Within Groups		272.735		8	34.092			
Total		3544.493		9				

$F(1,8) = 95.97$ ,  $p \leq 0.001$

Table B.2. One-way Anova analysis was conducted to evaluate the relationship for tCOD treatments in the UAF and UASB reactors in Phase 3 (HRT=2 h) for the acidogenic system (for days 176-223)

Descriptives	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
SCORES					Lower Bound	Upper Bound		
UAF	5	44.718	3.461	1.548	40.419	49.016	40.55	48.41
UASB	5	56.062	1.883	0.842	53.723	58.400	54.05	58.37
Total	10	50.390	6.530	2.065	45.718	55.061	40.55	58.37
ANOVA SCORES		Sum of Squares	df	Mean Square	F	Sig.		
Between Groups		321.716	1	321.716	41.424	.000		
Within Groups		62.131	8	7.766				
Total		383.847	9					

F(1,8)= 41.42,  $p < 0.001$ , the UASB reactor scores significantly higher ( $M = 56.06$ ,  $SD = 1.88$ ) than the UAF reactor scores ( $M = 44.71$ ,  $SD = 3.46$ ).

Table B.3. One-way Anova analysis was conducted to evaluate the relationship for BOD<sub>5</sub> removals

Descriptives	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
SCORES					Lower Bound	Upper Bound		
UAF	5	44.7180	3.461	1.548	40.419	49.016	40.55	48.41
UASB	5	56.0620	1.883	0.842	53.723	58.400	54.05	58.37
Total	10	50.3900	6.531			55.061	40.55	58.37
ANOVA SCORES		Sum of Squares	df	Mean Square	F	Sig.		
Between Groups		321.716	1	321.716	41.424	0.000		
Within Groups		62.131	8	7.766				
Total		383.847	9					

BOD<sub>5</sub> F(1,8)= 0.474,  $p = 0.511$ .

Table B.4. Bivaried Correlation Analysis was conducted to evaluate the correlation between sCOD(%)/ gVSS ratio changes and HRT for the UAF reactor in the acidogenic system

<i>Descriptive Statistics</i>		<i>Mean</i>	<i>Std. Deviation</i>	<i>N</i>
UAF		3.909	2.637	20
HRT		7.700	8.688	20
<i>Correlations</i>		<i>UAF</i>	<i>HRT</i>	
UAF	Pearson Correlation	1	.795(**)	
	Sig. (2-tailed)	.	.000	
	N	20	20	
HRT	Pearson Correlation	.795(**)	1	
	Sig. (2-tailed)	.000	.	
	N	20	20	

\*\* Correlation is significant at the 0.01 level (2-tailed),  $r(18)=0.795$ ,  $p<0.001$ .

Table B.5. Bivaried Correlation Analysis was conducted to evaluate the correlation between sCOD(%)/ gVSS ratio changes and HRT for the UASB reactor in the acidogenic system

	Mean	Std. Deviation	N
UASB	2.859	1.334	20
HRT	7.700	8.688	20
		UASB	HRT
UASB	Pearson Correlation	1	.686(**)
	Sig. (2-tailed)	.	.001
	N	20	20
HRT	Pearson Correlation	0.686(**)	1
	Sig. (2-tailed)	0.001	.
	N	20	20

\*\*Correlation is significant at the 0.01 level (2-tailed)  $r(18)=-0.686$ ,  $p=0.001$ .

Table B.6. Bivaried Correlation Analysis was conducted to evaluate the correlation between sCOD(%)/ gVSS ratio changes and OLR for the UAF reactor in the acidogenic system

	Mean	Std. Deviation	N
UAF	3.909	2.637	20
OLR	1.004	.784	20
		UAF	OLR
UAF	Pearson Correlation	1	-.843(**)
	Sig. (2-tailed)	.	.000
	N	20	20
OLR	Pearson Correlation	-.843(**)	1
	Sig. (2-tailed)	0.000	.
	N	20	20

\*\*Correlation is significant at the 0.01 level (2-tailed)  $R(18)=-0.843$ ,  $p<0.001$ .

Table B.7. Bivaried Correlation Analysis was conducted to evaluate the correlation between sCOD(%)/ gVSS ratio changes and OLR for the UASB reactor in the acidogenic system

Reactor	Correlations	UASB	OLR
UASB	Pearson Correlation	1	-.391
	Sig. (2-tailed)	.	.089
	N	20	20
OLR	Pearson Correlation	-0.391	1
	Sig. (2-tailed)	0.089	.
	N	20	20

$r(18)=-0.391$ ,  $p=0.089>0.05$ , non-significant.

Table B.8. One-way Anova analysis to evaluate the relationship for sCOD changes (%)/g VSS in the UAF and UASB reactors in Phase 3 for the acidogenic system

Descriptive	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
UAF	7	1.630	0.979	0.370	0.725	2.535	-0.07	3.02
UASB	7	2.754	0.603	0.228	2.197	3.312	1.79	3.36
Total	14	2.192	0.975	0.261	1.629	2.755	-0.07	3.36

ANOVA SCORES	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	4.424	1	4.424	6.698	0.024
Within Groups	7.927	12	.661		
Total	12.351	13			

F(1,12)= 6.69, p=0.024.

Table B.9. One-way Anova analysis to evaluate the relationship for VSS reduction in the UAF and UASB reactors in Phase 3 in the acidogenic system

Descriptive	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
UAF	8	77.750	2.550	0.902	75.618	79.881	73.00	82.00
UASB	8	82.875	3.044	1.076	80.329	85.420	77.00	86.00
Total	16	80.313	3.790	0.947	78.293	82.331	73.00	86.00
ANOVA	Sum of Squares	df	Mean Square	F	Sig.			
Between Groups	105.063	1	105.063	13.326	.003			
Within Groups	110.375	14	7.884					
Total	215.438	15						

F(1,14)=13.33, p=0.003<0.05.

Table B.10. One-way Anova analysis to evaluate the relationship for TS removal in the UAF and UASB reactors in Phase 3 (HRT=2 h) for the acidogenic system

Descriptives SCORES	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Mini mum	Maxi mum
					Lower Bound	Upper Bound		
UAF	10	16.3504	5.969	1.888	12.0803	20.6205	8.76	26.71
UASB	10	16.6195	8.684	2.746	10.4075	22.8314	3.86	27.52
Total	20	16.4849	7.254	1.622	13.0901	19.8797	3.86	27.52
ANOVA SCORES		Sum of Squares	Df	Mean Square	F	Sig.		
Between Groups		.362	1	0.362	0.007	0.937		
Within Groups		999.338	18	55.519				
Total		999.700	19					

F(1,18)=0.007, p=0.937, non-significant.

Table B.11. One-way Anova analysis to evaluate the relationship for TVS Removal in the UAF and UASB reactors in Phase 3 in the acidogenic system

Descriptives SCORES	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Mini mum	Maxim um
					Lower Bound	Upper Bound		
UAF	10	29.874	12.934	4.090	20.621	39.127	10.20	48.43
UASB	10	46.820	20.613	6.519	32.075	61.566	20.68	74.91
Total	20	38.347	18.870	4.220	29.516	47.179	10.20	74.91

F(1,18)=4.85, p=0.041, relationship is significant.

Table B.12. One-way Anova analysis to evaluate the correlation between tCOD removal and temperature for the two-phase system

Descriptives SCORES	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
35C	5	84.910	8.966	4.010	73.778	96.042	75.00	92.68
20C	4	82.304	5.615	2.808	73.368	91.239	75.47	88.48
Total	9	83.752	7.342	2.447	78.108	89.395	75.00	92.68
ANOVA SCORES		Sum of Squares	df	Mean Square	F	Sig.		
Between Groups		15.093	1	15.093	0.254	0.630		
Within Groups		416.117	7	59.445				
Total		431.210	8					

$F(1,7)=0.25, p=0.63$  , non-significant

Table B.13. One-way Anova analysis to evaluate the correlation between tCOD removal and temperature for the one-phase reactor

Descriptive SCORES	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
35C	5	73.191	6.706	2.999	64.864	81.517	62.00	79.39
20C	4	53.250	10.996	5.498	35.753	70.747	42.00	65.00
Total	9	64.328	13.352	4.451	54.065	74.592	42.00	79.39
ANOVA SCORES		Sum of Squares	df	Mean Square	F	Sig.		
Between Groups		883.614	1	883.614	11.399	0.012		
Within Groups		542.631	7	77.519				
Total		1426.245	8					

$F(1,7)= 11.40, p=0.012$ , significant.

Table B.14. One-way Anova analysis to evaluate the correlation between methane percentages and HRT for the two-phase reactor

Descriptives	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
HRT. (h)								
4	2	28.00	5.657	4.000	-22.825	78.825	24.00	32.00
6	3	31.00	3.606	2.082	22.043	39.957	28.00	35.00
8	2	47.00	2.828	2.000	21.588	72.412	45.00	49.00
10	6	37.67	13.765	5.619	23.222	52.112	22.00	54.00
Total	13	36.08	11.086	3.075	29.377	42.776	22.00	54.00
ANOVA		Sum of Squares	df	Mean Square	F	Sig.		
Between Groups		461.590	3	153.863	1.367	0.314		
Within Groups		1013.333	9	112.593				
Total		1474.923	12					

F(3,9)= 1.397, p=0.314 > 0.05 , non-significant.

Table B.15. One-way ANOVA & Post HOC Tests (TUKEY & SCHEFFE) to evaluate the correlation between methane percentages and HRT for the one-phase reactor According to Multiple Comparisons (POST HOC TEST), Tukey HSD Method

Descriptives SCORES	N	Mean	Std. Dev.	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
HRT=4 h	4	15.750	3.862	1.931	9.604	21.896	12.00	21.00
HRT=6 h	3	31.000	8.544	4.933	9.776	52.225	22.00	39.00
HRT=8 h	2	43.500	2.121	1.500	24.441	62.559	42.00	45.00
HRT=10 h	6	39.417	6.785	2.770	32.296	46.538	30.00	47.00
Total	15	31.967	12.18	3.146	25.218	38.716	12.00	47.00
ANOVA		Sum of Squares	df	Mean Square	F	Sig.		
Between Groups		1653.775	3	551.258	14.252	0.000		
Within Groups		425.458	11	38.678				
Total		2079.233	14					
Multiple Comparisons (POST HOC TEST) ependent Variable: SCORES								

\* The mean difference is significant at the .05 level. F(3,11)=14.252.

Table B.16. One-way ANOVA & Post HOC Tests (TUKEY & SCHEFFE) to evaluate the correlation between methane percentages and HRT for the one-phase reactor (Cont'd)

	(I) HRT	(J) HRT	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
		(h)				Lower B	Upper B.
Tukey HSD	4	6	-15.25 (*)	4.74997	.036	-29.545	-0.9547
		8	-27.75 (*)	5.386	.002	-43.960	-11.541
		10	-23.667(*)	4.0145	.001	-35.750	-11.585
	6	4	15.2500(*)	4.750	.036	0.955	29.545
		8	-12.5000	5.677	.183	-29.586	4.586
		10	-8.4167	4.398	.277	-21.652	4.818
	8	4	27.7500(*)	5.386	.002	11.541	43.960
		6	12.5000	5.677	.183	-4.586	29.5861
		10	4.0833	5.078	.851	-11.199	19.366
	10	4	23.6667(*)	4.015	.001	11.585	35.748
6		8.4167	4.398	.277	-4.818	21.652	
8		-4.0833	5.078	.851	-19.366	11.199	
Scheffe	4	6	-15.2500	4.750	.056	-30.833	.333
		8	-27.75 (*)	5.386	.003	-45.419	-10.081
		10	-23.667(*)	4.015	.001	-36.837	-10.497
	6	4	15.2500	4.750	.056	-.333	30.833
		8	-12.5000	5.677	.242	-31.125	6.125
		10	-8.4167	4.398	.348	-22.844	6.010
	8	4	27.7500(*)	5.386	.003	10.081	45.419
		6	12.5000	5.677	.242	-6.125	31.125
		10	4.0833	5.078	.884	-12.575	20.742
	10	4	23.6667(*)	4.015	.001	10.497	36.837
6		8.4167	4.398	.348	-6.010	22.844	
8		-4.0833	5.078	.884	-20.742	12.575	

$p=0.036$  , significant for HRTs of 6 h and 4 h at 35°C,

$p=0.002$  , significant for HRTs of 8 h and 4 h at 35°C,

$p=0.001$  , significant for HRTs of 10 h and 4 h at 35°C

and non-significant for HRts of 8-10 h and 6-8 h at 35°C.

Table B.17. One-way Anova analysis was conducted to evaluate the percent VSS reductions correlation between HRTs 8 h and 24 h for UAF reactor

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
8	3	98.6667	1.155	0.667	95.798	101.535	98.00	100.00
24	3	94.0000	7.000	4.042	76.611	111.389	86.00	99.00
Total	6	96.3333	5.164	2.108	90.914	101.753	86.00	100.00
ANOVA								
		Sum of Squares	df	Mean Square	F	Sig.		
Between Groups		32.667	1	32.667	1.298	0.318		
Within Groups		100.667	4	25.167				
Total		133.333	5					

F (1,4) = 1.298, p > 0.05, non-significant.

Table B.18. One-way Anova analysis was conducted to evaluate the percent VSS reductions correlation between HRTs 8 h and 24 h for UASB reactor

	N	Mean	Std. Deviation	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower Bound	Upper Bound		
8	3	96.333	3.5118	2.028	87.609	105.057	93.00	100.00
24	3	94.333	8.1445	4.702	74.101	114.566	85.00	100.00
Total	6	95.333	5.7155	2.333	89.335	101.331	85.00	100.00
ANOVA								
		Sum of Squares	df	Mean Square	F	Sig.		
Between Groups		6.000	1	6.000	0.153	0.716		
Within Groups		157.333	4	39.333				
Total		163.333	5					

F (1, 4) = 0.153, p > 0.05, non significant

Table B.19. One-way Anova analysis to evaluate the relationship for sCOD changes in the UAF and UASB reactors in Phase 3 (HRT=2 h)

	N	Mean	Std. Dev.	Std. Error	95% Confidence Interval for Mean		Minimum	Maximum
					Lower B.	Upper B.		
UAF	16	63.080	16.916	4.229	54.066	72.0948	36.75	85.21
UASB	16	61.972	14.321	3.580	54.341	69.6038	38.42	88.63
Total	32	62.526	15.428	2.727	56.964	68.0890	36.75	88.63
			Sum of Squares	Df	Mean Square	F	Sig.	
Between Groups			9.825	1	9.825	.040	0.843	
Within Groups			7368.987	30	245.633			
Total			7378.811	31				

$F(1, 30) = 0,040, p > 0.05.$ , non significant.

Table B.20. Correlation Analysis and One Way ANOVA test between sCOD reductions and temperature decreases from 35°C to 20°C for UAF and UASB system.

SUMMARY OUTPUT								
<i>Regression Statistics</i>								
Multiple R	0,915261							
R Square	0,837703							
Adjusted R Square	0,805244							
Standard Error	3,53837							
Observations	7							
ANOVA								
	<i>Df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>			
Regression	1	323,114	323,114	25,8077	0,003834			
Residual	5	62,6003	12,5201					
Total	6	385,7143						
	<i>Coeff</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95,0%</i>	<i>Upper 95,0%</i>
Intercept	-24,778	10,1681	-2,437	0,0589	-50,916	1,3601	-50,916	1,3604
X Variable 1	1,1286	0,22217	5,080	0,0038	0,5575	1,6997	0,5575	1,6998

$F(1,5)=25,81, R^2=0.840.843, p=0.0038.$

Table B.21. Correlation Analysis and One Way ANOVA test between sCOD changes and temperature decreases from 35°C to 20°C in the two-phase UAF and UASB system. (The Anova was nonsignificant and there was a weak correlation between sCOD removal and temperature).

SUMMARY OUTPUT								
Regression Statistics								
Multiple R	0.852							
R Square	0.726							
Adjusted R Square	0.526							
Standard Error	15.237							
Observations	6							
ANOVA								
	df	SS	MS	F	Significance F			
Regression	1	3072.185	3072.19	13.233	0.022			
Residual	5	1160.815	232.16					
Total	6	4233						
	Coefficients	Standard Error	t Stat	P-value	Lower 95%	Upper 95%	Lower 95,0%	Upper 95,0%
Intercept	0	-	-	-	-	-	-	-
X Variable 1	0,794	0.218	3.638	0.015	0.233	1.355	0.233	1.356

$F(1,5)=13.233, p=0.015, R^2=0.726, R^2=0.526$

Table B.22. The correlation analysis and ANOVA between TVS removal and sCOD changes, VFA production and CH<sub>4</sub> percentages at decreasing HRTs in two phase reactor

<i>Regression Statistics</i>	
Multiple R	0.980722
R Square	0.961815
Adjusted R Square	0.842218
Standard Error	10.6464
Observations	12

<i>ANOVA</i>					
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	3	25694.8	8564.935	75.56457	3.28E-06
Residual	9	1020.113	113.3459		
Total	12	26714.92			

	<i>Coeff.</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
X Variable 1	0.534	0.206	2.587	0.029	0.067	0.999	0.067	0.999
X Variable 2	-0.793	0.720	-1.101	0.299	-2.421	0.835	-2.421	0.835
X Variable 3	0.673	0.289	2.333	0.045	0.021	1.326	0.021	1.326

R<sup>2</sup>=0.96, adjusted R<sup>2</sup>=0.84, F(3,9)=75.56, p=0.029

Table B.23. The correlation analysis and ANOVA between TVS removal and sCOD changes, VFA production and CH<sub>4</sub> percentages at decreasing HRTs in one-phase reactor

<i>Regression Statistics</i>	
Multiple R	0.972183
R Square	0.94514
Adjusted R Square	0.806426
Standard Error	14.14031
Observations	11

ANOVA					
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	3	27558.28	9186.094	45.94233	5.67E-05
Residual	8	1599.587	199.9484		
Total	11	29157.87			

	<i>Coefficients</i>	<i>Std Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
X Variable 1	-0.118	0.236	-0.499	0.631	-0.663	0.427	-0.663	0.427
X Variable 2	1.351	0.422	3.200	0.013	0.378	2.325	0.378	2.325
X Variable 3	0.282	0.231	1.224	0.256	-0.250	0.814	-0.250	0.814

R<sup>2</sup> = 0.95. adjusted R<sup>2</sup> = 0.81. F(3,8) = 45.94. p < 0.05

Table B.24. The correlation analysis and ANOVA was performed between VSS removal and sCOD changes. tCOD removal and VFA production at decreased HRT from 8 h to 2 h in UAF reactor

<i>Regression Statistics</i>	
Multiple R	0.980
R Square	0.961
Adjusted R Square	0.928
Standard Error	2.920
Observations	12

ANOVA					
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	5	1255.754	251.151	29.453	0.000377
Residual	6	51.163	8.527		
Total	11	1306.917			

	<i>Coeff.</i>	<i>Std. Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
Intercept	128.83	23.662	5.4446	0.002	70.931	186.729	70.931	186.729
X Variable 1	1.058	2.077	0.509	0.629	-4.024	6.140	-4.024	6.139
X Variable 2	-0.256	0.282	-0.907	0.399	-0.947	0.435	-0.947	0.435
X Variable 3	0.147	0.157	0.942	0.382	-0.236	0.530	-0.236	0.530
X Variable 4	-0.154	0.099	-1.541	0.174	-0.398	0.091	-0.398	0.091
X Variable 5	-0.270	0.086	-3.136	0.020	-0.480	-0.059	-0.480	-0.059

$R^2=0.96$ . adjusted  $R^2=0.93$ .  $F(5,6)=29.45$ .  $p=0.001$

Table B.25. The correlation analysis and ANOVA between VSS removal and sCOD reductions. tCOD removal and VFA production at decreased HRT from 8 h to 2 h in UAF reactor ( $R^2=0.997$ . adjusted  $R^2=0.871$ .  $p=0.009$ )

<i>Regression Statistics</i>	
Multiple R	0.999
R Square	0.997
Adjusted R Square	0.871
Standard Error	6.077
Observations	12

ANOVA					
	<i>df</i>	<i>SS</i>	<i>MS</i>	<i>F</i>	<i>Significance F</i>
Regression	4	97442.34	24360.58	659.604	4.28E-09
Residual	8	295.457	36.932		
Total	12	97737.79			

	<i>Coeff</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>P-value</i>	<i>Lower 95%</i>	<i>Upper 95%</i>	<i>Lower 95.0%</i>	<i>Upper 95.0%</i>
X Variable 1	0.788	0.174	4.523	0.002	0.386	1.189	0.386	1.189
X Variable 2	0.058	0.190	0.306	0.767	-0.380	0.496	-0.380	0.4961
X Variable 3	0.053	0.250	0.212	0.837	-0.524	0.631	-0.524	0.631
X Variable 4	0.387	0.113			0.130	0.645	0.130	0.645

## APPENDIX C

### THE OPERATIONAL RESULTS OF SET 1 AND SET 2 EXPERIMENTS

Table C 1. Influent, effluent and percent reductions in suspended solid concentrations of UAF and UASB reactors

		Influent (mg/L)		UAF Reactor			UASB Reactor		
				Effluent (mg/L)		TSS Removal efficiency (%)	Effluent		TSS Removal efficiency (%)
Day	HRT (h)	VSS	TSS	VSS	TSS		VSS mg/L	TSS mg/L	
117	24	253	367	3 ±2.12	3±2.82	99	1 ±0.00	0.5±0.71	100
124	24	253	367	7 ±0.71	13±0.71	97	4 ±0.00	8 ±1.41	98
144	24	123	202	17±1.41	20±0.71	90	18±1.41	22 ±2.12	89
152	8	100	209	2±1.41	3.5±0.71	98	4 ±1.41	6 ±2.12	97
166	8	100	209	1±0.71	14±0.00	93	0 ±0.00	10 ±0.71	95
172	8	100	209	3±0.71	5±0.71	97	7.5±0.71	9 ±0.71	96
176	2	100	209	1±1.41	6 ±0.00	97	0 ±0.00	7 ±2.12	97
180	2	100	209	3±0.71	5.5 ±0.71	97	2.5±0.71	7 ±0.71	97
182	2	125	138	20±0.71	36±9.19	74	3 ±1.41	46 ±0.00	67
190	2	140	173	26±3.54	43 ±4.24	75	215±.95	58 ±16.3	67
192	2	140	173	31±2.83	56 ±2.83	68	24±2.83	65 ±14.8	63
196	2	144	170	32±4.24	44 ±5.66	74	27±4.24	64 ±4.24	62
201	2	144	170	30±2.83	68 ±1.41	60	28±1.41	73 ±9.19	57
206	2	158	173	34±20.5	44 ±2.83	75	23±5.66	42 ±5.66	76
211	2	158	173	34±1.41	38 ±7.07	78	23±1.41	44 ±2.82	75
226	2	136	165	37±2.12	46 ±3.54	72	31±4.24	42 ±7.07	75
237	2	136	165	32±1.41	40 ±5.66	76	18.5±11	38 ±2.83	77

Table C 2. The influent, effluent BOD<sub>5</sub> concentrations and removal performances of two-phase system and one-phase systems

Period	Day	Influent BOD <sub>5</sub> (mg/L)	Effluent BOD <sub>5</sub> (mg/L)			BOD <sub>5</sub> Removal Efficiency (%)			
			UAF R*	2 <sup>nd</sup> R.	UASB R*	UAF R*	2 <sup>nd</sup> R*	UAF+ 2 <sup>nd</sup> R*	UASB R*
1	274	185±05	76±2	24±0	35±3	58.92	68.42	87.03	81.08
3	288	185±25	95±5	25±1	23±1	48.65	73.68	86.49	87.57
4	295	65±25	32±4	24±2	25±4	50.77	25	63.08	61.54
5	304	200±10	55±5	20±0	41±2	72.50	63.64	90	79.50
5	309	135±20	63±3	20±0	46±2	53.33	68.25	85.19	65.93

R\*=Reactor

Table C 3. Average influent and effluent. pH. VFA. alkalinity contents of the UAF reactor. the 2<sup>nd</sup> phase reactor and the one-phase UASB reactor (VFA unit mg/L as Hac; Alkalinity unit mg/L as CaCO<sub>3</sub>; Values in parenthesis show the minimum and maximum values)

Parameter	Period 1			Period 2			Period 3			Period 4			Period 5		
	UAF	2 <sup>nd</sup>	UASB	UAF	2 <sup>nd</sup> R.	UASB									
Influent pH	7.71 (7.49-8.02)	7.17 (6.96-7.26)	7.71 (7.49-8.02)	7.4 (7.18-7.61)	7.05 (6.93-7.21)	7.4 (7.18-7.61)	7.62 (7.24-7.78)	7.14 (7.05-7.27)	7.62 (7.24-7.78)	7.71 (7.57-7.91)	7.10 (6.98-7.27)	7.71 (7.57-7.91)	7.55 (7.38-7.94)	7.09 (6.87-7.25)	7.55 (7.38-7.94)
Effluent pH	7.17 (6.96-7.26)	7.74 (7.53-8.06)	7.64 (7.45-7.84)	7.05 (6.93-7.21)	7.53 (7.23-7.75)	7.36 (7.04-7.66)	7.14 (7.05-7.27)	7.73 (7.53-8.02)	7.66 (7.47-7.78)	7.10 (6.98-7.27)	7.89 (7.84-8.02)	7.33 (7.24-7.47)	7.09 (6.87-7.25)	7.71 (7.48-7.85)	7.17 (6.91-7.41)
Influent VFA	19 (5-33)	48 (6-67)	19 (5-33)	44 (12-99)	54 (21-71)	44 (12-99)	55 (8-124)	49 (34-64)	55 (8-124)	11 (4-23)	38 (30-45)	11 (4-23)	16 (9-36)	45 (26-49)	16 (9-36)
Effluent VFA	48 (6-67)	31 (16-31)	19 (8-20)	54 (21-71)	14 (5-31)	26 (6-54)	49 (34-64)	10 (5-17)	14 (6-26)	38 (30-45)	11 (8-15)	11 (9-18)	45 (26-49)	24 (23-25)	20 (9-36)
Influent Alkalinity	121 (112-161)	81 (71-90)	121 (112-130)	91 (85-96)	77 (75-78)	91 (85-96)	119 (113-125)	76 (69-82)	119 (113-125)	117 (115-119)	70 (66-73)	117 (115-119)	126 (124-129)	86 (77-95)	126 (124-129)
Effluent Alkalinity	81 (71-90)	93 (82-105)	73 (55-91)	77 (75-78)	111 (108-113)	93 (80-106)	76 (69-82)	129 (128-129)	86 (75-96)	70 (66-73)	129 (128-130)	124 (121-126)	86 (77-95)	115 (112-119)	110 (98-131)

Table C 4. Average influent. effluent concentrations and removal efficiencies for VFA-COD in the UAF reactor

Period	Days	Influent VFA-COD (mg/L)	UAF React.	2 <sup>nd</sup> Phase React.	One Phase UASB React.	UAF React.	2 <sup>nd</sup> Phase React.	Two-phase UASB Reactor	One phase UASB React.
			Effluent VFA-COD (mg/L)			VFA-COD Removal Efficiency (%)			
Period 1	259-273	20	51	33	20	-153	35	-63	0
Period 2	274-280	47	58	15	28	-23	74	68	41
Period 3	281-288	59	52	11	15	11	80	82	75
Period 4	289-295	12	41	12	12	-245	71	0	0
Period 5	296-308	17	48	26	21	-181	47	-50	-25

Table C 5. Contents of Methane and Carbon dioxide and conversion rate of COD removed to methane during Periods 1-5

Period	CH4 (%)			CO2 (%)			N2(%)		
	UAF	2nd R.	UASB	UAF	2nd R.	UASB	UAF	2nd R.	UASB
1	23±6	43±11	37±7	24±6	4±1	6±2	53±11	53±11	57±2
2	29±5	47±4	44±2	25±5	3±1	9±2	46±4	50±4	47±2
3	17±2	32±1	36±1	21±3	3±1	8±1	62±1	65±1	56±1
4	21±4	31±5	22±5	16±3	3	8±2	63±6	66±5	70±2
5	13±4	24±2	14±2	7±3	4±1	12±4	80±6	72±2	74±4

R=Reactor

Table C 6. Methane Gas Production in studied HRTs/OLRs

	HRT (hour)			OLR=g sCOD/L.d (average)			mL CH <sub>4</sub> /day (average)		
	UAF R*	2 <sup>nd</sup> R*	UASB R*	UAF R*	2nd R*	UASB R*	UAF R*	2nd R*	UASB R*
Period 1	2	8	10	2.44	0.58	0.49	88.45	25.10	70.64
Period 2	2	6	8	3.10	0.70	0.77	85.90	57.20	119.40
Period 3	2	4	6	2.69	0.53	0.90	38.10	82.60	57.20
Period 4	2	2	4	1.22	0.38	0.61	20.18	73.70	45.90
Period 5	2	2	4	1.49	0.46	0.74	5.37	76.53	19.73

R\*=Reactor

Table C 7. Influent, effluent volatile suspended solids (VSS), total suspended solids (TSS) concentrations, TSS removal performances and VSS reductions of two-phase and one-phase systems

	Day	Influent (mg/L)	Effluent VSS (mg/L)			VSS Reductions (%)			
			UAF R*	2 <sup>nd</sup> R*	UASB R*	UAF R*	2 <sup>nd</sup> R*	UAF+ 2 <sup>nd</sup> R*	UASB R*
Period 1	261	93±2	58±2	24±3	22±2	38	59	74	76
Period 2	278	325±40	104±0	29±2	56±1	68	72	91	83
Period 3	288	80±0	64±4	35±2	42±3	20	45	56	48
Period 4	295	198	118±28	33±11	88±0	40	72	83	56
Period 5	305	122±34	88±3	34±20	73±5	28	61	72	40
	Day	Influent (mg/L)	Effluent TSS (mg/L)			TSS Removal Efficiency (%)			
			UAF R.	2 <sup>nd</sup> R.	UASB R.	UAF R.	2 <sup>nd</sup> R.	UAF+ 2 <sup>nd</sup> R.	UASB R.
Period 1	261	128±31	62±2	30±0	44±3	52	52	77	66
Period 2	278	403±46	125±0	38±3	92±6	69	70	91	77
Period 3	288	283±23	109±6	42±2	48±1	61	61	85	83
Period 4	295	254±14	190±46	44±0	127±6	25	77	83	50
Period 5	305	196±37	127±3	45±4	103±3	35	65	77	48

\*R= Reactor

Table C 8. The influent, effluent Total Volatile Solids (TVS), Total Suspended Solids (TS) concentrations and TS, TVS reductions of UAF, 2<sup>nd</sup> phase UASB, two-phase and one-phase reactors

TVS	Days	Influent(mg/L)	Effluent (mg/L)			Removal Efficiency (%)			
			UAF Reactor	2 <sup>nd</sup> phase Reactor	One-phase UASB Reactor	UAF Reactor	2 <sup>nd</sup> phase Reactor	Two-phase system	One-phase system
Period 1	259-273	233±12	133±9	117±8	119±9	43	11	50	49
Period 2	274-280	239±23	169±18	109±14	109±2	30	33	54	54
Period 3	281-288	170±6	130±9	74±11	78±2	19	42	55	52
Period 4	289-295	159±17	156±9	112±8	104±10	1	27	29	33
Period 5	296-308	171±25	160±28	100±13	98±9	7	37	59	61
TS	Day	Influent (mg/L)	Effluent (mg/L)			Removal Efficiency (%)			
			UAF Reactor.	2 <sup>nd</sup> phase Reactor	UASB Reactor	UAF Reactor	2 <sup>nd</sup> phase Reactor	Two-phase system	One-phase system
Period 1	259-273	1222±12	833±17	823±18	827±15	32	1	33	32
Period 2	274-280	1027±16	771±25	697±9	694±38	23	10	31	32
Period 3	281-288	990±11	851±50	749±30	790±57	14	12	24	20
Period 4	289-295	1044±13	927±14	833±10	926±7	11	10	20	11
Period 5	296-308	1194±21	1092±36	997±4	1036±55	8	9	16	13

Table C 9. Influent, effluent concentrations and removal percentages of protein for UAF, two-phase system and one-phase system (Periods 1- 4 at 35°C and Period 5 at 20°C)

Pe-riod	Day	Influent Protein (mg/L)	Effluent Protein (mg/L)			Protein Removal (%)			
			UAF	2 <sup>nd</sup>	UASB	UAF	2 <sup>nd</sup> phase	Two-phase	One-phase
1	267	132±20.99	58±35.72	10±0.84	10±0.63	56	83	92	92
1	273	68±5.52	32±0.72	9±0.09	8±0.19	53	72	87	88
2	278	228±24.71	51±9.54	11±2.66	11±2.28	78	78	95	95
3	287	25±7.60	14±6.46	4±0.38	6±0.76	44	71	84	76
4	295	56±0.38	15±0.76	4±1.14	13±0.38	73	73	93	77
5	304	105±30.03	22±22.81	4±3.42	14±24.3	79	82	96	87
5	309	73±21.29	21±47.52	5±0.76	15±2.66	71	76	93	79

Table C.10. Influent, effluent concentration and removal percentages of NH<sub>4</sub>-N, PO<sub>4</sub>-P and TP for UAF in two-phase UAF and UASB system and one-phase UASB system

NH <sub>4</sub> -N	Period	Influent (mg/L)	Effluent (mg/L)			Change (%)			
			UAF R.	2nd Phase R.	One phase Syst.	UAF.	2 <sup>nd</sup> Phase R.	Two-phase Syst.	One phase Syst.
	1	15±6.28	38±3.65	37±2.47	20±1.77	-153	3	-147	-33
	2	9±1.41	25±0	21±1.06	15±2.47	-172	22	-111	-67
	3	26±1.77	37±3.54	20±1.77	27±1.08	-40	45	22	-4
	4	27±1.06	33±0.71	13±15.91	32±1.06	-23	39	25	-19
	5	33±4.6	33±3.71	22±1.24	29±1.94	0	33	33	12
PO <sub>4</sub> -P	Period	Influent (mg/L)	Effluent (mg/L)			Removal (%)			
			UAF	2nd R.	UASB	UAF	2nd R.	UAF+2nd	UASB
	1	9±1.89	13±4.12	13±2.12	11±2.47	-44	0	-44	-22
	2	-	-	-	-	-	-	-	-
	3	12±1.77	13±2.83	12±2.12	13±1.77	-6	8	2	-6
	4	10±1.06	22±2.83	22±2.12	20±3.18	-110	0	-115	-95
	5	18±3.71	19±2.65	14±1.41	19±1.59	-6	26	22	-6
TP	Period	Influent	Effluent (mg/L)			Removal (%)			
			UAF	2nd Reac	UASB	UAF	2nd react	UAF+2nd	UASB
	1	22±1.10	20±0.23	19±0.59	16±0.48	9	5	14	27
	2	33±1.60	20±0.42	15±0.47	20±1.43	39	24	53	40
	3	28±0.24	24±0.35	18±1.78	18±0.83	17	24	37	38
	4	27±1.77	25±0.71	22±0.71	21±0.18	7	13	19	23
	5	35±0.47	31±0.71	30±21.25	29±0.48	11	3	14	17

## APPENDIX D

### SAMPLE CALCULATION

Table D.1. An example calculation for COD conversion

Influent Domestic Wastewater:
(Average values for Period 3)
tCOD= 509 mg/L
sCOD= 295 mg/L
VFA-COD (acidified fraction)= $1.066^1 * 0.04^{11} * \text{tCOD}$ =22 mg/L(Table 4.2)
sCOD/tCOD (hydrolyzed fraction)= $0.49^{11} * \text{tCOD}$ =249 mg/L(Table 4.2)
Effluent Domestic Wastewater:
(Average values for Period 3; 40% tCOD removal and 11% sCOD removal)
tCOD=305 mg/L
sCOD= 263 mg/L
VFA-COD= $1.066^1 * 145$ mg/L (average effluent VFA)= 155 mg/L
Results
Removed tCOD= influent tCOD- effluent tCOD=509-305= 204 mg/L
About 61% of influent sCOD (hydrolyzed) was removed.in effluent
Remaining COD( in inert/ particulate and slowly degradable )=305-263=42 mg/L

<sup>1</sup> VFA(as acetic acid) conversion rate to VFA-COD

<sup>11</sup> Table 4.2 winter time VFA/COD ratio

## APPENDIX E

### ENERGY GENERATED AND ENERGY NEEDED FOR HEATING

Assuming;

Wastewater flow = 1 MLD

Raw wastewater temperature= 20°C

Heat required (RH) to raise the incoming waste water temperature (T) to 35°C:

$$RH = Q_m \cdot W_s \cdot (T_2 - T_1)$$

Mass flow rate (Q<sub>m</sub>)

$$Q_m = 1 \cdot 10^6 \text{ (L/d)} \cdot (1 \text{ kg/1 L}) = 1 \cdot 10^6 \text{ (kg/L)}$$

W<sub>s</sub>= Specific heat of water (1 calorie/gram °C)

$$RH = 1 \cdot 10^6 \text{ (kg/L)} \cdot 1 \text{ (cal/g)} \cdot 10^3 \cdot (35 - 20) = 15 \cdot 10^9 \text{ cal/d}$$

Total available heat content of CH<sub>4</sub> produced:

*Period 4 at 35°C and 4 h HRT*

Influent tCOD=332 mg/L

UAF reactor effluent tCOD= 97 mg/L

2<sup>nd</sup> Phase UASB reactor effluent= 39 mg/L

One Phase UASB reactor=76 mg/L

COD removed in UAF reactor=(332-97) mg/L=235 mg/L=0.235 g/L

COD removed in 2<sup>nd</sup> Phase UASB reactor=58 mg/L=0.058 g/L

COD removed in one-phase reactor=256mg/L = 0.256 g/L

CH4 conversion rate at Period 4:

UAF reactor: 0.11 N m<sup>3</sup> CH4/kg COD removed

2nd Phase reactor= 0.202 N m<sup>3</sup> CH4/kg COD removed

One-phase UASB reactor= 0.045 N m<sup>3</sup> CH4/kg COD removed

Total methane produced in UAF reactor= CH4 conversion rate\* COD removed/day

Total methane produced in UAF reactor= 0.11\*0.235\*106

Total methane produced in UAF reactor=25850 L/d

Total methane produced in 2<sup>nd</sup> Phase reactor= 0.202\*0.058\*106

Total methane produced in 2<sup>nd</sup> Phase reactor=11716 L/d

Total methane produced in two-phase UAF and UASB system=37566 L/d

Total methane produced in one-phase system= CH4 conversion rate\* COD removed/day

Total methane produced in one-phase system= 0.045\*0.256\*106

Total methane produced in one-phase system=11520 L/d

*Period 5 at 20°C*

Influent tCOD=310 mg/L

UAF reactor effluent tCOD=166 mg/L

2nd Phase UASB reactor effluent=53 mg/L

One Phase UASB reactor=105 mg/L

COD removed in UAF reactor=(332-97)mg/L=144 mg/L

COD removed in 2<sup>nd</sup> Phase UASB reactor=257 mg/L

COD removed in one-phase reactor=205mg/L

CH4 conversion rate at Period 4:

UAF reactor: 0.005 N m<sup>3</sup> CH4/kg COD removed

2<sup>nd</sup> Phase reactor= 0.113 N m<sup>3</sup> CH4/kg COD removed

One-phase UASB reactor=  $0.029 \text{ N m}^3 \text{ CH}_4/\text{kg COD removed}$

CH<sub>4</sub> conversion rate at Period 5:

UAF reactor:  $0.005 \text{ N m}^3 \text{ CH}_4/\text{kg COD removed}$

2nd Phase reactor=  $0.113 \text{ N m}^3 \text{ CH}_4/\text{kg COD removed}$

One-phase UASB reactor=  $0.029 \text{ N m}^3 \text{ CH}_4/\text{kg COD removed}$

Total methane produced in UAF reactor= CH<sub>4</sub> conversion rate\* COD removed/day

Total methane produced in UAF reactor=  $0.005*0.144*106$

Total methane produced in UAF reactor=720 L/d

Total methane produced in 2<sup>nd</sup> Phase reactor=  $0.113*0.257*106$

Total methane produced in 2<sup>nd</sup> Phase reactor=29041 L/d

Total methane produced in two-phase UAF and UASB system=29761 L/d

Total methane produced in one-phase system= CH<sub>4</sub> conversion rate\* COD removed/day

Total methane produced in one-phase system=  $0.029*0.205*106$

Total methane produced in one-phase system=5945 L/d

Total available Heat content:

Assuming heat value of CH<sub>4</sub>=6475 cal/L

*At period 4*

Total methane produced in two-phase UAF and UASB system=37566 L/d

Total methane produced in one-phase system=11520 L/d

Two-phase UAF and UASB system available heat content:  $6475*37566=2.4*10^8$  cal/d

One-phase system available heat content:  $6475*11520=7.5*10^7$  cal/d

*At period 5*

Total methane produced in two-phase UAF and UASB system=29761 L/d

Total methane produced in one-phase system=5945 L/d

Two-phase UAF and UASB system available heat content:  $6475 \times 29761 = 1.9 \times 10^8$  cal/d

One-phase system available heat content:  $6475 \times 5945 = 3.9 \times 10^7$  cal/d

Comparison of available heat content and heat required to raise the incoming waste water temperature to 35°C:

Heat required (RH) =  $15 \times 10^9$  cal/d

Two-phase system available heat content =  $2.4 \times 10^8$  cal/d

One-phase system available heat content =  $7.5 \times 10^7$  cal/d

### **Result**

For two-phase system and one-phase system;

Heat required to raise the temperature to 35°C > Available heat content.

For dilute wastewaters, raising the temperature did not provide a good solution because of the available heat content was considerably less than the heat required to raise the temperature. Instead of raising the temperature, experimenting the system at 20°C gives a better result.

The two-phase system satisfies the effluent criteria at 4 h HRT and 20°C. Therefore, the application of two-phase system in low strength wastewater treatment would provide an extra  $1.9 \times 10^8$  cal/d (9201 Watt/day = 9.2 kW/day).

## CURRICULUM VITAE

### PERSONAL INFORMATION

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### EDUCATION

Degree	Institution	Year of Graduation
MS	Boğaziçi University Civil Engineering	1984
BS	Boğaziçi University Civil Engineering	1981
High School	Erdek Lisesi	1976

### WORK EXPERIENCE

Year	Place	Enrollment
2008- Present	BOTAŞ	Consultant
2001-2008	BOTAŞ	Head of Engineering and Contracts Dept.
1986-2001	BOTAŞ	Engineer and Head Engineer
1984-1984	M.Alptekin İnş.Ltd.	Engineer
1979-1984	Boğaziçi University	Student Asistant and Teaching Asistant