INVESTIGATION OF BIOGAS PRODUCTION POTENTIAL OF THREE DIFFERENT SLUDGES AFTER THERMAL HYDROLYSIS

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ECE ARI AKDEMİR

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submitted by **ECE ARI AKDEMİR** in partial fulfillment of the requirements for the degree of **Master of Science in Environmental Engineering Department, Middle East Technical University** by,

Prof. Dr. Halil Kalıpçılar
Dean, Graduate School of **Natural and Applied Sciences**

Prof. Dr. Bülent İçgen
Head of Department, **Environmental Eng.**

Prof. Dr. F. Dilek Sanin
Supervisor, **Environmental Eng., METU**

Assoc. Prof. Dr. L. Selim Sanin
Co-Supervisor, **Environmental Eng., Hacettepe Uni.**

Examining Committee Members:

Prof. Dr. İpek İmamoğlu
Environmental Engineering, METU

Prof. Dr. F. Dilek Sanin
Environmental Eng., METU

Assoc. Prof. Dr. Tuba Hande Bayramoğlu
Environmental Engineering, METU

Assoc. Prof. Dr. Emre Alp
Environmental Engineering, METU

Assist. Prof. Dr. Merve Görgüner
Environmental Engineering, Hacettepe Uni.

Date: 08.07.2019

I hereby declare that all information in this document has been obtained and presented in accordance with academic rules and ethical conduct. I also declare that, as required by these rules and conduct, I have fully cited and referenced all material and results that are not original to this work.

Name, Surname: Ece Arı Akdemir

Signature:

ABSTRACT

INVESTIGATION OF BIOGAS PRODUCTION POTENTIAL OF THREE DIFFERENT SLUDGES AFTER THERMAL HYDROLYSIS

Arı Akdemir, Ece
Master of Science, Environmental Engineering
Supervisor: Prof. Dr. F. Dilek Sanin
Co-Supervisor: Assoc. Prof. Dr. L. Selim Sanin

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Biogas production through anaerobic digestion using biological sludge is one the sustainable approaches. Hydrolysis is the first and rate limiting step during anaerobic digestion. For enhancement of hydrolysis of sludge, it is feasible to investigate pre-treatment before anaerobic digestion process. In this research, biogas and methane production potentials of municipal wastewater treatment plants sludge, membrane bioreactor sludge and organized industrial district sludge were investigated using small scale laboratory anaerobic digesters to enhance the rate limiting step through thermal hydrolysis. Sludge was pre-treated using thermal hydrolysis with different durations and temperatures. To obtain optimum anaerobic digestion triplicate BMP reactors were operated. The results of this study showed that, the methane production performance of biological sludge can be enhanced with thermal hydrolysis operated at low pressure. There is an optimum exposure time for effective thermal hydrolysis process. The results show that sludge pretreated at 127^{o}C for 60 minutes produced the highest gas volume and improved methane to carbon dioxide ratio, in biogas. For the municipal sludge (sludge from Tatlar WWTP). Methane content of biogas was 50%, 57% and 59% for non-processed, 30 minutes thermally hydrolyzed and 60 minutes thermally hydrolyzed operation, respectively. For the domestic sludge from MBR wastewater treatment process %53, %56, and %59 methane gas was obtained in biogas for non-processed, 30 minutes thermally hydrolyzed and 60 minutes thermally hydrolyzed operation, respectively. Methane generation of MBR, MBR127/60 and MBR90/30 were 53%, 59% and 56% respectively. For organized industrial district wastewater sludge %50, %55 and %58 methane was obtained for non-processed, 30 minutes thermally hydrolyzed and 60 minutes thermally hydrolyzed operation, respectively. Non-pretreated sludge had had the lowest methane percentage in biogas for all sludge types.

Keywords: Anaerobic digestion, Sludge management, Thermal hydrolysis, Biochemical methane potential test, Methane production

ÖZ

ÜÇ FARKLI ÇAMURUN BİYOGAZ ÜRETİM POTANSİYELİNİN TERMAL HİDROLİZ SONRASI İNCELENMESİ

Arı Akdemir, Ece
Yüksek Lisans, Çevre Mühendisliği
Tez Danışmanı: Prof. Dr. F. Dilek Sanin
Ortak Tez Danışmanı: Doç. Dr. L. Selim Sanin

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Biyolojik çamur kullanarak anaerobik çürütme yoluyla biyogaz üretimi sürdürülebilir yaklaşımlardan biridir. Hidroliz, anaerobik çürütme sırasındaki ilk ve hız sınırlayıcı adımdır. Çamur hidrolizinin iyileştirilmesi için, anaerobik çürütme işleminden önce ön arıtımın incelenmesi mantıklıdır. Bu araştırmada, evsel atıksu arıtma tesisi çamuru, membran biyoreaktör çamuru ve organize sanayi bölgesi çamurunun biyogaz ve metan üretim potansiyelleri, termal hidroliz kullanılarak hız sınırlayan adımı iyileştirmek için, küçük ölçekli laboratuvar anaerobik çürütücüleri kullanılarak araştırılmıştır. Çamur, farklı süre ve sıcaklıklardaki termal hidroliz yöntemi kullanılarak ön arıtıma tabi tutulmuştur. Optimum anaerobik çürütme elde etmek için üçlü BMP reaktörleri çalıştırılmıştır. Bu çalışmanın sonuçları, biyolojik çamurun metan üretim performansının, düşük basınçta uygulanan termal hidroliz ile arttırılabileceğini göstermiştir. Etkili termal hidroliz işlemi için optimum maruz kalma süresi vardır. Sonuçlar, 127 ^{o}C'de 60 dakika boyunca ön işleme tabi tutulan çamurun, evsel Tatlar atıksu arıtma tesisi çamuru için en yüksek gaz hacmini ürettiğini ve biyogazda metan-karbondioksit oranını geliştirdiğini göstermektedir. Biyogazın metan içeriği, işlenmemiş, 30 dakika termal hidroliz edilmiş ve 60 dakika termal hidroliz edilmiş örnekler için sırasıyla %50, %57 ve %59'dir. MBR atık su arıtma işleminden elde edilen evsel çamurdan, işlenmemiş, 30 dakika termal hidroliz edilmiş ve 60 dakika termal hidroliz edilmiş örnekler için sırasıyla biyogazda %53, %56 ve %59 metan gazı elde edilmiştir. MBR numunesi için MBR, MBR127/60 ve MBR90/30'un metan üretimi sırasıyla 53%, 59% ve 56%'dır. Organize sanayi bölgesi atık su çamuru için, işlenmemiş, 30 dakika termal hidroliz edilmiş ve 60 dakika termal hidroliz edilmiş örnekler için sırasıyla %50, %55 ve % 58 metan elde edilmiştir. Tüm çamur tipleri içinde, biyogazda metan yüzdesinin en düşük olduğu çamur, ön arıtıma tabi tutulmamış çamur çıkmıştır.

Anahtar Kelimeler: Anaerobik stabilizasyon, Çamur yönetimi, Termal hidroliz, Biyokimyasal metan potansiyeli testi, Metan üretimi

To my family and my love…

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LIST OF ABBREVIATIONS

BMP: Biochemical methane potential

CAS: Conventional activated sludge

COD: Chemical oxygen demand

CST: Capillary suction time

DS: Dry solids

GC: Gas Chromatograph

LWA: Lightweight aggregate

MBR: Membrane bioreactor

METU: Middle East Technical University

MF: Microfiltration

OID: Organized industrial district

RAS: Return activated sludge

SCOD: Soluble chemical oxygen demand

SRT: Solid retention time

TCD: Thermal conductivity detector

TCOD: Total chemical oxygen demand

THP: Thermal hydrolysis process

TS: Total solids

TSS: Total suspended solids

UF: Ultrafiltration

VFA: Volatile fatty acids

VS: Volatile solids

VSS: Volatile suspended solids

WAS: Waste activated sludge

WWTP: Waste water treatment plant

CHAPTER 1

INTRODUCTION

Environmental pollution and how to deal with it using effective treatment methods are among the crucial problems of the industrialized world. Feasibility of utilizing different types of sludge produced from wastewater plants is extensively studied for the last decades (Barber, 2016; Abelleira-Pereira et. al., 2015). For instance, activated sludge is widely applied for treating domestic and industrial wastewater biologically. Activated sludge enhances transformation of dissolved organic matter in wastewater to biomass, which culminates into water and carbon dioxide (Liu et. al., 2012; Xue et. al., 2015). WAS is formed throughout biological wastewater treatment. Since more wastewater is produced with the increasing population and water demand, so is the amount of WAS (Liao et al., 2016; Kim et al., 2003).

Feasible management of sewage sludge is crucial but costly operation untreated sludge has a potential to damage the environment. Sludge management accounts for around 30-40% of the capital cost and up to 50% of the operating cost of wastewater treatment plants (Vlyssides and Karlis, 2004). Furthermore, due to heavy metals, volatile micropollutants, and pathogen presence in sewage sludge, it requires a meticulous treatment (Serrano et al., 2015; Doğan and Sanin, 2009). In European Union, sewage sludge is regarded as hazardous waste. Since sewage sludge produces odorous emissions and enhances generation of highly polluted leachate, landfill disposal is not suggested as a proper management practice. In addition, sewage sludge has resources (organic matter and nutrients) to be recovered. Therefore, it is not acceptable to simply dispose it of to the landfills. In this regard, European Union member states had the obligation to reduce sewage sludge disposal to landfills by 35% in the year 2016 compared to the year 2000 (Lundin et al., 2004).

To preclude the environmental drawbacks of sludge and minimize the potential danger for public health numerous sludge stabilization options are applied such as composting, aerobic digestion, and anaerobic digestion. Anaerobic digestion is the most widely applied method since it offers the possibility to produce biogas (Baier and Schmidheiny, 1997; Doğan and Sanin, 2009). Produced biogas is used for generating electricity, which is commonly used to meet the demand of wastewater treatment plant (Hendriks et al., 2014).

Anaerobic digestion is composed of four steps, which are hydrolysis, acidogenesis, acetogenesis, and methanogenesis (Kim et al., 2003). In the hydrolysis step, solubilisation of particulate matter occurs, and organic polymers are decomposed biologically. Hydrolysis takes place very slowly, which makes this step the rate-limiting step of anaerobic digestion. Hydrolysis is followed by acidogenesis and acetogenesis. Lastly, methane is produced by methanogenic archaea in the final step, which is called as methanogenesis (Gavala et al., 2003).

There are numerous advantages of anaerobic digestion such as volume reduction and enhancement on the dewaterability of sludge, and production of biogas which is used for energy generation (Abe et al., 2013). Also, the cost of anaerobic digestion is less than aerobic digestion. Lastly, the impact of anaerobic digestion on the environment is relatively less (Xue et. al., 2015). Even though anaerobic digestion is regarded as cheap and clean technology, it has various disadvantages and limitations as well. For instance, anaerobic digestion requires high retention time, and it has low efficiency on the degradation of organic solids (around 30-50% for mesophilic anaerobic digestion)

(Albelleira-Pereira et. al., 2015). Moreover, anaerobic digestion is not efficient for the degradation of facultative anaerobic biomass. Anaerobic digestion also results in less efficient volatile solid degradation, and it may lead to the production of inadequate quantity and quality biogas (Doğan and Sanin, 2009; Bolzonella et al., 2005; Serrano, et al., 2015).

As mentioned before, hydrolysis is the rate-limiting step of anaerobic digestion (Ariunbaatar et al, 2014; Kim et al., 2003). Insoluble organics could not solubilize completely, influencing adversely the following steps, which results in low digestion efficiency. There are different pretreatment methods which are applied on WAS to decrease the effect of rate limiting step. These pretreatment methods include mechanical disintegration and ultrasonic, alkaline, and thermal pretreatment (Müller,2001; Köksoy and Sanin, 2010). Pretreatment methods may expedite the hydrolysis of sewage sludge and results in better anaerobic digestion efficiency by reducing particle size and increasing soluble COD amount (Da Silva et al., 2018; Kim et al., 2003). Thus, applying pretreatment is a feasible way to increase anaerobic digestion efficiency. Full-scale implementation of pretreatment methods is widely applied on sewage sludge in terms of physical, alkaline, heat-shock, biochemical, freezing, and thawing means (Carrere et al., 2010; Cesaro and Belgiorno, 2014; Serrano et al., 2015).

Various types of pretreatment processes have been studied to increase the solubilisation of solids in sewage sludge (Gavala et al., 2003). Along the anaerobic digestion process, biogas is produced regardless of the pretreatment.

Nevertheless, since hydrolysis is the rate-limiting step, biogas and in turn energy production are limited. In other words, the hydrolysis step is expedited by using pretreatment methods (Ariunbaatar et al., 2014; Doğan & Sanin, 2009). Also, pretreatment methods result in the sterilization of sludge, which is beneficial due to the elimination of the sterilization cost (Ariunbaatar et al., 2014).

Although, thermal hydrolysis is investigated for municipal sludge as a pretreatment method, in the literature, application of thermal hydrolysis on industrial sludge is uncommon. Research on low pressure and temperature thermal hydrolysis application is also limited. Additionally, efficiency of thermal hydrolysis is not investigated and compared for different sludge types. This research focused on investigating the efficiency of thermal hydrolysis on different sludge types, their performance differences at low pressure conditions.

There are three main objectives of this study:

1. To compare the effects of thermal pretreatment of conventional wastewater treatment sludge, membrane bioreactor sludge, and sludge from an organized industrial district at different times and temperatures,
2. For three different types of sludge, to investigate optimum thermal pretreatment condition for solubilisation of COD and
3. To investigate the methane production potential of these three sludge that underwent thermal pretreatment.

This study comprises of two main parts, which are a preliminary pretreatment experiment of thermal hydrolysis to enhance solubility for three different sludge types and to determine the effect of thermal hydrolysis pretreatment on biogas production and composition. First selected sludge was a WAS sample from a typical municipal WWTP which was also used as a reference sludge. Thermal hydrolysis is commonly applied to domestic and municipal sludge, worldwide. The second sludge was an MBR sludge. MBR sludge has longer solid retention time with respect to typical domestic

WWTP which means that typical domestic WWTP has less initial stability. Therefore, lower methane generation can be expected from MBR sludge. Last selected sludge samples obtained from an OID. OID has vast amount of sludge after treatment process. In addition, ID sludge possibly contain toxic compounds due to various sectoral wastewater. Therefore, management of OID sludge has crucial for OID.

CHAPTER 2

LITERATURE REVIEW

2.1. Sludge Management

Sludge is the solid residue generated along wastewater treatment as well as numerous environmental processes (Gurjar et al., 2017; Gavala et al., 2003). Increasing population in residential areas causes significant increase in municipal sludge formation, which leads to accumulation of vast amounts. For instance, approximately 3.5 million tons of sludge production annually, in terms of dry solids, has been reported in the countries located on the Baltic Sea basin, and this value is predicted to be 4 million tons by 2020 (Pure, 2012).

On the other hand, in Turkey, approximately 5,500 ton DS/day municipal sludge is produced at a production rate of 35 g DS/capita/day (Ministry of Environment and Urbanization, 2013). However, sludge production rate varies from country to country, which ranges from 25 to 107 g DS/capita day (Sanin et al. 2011). Table 2-1 shows the sludge production rates of some countries for 2015.

Table 2-1. *Total sludge production for European Countries in 2015 (Eurostat, n.d.).*

Country	Total Sludge Production (thousand tonnes/year)
Germany	1,820.6
Poland	568.0
Romania	210.4
Czechia	210.2
Sweden	197.5
Hungary	156.8
Albania	91.5
Ireland	58.4
Bulgaria	57.4
Slovakia	56.2
Lithuania	42.8
Slovenia	29.1
Croatia	17.9
Serbia	10.8
Luxembourg	9.1
Malta	8.4
Cyprus	6.6

Due to the amount of sludge, the sludge treatment process is a crucial part of wastewater treatment. Also, sludge is can be a significant resource for the recovery of various nutrients in its composition and its energy potential. Due to the high volume of sludge generated in WWTPs, sludge management consists of 30-40% of capital costs and approximately 50% of operating cost of the plant (Haug et al., 1978; Vlysside and Karlis, 2004). Sludge management gains importance in each passing day and becoming a serious environmental problem as secondary treatment is being applied more and more with stricter national standards (Abelleira-Pereira et al., 2015, Haug et al., 1978).

Challenges and the benefits of sludge management can be handled together since responsible and sustainable removal is needed to preserve the environment from serious drawbacks (Pure, 2012). While drawbacks of sludge such as large volume and contaminants in sludge (heavy metals, pathogens, and trace organics) can be a starting point for the nutrients in sludge, which is suitable for agricultural use, and organic matter, which is good for enriching soil and beneficial for energy production (Pure, 2012).

2.1.1. Sludge Types

In wastewater treatment plants, there are various sources of sludge generation. Depending on the plant type and applied systems, main sludge sources of a wastewater treatment plant are primary sedimentation, biological treatment, secondary sedimentation, and sludge processing facilities. Characteristics of the sludge resulted from these sources may differ to a great extent (Tchobanoglous et. al., 2003).

Properties of municipal sludge and industrial sludge are different. These properties vary greatly with respect to wastewater properties and applied process. Generally, specific gravity of industrial sludge is higher than municipal sludge. On the other hand, pH and moisture content of municipal sludge potentially higher than industrial sludge. Lastly, municipal sludge has more loss on ignition percent than industrial sludge since municipal sludge consist of more organic substances. Inorganic materials and metals might be higher in industrial wastewaters (KuanYeow et. al., 2018).

Biological treatment process affects the sludge properties to a great extent. For example, MBR sludge has a lower particle size with respect to sludge resulting from CAS systems. This situation leads MBR sludge to have worse dewaterability properties. Moreover, MBR sludge has longer SRT than CAS sludge, which means that CAS sludge has less initial stability. (Pontoni et. al., 2017). With respect to CAS sludge, it is expected that MBR sludge may lead to lower methane generation due to its longer SRT and more initial stability. This situation is caused by endogenous microorganisms in the reactor, which could lead to formation of slowly degrading or non-biodegradable compounds. Moreover, these compounds may inhibit methane production due to the presence of aromatic moieties. Whereas digestion of CAS sludge generally follows a first order kinetic, digestion of MBR sludge follows sigmoidal-like trend which include longer lag phase due to less hydrolysable compounds (Pontoni et. al., 2015).

There are various sludge disposal methods but incineration, landfilling, land application and use of sludge as raw material are the most used ones. Before disposal, anaerobic digestion has been widely applied as a convenient method of sludge stabilization, which is also among the oldest stabilization methods (Gavala et al., 2003). Sludge stabilization may lead to sludge minimization, which further decreases the transportation and disposal costs. Thus, sludge minimization has notable significance in sludge treatment processes (Morosini et al., 2016).

2.1.2. Sludge Treatment Methods

Before sludge can be disposed in an appropriate way, it needs to be treated properly. Major steps of sludge treatment include thickening, stabilization, dewatering, and drying. After applying these steps, effective and feasible sludge disposal can be carried out (Tchobanoglous et. al., 2003).

2.1.2.1. Methods That Reduce the Water of Sludge

There are three treatment methods which are used to reduce the water content of the sludge: Thickening, dewatering, and drying.

In order to remove suspended solids in sludge, thickening is applied. Main objective of thickening process is to concentrate suspended solids in sludge and to separate these solids from liquid. By using thickening, process volume is decreased which means there is less sludge to be handled by the downstream processes. Thickening is beneficial for wastewater treatment plants since it leads to decrease water content of the sludge and in turn reduction in cost due to smaller sludge volume (WEF, 1996). Generally physical forces are used in thickening such as gravity settling, flotation, co-settling, and centrifugation (Tchobanoglous et. al., 2003). Some examples of thickening methods such as gravity thickening, dissolved air flotation, centrifugation, gravity belt thickener, and rotary drum thickener (Ontario, 2019).

Dewatering is also a physical process in which solids are separated from water in sludge. Dewatering is applied to further reduce water content of sludge. After applying dewatering, part of sludge having the high solid concentration is called “cake”. Furthermore, dewatering is advantageous since sludge volume is decreased and inturn transportation and further sludge operation costs are reduced. Solid bowl centrifuge, and belt filter press are two examples of the sludge dewatering methods (Tchobanoglous et. al., 2014).

Sludge drying includes using thermal energy to evaporate remaining water content of sludge after thickening and dewatering. Since sludge volume is decreased by drying, storage, transportation, and disposal costs of sludge reduces (IWA Publishing, 2019). Direct dryers, which contains flash dryer, rotary dryer, and fluidized-bed dryer, and indirect dryers are two types of drying applications (Tchobanoglous et. al., 2003).

2.1.3. Beneficial Use of Sludge and Disposal Methods

2.1.3.1. Incineration

Incineration is regarded as the most feasible method for waste disposal, especially in Europe. Tightening regulations on landfilling, agricultural land application and sea disposal led to an increase in the tendency to use incineration as a disposal method in the long term (Malerius, 2003). In the last decades, incineration technology in terms of engineering and energy efficiency has improved to a great extent (Minimi et al., 1997; Fytili et al., 2008). It is possible to incinerate sludge solely as well as simultaneously with other fuels. For instance, sludge can be incinerated with municipal solid waste (Tchobanoglous, Stensel, Tsuchihashi, & Burton, 2014).

The literature has research results which indicate the advantages of using incineration as a beneficial use/disposal method. According to Sanin et al., 2011; Khiari et al., 2004; Fytili et al., 2008, sludge volume decreases when combustion is used as a disposal method and dangerous pathogenic microorganism and a list of organic compounds which has toxic effects are destroyed along incineration process. The energy content of the waste can be recovered and odor problems are significantly reduced by using incineration for sludge management (Fytili et al., 2008).

On the other hand, incineration has limitations as a process. Ash, which is the main end product of incineration, is needed to be disposed properly (Menendez, et al., 2002; Fytili et al., 2008; Sanin et al., 2011). Inorganic compounds may change forms under high temperature bur are not destroyed during incineration, and their concentration is enhanced in the bottom ash, which may lead to hazards by leaching underground level (Sanin et al., 2011). There are severe public concerns on potential hazardous emissions generated from incineration processes (Fytili et al., 2008; Sanin et al., 2011).

One approach of eliminating ash from incineration is to co-combust sludge in cement production. This way sludge ash mixes into the cement material and ends-up producing no ash. Incineration based on refuse-derived fuel is another type of incineration. This type of incineration is favored by cement factories, and less air is required than mass incineration. Consequently, emission control is much easier and cost is lower than mass incineration (Worrell & Vesilind, 2012).

2.1.3.2. Land Application

Land application of sludge consists of spreading, injecting or spraying sludge onto the ground level to benefit from sludge's fertilizer qualities (EPA, 1994). Properties of sludge depend on the treatment type and source of sludge. Since sludge has necessary nutrients both organic and inorganic, it possibly used as fertilizer. In this sense, land application of sludge is a promising disposal method (Singh & Agrawal, 2008). Sludge might be applied to land used for agricultural purposes, reclamation areas, forest, areas which may include public contact such as golf courses and parks and gardens (EPA, 1994; Sanin et al., 2011). On the other hand, the application of sludge on land is restricted due to the presence of potentially toxic materials and microorganisms in sludge (Singh & Agrawal, 2008). One critical issue about land application is that raw sludge is not allowed to be used on land worldwide. Sludge is required to be stabilized biologically or chemically before it is allowed to be and applied.

2.1.3.3. Usage as Raw Material

Sludge possibly used as a raw material for making bricks, concrete filling and LWA concrete. In this sense, using sludge as a construction material presents a technical alternative for sludge disposal (Jiang, Ni & Ma, 2011). Moreover, residual ash from sludge incineration might be utilized as construction or filling material as well. In this sense, remaining ash which constitutes approximately 30% of the initial waste, possibly disposed properly (Fytili et al., 2008, Abiero & Owili, 2016). Residual ash from incineration of sludge is stored by mixing with asphalt or cement (Sato, Oyamada & Hanehara, n.d.). Furthermore, material produced by mixing soil with sludge is used for road building, foundation filling of a building and enhancing soil bearing capacity of any structure (Portela et al., 2014).

2.1.3.4. Landfilling

Wastes are compressed and situated in landfills; afterward, landfill sites are covered to prevent water and air exposure (Mihelcic & Zimmerman, 2010). In many countries, landfilling offers an economical and easy way to dispose wastes. Many developed and developing countries use landfills for domestic and industrial waste disposal (Singh & Agrawal, 2008, Seshadri et al., 2016). However, landfills lead to generation of highly concentrated and polluted leachate which can mix with groundwater and cause serious environmental and health problems (Bjerg et al., 2003). Leachate can be defined as a liquid which gets in contact with wastes and turns into highly polluted wastewater. Since organic substances in landfills decompose biologically by microorganisms that use oxygen and produce carbon dioxide, anaerobic conditions arise in time which in turn leads to production of methane by anaerobic digestion (Mihelcic & Zimmerman, 2010). Though landfills may seem like an economical way of waste disposal, it can cause serious environmental problems. Main environmental drawbacks of landfilling include surface and groundwater pollution, and greenhouse gas and odor emissions (Albright et al., 2006; Seshadri et al., 2016).

2.2. Anaerobic Digestion

Anaerobic digestion, which takes place in the absence of free oxygen, is a biological process that contains organic substance break-down into useful biogas, solid and liquid end products (Chen and Neibling, 2014). Biogas comprises of mainly methane (approximately 60-70% by volume) and carbon dioxide (around 30-40% by volume). Small quantities of trace gases and H_2S possibly contained in biogas as well. Biogas is a valuable end product, which might be combusted to produce electricity and heat. Biogas might be utilized as renewable natural gas and fuel for transportation purposes (Chen and Neibling, 2014).

Anaerobic digestion is among the oldest methods used for organic substance stabilization (Ariunbaatar et al., 2014). Furthermore, anaerobic digestion is a feasible method, which is widely used for sludge stabilization as well. There are numerous advantages of anaerobic digestion such as (Chen and Neibling, 2014; Sapkaite et al., 2017):

- Minimal environmental drawbacks,
- Potential electricity generation from biogas,
- Decreases volume of sludge to be disposed of,
- Increases dewaterability of sludge,
- Decreases odor problems which enhance air quality,
- Leads to the enhancement of water quality by preventing pathogenic microorganisms from entering watershed,
- Effective control of pathogenic microorganisms,
- Beneficial in terms of nutrient recovery.

As shown in Figure 2-1, anaerobic digestion consists of four main stages which are hydrolysis, acidogenesis, acetogenesis, and methanogenesis.

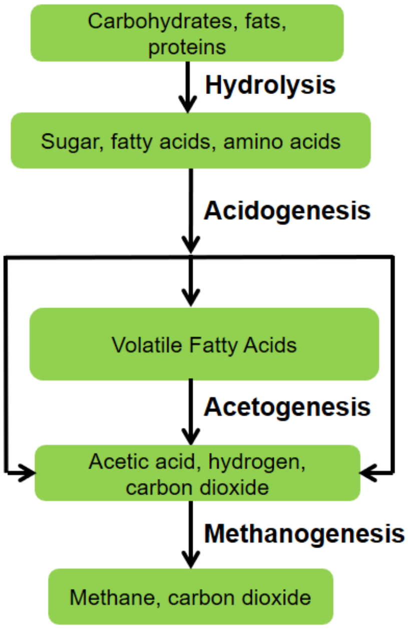


Figure 2-1. Anaerobic Digestion Steps (Chen and Neibling, 2014)

- **Hydrolysis:** The first stage of anaerobic digestion, hydrolysis, takes place by the help of extracellular enzymes of hydrolytic microorganisms. This stage includes decomposition of complex organic polymers into soluble and simpler monomers (Chen and Neibling, 2014).

- **Acidogenesis:** In this step, simpler and soluble monomers from hydrolysis are transformed by acidogens (fermentative microorganism) into an assortment of volatile fatty acids (VFA) and other ends products such as carbon dioxide, acetic acid, and hydrogen. Acidogenesis is generally the fastest step of the anaerobic digestion. (Chen and Neibling, 2014).

- **Acetogenesis:** In this stage, VFAs are converted into acetate, hydrogen and carbon dioxide by acetogenic bacteria (Chen and Neibling, 2014).

- **Methanogenesis:** In the last step of anaerobic digestion, methanogenesis, end products from acetogenesis are converted into mainly methane and other ends products such as carbon dioxide and trace gases (Chen and Neibling, 2014).

Rate limiting step for anaerobic digestion is hydrolysis (liquefaction or solubilisation) stage. In this sense, pre-treatment methods in terms of physical, chemical and biological means are used to enhance hydrolysis performance (Sapkaite et al., 2017). Most widely used pretreatment methods are ultrasonic pretreatment, thermal pretreatment (THP), alkali pretreatment and ozone pretreatment (Mills, 2015).

In the last years, THP system has become widely used mainly in the United States. When THP is used before anaerobic digestion, temperature, pressure and reactive properties of water lead to shorter sludge decomposition time, making a significant positive impact on biogas generation. Furthermore, another significant consequence is that applying THP before anaerobic digestion results in lower sludge amount after digestion. Compared to single anaerobic digesters, THP-anaerobic digestion system may produce approximately 34% more energy for 1 ton of sludge. By using THP-anaerobic digestion system, 1020 kWh of energy might be produced out of 1 ton of sludge (Mills, 2015). Thus, thermal pretreatment is a promising technique which results in lower solid retention times in digesters by boosting methane generation. Also, thermal pretreatment may result in making sludge more appropriate for land application (Xue et al., 2015).

It is recommended that anaerobic digesters after thermal pretreatment process have the best results with a retention time of 10 to 12 days since 95% of biogas potential possibly utilized after 20 days. Longer retention times result in protein decomposition which in turn enhance ammonia, alkalinity and pH increase and do not cause noticeable growth in biogas generation (Barber, 2016).

In order to increase the efficiency of AD, there are some processes which can be applied. First of all, thickening sludge before AD is beneficial since it leads to a reduction in heating requirement. Secondly, screening of the primary solids may be applied prior to AD so as to prevent digester volume loss and clogging problems. Various technologies are offered for screening such as rotary drum screens and strain presses. Lastly, if toxic materials are present in the sludge, AD efficiency and methane generation may be affected negatively which may result from upset of digester, unfavorable pH conditions and high volatile acid concentrations. Toxic materials may have acute impacts such as instant failure of the process and chronic impacts such as low performance. In order to prevent the negative impacts of toxic materials, chemicals may be applied to control dissolved concentrations of toxicants. An example may be to use iron salts to control sulfide (WEF, 2009).

Thermal hydrolysis is mostly affected by temperature whereas reaction time has lower impact. Using steam explosion pretreatment may increase the efficiency of thermal hydrolysis in turn AD. For instance, methane generation was improved 50% when steam explosion pretreatment is applied at 140-170◦C and for 5-35 minutes (Sapkaite, Barrado, Fdz-Polanco & Pérez-Elvira, 2017). Moreover, in order to increase biogas production, steam accumulators may be used. In steam accumulators, steam is stored at a higher pressure than the required in thermal pretreatment. In this way, energy contained in exhaust gases can be used. For instance, using steam accumulator in

Burgos WWTP leads to 20% improvement in biogas production (García-Cascallana, Borge-Díez & Gómez, 2018).

2.2.1. Methods That Improves the Anaerobic Digestion

Stabilization of sludge is carried out in order to reduce pathogens, to exterminate odor problems, and to eliminate putrefication problems (WEF, 2010). The efficiency of stabilization methods depends on the stabilization technique used and operations applied on organic fraction of sludge. Stabilization methods are also beneficial in terms of sludge volume reduction, valuable biogas generation and improvement in dewaterability of sludge (Tchobanoglous et. al., 2014). Most widely applied stabilization methods are aerobic digestion, alkaline stabilization, anaerobic digestion, and composting (Tchobanoglous et. al., 2003).

Aerobic digestion includes biological degradation of organic substances in sludge, in the presence of oxygen or air. This process is usually carried out in an open top tank (Tchobanoglous et. al., 2003).

Alkaline stabilization consists of adding alkaline materials, such as lime or another alkaline additive, to increase the pH to a certain level which inhibits the pathogen growth. Since lime and alkaline additive are relatively inexpensive, alkaline stabilization is among the cheapest choices of sludge stabilization processes (EPA, 2000).

In anaerobic digestion, organic materials in sludge are degraded in an environment where there is no oxygen. Anaerobic digestion is carried out in closed tank which is called a digester. Biogas, which can be used for energy generation, is generated throughout anaerobic digestion process. Biogas mainly comprises of methane, carbon dioxide, and small amounts of water vapor and trace gases (EPA, 2018).

Composting includes also a biological degradation of sludge to have a stable end product. If composting is carried out properly, end product will be odor free and a humus-like substance. In composting, approximately 20-30% of organics in sludge possibly decomposed into carbon dioxide and water. Though composting can be conducted both aerobic and anaerobic environments, aerobic composting is more common among wastewater treatment plants. However, it should be noted that even if composting is done under aerobic conditions, it is never completely aerobic like aerobic digestion (Tchobanoglous et. al., 2003).

2.2.2. Sludge Minimization Processes

Sludge disposal constitutes a major part of a wastewater treatment plant operation cost. Therefore, sludge minimization has notable significance in sludge treatment processes (Morosini et al., 2016). In Figure 2-2, sludge treatment processes are shown. Sludge volume decreasing processes are pretreatment methods, anaerobic and aerobic digestion (Tchobanoglous, Stensel, Tsuchihashi, & Burton, 2014).

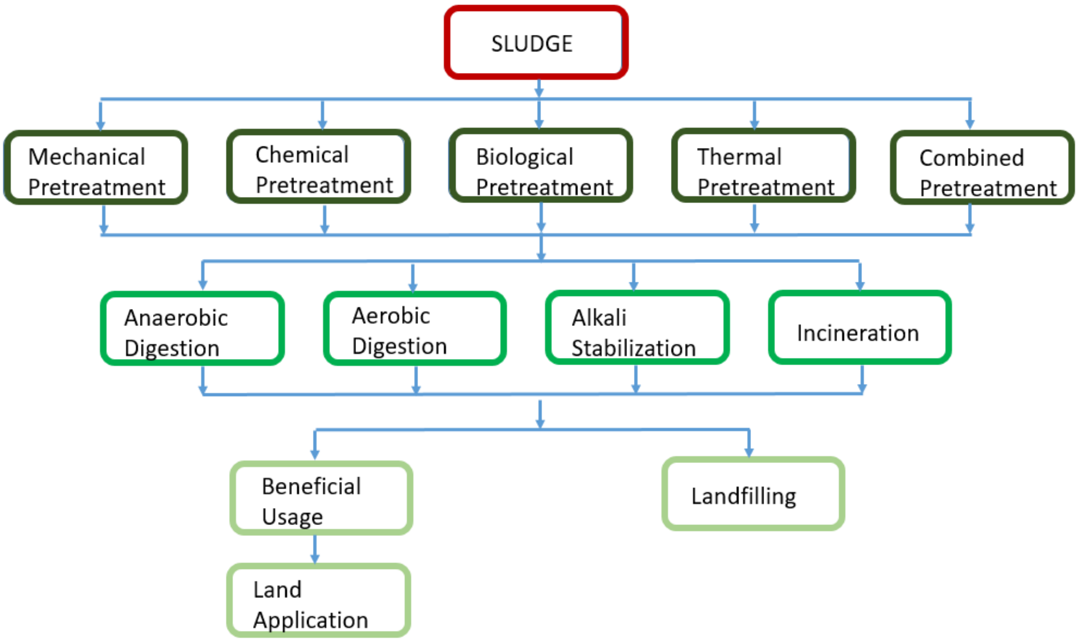


Figure 2-2. Sludge minimization and disposal processes (Tchobanoglous, Stensel, Tsuchihashi, & Burton, 2014)

Pretreatment methods are crucial for effective minimization of sludge. For instance, these methods are widely used for enhancing digestibility and solubilisation of sludge (Vlyssides and Karlis, 2004; Gavala et al., 2003).

2.2.2.1. Pretreatment Methods

In sludge management, pretreatment methods have gained importance after anaerobic digestion process was established (Abelleira-Pereira et al., 2015). Even without pretreatment step, energy can be generated from anaerobic digestion process by using methane production. However, hydrolysis acts like the rate-limiting step (Doğan & Sanin, 2009). On the other hand, hydrolysis steps are accelerated by pretreatment methods (Ariunbaatar et al., 2014; Doğan & Sanin, 2009). Additionally, pretreatment methods sterilize the sludge which is useful for eliminating sterilization cost (Ariunbaatar et al., 2014).

Mechanical Pretreatment

Aim of mechanical pretreatment is disintegrating solid particles and cells in sludge by physical means; thus, rendering sludge more degradable. After mechanical pretreatment, cells disintegrate, and substances contained in cells set free, which leads to an increase in surface area (Ministry of Environment and Urbanization, 2013, Ariunbaatar et al., 2014, Kopplow & Barjenbruch, 2003). As surface area increases, contact between anaerobic bacteria and substrate enhances which has a positive impact on anaerobic digestion (Ariunbaatar et al., 2014). There are various types of mechanical pretreatment such as high-pressure homogenizer and ultrasound.

Homogenizer

Homogenizers comprise of a homogenizing valve and multistep high-pressure pump. The suspension is constricted by the pump (Perez-Elvire et al., 2006). After pressure on the sludge is beyond 900 bar, sludge goes across a homogenization valve beneath strong depressurization (Carrèrea et al., 2010, Perez-Elvire et al., 2006).

Ultrasound

In ultrasound systems, the aim is to disintegrate sludge by using sound energy between 20 kHz and 10 MHz (Ministry of Environment and Urbanization, 2013). Through ultrasound, local high temperatures possibly observed due to cavitations. Moreover, high-speed reactions, disintegration, and mass and heat transfer are kept in high-temperature spots. (Fang, et al., 2018). Ultrasound treatment leads to mechanical interruption on the structure of cells and floc matrix (Carrèrea et al., 2010). Sludge volume is decreased, and biogas production is enhanced when ultrasound treatment is applied before anaerobic digestion (Apul & Sanin, 2010).

Chemical Pretreatment

Chemical pretreatment is basically transforming complex organic compounds to a more degradable form by using chemicals such as ozone, chlorine, strong minerals and acids, alkalis or oxidants (Tanaka et al., 1997, Ministry of Environment and Urbanization, 2013, Perez-Elvire et al., 2006, Ariunbaatar et al., 2014). Ozone is the most widely used treatment method since no chemicals are added (Perez-Elvire et al., 2006). Used method and properties of the substrate affect the efficiency of the chemical pretreatment. For instance, chemical pretreatment is not feasible for readily biodegradable substances as these substances degrade rapidly causing VFA accumulation which adversely affect methanogenesis step. On the other hand, chemical pretreatment possibly used effectively for substrates including excessive lignin amount (Ariunbaatar et al., 2014).

Alkali pretreatment

Alkali pretreatment is a straightforward and effective pretreatment type which is applied for sludge disintegration. In alkali pretreatment, pH of sludge is increased up to 12 leading to decomposition of cells that makes cells easier to disintegrate (Perez-Elvire et al., 2006, Ministry of Environment and Urbanization, 2013). Also, interactions between microorganism and flora structure are altered by alkali pretreatment (Huang et al., 2018). Biogas production rate and volume are enhanced when alkali pretreatment is applied before anaerobic digestion (Doğan & Sanin, 2009).

Ozonation

The most widely applied chemical pretreatment method is ozonation which induces solubilisation of sludge and enhancement of yield (Yeom et al., 2002; Carrèrea et al., 2010). Ozonation is used as a pretreatment method before anaerobic digestion or a posttreatment method where sludge is recycled back to the anaerobic digester (Weemaes et al., 2000, Yeom et al., 2002, Bougrier et al., 2007, Carrèrea et al., 2010).

Ozone is an oxidant having high reactivity which enhances decomposition of substances and exterminates odor problem before stabilization (Ministry of Environment and Urbanization, 2013). However, full-scale application of ozonation on sludge is limited due to its high cost (Chu et al., 2009).

Biological Pretreatment

Biological pretreatment provides disintegration of cell walls, rendering the sludge more biodegradable. (Ministry of Environment and Urbanization, 2013). This method includes aerobic and anaerobic means as well as enzymatic reactions by adding particular enzymes to the anaerobic digestion system (Ariunbaatar et al., 2014, Carrèrea et al., 2010). These enzymes could be either generated in the system (internally) or externally (Perez-Elvire et al., 2006).

Thermal Pretreatment

Thermal pretreatment is the most widely applied and studied pretreatment method (Ariunbaatar et al., 2014). Aim of thermal pretreatment is increasing biodegradable portion of sludge by using different temperatures under appropriate pressures (Gavala et al., 2003). Moreover, removal of pathogenic microorganisms, enhancement of dewatering potential and reduction in the digestive viscosity possibly achieved by using thermal pretreatment (Ariunbaatar et al., 2014; Higgins et al., 2017).

Thermal hydrolysis

Applying heat and pressure for a defined time is called thermal hydrolysis. Thermal hydrolysis is preferred especially before anaerobic digestion (Barber, 2016). Treatment duration is less important than treatment temperature in thermal treatment of sludge. Organic substances in sludge solids mostly dissolve after applying the thermal treatment at low temperature, which leads to a significant increase in the liquidity of sludge (Liao et al., 2016). Using high temperatures and corresponding high pressures to sludge leads to the disintegration of cells which in turn reduce the time needed for the hydrolysis step of anaerobic digestion (Elliot and Mahmood, 2007). In this sense, the main variables and processes of thermal hydrolysis are heating, which includes corresponding temperature and time, and pressure reduction (Sapkaite et al., 2017).

Thermal hydrolysis is feasible for substrates rich in protein and carbohydrates whereas it has significantly less impact on substrates rich in lipids (Barber, 2016). Thermal hydrolysis process affect both physical and chemical properties, which accelerates hydrolysis step, and improves dewatering of sludge, viscosity reduction, organic compound solubilisation, anaerobic biodegradability and production of biogas (Higgins et al., 2017; Liu et al., 2012; Liao et al., 2016; Oosterhuis et al., 2014). Due to the expedited hydrolysis step and reduced viscosity of the sludge, biogas production possibly enhanced by 10% and digestion time might be significantly reduced after applying thermal hydrolysis (Liao et al., 2016). With improved dewaterability of sludge, transport and processing costs of sludge are decreased. Additionally, with improved anaerobic biodegradability, thermal hydrolysis increases energy production (Barber, 2016; Liao et al., 2016). Although there are other positive effects such as increased dewaterability of sludge, the primary purpose of the thermal hydrolysis is enhancing energy production and having a better quality sludge after anaerobic digestion (Oosterhuis et al., 2014; Sapkaite et al., 2017). Thermal hydrolysis considerably scales up methane production compared to no thermal pretreatment (Liao et al., 2016; Abelleira-Pereira et al., 2015; Liu et al., 2012; Xue et al., 2015; Ariunbaatar et al., 2014).

Temperature is a vital operating parameter for thermal hydrolysis (Higgins et al., 2017). Recent studies on the sludge from food industry suggest that carrying out thermal pretreatment at moderate temperatures, around 50-90°C, enhances solubilisation of sludge but require approximately 10% more process times (Vlyssides and Karlis, 2004, Elliott and Mahmood, 2007). In other respects, thermal pretreatment is conducted at much higher temperatures (150-200°C) in most of the previous studies (Elliott and Mahmood, 2007). When higher temperatures than 200°C are used in thermal pretreatment, the Maillard reactions start to take place which includes the production of melanoidins and reduction of amino acids and sugars. Melanoidins are heterogenous polymers which have high molecular weights and therefore, very problematic to be degraded in digestion processes. Furthermore, melanoidins may even have adverse effects on the degradation of other organic substances (Xue et al., 2015).

In one of the studies, soluble COD is increased by 25%, 44%, and 60% by conducting thermal hydrolysis at 130°C, 150°C, and 170°C, respectively (Elliott and Mahmood, 2007). Moreover, around 62% more reduction in volatile solids is observed when thermal hydrolysis is applied to WAS (Oosterhuis et al., 2014). The optimal conditions for thermal hydrolysis vary by the type of sludge (Gavala et al., 2003, Liao et al., 2016, Elliott and Mahmood, 2007, Barber, 2016).

By using thermal hydrolysis, around 11% more biogas possibly produced and anaerobic digestion times might decrease from 22 to even 15 days (Liao et al., 2016). In this sense, thermal pretreatment is the most widely used process in industrial applications since it offers a profitable alternative method (Sapkaite et al., 2017).

As temperature of thermal hydrolysis increases, biogas generation also increases to a certain extent. Though numerous different results are reported, biogas generation starts to decrease after a particular temperature threshold. The reason behind the differences in these results is not evident. However, these differences may result from a characteristic of sludge or testing environment (Higgins et al., 2017).

There are several advantages of using thermal hydrolysis as a pretreatment method; considerably enhances the degradability of both primary sludge and activated sludge (Haug et al., 1978, Stuckey and McCarty 1978, Liao et al., 2016, Xue et al., 2015, Barber, 2016, Wilson and Novak, 2009),significantly increases biogas production rate is observed (Barber, 2016, Perez-Elvire et al., 2006),THP enhances the dewaterability of sludge (Oosterhuis et al.,2014; Barber, 2010; Haug et al., 1978; Barber, 2016; Perez-Elvire et al., 2006),decreases the viscosity of sludge (Liu et al., 2012; Oosterhuis et al., 2014; Bougrier et al., 2006; Barber, 2016),loading rate of anaerobic digestion significantly increases (Xue et al., 2015; Barber, 2016),has sterilization impact on sludge by deactivating pathogenic microorganisms (Barber, 2016, Perez-Elvire et al., 2006; Oosterhuis et al., 2014, Liu et al., 2012),eliminates odor problems,reduces scum and foaming formation and decreases the need for downstream thermal processes such as drying (Barber, 2016).

There are some limitations for the application of thermal hydrolysis, it requires heat energy and use of boilers (Haug et al., 1978; Barber, 2016), whole process becomes more complex processes than standard anaerobic digestion (Barber, 2016),more ammonia production and concentration than standard anaerobic digestion is observed (Oosterhuis et al., 2014; Wilson and Novak, 2009),production of refractory and slowly degradable materials is likely with food waste (Liu et al., 2012, Barber, 2016), the process may require more polymers for dewatering (Oosterhuis et al., 2014; Barber, 2016),odors are generally produced along with thermal hydrolysis (Haug et al., 1978; Perez-Elvire et al., 2006), fouling and corrosion problems in heat exchanger tubes have been observed (Haug et al., 1978; Perez-Elvire et al., 2006) and finally,methanogenic archaea possibly damaged by the use of thermal pretreatment techniques (Liu et al., 2012).

Influence of Thermal Hydrolysis

Thermal hydrolysis can be applied at different steps of sludge processing and influences the downstream sludge processing in several ways. Summarized figure of thermal hydrolysis effects adopted from Barber (2016) is given in Figure 2-3. These effects and their consequent impacts are shown. One of the most important effect of thermal hydrolysis is increase in solubility of organic contents. This improves the biogas production with respect to volatile solids. Reduction of particle size during thermal hydrolysis contributes to transport of water and chemicals in sludge.

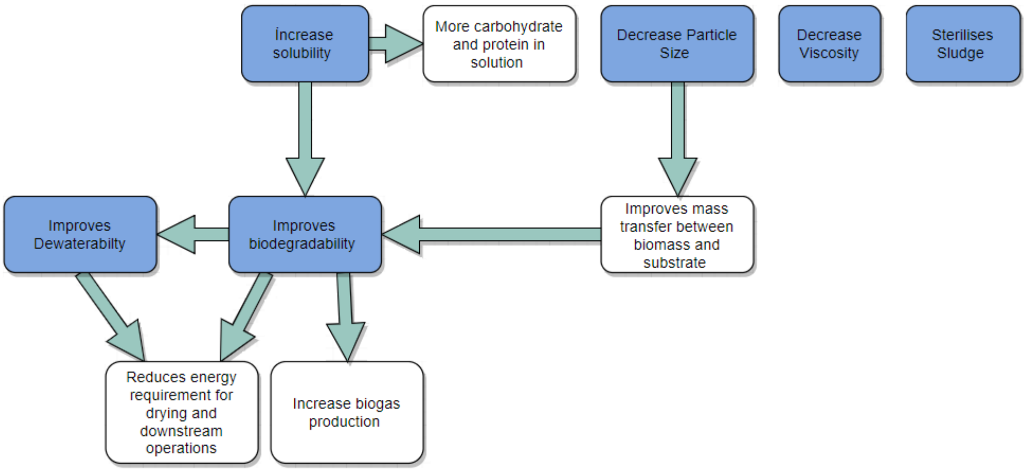


Figure 2-3. Some effects of thermal hydrolysis modified from Barber, 2016

Influence of Thermal Hydrolysis on COD Solubility

Studies involving thermal hydrolysis suggest that using thermal hydrolysis results in more soluble COD in the sludge (Farno et al., 2014, Barber, 2016, Everett, 1972, Bougrier et al., 2007, Liu et al., 2012).

Influence of Thermal Hydrolysis on Viscosity

Viscosity is reduced as the temperature is increased by thermal hydrolysis. First of all, the viscosity of free water within sludge is reduced reversibly concerning Arrhenius' law (Eshtiaghi et al., 2013). Furthermore, substances in sludge might be thermally exterminated such as protein denaturation (Farno et al., 2015). Lastly, interactions between substances are weakened since thermal treatment alters rheology. All in all, thermal pretreatment reduces viscosity more than ultrasonic or ozone pretreatment (Barber, 2016).

Influence of Thermal Hydrolysis on Anaerobic Biodegradability

WAS digestion are enhanced and biogas generation considerably up to 75-80% with respect to non-pretreated ones (Phothilangka et al., 2008; Barber, 2016). According to Haug et al. (1978), energy generation increases by 25% when thermal hydrolysis is applied before anaerobic digestion. Using thermal hydrolysis increases biogas production along anaerobic digestion after ten days whereas there is negligible difference in biogas production throughout anaerobic digestion in the first 24 hours (Barber, 2016).

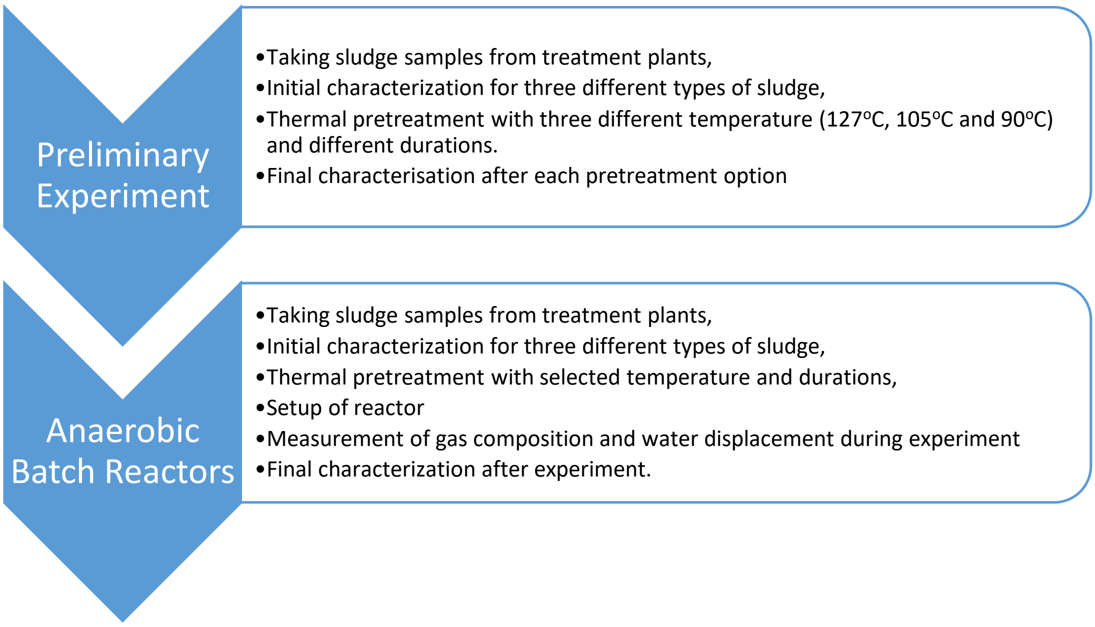
Influence of Thermal Hydrolysis on Dewaterability

Mesophilically digested sludge dewaterability is increased up to 10% by applying thermal hydrolysis before anaerobic digestion. Generally, thermal hydrolysis has a positive impact on sludge dewaterability (Barber, 2016, Everett, 1972).

CHAPTER 3

MATERIALS AND METHOD

A range of experiments was designed and carried out to understand the effect of thermal pretreatment on biogas production from different sludges by applying anaerobic digestion. First of all, the series of thermal pretreatment experiments were conducted under different conditions to determine the best solubilization of sludge and hence the best possible conditions for biogas production. Then, BMP experiments were conducted to analyze the impact of discrete pretreatment levels on anaerobic digestion efficiency. Major steps of the three trials are summarized in Figure 3-1. Experimental procedures are elaborated along this section.



- Taking sludge samples from treatment plants,
- Initial characterization for three different types of sludge,
- Thermal pretreatment with three different temperature (127°C, 105°C and 90°C) and different durations.
- Final characterisation after each pretreatment option

- Taking sludge samples from treatment plants,
- Initial characterization for three different types of sludge,
- Thermal pretreatment with selected temperature and durations,
- Setup of reactor
- Measurement of gas composition and water displacement during experiment
- Final characterization after experiment.

Figure 3-1. Main steps followed in this study.

3.1. Sludge Samples

In this study three types of biological sludges were used. First sludge is that a WAS sample from a domestic wastewater treatment plant (Tatlar WWTP, Ankara) which was investigated as a standard sludge on which thermal hydrolysis is most commonly applied; The second sludge was an MBR sludge. MBR sludge has longer solid retention time with respect to typical domestic WWTP which means that typical domestic WWTP has less initial stability. Therefore, MBR sludge possibly has lower methane generation. Last selected sludge samples obtained from an OID. OID has vast amount of sludge after treatment process. However, OID sludge possibly contain toxic compounds due to containing various sectoral wastes in it. Therefore, management of OID sludge has crucial for OID. The sludge samples and the treatment plants that they were collected from are explained below.

3.1.1. Ankara Central Wastewater Treatment Plant

Having a capacity of 765,000 m^3/day, Tatlar Wastewater Treatment Plant (Tatlar), which utilize Conventional Activated Sludge Process, is the largest wastewater treatment plant in Turkey. In this plant, 250 million m^3 of wastewater is being treated annually. Treated wastewater is discharged to Ankara Creek while ultimate discharge location is Sakarya River. The initial wastewater characteristics of Tatlar is given in Table 3-1.

Table 3-1. *Initial characterization of the wastewater from Tatlar Wastewater Treatment Plant*

Parameter	**Unit**	**Value**
pH	-	8.06
COD	mg/L	356
BOD	mg/L	184
COD/BOD	-	1.9
Temperature	◦C	12.1

COD value for low, medium and high strength domestic wastewaters are 250, 430 and 800 mg/L, respectively. Additionally, BOD value for low, medium and high strength domestic wastewater are 110, 190 and 350 mg/L, respectively (Tchobanoglous et.al., 2014). Therefore, Tatlar wastewater is typical low strength wastewater. COD/BOD ratio represent the biodegradability of the wastewater and selection of treatment process to be done with respect to COD/BOD ratio. For Tatlar wastewater COD/BOD ratio is 1.9 which is between 1.7 and 2.4. Therefore, the biodegradable fraction of Tatlar wastewater is high (Sperling, 2007). With respect to that ratio, biological treatment and anaerobic digestion system is applicable to Tatlar wastewater. Additionally, pH value is suitable for anaerobic digestion operation.

Throughout Ankara, collected wastewater is transported to the plant only by gravity. Tatlar Wastewater Treatment Plant includes pretreatment units, primary clarifiers, aerations tanks, secondary clarifiers, return activated sludge (RAS) pumping station, raw sludge thickening units, anaerobic digesters, biogas station, biogas storage units, digested sludge thickening units and sludge dewatering units (ASKİ Genel Müdürlüğü, 2019). First unit in the sludge treatment line is thickener, after which, excess sludge is sent to anaerobic digestion units. In this way, biogas, a valuable source, is produced. Moreover, electricity is generated by using biogas, which is used to meet the partial electricity demand of the plant.

Figure 3-2. Air View of Tatlar Wastewater Treatment Plant (ASKİ Genel Müdürlüğü, 2019)

Characteristics of Tatlar Wastewater Treatment Plant’s sludge are shown in Table 3-2.

Table 3-2. *Initial characterization of the sludge from Tatlar Wastewater Treatment Plant*

Parameter	Unit	Value
TCOD	mg/L	38,538
Soluble COD	mg/L	3,457
COD solubilization (total COD being soluble COD) (Soluble COD/TCOD)	%	8.97
TS	mg/L	27,633
TSS	mg/L	26,583
VS	mg/L	21,183
VSS	mg/L	19,333
pH	-	7.49

3.1.2. Middle East Technical University Treatment Plant (MBR Sludge)

Domestic and industrial wastewater are treated in MBR system for wastewater recovery. In MBR system required area for treating wastewater is reduced (Baysal, 2017). MBR are an improved form of conventional activated sludge systems. It is a combination of biological reactors and membrane technology. After biological treatment, separation is carried out using ultrafiltration (UF) or microfiltration (MF) membranes instead of the clarifier (Aslan, 2015). In the Middle East Technical University. METU Treatment Plant domestic wastewater which are coming from Middle East Technical University campus is pretreated. The wastewater treatment plant comprises a vacuum rotating membrane bioreactor in two tanks. Biological treatment occurs in the first tank which aeration is implemented. Additionally, filtration occur in the second tank. The initial wastewater characteristics of METU MBR treatment Plant is given in Table 3-6.

Table 3-3. *Initial characterization of the wastewater from MBR*

Parameter	**Unit**	**Value**
pH	-	7.1-7.5
COD	mg/L	426
BOD	mg/L	250
COD/BOD	-	1.7
Temperature	◦C	12-23

As discussed for Tatlar WWTP BOD, COD, COD/BOD ratio are evaluated for WWTP from MBR wastewater the COD/BOD ratio is 1.7 which is between 1.7 and 2.4. Therefore, the biodegradable fraction of MBR wastewater is high (Sperling, 2007). Therefore, anaerobic digestion system is an alternative for MBR sludge. Additionally, pH value is suitable for anaerobic digestion operation.

Having a 540 m^2 membrane surface area, treated wastewater is utilized for irrigating grass in the Middle East Technical University. METU Treatment Plant has a 200 m^3/day capacity. The plant treats 8.5 m^3 wastewater per hour which corresponds to the demand of approximately 2000-2500 people (Su ve Çevre, 2007). While using similar approaches in biological wastewater treatment, sludge at the outlet is filtered by membranes, rather than having it settled. While sludge is treated in 3 weeks by 40-50% in conventional treatments plants, membrane technology offers 80-90% sludge treatment in 4 days. In this way, treated wastewater from METU Treatment Plant can be used for irrigational purposes of parks and agricultural purposes except for drinking (ODTÜ, 2013).

Figure 3-3. Membrane Bioreactor from Middle East Technical University (ODTÜ, 2013)

Characteristics of Middle East Technical University (MBR) sludge are shown in Table 3-4.

Table 3-4. *Initial characterization of MBR sludge from Middle East Technical University*

Parameter	Unit	Value
TCOD	mg/L	21,754
Soluble COD	mg/L	663
COD solubilization (total COD being soluble COD) (Soluble COD/TCOD)	%	3.04
TS	mg/L	27,866
TSS	mg/L	22,516
VS	mg/L	20,883
VSS	mg/L	18,233
pH	-	7.17

3.1.3. Organized Industrial District Treatment Plant

In OID there are eleven different industries which are machine manufacturing, forest products, furniture and paper, metal goods, electrical equipment manufacturing, food, beverage, woven and ready-made clothing, metal, chemistry, mining and stone and soil based manufacturing. Among these industries five of them are more dominant than others, which are metal, machine manufacturing, food, beverage, and chemistry. OID wastewater treatment plant has a capacity of 36,000 m^3/day. The wastewater treatment plant in OID comprises of physical, chemical and biological treatment with nitrogen removal. Additionally, for sludge treatment, there are sludge thickener, dewatering, and solar drying unit. Sludge from OID is regarded as hazardous sludge (Eskişehir Organize Sanayi Bölgesi, 2019). (Figure 3-4). The initial wastewater characteristics is given in Table 3-8.

Table 3-5. *Initial characterization of the wastewater from OID*

Parameter	**Unit**	**Value**
pH	-	6.58
COD	mg/L	1018,2
Temperature	◦C	20-25

As discussed for Tatlar and MBR WWTP BOD,COD, COD/BOD ratio are evaluated for WWTP Therefore, OID wastewater is high strength wastewater. COD/BOD ratio represent the biodegradability of the wastewater and selection of treatment process to be done with respect to COD/BOD ratio. For OID wastewater, measurement of BOD is not done. However, expected COD/BOD ratio is greater than 3.5 Therefore, the non-biodegradable fraction of OID wastewater is high (Sperling, 2007). With respect to that ratio, physical and chemical treatment system is applicable to is applied to OID wastewater. Additionally, pH value is suitable for anaerobic digestion operation.

Figure 3-4. Air View of Organized Industrial District Treatment Plant (Eskişehir Organize Sanayi Bölgesi, 2019)

There are three preliminary experiments established with sludge, which was taken from OID Treatment Plant Characteristics of the sludge from OID Treatment Plant are shown in Table 3-6.

Table 3-6. *Initial characterization of the sludge from OID Treatment Plant*

Parameter	Unit	Value
TCOD	mg/L	44,565
Soluble COD	mg/L	442
COD solubilization (total COD being soluble COD) (Soluble COD/TCOD)	%	0.99
TS	mg/L	37,083
TSS	mg/L	36,033
VS	mg/L	30,117
VSS	mg/L	31,433
pH	-	6.45

3.2. Thermal Pretreatment

To increase anaerobic digestibility and biogas generation from sludge, thermal pretreatment was applied by using an autoclave. After having it settled, concentrated sludge from three plants mentioned above was distributed into beakers as 100 mL portions. These beakers were put into an autoclave machine. Used autoclave can be seen from Figure 3-5.

Figure 3-5. Autoclave machine used in the experiments

Autoclave used was a HICLAVE HV-85L. There are four separate programs in this autoclave, which is presented in Table 3-7 in detail.

Table 3-7. *Different programs of the HICLAVE HV-85L*

Program	Mode	Temperature Interval (ºC)	Duration Interval (min)	Pressure (bar)
1	Liquid	105-127	1-250	0.3-1.7
2	Liquid	105-127	1-250	0.3-1.7
3	Solid	105-127	1-250	0.3-1.7
4	Agar	60-100	1-60	-

Program 1 and Program 4 are used for the preliminary experiments. Throughout preliminary experiments, different durations of autoclaving and different autoclaving temperatures were tested. The solubilization was monitored by measuring soluble COD. According to outcomes of these preliminary experiments, a unique thermal hydrolysis temperature and duration were determined for anaerobic batch reactors. Impacts of the thermal hydrolysis are analyzed by measuring soluble COD concentration before and after the application of thermal pretreatment.

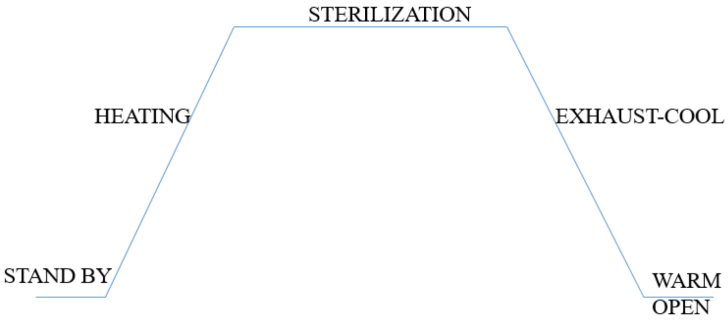


Figure 3-6. The working steps of the autoclave (Operating Manual,2003)

Stages of autoclaving are summarized in Figure 3-6. The first step of thermal hydrolysis is heating. In this step, the temperature in autoclave rises to the intended thermal hydrolysis temperature. Thermal hydrolysis is conducted with the intended temperature and duration in the second step, which is sterilization. Lastly, throughout exhaust-cool step, the temperature in autoclave decreases until it can be opened.

Impacts of thermal hydrolysis and related enhancement of soluble COD concentrations are discussed in Chapter 4. In this study, thermal hydrolysis was only applied to the WAS since excess temperature would damage the microorganisms in seed sludge, which is crucial for anaerobic digestion.

3.3. Pretreatment Studies

In this part, to investigate optimum conditions, thermal hydrolysis was analyzed with different durations and temperatures for each sludge. To determine optimum conditions, soluble COD, pH, TS, TSS, VS and VSS are investigated. First triple set of pretreatment was examined with Tatlar Wastewater Treatment Plant sludge. Second triple set of pretreatment was examined with Middle East Technical University Wastewater Treatment Plant sludge. Final triple set of pretreatment was examined with OID Treatment Plant sludge. Before both pretreatment experiments and anaerobic digestion, to normalize the sludge sample, Tatlar and MBR samples were centrifuged.

3.3.1. Pretreatment Experiment with Tatlar Sludge

WAS was collected from the Tatlar Wastewater Treatment Plant. TSS and VSS were determined as 26,583 and 21,183 mg/L respectively, whereas the total COD was 38,538 mg/L.

To determine the optimum pretreatment conditions, in the first set, highest temperature (127^{o}C) and pressure (1.7 bar) were analyzed to investigate the effect of thermal hydrolysis with controlled temperature at six different durations (15, 30, 45, 60, 75 and 90 minutes). After thermal hydrolysis was completed, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was centrifuged and separated from its supernatant at 4000 rpm and for 15 minutes.

In the second set, medium temperature (still above boiling temperature), of 105^{o}C and medium pressure (0.3 bar) were analyzed to investigate the effect of thermal hydrolysis with moderate temperature and seven different durations which are 15, 30, 45, 60, 75, 90 and 105 minutes. After thermal hydrolysis was conducted, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was centrifuged at 4000 rpm and for 15 minutes to separate it from its supernatant.

In the third set, low temperature (90^{o}C) was analyzed to investigate the effect of thermal hydrolysis with low temperature and four different durations which are 15, 30, 45 and 60 minutes. After thermal hydrolysis was done, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was separated from its supernatant by centrifuging at 4000 rpm for 15 minutes.

3.3.2. Pretreatment Experiment with MBR Sludge

WAS was collected from the Middle East Technical University MBR System. TSS and VSS were determined as 22,516 and 18,233 mg/L respectively, whereas the total COD was 21,754 mg/L.

To determine the optimum pretreatment conditions, in the first set, highest temperature (127^{o}C) and pressure (1.7 bar) were analyzed to investigate the effect of thermal hydrolysis with controlled temperature and six different durations which are 15, 30, 45, 60, 75 and 90 minutes. After thermal hydrolysis was completed, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was centrifuged and separated from its supernatant at 4000 rpm and for 15 minutes.

In the second set, medium temperature (still above boiling temperature), of 105°C and medium pressure (0.3 bar) were analyzed to investigate the effect of thermal hydrolysis with moderate temperature and seven different durations which are 15, 30, 45, 60, 75, 90 and 105 minutes. After thermal hydrolysis was conducted, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was centrifuged at 4000 rpm and for 15 minutes to separate it from its supernatant.

In the third set, low temperature (90°C) was analyzed to investigate the effect of thermal hydrolysis with low temperature and four different durations which are 15, 30, 45 and 60 minutes. After thermal hydrolysis was done, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was separated from its supernatant by centrifuging at 4000 rpm for 15 minutes.

3.3.3. Pretreatment Experiment with OID Sludge

WAS was collected from the OID Treatment Plant. TSS and VSS were determined as 36,033 and 31,433 mg/L respectively, whereas the total COD was 44,565 mg/L.

To determine the optimum pretreatment conditions, in the first set, highest temperature (127°C) and pressure (1.7 bar) were analyzed to investigate the effect of thermal hydrolysis with controlled temperature and six different durations which are 15, 30, 45, 60, 75 and 90 minutes. After thermal hydrolysis was completed, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was centrifuged and separated from its supernatant at 4000 rpm and for 15 minutes.

In the second set, medium temperature (still above boiling temperature), of 105°C and medium pressure (0.3 bar) were analyzed to investigate the effect of thermal hydrolysis with moderate temperature and seven different durations which are 15, 30, 45, 60, 75, 90 and 105 minutes. After thermal hydrolysis was conducted, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was centrifuged at 4000 rpm and for 15 minutes to separate it from its supernatant.

In the third set, low temperature (90°C) was analyzed to investigate the effect of thermal hydrolysis with low temperature and four different durations which are 15, 30, 45 and 60 minutes. After thermal hydrolysis was done, soluble COD, pH, CST, TS, TSS, VS, and VSS values were determined. To determine soluble COD, pretreated sludge was separated from its supernatant by centrifuging at 4000 rpm for 15 minutes.

3.4. Anaerobic Batch Reactor

At the beginning of this part, fresh sludge samples from Tatlar WWTP digestion unit as seed sludge and three different types of WAS from the three studied plants. All sludges were first settled to the desired concentration. In this part 10 different reactors were prepared which were seed, non-pretreated, pretreated at 60 minutes under 127°C and pretreated at 30 minutes under 90°C. Applying 127°C for 60 minutes was selected since this condition resulted in the highest soluble COD among all samples. Moreover, pretreatment at 90°C for 30 minutes was selected as this condition leads to highest soluble COD among the samples pretreated at 90°C for different durations. Each reactor was prepared with three replicates. Therefore, there were 30 reactors. For all reactors, TS, TSS, VS, VSS were investigated. To specify sludge volume samples for each reactor, F/M ratio (g VS WAS/g VS Seed) was selected as 2. In previous studies, this ratio was tested for ratios between 0.5 and 1 (Köksoy and Sanin, 2010) and used as 0.5 and 1 in different studies (Köksoy and Sanin, 2010; Çelebi, 2015). Since keeping the WAS from different plants at the highest amount possible was the purpose, seed from Tatlar WWTP was minimized by adjusting the F:1 as 2. As shown in Table 3-8, concentrations of solids were measured for each BMP assays.

Table 3-8. *Set-up concentration of solids for each BMP assays.*

Type of Sludge	TS (mg/L)	TSS (mg/L)	VS (mg/L)	VSS (mg/L)
Anaerobic Seed	10,300	8,722	6,761	5,455
Tatlar WAS – Non-pretreated	23,955	21,488	17,266	15,205
MBR WAS - Non-pretreated	23,322	20,644	17,388	14,427
OID WAS - Non-pretreated	24,222	20,800	16,800	14,927
Tatlar WAS – after 127^{o}C with 60 min. pretreatment	23,722	20,833	17,094	14,755
MBR WAS – after 127^{o}C with 60 min. pretreatment	23,976	21,122	17,550	15,261
OID WAS – after 127^{o}C with 60 min. pretreatment	23,611	21,100	16,705	15,555
Tatlar WAS – after 90^{o}C with 30 min. pretreatment	23,350	20,472	16,844	14,994
MBR WAS – after 90^{o}C with 30 min. pretreatment	23,122	20,844	16,661	14,794
OID WAS – after 90^{o}C with 30 min. pretreatment	23,783	20,433	15,611	14,838

Each BMP bottle had 275 mL volume. However, first of all, 250 mL of sample was prepared and filled to each bottle and then 50 mL of this volume is used for initial characterization. Thus, 200 mL of sample used as a working volume for anaerobic digestion process. The headspace for gas accumulation was 75 mL. For ten reactors, mixing ratios were presented in Table 3-9.

Table 3-9. *Volumetric additions of each sludge into BMP reactors*

Reactors	Label	Seed Vol. (mL)	Food Vol. (mL)	Distilled Water (mL)	Total Vol. (mL)
Anaerobic Seed	S	128	0.00	122	250
Tatlar WAS - No pretreatment	T	128	120.6	1.4	250
MBR WAS - No pretreatment	M	128	117.2	4.8	250
OID WAS - No pretreatment	E	128	115.0	7.0	250
Tatlar WAS – after 127°C with 60 Min. pretreatment	T127/60	128	118.4	3.6	250
MBR WAS – after 127°C with 60 Min. pretreatment	M127/60	128	120.1	1.9	250
OID WAS – after 127°C with 60 Min. pretreatment	E127/60	128	116.5	5.5	250
Tatlar WAS – after 90°C with 30 Min. pretreatment	T90/30	128	119.5	2.5	250
MBR WAS – after 90°C with 30 Min. pretreatment	M90/30	128	116.0	6.0	250
OID WAS – after 90°C with 30 Min. pretreatment	E90/30	128	120.8	1.2	250

After WAS and seed sludge samples were filled into the bottles, each of them was sparged with nitrogen gas with 99% purity for ten minutes. In this way, existing oxygen was removed from the bottles. Then, each bottle was sealed and put into the incubator at 35°C as shown in the Figure 3-7. In Table 3-10, measured parameters, their units and their measurement frequencies are shown. The BMP assays were terminated at day 29.

Figure 3-7. Incubator used for BMP assays

Table 3-10. *Investigated parameters for set-up BMP assays with their units and measurement frequencies*

Parameters	Units	Measurement Frequency
TS	mg/L	Day 0 and day 29
TSS	mg/L	Day 0 and day 29
VS	mg/L	Day 0 and day 29
VSS	mg/L	Day 0 and day 29
Soluble COD	mg/L	Day 0 and day 29
pH	-	Day 0 and day 29
CST	seconds	Day 0 and day 29
Biogas generation	mL	Every day (0-29)
Methane generation	mL	Days 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 17, 19, 21, 23, 25, 27, 29

3.5. Characterization Procedure

During pretreatment experiment, pH, TSS and VSS, total COD, soluble COD, TS and VS concentrations as well as CST of sludge were analyzed. These analyses were carried out for all types of sludge samples. Then for the BMP tests in addition to the above parameters, produced biogas volume and biogas composition were also measured. Methods are explained below.

3.5.1. pH

By utilizing Standard Method 4500H (APHA, AWWA, WEF, 2005), pH of the sludge samples was measured by CyberScan PC 510 pH-meter with an EC-PH510/21S probe (Eutech Instruments Pte Ltd., Spain).

3.5.2. Solids Analysis

TSS was determined by using Method 2540D (APHA, AWWA, WEF, 2005). Moreover, VSS was analyzed in conformity with Method 2540E (APHA, AWWA, WEF, 2005). TS was determined by following Method 2540B (APHA, AWWA, WEF, 2005). Furthermore, VS was measured by using Method 2540E (APHA, AWWA, WEF, 2005). To determine TS and VS, firstly, crucible dare (C g) was measured. Secondly, determined amount of sample (X mL) was put into the crucible and waited overnight (approximately 20 hours) in oven at 105◦C. Then, weight of crucible with sample (C_{105} g) was measured. Finally calculated TS value as shown in below.

$$TS = \frac{(C_{105} - C) * 1000 * 1000}{X}$$

To determine VS, crucible with sample (C_{105} g) waited approximately 1 hour in oven at 550◦C. Then, weight of crucible with sample (C_{550} g) was measured. Finally calculated VS value as shown in below.

$$VS = \frac{(C_{550} - C_{105}) * 1000 * 1000}{X}$$

To determine TSS and VSS, sample was filtered through glassfiber filter papers. Then, protocols and formulas used for TS and VS, are applied for TSS and VSS calculations.

To determine removal of TS and VS, initial and final TS and VS amount were measured which are TSi, TSf, VSi and VSf, respectively. Then calculation of TS removal as shown in below.

$$TS\% = \frac{\left(TS_i - TS_f\right) * 100}{TS_i}$$

Calculation of VS removal as shown in below.

$$VS\% = \frac{\left(VS_i - VS_f\right) * 100}{VS_i}$$

3.5.3. COD

Total COD, as well as soluble COD, were measured by using HACH LCK-514 kits (100-2000 mg/L) and HACH DR 3900 spectrophotometer (Hach Company, Colorado, USA). These measurements were done in conformity with the US Environmental Protection Agency approved dichromate method (Jirka and Carter, 1975). When measuring soluble COD, sludge was filtered by Millipore (Merck Millipore, Massachusetts, USA) filter papers having 0.45 μm-pores. After filtration, the filtrate was utilized for COD measurements. All analyses were conducted in triplicates.

3.5.4. Capillary Suction Time

CST test was conducted to analyze the dewaterability of sludge. The purpose of conducting this analysis, along preliminary studies, was to determine how the dewaterability of pretreated and non-pretreated sludge are different from each other. CST analyses were also carried out after the anaerobic digestion step, which provided more credible results as dewatering is conducted after anaerobic digestion in large-scale applications. CST experiment was performed by using Type 304 M Triton Electronics Capillary Suction Timer and following Method 2710G (APHA, AWWA, WEF, 2005). Samples from different sludge types were put in a cylindrical sample holder which was placed on a Whatman 17 chromatography paper sheet. The time needed for sludge to travel a particular distance was recorded. Lower travel time indicates shorter CST, which means better dewaterability of the sample. CST analyses were also conducted in triplicates.

3.5.5. Biogas Volume

The generated volume of biogas is determined by water displacement unit. As more and more biogas is generated, the pressure inside of the bottle rises since the sealed bottle has constant volume. The sealed bottle is connected to the water displacement unit by using a needle, and gas pressure is reduced to atmospheric pressure. By using the volume of water displaced in the water displacement unit, generated biogas volume was measured.

3.5.6. Biogas Composition

Biogas composition was analyzed by using Agilent Technologies 6890N Gas Chromatograph (Agilent Technologies, California, USA) with thermal conductivity detector (TCD). Before injections, calibration of GC was conducted with two standards which are %20 nitrogen with %25 methane and %10 nitrogen, %25 carbon dioxide and %65 methane. All data were calibrated these gas mixture. While operating GC, the room temperature to fixed 25◦C. Injections were conducted by a Hamilton Samplelock syringe (Hamilton Company, Nevada, USA), having a 250 μL volume. Measurements were conducted in duplicates.

To determine generated methane volume in each reactor, average methane percentage, which is determined by GC, was multiplied with the total volume of the gas generation of that specific day and daily methane production was calculated. After daily methane production was determined, methane production was added up, and cumulative methane production was calculated.

CHAPTER 4

RESULTS AND DISCUSSION

4.1. First Pretreatment: Pretreatment with WAS from Domestic WWTP

In the first pretreatment study, sludge from Tatlar Wastewater Treatment Plant was pretreated at different temperatures and durations. Each time a 100 mL of sludge was pretreated to see the effect. Three different sets were conducted under this group.

First set conducted with WAS from the domestic WWTP (Tatlar) was pretreated at 127°C and for six different durations which were 15, 30, 45, 60, 75 and 90 minutes. It was found that initial total COD of sludge was 38,540 mg/L and initial soluble COD of sludge was 3,457 mg/L with 8.97% of total COD being soluble COD. Initial data for this set can be seen in Table 3-2. Despite being pretreated for up to 90 minutes, maximum soluble COD of sludge was 9,118 mg/L with a total of 23.66% solubilisation (of TCOD) achieved at 60-minutes duration. This means that, longer duration did not lead to further solubilisation of COD. For the first set, average TS and VS were determined as 31,386 mg/L and 23,519 mg/L, respectively following pretreatment. Due to thermal pretreatment, TS and VS of pretreated samples increased slightly due to evaporation of water with respect to non-pretreated sample. Additionally, average pH for this experiment was 7.23.

Table 4-1. *Initial conditions of sludge for Tatlar Wastewater Treatment Plant and best values after pretreatment*

Parameter	Unit	Initial value	Maximum value after 127°C	Maximum value after 105°C	Maximum value after 90°C
Total COD	mg/L	38,540			
Soluble COD	mg/L	3,457	9,118	7,683	5,407
COD solubilization (solubilized total COD)	%	8.97	23.66	19.94	14.03
TS	mg/L	27,633	31,386	28,600	28,566
VS	mg/L	21,183	23,519	21,716	21,075
pH	-	7.49	7.23	7.5	7.36

Second set conducted with WAS from the domestic WWTP was pretreated at 105°C and for seven different durations which were 15, 30, 45, 60, 75, 90 and 105 minutes. Maximum soluble COD of sludge was 7,683 mg/L with 19.94% solubilisation at 75 minutes duration. For the second set, average TS and VS were determined as 28,600 mg/L and 21,716 mg/L, respectively. Moreover, average pH for this step was 7.5. The best results obtained for COD are solubilisation in Table 3-2.

Third set conducted with WAS from the domestic WWTP was pretreated at 90°C and for four different durations which were 15, 30, 45 and 60 minutes. Maximum soluble COD of sludge was 5,407 mg/L with 14.03% solubilisation at 30 minutes duration. According to Liao et. al. (2016), best condition for pretreatment at low heating temperature was also 30 minutes. For the third set, average TS and VS were determined as 28,566 mg/L and 21,075 mg/L respectively. Furthermore, average pH for this step was 7.36. All the results from the first set is summarized in Figure 4-1. The graph shows averages of triplicate analyses and the error bars indicate the standard deviations.

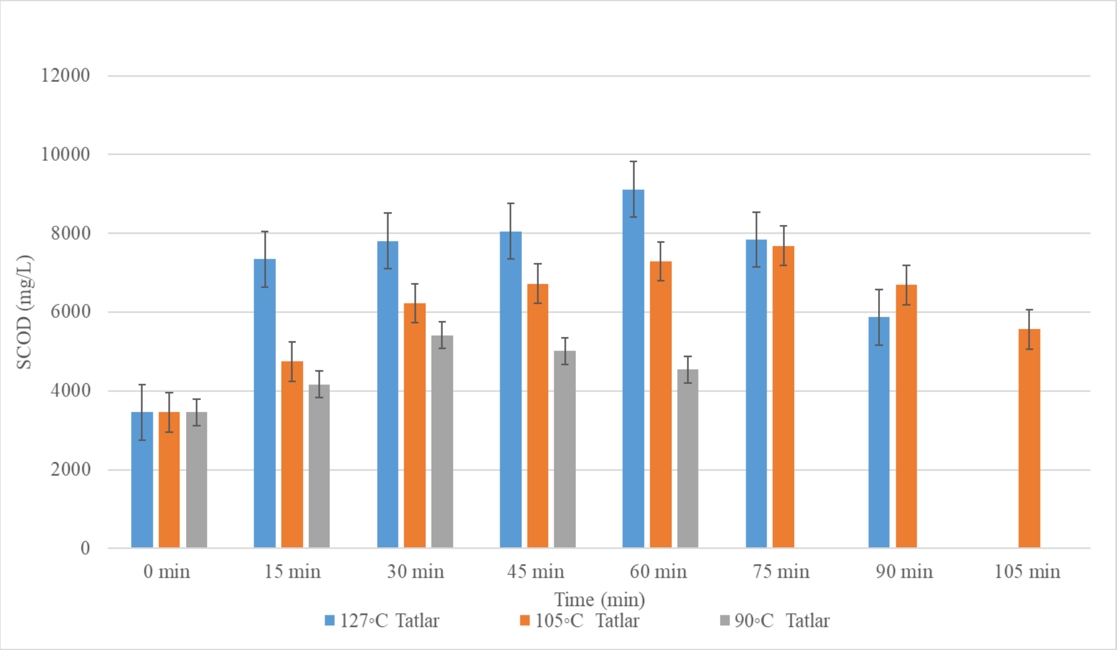


Figure 4-1. Soluble COD concentrations of Tatlar WAS sludge with respect to pretreatment temperature and time

As shown in Figure 4-1, samples which were pretreated at higher temperatures have higher soluble COD concentration. In the study of Xue et. al. (2015), soluble COD concentration at low pretreatment temperatures were compared to high pretreatment temperatures. It was found that higher pretreatment temperatures led to higher soluble COD concentrations. Additionally, in literature, with respect to time and temperature of pretreatment, change in soluble COD amount which increased from 7.8% to 29% was observed (Valo et al., 2004; Carr`ere et al., 2008; Bougrier et al., 2006). In the study of Carrere et. al. (2008), similar results were also observed.

At 127^{o}C, SCOD/TCOD percent increased from 8.97 to 19.06, 20.25, 20.89, 23.66, 20.34 and 15.23 for 15, 30, 45, 60, 75, and 90 minutes, respectively (Figure 4-2). This trend can be observed from Figure 4-2 (as averages with standard deviations). It is seen that at 90 min, the increase in soluble COD relative to the initial COD falls to a rather minimum. At 105^{o}C, SCOD/TCOD percent increased from 8.97 to 12.31, 16.16, 17.45, 18.92, 19.94, 17.36, and 14.44 for 15, 30, 45, 60, 75, 90, and 105 minutes, respectively (Figure 4-2). Additionally, at 90^{o}C, SCOD/TCOD percent increased from 8.97 to 10.81, 14.03, 13.00, and 11.77 for 15, 30, 45, and 60 minutes, respectively. To conclude, although longer durations at lower temperatures show an increase in soluble COD concentration, solubilisation percentage is relatively low. On the other hand, shorter durations at higher temperatures came out to be more beneficial to achieve high COD solubilisation. As discussed here and it is seen in graph (Figure 4-2) there is a maximum value. Decreasing of soluble COD concentration is observed in each graph following maximum soluble COD value. This could be explained by melanoidins formation (Liu et. al., 2006). When sugars and amino acids combine at high temperatures and low water activity, melanoidins which are high molecular weight heterogeneous polymers are formed (Ariunbaatar et. al., 2014). These polymers are more difficult to degrade so the soluble COD cannot increase further.

According to Haug et. al. (1978), pretreatment for 30 minutes at 100^{o}C was mentioned as the optimum condition and was similar to the observed results from pretreatment for 30 minutes at 90^{o}C. However, in this paper only optimum conditions which would yield the best solubilisation were examined. Additionally, according to Valo et. al. (2004), pretreatment for 60 minutes at 130^{o}C were mentioned as the best conditions. After this temperature and duration solubilisation of COD slows down. Results of this study was similar to the observed results from pretreatment for 60 minutes at 127^{o}C.

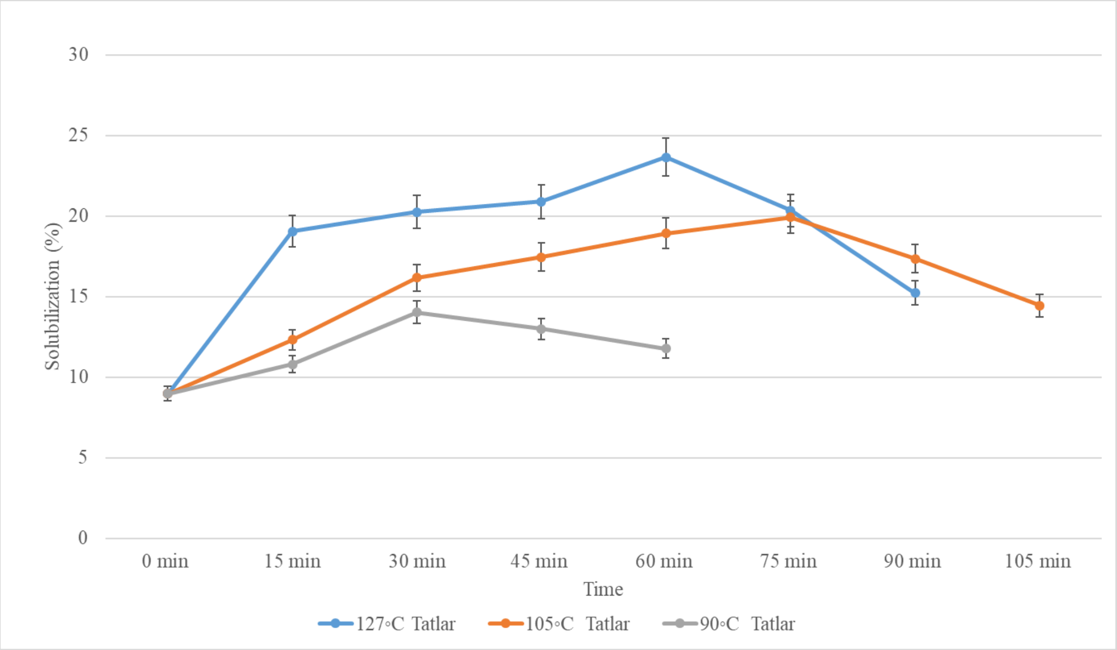


Figure 4-2. Solubilisation of Tatlar WAS sludge with respect to pretreatment temperature and time

In the study of Wang et al. (1997b), solubilisation of organic substances were analyzed at 60°C to 120°C and for 5 to 60 minutes durations. It was found that COD solubilisation reaches to maximum 30 minutes and 60°C for sludge samples. Similar results were also observed in other studies (Brooks and Grad, 1968; Wang et al., 1988). Analogically, according to Appels et al. (2010), improvement of soluble COD amount decreased after pretreatment for 60 minutes at 70°C. In this work similar results were observed. Higher solubilisation for lower temperatures was observed in 30 minutes duration rather than 60 minutes duration.

After pretreatment, CST measurements were also done in order to assess the change in sludge dewaterability. Dewaterability of WAS deteriorated as it is shown by the increase in the CST value in Figure 4-3. CST was 835.9 sec for control (non-pretreated sample) and it increased with thermal hydrolysis to nearly 2000 sec. Even though there was a general increase of CST with pretreatment, the higher temperatures generally caused the lower CST value compared to lower pretreatment temperatures. Pretreatment methods have an adverse influence on the dewaterability possibly because of cell and floc disintegration (Müller et al., 2004). Due to the fact that higher cell and floc disintegration cause higher COD solubilisation, samples which have higher soluble COD has worse CST.

As shown in Figure 4-3, CST increased up to 90 minutes at 127^{o}C from 835.87 sec to 1856.27 sec. For pretreated sludge samples at 127^{o}C, CST increased with pretreatment duration increase. On the other hand, CST increased up to 90 minutes at 105^{o}C from 835.87 sec to 1982.27 sec. For pretreated sludge samples at 105^{o}C, CST increased up to 90 minutes and then decreased. Additionally, CST increased up to 45 minutes at 90^{o}C from 835.87 sec to 1960.27 sec. For pretreated sludge samples at 90^{o}C, CST increased up to 45 minutes and then decreased.

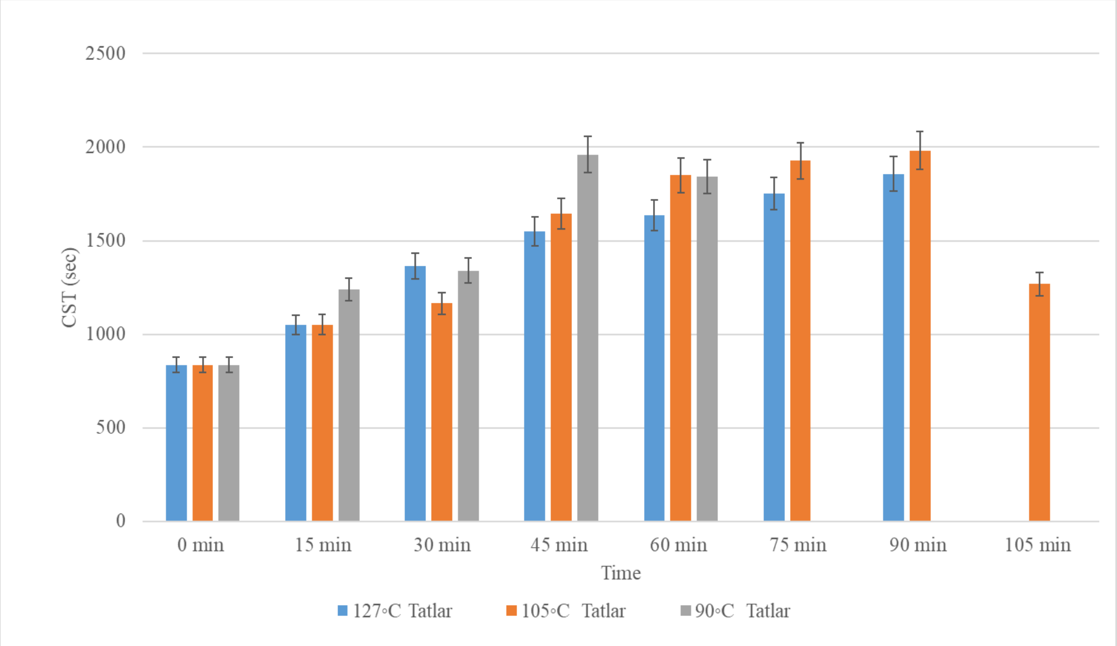


Figure 4-3. CST values of Tatlar WAS sludge with respect to pretreatment temperature and time

Three worst dewaterability results listed below for each temperature:

- Pretreatment for 90 minutes at 127°C resulted in CST of 1856.3 sec,
- Pretreatment for 90 minutes at 105°C resulted in CST of 1982.3 sec,
- Pretreatment for 45 minutes at 90°C resulted in CST of 1960.3 sec.

According to Şahinkaya (2018), initial characterization of WAS sample was determined as 8.5 second. After thermally pretreated, CST increased to 30.4, 41.7 and 50.4 respectively at 80°C, 100°C 120°C. This study concludes that particles size decreased and polymeric substances increased due to floc dissolution; therefore, the CST values deteriorated.

4.2. Second Pretreatment: Pretreatment with MBR Sludge

In the second pretreatment study, sludge from Middle East Technical University MBR Treatment Plant was pretreated with different temperatures and durations. Each time 100 mL of sludge was pretreated to see the effect of temperature and time on COD solubilisation. Three different sets were conducted under this group.

First set conducted with WAS from MBR was pretreated at 127°C and for six different durations which were 15, 30, 45, 60, 75 and 90 minutes. Results are summarized in Table 4-2. It was found total COD of sludge was 21,754 mg/L initially and initial soluble COD of sludge was 663 mg/L with 3.04 % of total COD being soluble COD. Despite being pretreated for up to 90 minutes, maximum soluble COD of sludge was 7,435 mg/L with 34.18% solubilisation at 60-minute duration (Figure 4-4). For the first set, average TS and VS were determined as 27,605 mg/L and 18,291 mg/L, respectively. On the other hand, initial condition TS and VS were determined as

27,866 mg/L and 20,833 mg/L, respectively. Due to thermal pretreatment, TS and VS of pretreated samples increased slightly with respect to non-pretreated sample similar to the previous set. Additionally, average pH for this experiment was 7.63.

Table 4-2. *Initial conditions of sludge for MBR and best values after pretreatment*

Parameter	Unit	Initial value	Maximum value after 127°C	Maximum value after 105°C	Maximum value after 90°C
Total COD	mg/L	21,754			
Soluble COD	mg/L	663	7,435	4,645	3,002
COD solubilization (solubilized total COD)	%	3.04	34.18	21.35	13.80
TS	mg/L	27,866	27,605	28,873	27,987
VS	mg/L	20,883	18,291	19,345	21,891
pH	-	7.17	7.63	7.78	7.44

Second set was pretreated at 105°C and for seven different durations which were 15, 30, 45, 60, 75, 90, and 105 minutes. Maximum soluble COD of sludge was achieved as 4,645 mg/L with 21.35% solubilisation at 30-minute duration. For the second set, average TS and VS were determined as 28,873 mg/L and 19,345 mg/L, respectively. Additionally, average pH for this step was 7.78 (Table 4-2).

Third set was pretreated at 90°C and for four different durations which were 15, 30, 45 and 60 minutes. Maximum soluble COD of sludge was 3,002 mg/L with 13.80% solubilisation at 45-minute duration (Table 4-2). As can be seen, as the temperature dropped (from 127 to 105 and then to 90°C), the solubilisation level decreased. For the third set, average TS and VS were determined as 27,987 mg/L and 21,891 mg/L respectively. Additionally, average pH for this step was 7.44. All the results from the first set is summarized in Figure 4-4. The graph shows averages of triplicate analyses and the error bars indicate the standard deviations.

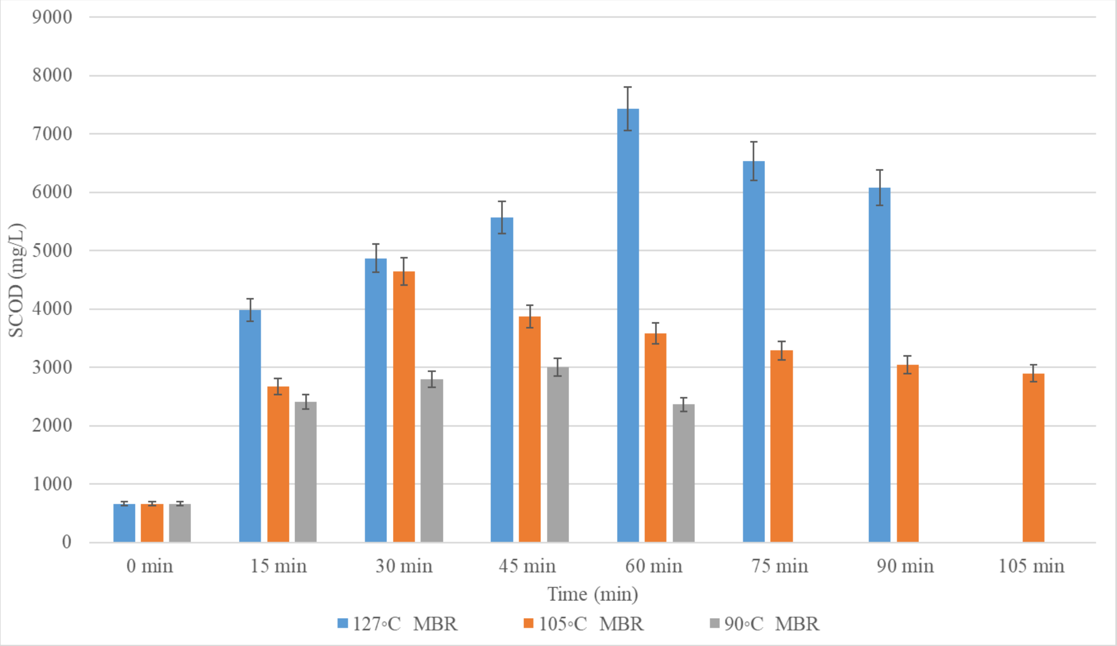


Figure 4-4. Soluble COD concentrations of MBR sludge with respect to pretreatment temperature and time

At 127°C, SCOD/TCOD percent increased from 3.04 to 18.31, 22.38, 25.58, 34.18, 30.03 and 27.94 for 15, 30, 45, 60, 75, and 90 minutes, respectively (Figure 4-5). At 105°C, SCOD/TCOD percent increased from 3.04 to 12.30, 21.35, 17.81, 16.44, 15.11, 14.01 and 13.32 for 15, 30, 45, 60, 75, 90, and 105 minutes, respectively. Lastly, at 90°C, SCOD/TCOD percent increased from 3.04 to 11.08, 12.87, 13.80 and 10.89 for 15, 30, 45, and 60 minutes, respectively. As can be seen similar to the study for Tatlar WAS, there was a peak of solubilisation at an intermediate time period for each temperature studied. The times of this peak depended on the temperature of pretreatment and were observed as 60 minutes, 30 minutes and 45 minutes for 127°C, 105°C and 90°C, respectively.

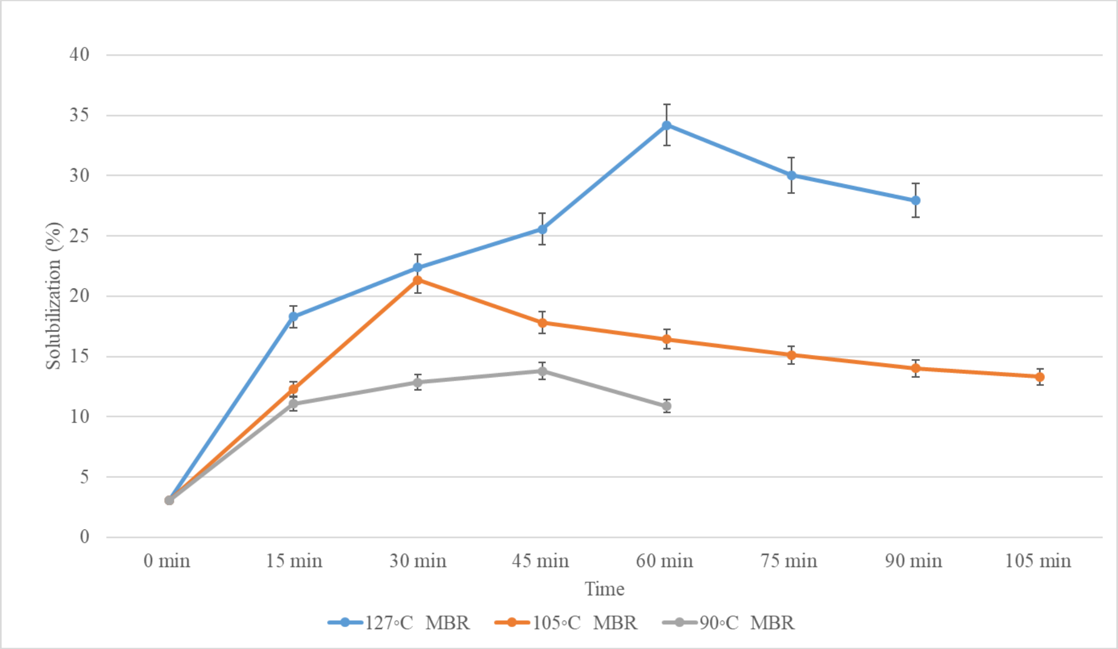


Figure 4-5. Solubilisation of MBR sludge with respect to pretreatment temperature and time

After pretreatment, CST measurements were done in order to assess the change in sludge dewaterability and presented in Figure 4-6 as averages of triplicate analyses with standard deviations. Dewaterability of WAS deteriorated as it is shown by the increase in the CST value with pretreatment. CST was 28.1 sec for control and it increased with thermal hydrolysis. Generally, the increases did not follow a particular trend.

As shown in Figure 4-6, CST were increased up to 15 minutes at 127^{o}C from 28.10 sec to 496.47 sec. On the other hand, CST increased up to 75 minutes of pretreatment at 105^{o}C from 28.10 sec to 484.43 sec. For pretreated sludge samples at 105^{o}C, CST increased up to 75 minutes of pretreatment and then decreased. Additionally, CST increased up to 60 minutes at 90^{o}C from 28.10 sec to 194.43 sec. For pretreated sludge samples at 90^{o}C, CST increased with duration increase. Again in this part of the study pretreatment caused significant deterioration of sludge CST. However, the increases were not as high as it was for Tatlar WWTP. This possibly due to the floc structure of MBR and its dissolution. One can see that always the maximum soluble COD achieved was higher for Tatlar sludge compared to MBR sludge. This possibly has a reflection of CST as lower values.

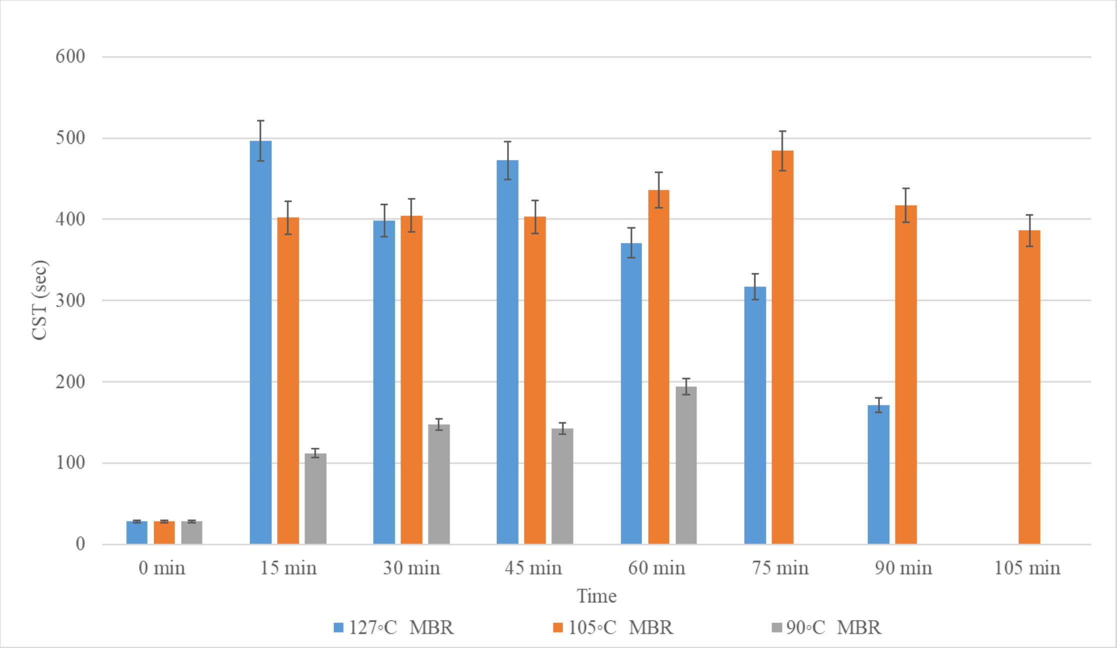


Figure 4-6. CST values of MBR sludge with respect to pretreatment temperature and time

Three worst dewaterability results are listed below for each temperature:

- Pretreatment for 15 minutes at 127^{o}C resulted in CST of 496.5 sec,
- Pretreatment for 75 minutes at 105^{o}C resulted in CST of 484.4 sec,
- Pretreatment for 60 minutes at 90^{o}C resulted in CST of 194.4 sec.

4.3. Third Pretreatment: Pretreatment with an OID WWTP

In the third pretreatment study, sludge from an OID Wastewater Treatment Plant was pretreated with different temperatures and durations. 100 mL of sludge was pretreated to increase soluble COD amount. Figure 4-7 shows the averages of solubilized COD along with the standard deviations.

First set conducted with WAS from an OID was pretreated at 127°C and for six different durations which were 15, 30, 45, 60, 75 and 90 minutes. It was found total COD of sludge was 44,565 mg/L and initial soluble COD of sludge was 442 mg/L with 0.99% of total COD being soluble COD (Table 4-3). Despite being pretreated for up to 90 minutes, maximum soluble COD of sludge was 8,650 mg/L with 19.41% solubilisation at 60-minute duration. For the first set, average TS and VS were determined as 35,333 mg/L and 23,933 mg/L, respectively. On the other hand, initial condition TS and VS were determined as 37,083 mg/L and 30,116 mg/L, respectively. During thermal pretreatment, TS and VS of pretreated samples increased with respect to non-pretreated sample as mentioned previously. Additionally, average pH for this experiment was 7.23.

Table 4-3. *Initial conditions of sludge for OID and best values after pretreatment*

Parameter	Unit	Initial value	Maximum value after 127°C	Maximum value after 105°C	Maximum value after 90°C
Total COD	mg/L	44,565			
Soluble COD	mg/L	442	8,650	4,719	2,789
COD solubilization (solubilized total COD)	%	0.99	19.41	10.59	6.26
TS	mg/L	37,083	35,333	34,211	35,870
VS	mg/L	30,117	23,933	27,533	23,912
pH	-	6.45	7.23	8	7.41

Second set conducted with WAS from OID was pretreated at 105°C and for seven different durations which were 15, 30, 45, 60, 75, 90 and 105 minutes. Maximum soluble COD of sludge was 4,719 mg/L with 10.59% solubilisation at 60-minute duration. For the second set, average TS and VS were determined as 34,211 mg/L and 27,533 mg/L, respectively. Additionally, average pH for this step was 8.

Third set conducted with WAS from OID was pretreated at 90°C and for four different durations which were 15, 30, 45 and 60 minutes. Maximum soluble COD of sludge was 2,789 mg/L with 6.26% solubilisation at 30-minute duration. For the third set, average TSS and VSS were determined as 35,870 mg/L and 23,912 mg/L, respectively. Additionally, average pH for this step was 7.41. On the overall the effect of 127°C on the solubilisation of COD was predominantly stronger compared to the effect of other temperatures as seen from Figure 4-7.

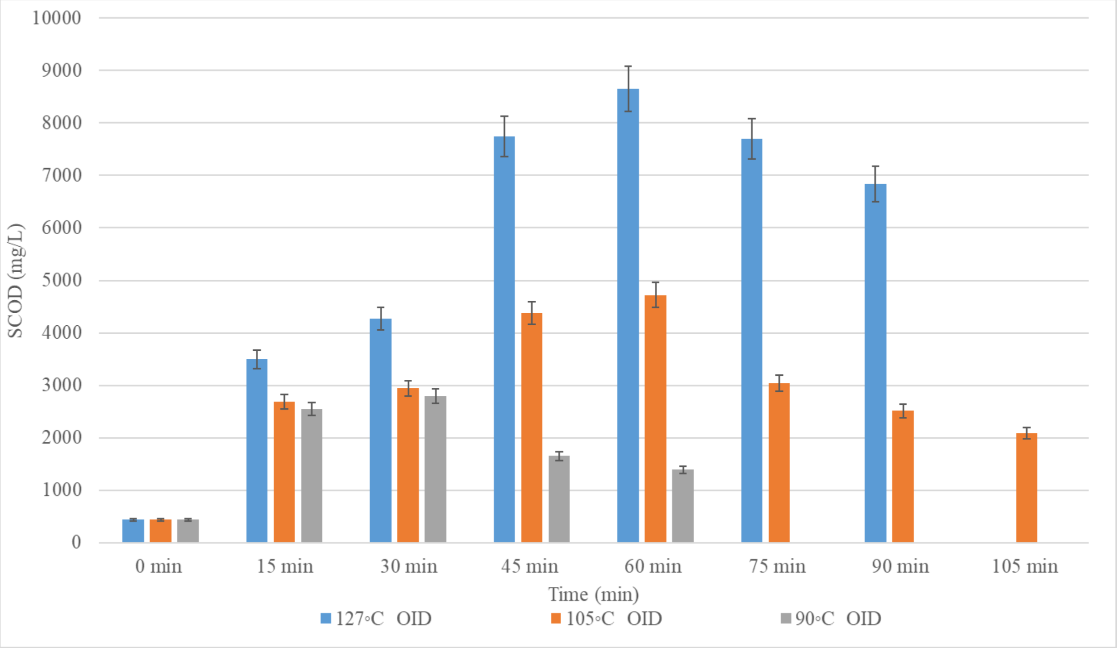


Figure 4-7. Soluble COD concentrations of OID sludge with respect to pretreatment temperature and time

At 127^{o}C, SCOD/TCOD percent increased from 0.99 to 7.85, 9.58, 17.37, 19.41, 17.28 and 15.35 for 15, 30, 45, 60, 75, and 90 minutes, respectively. At 105 ^{0}C, SCOD/TCOD percent increased from 0.99 to 6.04, 6.61, 9.82, 10.59, 6.83, 5.63 and 4.68 for 15, 30, 45, 60, 75, 90, and 105 minutes, respectively. Moreover, at 90^{o}C, SCOD/TCOD percent increased from 0.99 to 5.72, 6.26, 3.71, and 3.12 for 15, 30, 45, and 60 minutes, respectively. Results are shown in Figure 4-8 as the averages of triplicate measurements. This Figure 4-8 too shows the distinctly different solubilisation effect of 127^{o}C compared to other two lower temperatures.

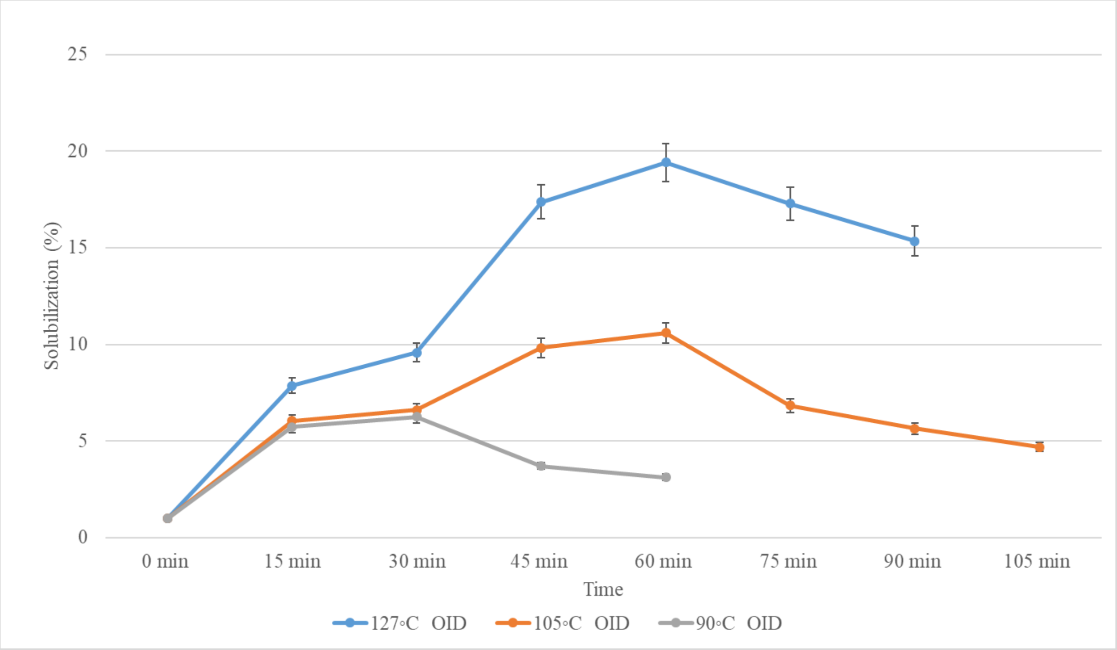


Figure 4-8. Solubilisation of OID sludge with respect to pretreatment temperature and time

After pretreatment, CST measurements were conducted in order to assess the change in sludge dewaterability. Dewaterability of WAS deteriorated as it is shown by the increase in CST value with pretreatment. This is similar to the case of previous sludges. CST was 15.5 sec for control and it increased with thermal hydrolysis up to a value of over 250 sec. It seems that the higher the pretreatment temperature, higher the increase in CST. So this must be an outcome of COD dissolution and floc break-up which happens higher at higher temperatures.

As shown in Figure 4-9, CST increased up to 30 minutes at 127°C from 15.5 sec to 273.0 sec. For pretreated sludge samples at 127°C, CST increased up to 30 minutes of pretreatment and then decreased slightly. On the other hand, CST increased up to 90 minutes at 105°C from 15.5 sec to 136.2 sec. For pretreated sludge samples at 105°C, CST increased up to 90 minutes and then decreased. Additionally, CST increased up to 60 minutes at 90°C from 15.50 to 59.93. For pretreated sludge samples at 90°C, CST increased with duration increase. Considering the results up to this point, it is expected that higher solubilisation to cause worse CST. However, the trend was slightly different in this set.

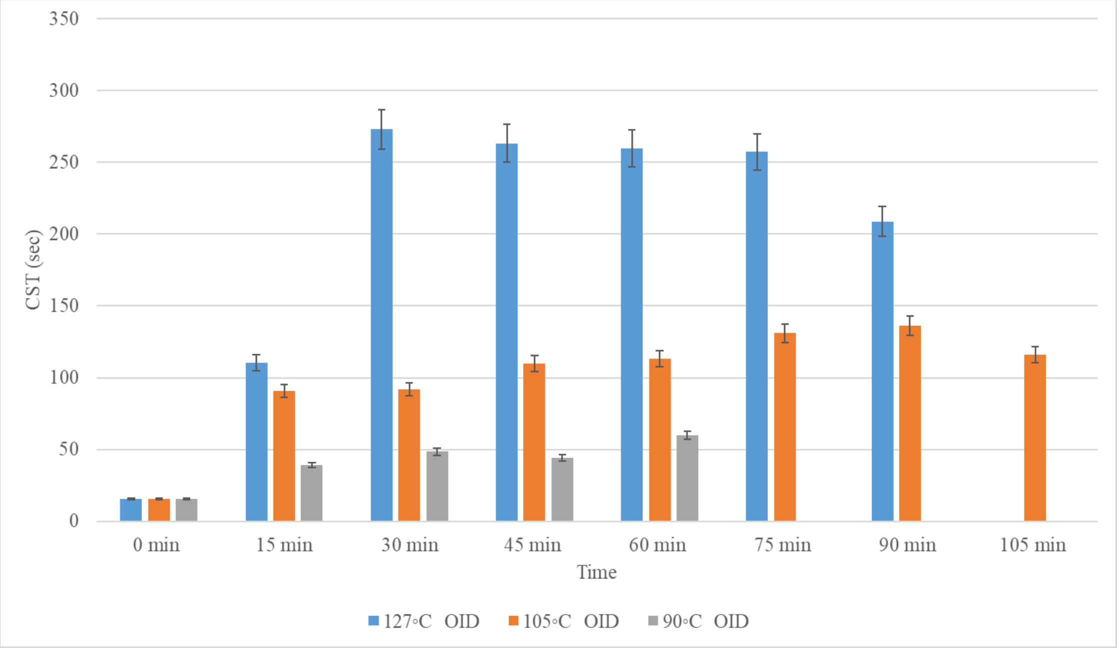


Figure 4-9. CST of OID sludge with respect to pretreatment temperature end time

Three worst dewaterability results are listed below for each temperature:

- Pretreatment for 30 minutes at 127°C resulted in CST of 273.0 sec,
- Pretreatment for 90 minutes at 105°C resulted in CST of 136.2 sec,
- Pretreatment for 60 minutes at 90°C resulted in CST of 59.9 sec.

Among the studied pretreatment methods, 60 minutes at 127°C and 30 minutes at 90°C were chosen for the second part of anaerobic biodegradability study. Since 60 minutes at 127°C resulted in the highest amount of soluble COD release, it was decided to be investigated in the second part. In addition, 30 minutes at 90°C was also selected to investigate the lower temperature effects to be able to do a comparative analysis. Additionally, this second set of condition demonstrates a low energy pretreatment.

4.4. Anaerobic Batch Reactors

For setting up BMP assays, 750 mL sludge from WWTPs of Tatlar, METU and OID were pretreated at 60 minutes at 127°C and 30 minutes at 90°C separately. Before anaerobic batch reactor set-up, preliminary pretreatment experiments were conducted with 100 mL sludge samples. However, to minimize experimental error that would be introduced due to having a number of small individual samples, 750 mL samples were prepared and pretreated for each type of BMP assays. Before setting up the reactors, total COD, pH, TS, TSS, VS, VSS and CST of the applied pretreatment conditions were again measured for the same sludge samples. These results are summarized below.

In Figure 4-10, Figure 4-11 and Figure 4-12, soluble COD concentration normalized with average VS for each reactor set-up is given. SCOD/VS ratio is calculated since after pretreatment solids content changes to some degree as explained previously. So normalization with VS makes a healthier comparison possible.

As seen in the Figure 4-10, for Tatlar non-pretreated sample has the lowest SCOD/VS ratio. On the other hand, as expected, sample which was pretreated for 60 minutes at 127^{o}C has the highest SCOD/VS ratio. Additionally, sample which was pretreated for 30 minutes at 90^{o}C has the moderate SCOD/VS ratio. The increase is 59% from the original sample to 127^{o}C pretreated sample; on the other hand, the same value is about 37% for 90^{o}C pretreated sample.

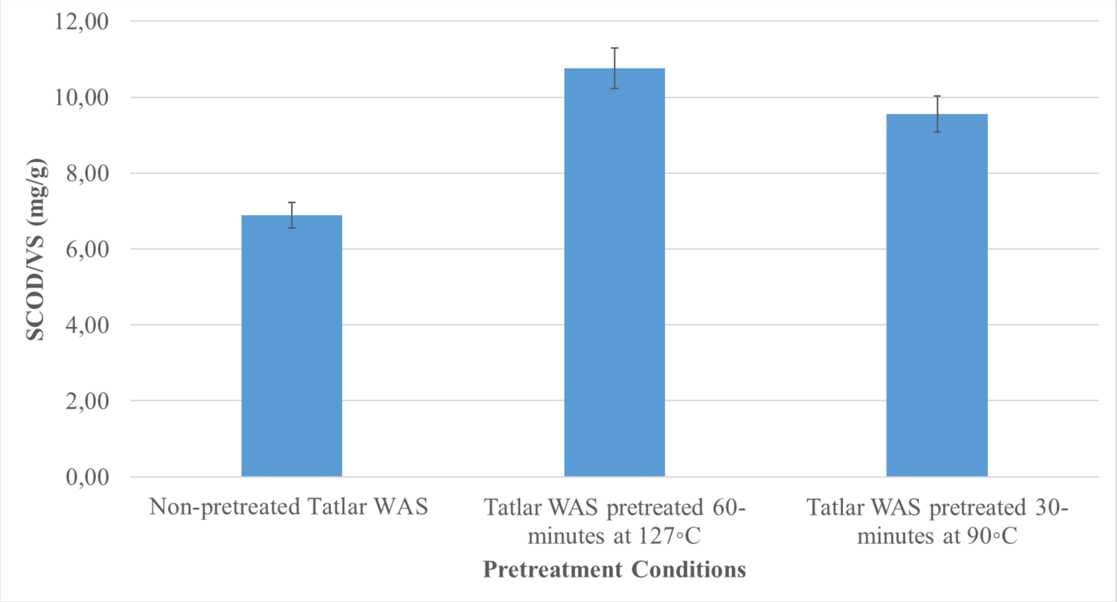


Figure 4-10. Ratio of SCOD/VS for Tatlar WAS sample

For MBR sludge, similar graph was plotted. As seen in the Figure 4-11, SCOD/VS ratio of the sample which was pretreated for 30 minutes at 90^{o}C was improved but to a limited extent compared with the sample which was pretreated for 60 minutes at 127^{o}C. The increase is about 118% from the non-pretreated sample to 127^{o}C pretreated sample; on the other hand, the same value is about 42% for 90°C pretreated sample.

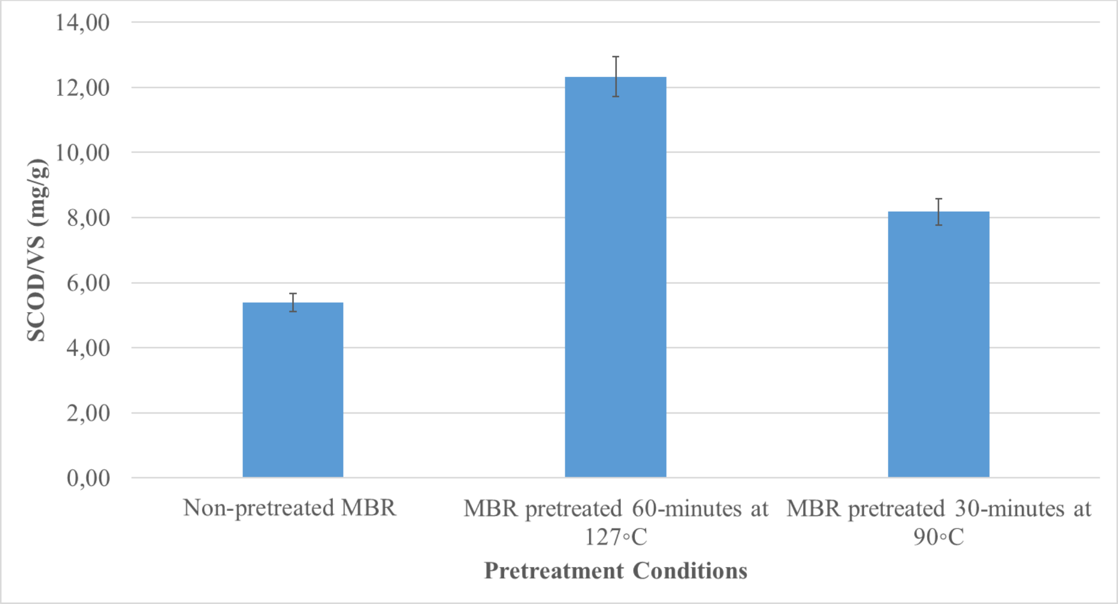


Figure 4-11. Ratio of SCOD/VS for MBR sample

On the other hand, as shown in Figure 4-12, SCOD/VS ratio of the sample which was pretreated for 30 minutes at 90°C is about 34% higher compared to the non-treated OID sludge. However, SCOD/VS ratio of the sample which was pretreated for 60 minutes at 127°C is approximately 2.5 times higher than the sample which was pretreated for 30 minutes at 90°C. Furthermore, the sample which was pretreated for 60 minutes at 127°C is approximately three times higher than the non-pretreated sample.

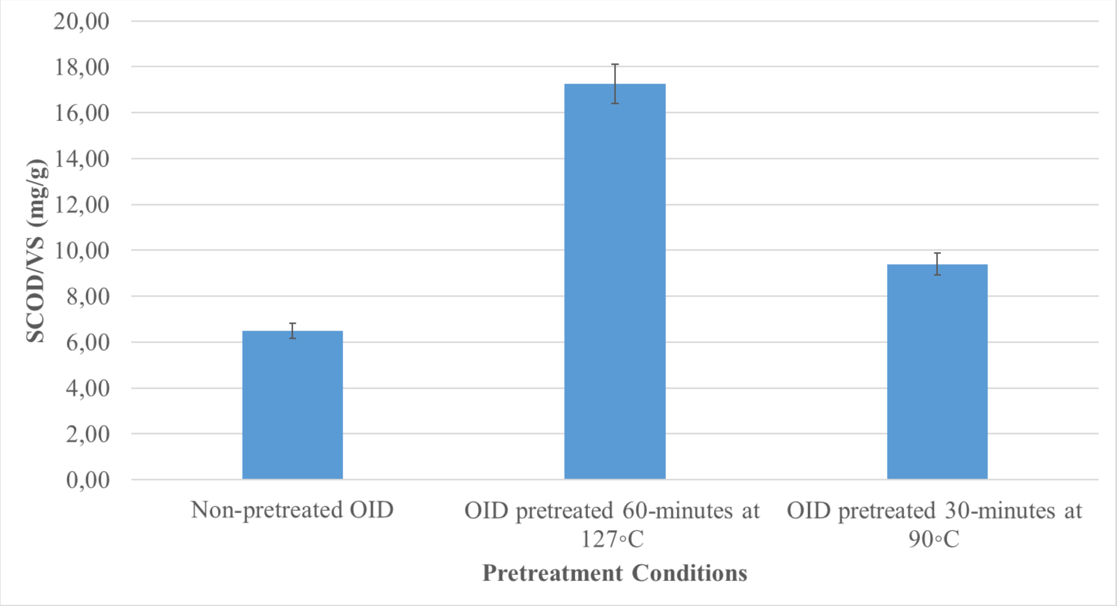


Figure 4-12. Ratio of SCOD/VS for OID sample

As seen in the Figure 4-13, COD solubilisation was improved with higher temperatures. In control samples, soluble COD concentrations were 7.8, 6.5 and 7.3 mg/g for Tatlar WAS, MBR and OID, respectively. After applying pretreatment for 60 minutes at 127^{o}C, these values increased to 10.76, 12.33 and 17.27 mg/g. Pretreatment for 60 minutes at 127^{o}C was the most effective on OID sample. On the other hand, pretreatment for 30 minutes at 90^{o}C was the most influential on Tatlar sample.

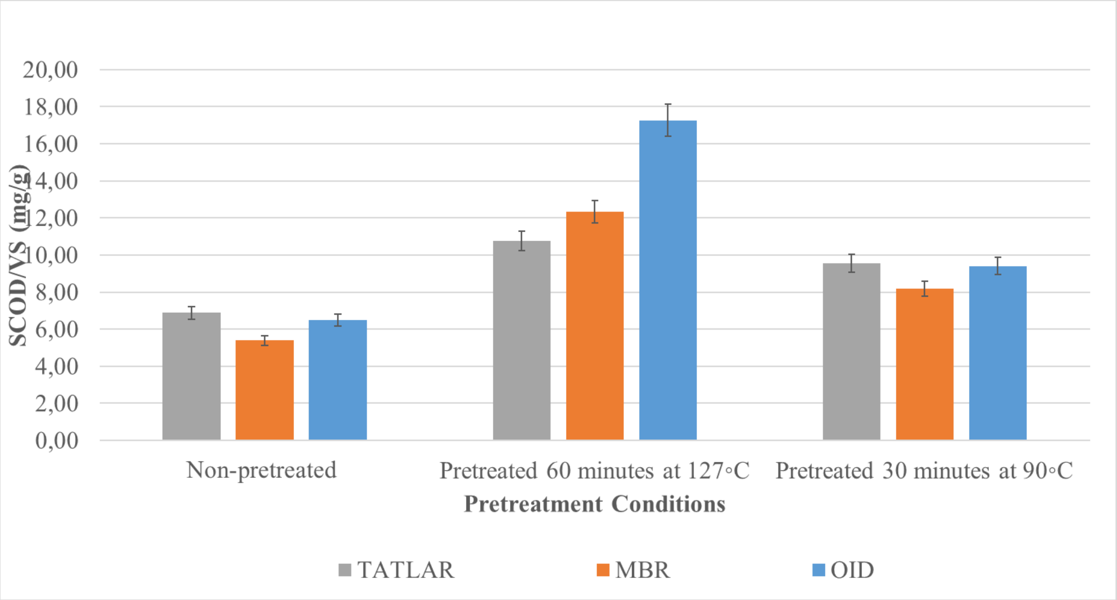


Figure 4-13. Ratio of SCOD/VS for each type of reactor

4.4.1. pH of Samples before Reactor Set-Up

In general, anaerobic digestion is operated under neutral pH such as 6.5 to 7.6. Methanogenic processes will be affected negatively if pH is less than 6.3 or higher than 7.8, which will decrease biogas production (Leitao et al., 2006).

In BMP reactors, initial pH was around 7-8.5. With respect to that, no problems are expected to be observed during the experiment.

4.4.2. Soluble COD Concentration in Reactors before Anaerobic Digestion

For the BMP reactors, soluble COD concentrations are shown in Figure 4-14 with respect to pretreatment conditions.

Soluble COD of non-pretreated Tatlar digester (seed), Tatlar WAS, Tatlar sample which was pretreated for 60 minutes at 127°C and Tatlar sample which was pretreated for 30 minutes at 90°C are 1,775 mg/L, 2,379 mg/L, 3,679 mg/L and 3,220 mg/L, respectively. Here again one can see that the higher the temperature and the time (generally), the higher the COD release.

Similarly, soluble COD of MBR, MBR sample which was pretreated for 60 minutes at 127°C and MBR sample which was pretreated for 30 minutes at 90°C were 1,874 mg/L, 4,329 mg/L and 2,725 mg/L, respectively. An increasing of soluble COD is for MBR sample at 127 °C and 60 min similarly as compared to Tatlar sample. This is similar to the results obtained during preliminary study.

Finally, soluble COD of OID, OID sample which was pretreated for 60 minutes at 127°C and OID sample which was pretreated for 30 minutes at 90°C were 2,185 mg/L, 5,769 mg/L and 2,934 mg/L, respectively. OID sludge had the sharpest increase at 127°C, 60 min treatment compared to other two sludges.

As an overall evaluation one can see that, the highest soluble COD concentration was observed for the sample pretreated at 127°C for 60 minutes as expected. For the sample pretreated at 90°C for 30 minutes, soluble COD concentrations were improved in regard to non-pretreated reactors. However, improvement of soluble COD concentrations was limited with respect to the sample pretreated at 127°C for 60 minutes. This situation was expected in parallel to earlier the results obtained from earlier experiments. Additionally, among all the samples pretreated at 127°C for 60 minutes, OID had shown the highest improvement compared to the others. On the other hand, among all the samples pretreated for 30 minutes at 90°C, Tatlar WAS had shown the highest improvement compared to the others.

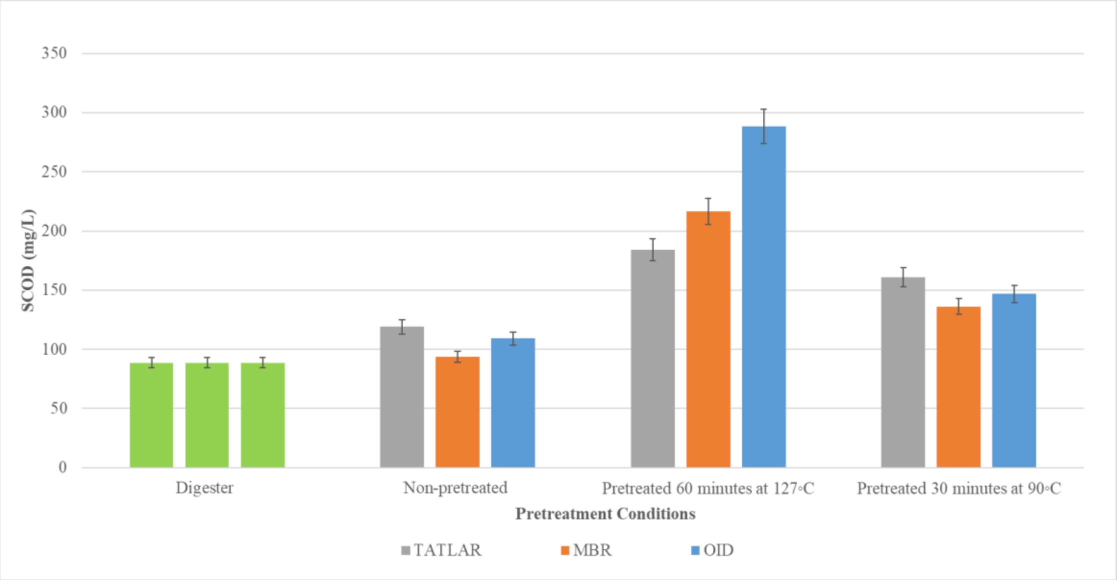


Figure 4-14. Concentrations of soluble COD for all samples

4.4.3. Capillary Suction Time for Reactors before Anaerobic Digestion

In this study, impact of pretreatment on dewaterability is determined by using CST. As pretreatment leads to cell disintegration and disturbance on the floc structures in sludge, CST is used to assess whether structure of sludge is disrupted or not. If dewaterability is getting harder, it shows that sludge is being disrupted more and more. Thus, in this part, CST results obtained right before reactor set-up are used to assess cell disintegration indirectly. The aim is not only analyzing dewaterability but also having a general idea on the cell and floc disintegration. Figure 4-15 shows the results for all the samples in a comparative manner following pretreatment and right before reactor set-up.

Pretreating Tatlar sample for 30 minutes at 90^{o}C resulted in the worst dewaterability, which was not expected. Due to disrupting effect, Tatlar sample pretreated for 60 minutes at 127^{o}C should have had the worst dewaterability. Besides this trend has not the same as observed in the preliminary experiments. One thing to note during preliminary experiments the CST values were exceptionally high. This is possibly due to the characteristics of sludge sample collected from WWTP; which may show drastic changes time to time. MBR sample pretreated for 60 minutes at 127°C resulted in the worst dewaterability, as observed previously. Similar to the MBR sample, OID sample pretreated for 60 minutes at 127°C had the worst dewaterability due to worse floc disruption effect.

As a general comparison, the highest impact of pretreatment on CST was observed for two types of pretreated sludge samples for 60 minutes at 127°C as expected, which were MBR and OID samples. Because these sludge samples were disrupted more than the other ones in this process. On the other hand, non-pretreated sludge samples had lowest CST. However, Tatlar samples had high CST, especially the sample pretreated for 30 minutes at 90°C. One thing to note is that Tatlar sample had the highest CST for all conditions among all the sludge samples studied.

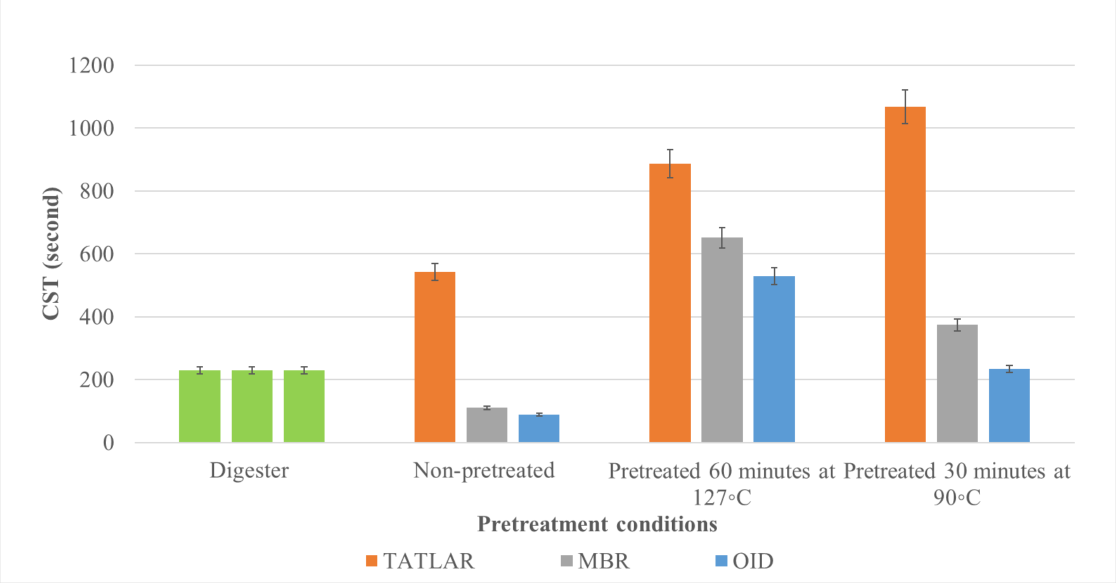


Figure 4-15. CST values for each pretreatment and for all sludge samples

4.4.4. BMP Results

4.4.4.1. Total Gas Production

The BMP test was then set up and continued for 29 days. The reactor operation time in this study was in line with previous work. Batch anaerobic digestions were proceeded along 17-24 days in the study of Carrere et. al., (2008).

Total biogas production was measured daily by a water displacement device during whole experiment (29 days). Biogas production increased for the first half of reactor operation (first 15 days). For the second half of reactor operation, which covers the last 14 days, production of gas decreased. So the, frequency of gas production measurements was reduced. All data shown in this section were calculated by taking the average of three replicate measurements. Graphs also show standard deviations. Gas production for Tatlar WAS is presented in Figure 4-16. Reactor names in this part are in abbreviations as described in the text.

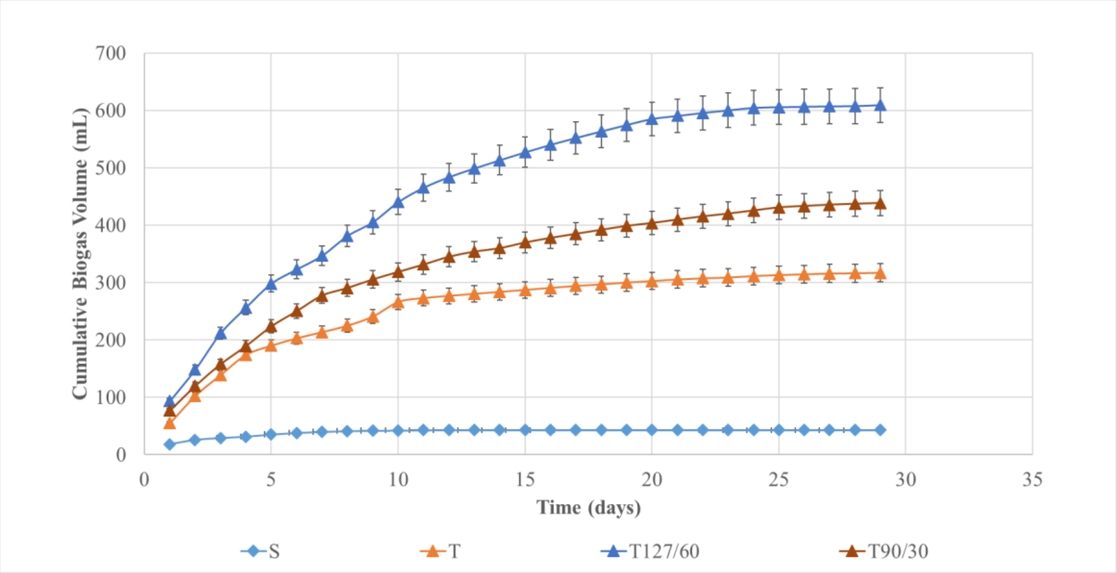


Figure 4-16. Generation of cumulative biogas from Tatlar WAS samples

For Tatlar WAS, which was pretreated at 127^{o}C and for 60 minutes, the cumulative biogas generation was 608.6 mL. T127/60 has the highest biogas generation amount, when it is compared to the Tatlar sample pretreated at 90^{o}C and for 30 minutes and non-pretreated sample. For Tatlar WAS, which was pretreated at 90^{o}C and for 30 minutes, the cumulative biogas generation was 438.43 mL. This means that, pretreated sludge resulted in more biogas generation than non-pretreated sample. With respect to non-pretreated reactor (T), approximately 92% and 39% higher biogas generation were observed from T127/60 and T90/30, respectively. This increase was parallel to the increase in soluble COD following pretreatment (Figure 4-10).

According to Haug et.al. (1978), sludge sample pretreated for 30 minutes at 100^{o}C produces 14% more biogas than the control sample. Furthermore, various studies also show similar results such as sludge samples pretreated thermally at 100^{o}C generated 30% more biogas. However, this type of pretreatment needs longer treatment durations with respect to pretreatments applied at higher temperatures (Hiraoka et.al., 1985).

Biodegradability of anaerobic sludge increases with increasing temperature of pretreatment (Carrere et al., 2008). However, WAS biodegradability reduced after a certain temperature threshold (>200 ^{0}C) because of melanoidins compounds. Furthermore, in the study of Bougrier et al. (2008), a slight reduction in biodegradability of WAS was observed at temperatures higher than 190^{o}C due to toxic substance formation mentioned above.

Figure 4-17 shows the biogas results for MBR sludge. For MBR sample which was pretreated at 127^{o}C for 60 minutes, the cumulative biogas generation was 498.01 mL. In addition, for MBR sample which was pretreated at 90^{o}C at 30 minutes, the cumulative biogas generation was 389.43 mL. Non-pretreated reactor (MBR) generated 365.04 mL biogas. Thus, with respect to non-pretreated MBR reactor, in MBR127/60 and MBR90/30 approximately 37% and 7% higher biogas generation were observed, respectively. This was similar to the soluble COD release with pretreatment as given in Figure 4-11.

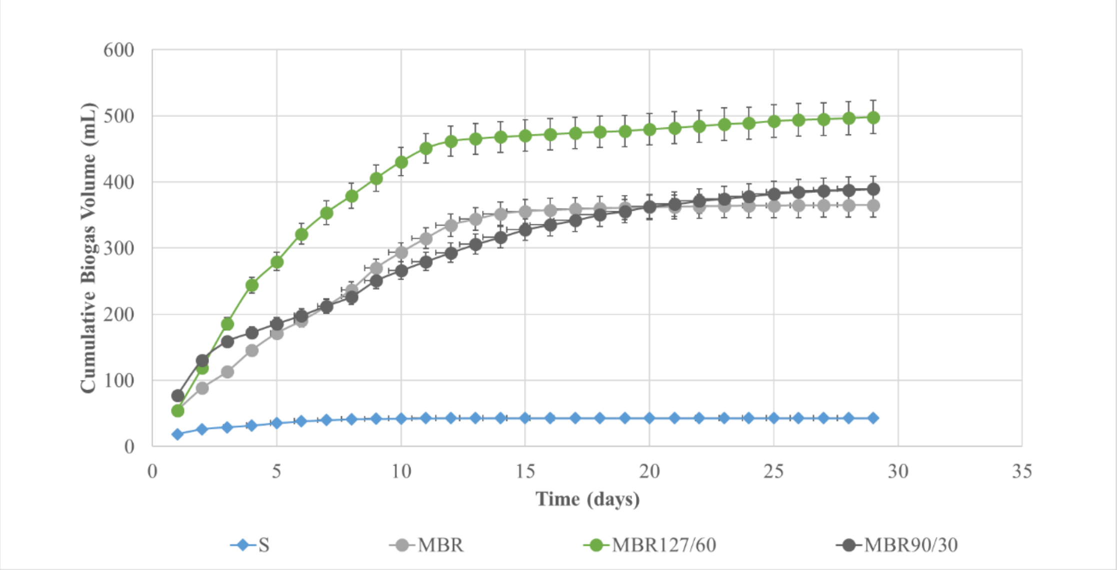


Figure 4-17. Generation of cumulative biogas from MBR samples

Figure 4-18 shows the biogas results for OID sludge. For OID samples which was pretreated at 127°C and for 60 minutes, production of cumulative biogas was 368.31 mL. OID127/60 has the highest biogas generation amount, when it is compared to the sample pretreated at 90°C and for 30 minutes and non-pretreated sample. For OID90/30, generation of biogas was 324.10 mL. For non-pretreated reactor (OID), generated biogas amount was 170.59 mL. Thus, with respect to non-pretreated OID reactor, approximately 116% and 89% higher biogas generation were observed in OID127/60 and OID90/30, respectively. This trend was similar to the soluble COD increase with pretreatment (Figure 4-12).

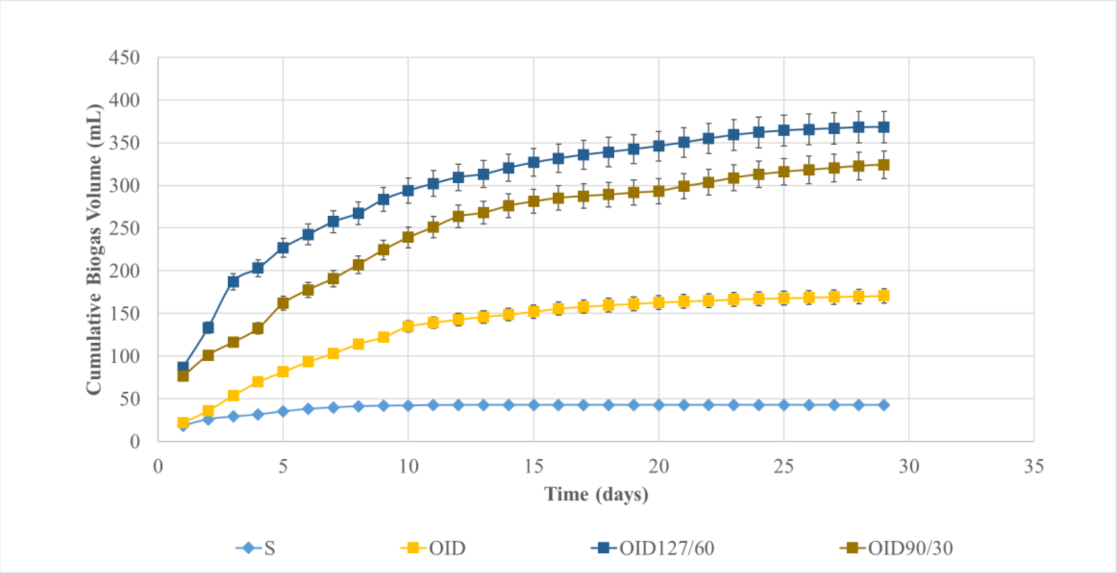


Figure 4-18. Generation of cumulative biogas from OID samples

Figure 4-19 summarizes the biogas results for all sludges studied. As shown in the Figure 4-19, production of total biogas was dependent on pretreatment and sludge type. Non-pretreated samples generated less amount of biogas. On the other hand, highest biogas amounts were observed from samples pretreated at 127^{o}C and for 60 minutes.

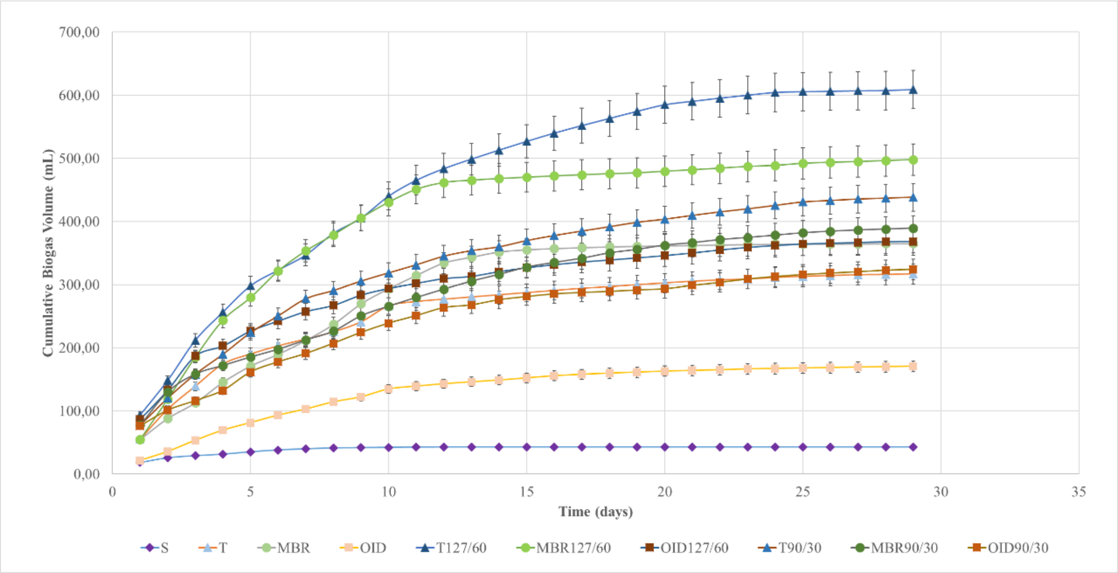


Figure 4-19. Generation of cumulative biogas from each sample

As can be seen in the Figure 4-19, OID sludge was always lower in biogas productions compared to the other sludges. In addition, even though pretreatment causes the release of soluble COD, biogas production was not observed in parallel. This is thought to be due to a possible toxicity of the sludge, the biogas generation was limited for all OID reactors. On the other hand, Tatlar sample pretreated at 127^{o}C and for 60 minutes generates highest biogas with respect to other reactors. This is parallel to the earlier results as started above.

In addition to cumulative biogas production, daily biogas productions from reactors are also plotted (Figure 4-20) and discussed. Daily biogas generation from Tatlar sample is shown in Figure 4-20. According to Figure 4-20, highest biogas generation occurred on the first day. However, this could be caused by the nitrogen gas used in sparging the reactors. As mentioned before, when BMP assays were set up, nitrogen gas was used to sparge the bottles for 10 minutes. Other than the first day, highest biogas generation was seen on the third day for Tatlar sample pretreated for 60 minutes at 127^{o}C. And after that day, generation of daily biogas decreased gradually. Hydrolysis of organic substances possibly the reason of the peak observed on day 3. In the earlier days, Tatlar sample pretreated for 60 minutes at 127^{o}C has been generating the highest daily biogas volume. Sample pretreated for 30 minutes at 90^{o}C and non-pretreated sample (T) followed T127/60. However, in the final days, daily biogas generation were approximately the same and very low. From the graph it is clearly seen that the peaks observed moved to earlier days with pretreatment (non-pretreated Tatlar at 4 and 10 days; 127^{o}C pretreated Tatlar at 3 days and 90^{o}C pretreated Tatlar on 5 and 7 days). This is an expected result of pretreatment; since the purpose is to solubilize organics, the result is clearly seen from the Figure 4-20. This also indicates that the rate of hydrolysis increased and the reactors sped-up.

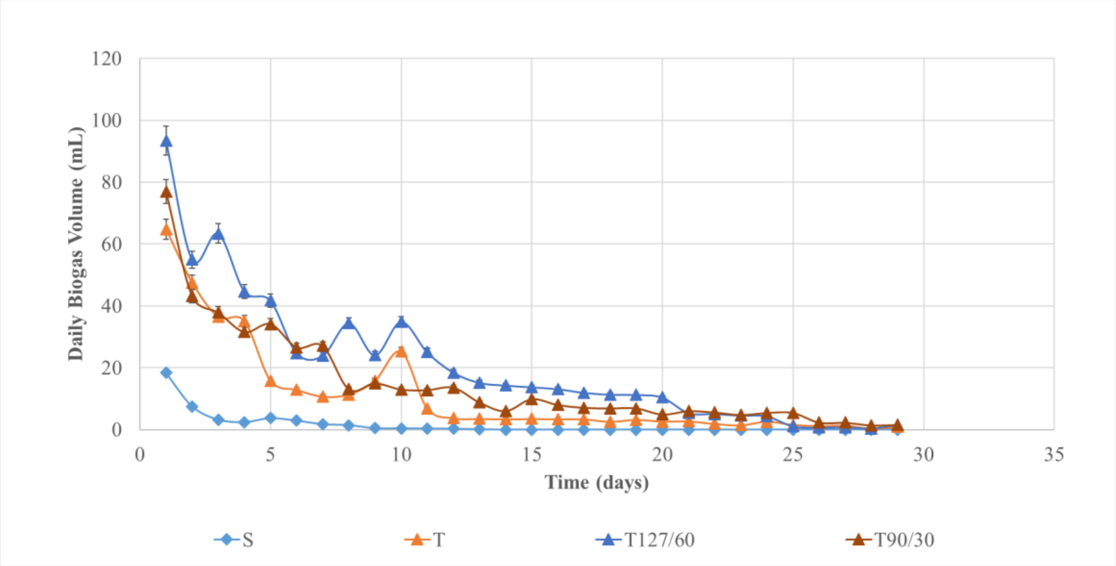


Figure 4-20. Generation of daily biogas from Tatlar WAS samples

Daily biogas generation for MBR samples is shown in Figure 4-21. In the early days of anaerobic digestion, MBR sample pretreated for 60 minutes at 127^{o}C has been generating the highest daily biogas volume. However, in the final days, sample pretreated for 30 minutes at 90°C has generated more biogas than the other samples. Also from the graph it is clearly seen that the peaks observed moved to earlier days with pretreatment (non-pretreated MBR at 4 and 8 days; 127°C pretreated MBR at 6 days and 90°C pretreated MBR on 9 days). This is an expected result of pretreatment; since the reactors got faster and organic degradation sped-up.

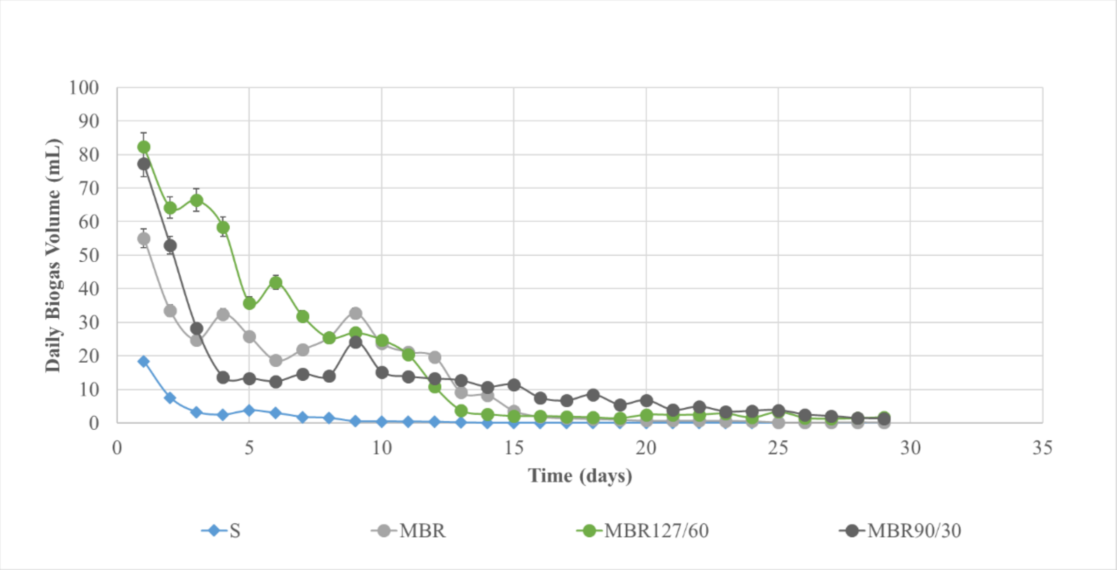


Figure 4-21. Generation of daily biogas from MBR samples

Daily biogas generation for OID samples are shown in Figure 4-22. In the early days of anaerobic digestion, OID sample pretreated for 60 minutes at 127°C has been generating the highest daily biogas volume. Additionally, sample pretreated for 30 minutes at 90°C also has been producing considerable amount of daily biogas. Peaks are observed at the third, fifth, and the third day (much smaller peak) for OID127/60, OID90/30, and non-pretreated sample (OID), respectively. However, in the final days, biogas volume was approximately the same. In addition, daily biogas generation in the final days were less than 5 mL. Similar to the other sludges, from the graph it is clearly seen that the peaks observed moved to earlier days with pretreatment (non-pretreated OID at 3, 8 and 10 days; 127°C pretreated OID at 3 and 5 days and 90°C pretreated OID on 5 and 9 days). This is an expected result of pretreatment; since the purpose is to solubilize organics, purpose is achieved and the result is clearly seen from Figure 4-23.

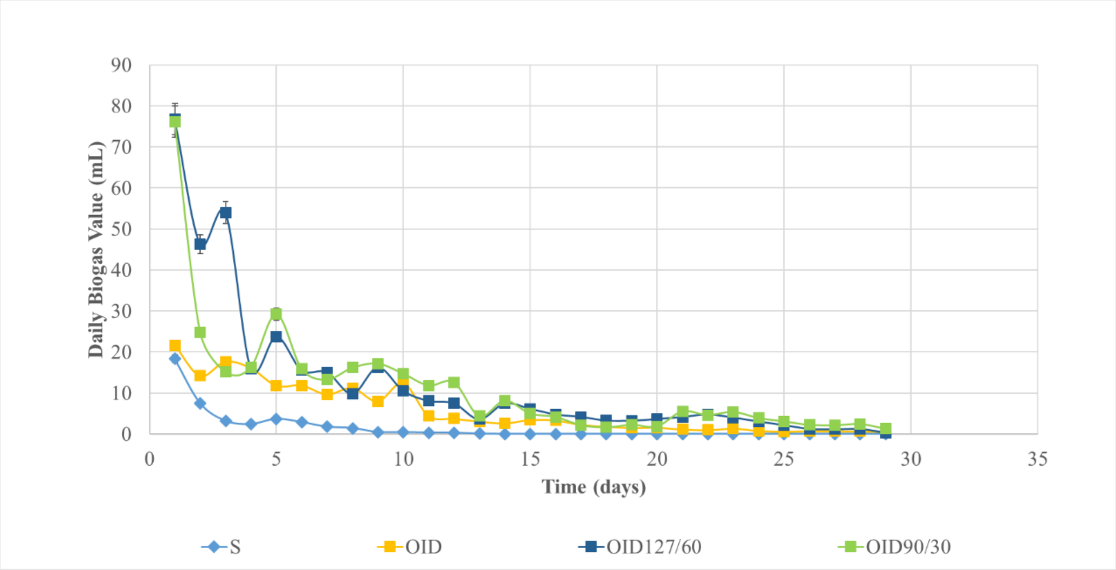


Figure 4-22. Generation of daily biogas from OID samples

Figure 4-23 gives an overall summary of all the obtained results for daily biogas generation. Peaks were observed in different days for different types of pretreated and non-pretreated samples as mentioned above indicating clearly the improvement. As seen in Figure 4-23, all of the samples managed to adapt to the environment. Due to soluble COD concentrations of pretreated samples 127°C and for 60 minutes were much higher than the other ones; their peaks were also observed more frequent during the first 15 days. Thus, their biogas generation were also more than the other samples. Furthermore, T127/60 sample had the highest peak compared to other samples.

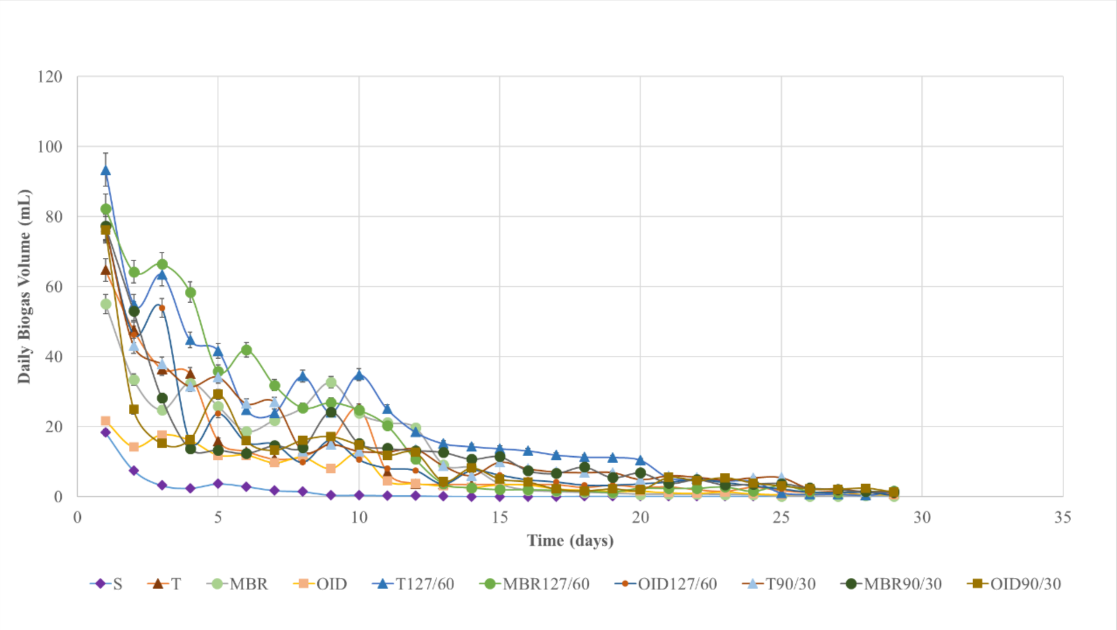


Figure 4-23. Generation of daily biogas from each sample

4.4.4.2. Methane Production

In addition to biogas volume, biogas composition was also analyzed by gas chromatography. Then, methane production was determined. During 29 days, cumulative methane generation was investigated. For each sample, measurements were done with two replicates. Methane generation amounts and their standard deviations were calculated from these two measurement replicates and triplicate reactors (a total of six measurements). Cumulative methane generation are shown in the following figures (Figure 4-24 for Tatlar sample, Figure 4-25 for MBR sample and Figure 4-26 for OID sample).

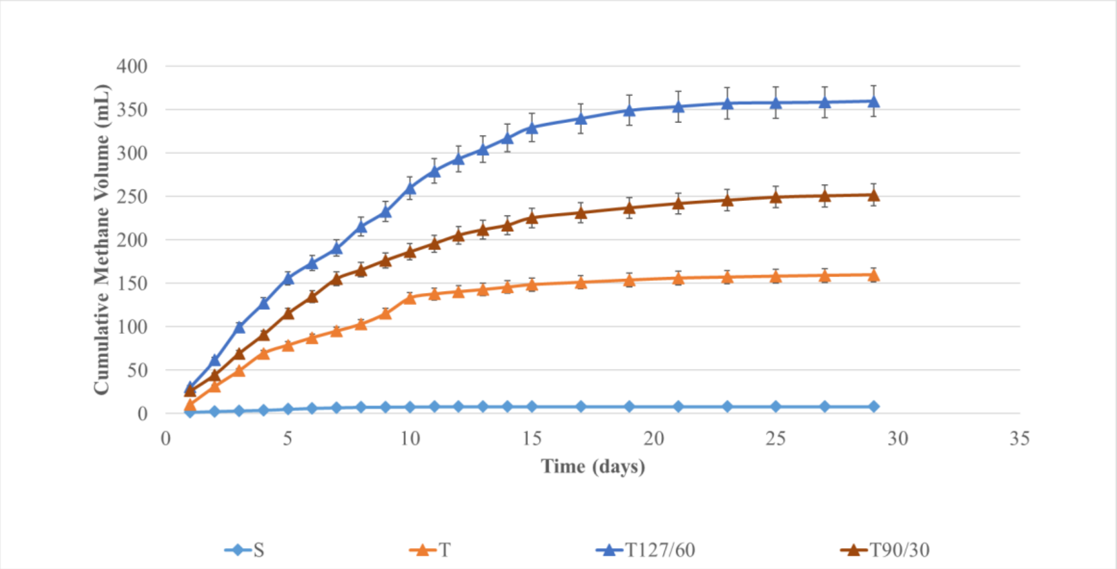


Figure 4-24. Generation of cumulative methane from Tatlar WAS samples

Methane productions were 50%, 59%, and 57% of the total biogas from reactors T, T127/60, and T90/30, respectively (Figure 4-24). On the other hand, with respect to non-pretreated BMP reactor (T), approximately 125% and 57% more methane was produced in T127/60 and T90/30 reactors, respectively. This means that, methane generation was improved in the pretreated samples.

Results from methane analyses indicated that %53, %59, and %56 of the total biogas from MBR, MBR127/60, and MBR90/30 was methane, respectively (Figure 4-25). Moreover, with respect to non-pretreated sample (MBR), approximately 49% and 13% more methane were produced from MBR127/60 and MBR90/30, respectively. These results are also in line with the performances that are reported in literature.

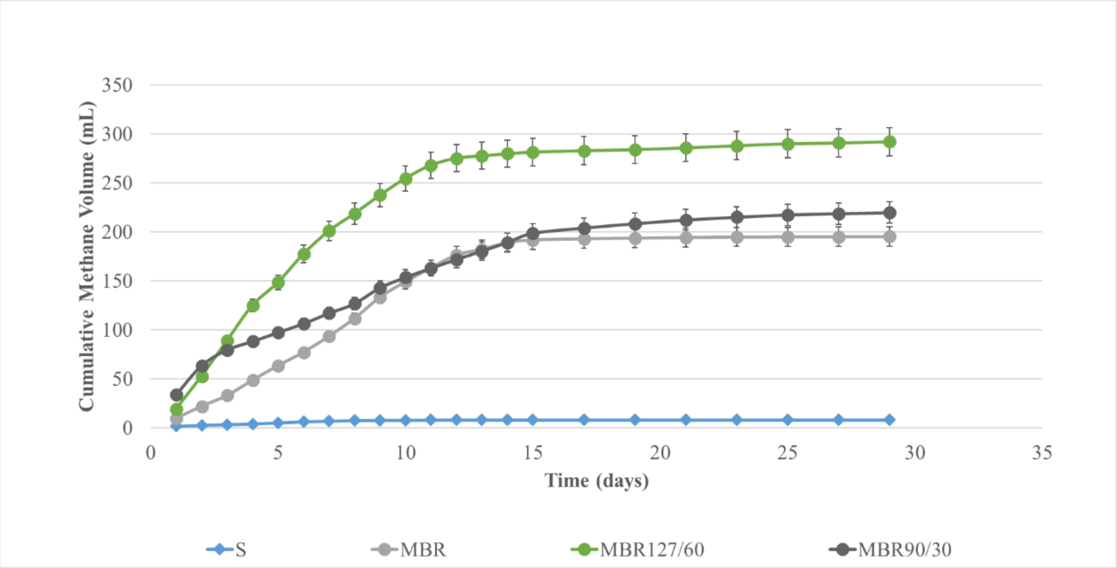


Figure 4-25. Generation of cumulative methane from MBR samples

Methane generated was %50, %58, and %55 of the total biogas generation from reactors OID, OID127/60 and OID90/30, respectively (Figure 4-26). Furthermore, with respect to non-pretreated sample (OID), approximately 150% and 109% more methane were produced from OID127/60 and OID90/30 respectively. These results indicate that the performance of reactors operated are better compared to the ones reported in literature. The improvement is also parallel to the ones observed in soluble COD release.

According to Haug et. al. (1978), sludge sample pretreated for 30 minutes at 100°C produced 13% more methane than the control sample. Additionally, methane generation from the sludge sample pretreated for 30 minutes at 120°C increased by 25% with respect to control sample (Jeong et.al., 2007). These results indicate that the performance of our reactors are better compared to the ones reported in some literature.

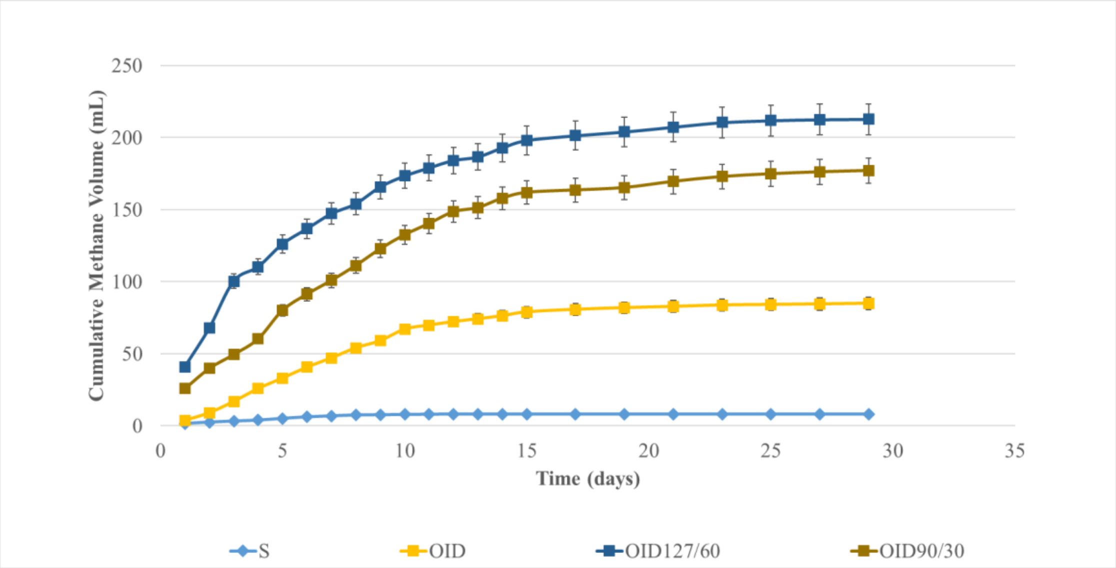


Figure 4-26. Generation of cumulative methane from OID samples

Figure 4-27 summarizes the results from all the reactors for their methane productions during the reactor operation period. As seen in Figure 4-27, highest cumulative methane generation was observed from T127/60 (359 mL). On the other hand, non-pretreated OID sample generated lowest methane volume (85 mL) except for seed sludge sample (S). Till the end of reactor termination 160 mL, 195 mL, 85 mL, 359 mL, 292 mL, 213 mL, 252 mL, 220 mL, 177 mL, and 9 mL methane were generated during anaerobic digestion from T, MBR, OID, T127/60, MBR127/60, OID127/60, T90/30, MBR90/30, OID90/30, and S, respectively. Although soluble COD amount in OID and MBR samples were much higher than the others, methane generations were not observed up to this level. Methane generation does not only depend on solubilisation but also depend on adaptation of microorganisms to the environment. Therefore, this is not an unexpected result, considering especially the industrial sludge.

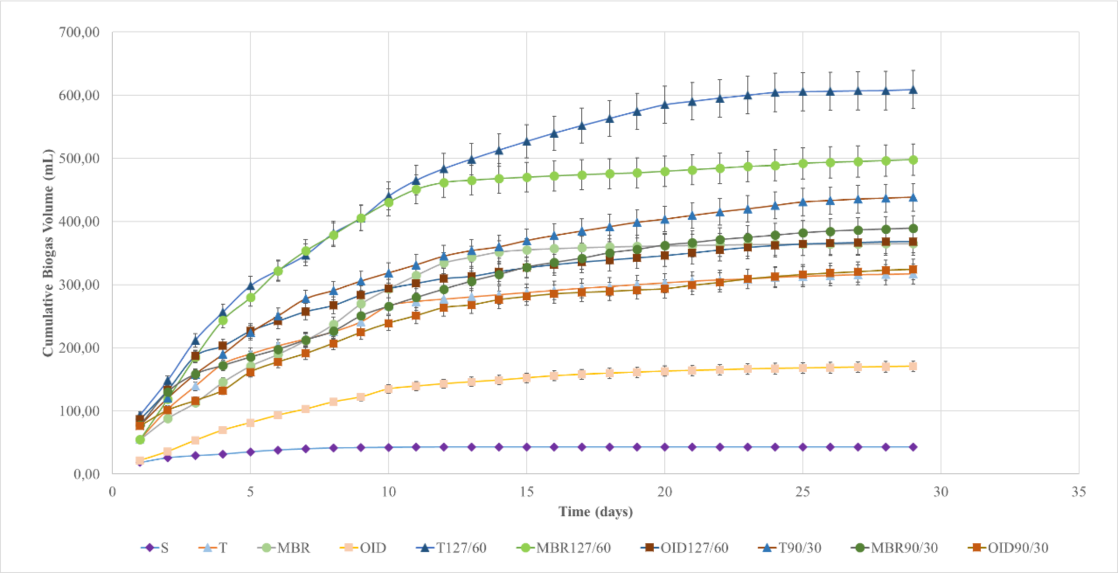


Figure 4-27. Generation of cumulative methane from each sample

According to Takashima (2008), cumulative methane generation from sludge sample pretreated for 60 minutes at 120ºC was 2.4 to 3.0 times higher with respect to non-pretreated sample. According to Eskicioğlu and Kor-Bicakci (2019), biogas generation from samples which were pretreated at higher temperatures are more than the samples pretreated at lower temperatures. These results are similar to the results observed in this part. All of the samples pretreated for 60 minutes at 127ºC generated higher amount of methane than the other samples.

Daily methane generation from Tatlar samples are shown in Figure 4-28. In the first day, daily methane generation from T90/30 was slightly lower than T127/60. However, after that day, T127/60 kept the highest methane amount in the early days. T90/30 and non-pretreated sample (T) followed T127/60. Daily methane generation peaks were observed at days 3, 8 and, 10 for T127/60. On the other hand, methane generation peaks for T90/30 were observed at days 3, 5, and 7. In the final days, daily methane generation were nearly the same.

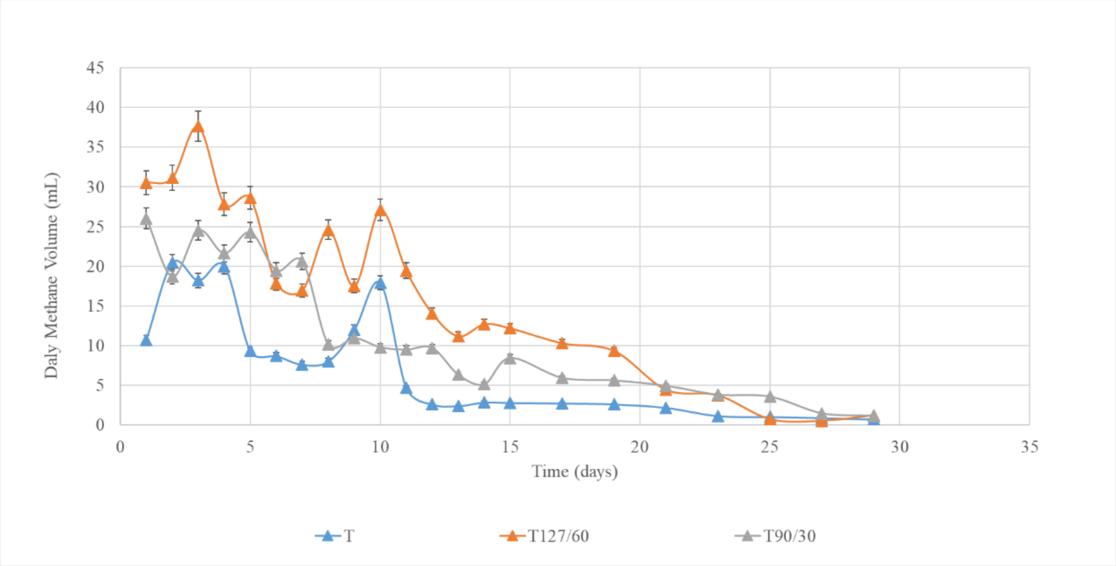


Figure 4-28. Generation of daily methane from Tatlar WAS samples

Daily methane generation from MBR samples are shown in Figure 4-29. In the first day, daily methane generation from MBR127/60 was nearly the same with non-pretreated MBR sample. In addition, methane generation amount from MBR90/30 was more than the other samples. After first day, in the early days of anaerobic digestion, MBR127/60 had the highest methane generation amount. However, in the final days, MBR90/30 generated more methane than the other samples. Peaks for MBR127/60 were observed at days 3, 4 and, 6. Moreover, peaks for MBR90/30 were observed on day 2, 3 and 9.

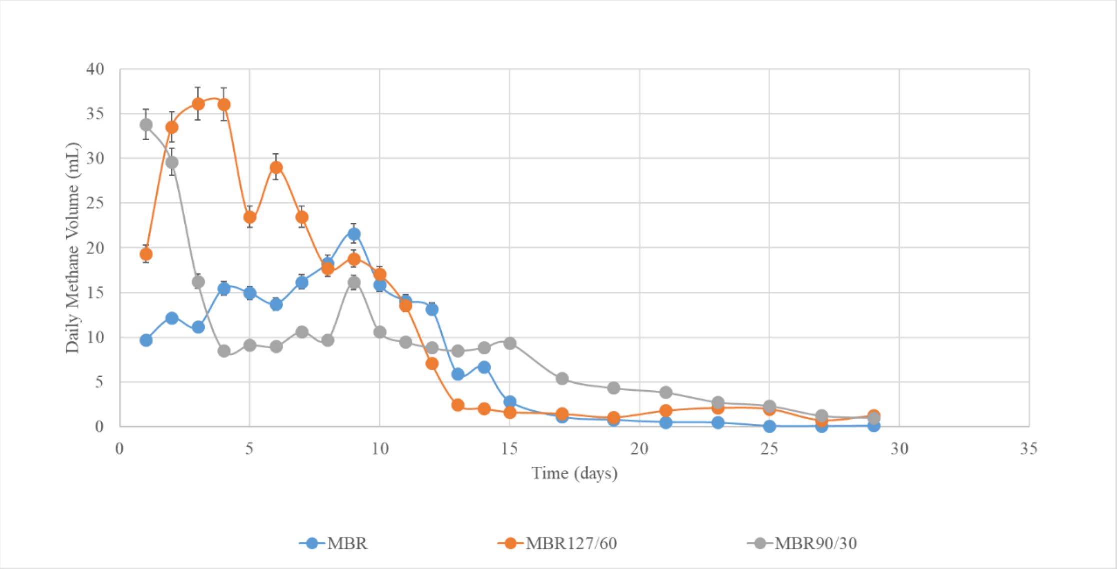


Figure 4-29. Generation of daily methane from MBR samples

Daily methane generation from OID samples are shown in Figure 4-30. In the early days of anaerobic digestion, OID127/60 had the highest daily biogas generation volume. However, in the final days, daily biogas generation from each sample were approximately the same. In addition, volume of generated biogas in the final days was less than 3 mL. It can be seen from Figure 4-30 that non-pretreated OID sludge has adverse effect on hydrolysis step of anaerobic digestion. Since industrial OID sludge leads to lower degradation and organic conversion efficiency, this trend is expected and similar with the studies in literature (Feng et al., 2013; Elbeshbishy et al., 2012).

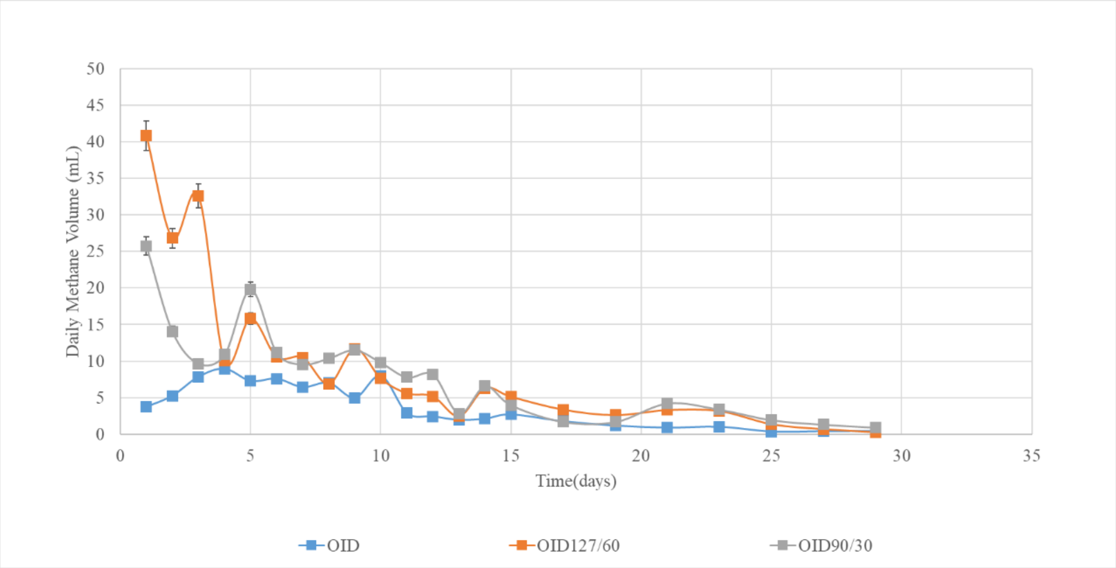


Figure 4-30. Generation of daily methane from OID samples

Daily methane generation from all studied samples are summarized in Figure 4-31. Peaks were observed at different days for different types of pretreated and non-pretreated samples. Due to soluble COD concentrations of pretreated samples at 127^{o}C and for 60 minutes, peaks were much higher than the other samples; the peaks were observed more frequent as well during anaerobic digestion. Thus, their biogas generation were also more than the other samples.

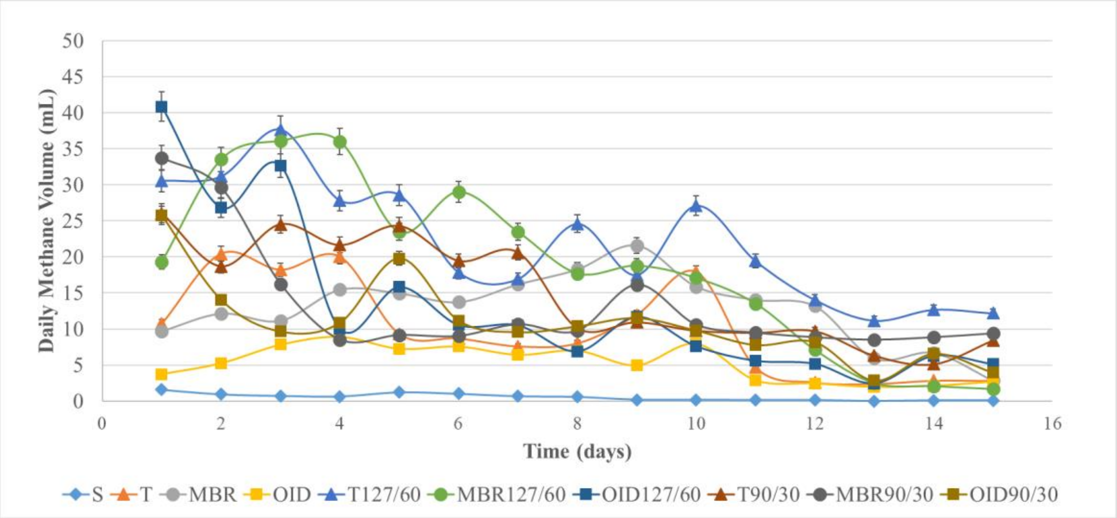


Figure 4-31. Generation of daily methane from each sample

4.4.4.3. Changes in Solids Concentrations and pH

To determine the performance of the reactors, TS and VS reduction during anaerobic digestion was investigated.

Table *4-4* summarizes the TS and VS concentrations at the beginning and end of the reactor operation as well as their percent removals. The TS and VS values are measured in triplicates; so, the data in tables show the standard deviations calculated as well. For seed sample removal of TS was 6.3% and removal of VS was 25,2%. This sample was one of the non-pretreated samples and examined for seed control.

TS and VS reduction percentages for Tatlar samples are shown in Table 4-4Table *4-4*. Reduction rates of TS was 16.4%, 17.4% and 17.5%, respectively for T, T127/60 and T90/30 samples. Additionally, reduction rates of VS were 46.8%, 48.0% and 47.4%, respectively for T, T127/60 and T90/30 samples. TS removal during anaerobic digestion changes between 20 and 25 percent in literature (Köksoy and Sanin, 2010; Bougrier et al., 2006). On the other hand, according to Bahçeci and Sanin, for municipal WWTP, TS removal during anaerobic digestion 10 and 25 percentage.

Therefore, the reductions in this study seems to be acceptable compared to the values in literature.

Table 4-4. *Initial and final TS and VS concentrations and reductions after anaerobic digestion for Tatlar WAS sample*

Sample	**TSi (mg/L)**	**TSf (mg/L)**	**TS Removal (%)**	**VSi (mg/L)**	**VSf (mg/L)**	**VS Removal (%)**
T	23,955±487.5	20,022±668.5	16.4	17,266±337.1	9,183±106.6	46.8
T127/60	23,722±554.3	19,588±311.2	17.4	17,094±702.1	9,138±445.9	48
T90/30	23,350±432.1	19,255±327.5	17.5	16,844±632.6	9,844±365	47.4

In addition to TS and VS, TSS, VSS and pH values were also measured in the reactors (Table 4-5). The TSS, VSS and pH values were measured in triplicates. The data shown in the Table 4-5 has standard deviations calculated as well.

For Tatlar seed sludge sample removal of TSS was 5.4% and removal of VSS was 21.6%. This sample was one of the non-pretreated sample and examined for control. TSS and VSS reduction percentages for Tatlar samples are shown in the Table 4-5. Reduction rates of TSS was 15.9%, 15.8% and 15.5%, respectively for T, T127/60 and T90/30 samples. Additionally, reduction rates of VSS was 45.3%, 46.2% and 44.4%, respectively for T, T127/60 and T90/30 samples.

Table 4-5. *TSS and VSS amounts before and after anaerobic digestion and removal percents for Tatlar WAS sample*

Sample	TSSi (mg/L)	TSSf (mg/L)	TSS Removal (%)	VSSi (mg/L)	VSSf (mg/L)	VSS Removal (%)	pHi	pHf
T	21,488±653	18,061±251	15.9	15,205±212.4	8,300±253.4	45.3	7.49	7.45
T127/60	20,833±899	17,544±292	15.8	14,755±457.2	7,938±501.6	46.2	7.66	7.48
T90/30	20,472±645	17,305±163	15.5	14,994±582.5	8,333±266.8	44.4	7.29	7.50

Although sample S has much lower TSS and VSS value, other samples are similar in initial and final TSS and VSS magnitudes. Last parameter (pH) has almost similar values for each reactor, both at the reactor start-up and reactor take-down.

TS and VS reduction percentages for MBR samples are shown in the Table 4-6. Reduction rates of TS was 14.4%, 14.7% and 16.4%, respectively for MBR, MBR127/60 and MBR90/30 samples. Additionally, reduction rates of VS were 42.3%, 44.8% and 42.5%, respectively for MBR, MBR127/60 and MBR90/30 samples. As can be seen from table reductions are increasing with pretreatment for both VS and TS. This is as expected due to COD solubilisation and conversion of COD to methane during reactor operation.

Table 4-6. *Initial and final TS and VS concentrations and reductions after anaerobic digestion for MBR sample*

Sample	TSi (mg/L)	TSf (mg/L)	TS Removal (%)	VSi (mg/L)	VSf (mg/L)	VS Removal (%)
MBR	23,322±639.8	19,966±360.5	14.4	17,388±390.7	10,022±411.9	42.3
MBR127/60	23,976±668.5	20,433±530.1	14.7	17,550±435.9	9,677±386.5	44.8
MBR90/30	23,122±408.6	19,327±439.1	16.4	16,661±649.5	9,555±415.9	42.5

In addition to TS and VS, TSS, VSS and pH are also measured in the reactors (Table 4-7). The TSS, VSS and pH values are measured in triplicates. The data show in the Table 4-7 has standard deviations calculated as well.

TSS and VSS reduction percentages for MBR samples are shown in the Table 4-7. Reduction rates of TSS was 13.3%, 13.5% and 13.7%, respectively for MBR, MBR127/60 and MBR90/30 samples. Additionally, reduction rates of VSS was 41.4%, 41.6% and 39.2%, respectively for MBR, MBR127/60 and MBR90/30 samples. The reductions of TSS and VSS increases with the pretreatment application.

Last parameter (pH) has almost similar values for each reactor, both at the reactor start-up and reactor take-down.

Table 4-7. *TSS and VSS amounts before and after anaerobic digestion and removal percents for MBR WAS sample*

Sample	**TSSi (mg/L)**	**TSSf (mg/L)**	**TSS Removal (%)**	**VSSi (mg/L)**	**VSSf (mg/L)**	**VSS Removal (%)**	**pHi**	**pHf**
MBR	20,644±888.1	17,883±394.1	13.3	14,427±366.4	8,433±581.5	41.4	8.14	7.28
MBR127/60	21,122±573.4	18,266±272	13.5	15,261±382.7	8,911±716.7	41.6	7.57	7.34
MBR90/30	20,844±619	17,988±265.2	13.7	14,794±219.2	8,977±502.1	39.2	7.52	7.27

TS and VS reduction percentages for OID samples are shown in the Table 4-8. Reduction rates of TS was 19.0%, 19.0% and 18.2%, respectively for OID, OID127/60 and OID90/30 samples. Additionally, reduction rates of VS were 27.7%, 29.4% and 28.2%, respectively for OID, OID127/60 and OID90/30 samples. This is as expected and parallel to the results obtained for other sludges. Since with pretreatment COD is solubilized, it is converted to methane and cause an increased reduction of TS an VS.

Table 4-8. *TS and VS amount and removal rate both of them before and after anaerobic digestion for OID sample*

Sample	**TSi (mg/L)**	**TSf (mg/L)**	**TS Removal (%)**	**VSi (mg/L)**	**VSf (mg/L)**	**VS Removal (%)**
OID	24,222±459.1	19,611±191.8	19.0	16,800±857.2	12,138±767.2	27.7
OID127/60	23,611±639.4	19,111±943.2	19.0	16,705±554.1	11,794±215.6	29.4
OID90/30	23,783±485.5	19,450±612.2	18.2	15,611±759	11,205±473.3	28.2

In addition to TS and VS, TSS, VSS and pH are also measured in the reactors Table 4-9. The TSS, VSS and pH values are measured in triplicates. The data in show the Table 4-9 standard deviations calculated as well.

TSS and VSS reduction percentages for OID samples are shown in the Table 4-9. Reduction rates of TSS was 16.7%, 16.5% and 16% respectively OID, OID127/60 and OID90/30 samples. Additionally, reduction rates of VSS was 20.2%, 22.1% and 21.9% respectively OID, OID127/60 and OID90/30 samples. Again similar increases in reduction of VSS can be seen due to pretreatment. Last parameter (pH) has almost similar values for each reactor, both at the reactor start-up and reactor take-down.

Table 4-9. *Removal rate of TSS and VSS amount and amount both of them before and after anaerobic digestion for OID WAS sample*

Sample	**TSSi (mg/L)**	**TSSf (mg/L)**	**TSS Removal (%)**	**VSSi (mg/L)**	**VSSf (mg/L)**	**VSS Removal (%)**	**pHi**	**pHf**
OID	20,800±685.3	15,300±326.8	16.7	14,927±506.1	10,150±781.8	20.2	7.9	7.3
OID127/60	21,100±448.3	15,111±961.2	16.5	15,555±714	9,844±430	22.1	7	7.3
OID90/30	20,433±858.8	15,055±375.1	16	14,838±625.2	9,144±451.8	21.9	7.4	7.3

4.4.5. Soluble Chemical Oxygen Demand Concentration for Reactors Before and After Anaerobic Digestion

When anaerobic digestion was terminated, soluble COD concentrations were measured for each BMP assay. To determine the reductions in soluble COD in the bottles during anaerobic digestion their concentrations were investigated before reactor set-up and after reactor termination.

In Table 4-10, soluble COD reduction percentages for Tatlar samples are shown. For Tatlar seed sample removal of soluble COD was 8.6%. Removal efficiency of soluble COD was 19,9%, 47,9% and 29,5%, respectively for T, T127/60 and T90/30 samples. As can be seen pretreatment at higher temperature and time enhances COD reduction greatly. On the other hand, intermediate temperatures and times cause intermediate level of COD reduction.

Table 4-10. *Concentration of soluble COD and removals during anaerobic digestion for Tatlar sample*

Sample	**Initial SCOD (mg/L)**	**Final SCOD (mg/L)**	**SCOD Removal (%)**
T	2,378±210.9	1,904±84.9	19.9
T127/60	3,678±240.1	1,917±111.2	47.9
T90/30	3,219±467.3	2,270±137.5	29.5

In Table 4-11, soluble COD reduction percentages for MBR samples were shown. For Removal efficiency of soluble COD was 38.4%, 51.6% and 44.5% respectively MBR, MBR127/60 and MBR90/30 samples. As can be seen pretreatment at higher temperature and time enhances COD reduction greatly. On the other hand, intermediate temperatures and times cause intermediate level of COD reduction.

Table 4-11. *Concentration of soluble COD and removals during anaerobic digestion for MBR sample*

Sample	**Initial SCOD (mg/L)**	**Final SCOD (mg/L)**	**SCOD Removal (%)**
MBR	2,355±203.2	1,155±110.4	38.4
MBR127/60	3,456±256.8	2,279±172.1	51.6
MBR90/30	3,230±297.1	2,060±103.4	44.5

In Table 4-12, soluble COD reduction percentages for OID samples were shown. For Removal efficiency of soluble COD was 27.3%, 58.9% and 47.6% respectively OID, OID127/60 and OID90/30 samples. As can be seen pretreatment at higher temperature and time enhances COD reduction greatly. On the other hand, intermediate temperatures and times cause intermediate level of COD reduction.

Table 4-12. *Concentration of soluble COD and removals during anaerobic digestion for OID sample*

Sample	**Initial SCOD (mg/L)**	**Final SCOD (mg/L)**	**SCOD Removal (%)**
OID	2,185±190.4	1,588±98.3	27.3
OID127/60	5,768±130.1	2,370±108.3	58.9
OID90/30	2,934±69.4	1,538±259.6	47.6

4.4.6. Methane Produced with respect to Added VS in Anaerobic Digestion

Methane yield normalized with the amount of added VS is shown in Figure 4-32. Reactor S demonstrate only domestic digester sludge. On the other hand, reactor OID consist of organized industrial district sludge. Reactors T127/60, OID127/60 and MBR127/60 had the highest methane yield, respectively. The methane yield of reactor S, T, MBR, OID, T127/60, MBR127/60, OID127/60, T90/30, MBR90/30, OID90/30 were 0.02 L/g, 0.1 L/g, 0.13 L/g, 0.09 L/g, 0.22 L/g, 0.18 L/g, 0.21 L/g, 0.16 L/g, 0.15 L/g, and 0.20 L/g, respectively. As expected by the results of preliminary study, methane yields increased for pretreated samples with respect to non-pretreated samples.

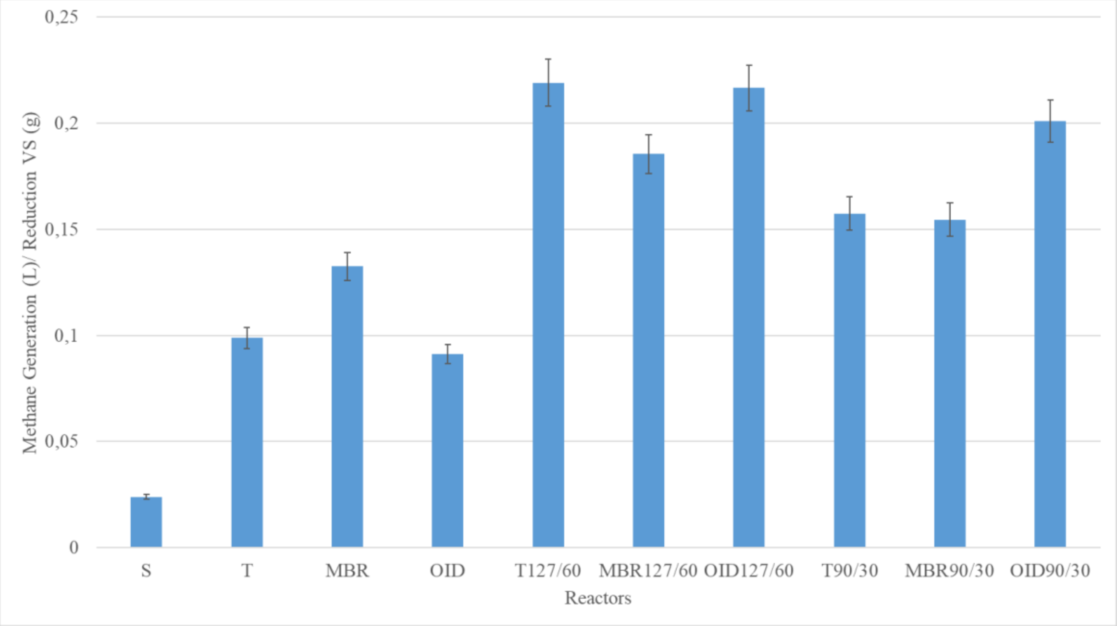


Figure 4-32. Methane yield with respect to added VS in BMP assays

4.4.7. Capillary Suction Time for Reactors After Anaerobic Digestion

When anaerobic digestion was terminated, CST was measured for each BMP bottle at reactor take-down. These values were compared to the ones measured after pretreatment but before anaerobic digestion started for the same reactors. In Table 4-13, all these changes for all reactors are given. When compared pretreated and non-pretreated samples, CST affected negatively with respect to pretreatment for all types of sludge.

It is very clear that CST decreased after BMP assays. For Tatlar WWTP sample, CST of pretreated samples were lower than the CST of non-pretreated samples. On the other hand, for MBR and OID, CST of pretreated samples were higher than the CST of non-pretreated samples. This condition possibly caused by particle size. Municipal WWTP sludges are more fragile than the MBR and OID sludges.

Table 4-13. *Change of CST for each sample*

Sample	CST before pretreatment (sec)	Initial CST (sec)	Final CST (sec)
S	-	228.6±19.5	213.4±9.7
T	835.6±61.4	542.1±21.1	409.3±17.4
T127/60	-	886.6±73.6	389.5±16.0
T90/30	-	1068±534.7	336.1±23.2
MBR	28.1±1.9	109.9±8.2	93.4±9.6
MBR127/60	-	650.7±13.2	360.7±21.4
MBR90/30	-	374.2±5.9	168.5±5.8
OID	15.5±0.4	87.9±5.0	73.1±2.2
OID127/60	-	529.4±32.7	283.6±21.4
OID90/30	-	234.1±5.0	136.3±8.2

4.4.8. Performance of the Three Different Sludges

In preliminary experiments, for three different types of sludge, pretreatment for 60 minutes at 127◦C gave the best results in terms of soluble COD. Solubilisation for T, MBR and OID were 8.97%, 3.05% and 0.99%, respectively. On the other hand, solubilisation for T127/60, MBR127/60 and OID127/60 were 23.66%, 34.18% and 19.41%, respectively. Additionally, solubilisation for T90/30, MBR90/30 and OID90/30 were 14.03%, 13.80% and 6.26%, respectively. As can be seen above, thermal hydrolysis improved of solubilisation in terms of soluble COD. Therefore, methane generation from sludge pretreated 60 minutes at 127◦C and sludge pretreated 30 minutes at 90◦C were expected higher than the non-pretreated samples. In addition to this, methane generation of sludge pretreated 60 minutes at 127◦C should be higher than sludge pretreated 30 minutes at 90◦C. In Table 4-14, initial and final soluble COD of BMP assays are shown.

Table 4-14. *Initial and final soluble COD amount for BMP assays*

Parameter	Sample Code	Measurement
Initial soluble COD (mg/L)	S	1,774
	T	2,378
	MBR	1,874
	OID	2,185
	T127/60	3,678
	MBR127/60	4,328
	OID127/60	5,768
	T90/30	3,219
	MBR90/30	2,725
	OID90/30	2,934
Final soluble COD (mg/L)	S	1,621
	T	1,904
	MBR	1,155
	OID	1,588
	T127/60	1,917
	MBR127/60	2,095
	OID127/60	2,370
	T90/30	2,270
	MBR90/30	1,511
	OID90/30	1,537

When initial and final COD of BMP assays were compared with respect to preliminary experiments, expected results have been observed. Sludge pretreated 60 minutes at 127◦C has highest soluble COD initially and sludge pretreated 30 minutes at 90◦C follows this sample. Additionally, non-pretreated samples have the lowest soluble COD amount as expected.

In Table 4-15 removal percentages of TS, VS and COD and percentage of methane with respect to cumulative biogas for three types of sludge are shown.

Table 4-15. *Removals after BMP assays*

Parameter	**Sample Code**	**Measurement**
TS Removal (%)	S	6.3
	T	16.4
	MBR	14.4
	OID	19.0
	T127/60	17.4
	MBR127/60	14.7
	OID127/60	19.0
	T90/30	17.5
	MBR90/30	16.4
	OID90/30	18.2
VS Removal (%)	S	25.2
	T	46.8
	MBR	42.3
	OID	27.7
	T127/60	48.0
	MBR127/60	44.8
	OID127/60	29.4
	T90/30	47.4
	MBR90/30	42.5
	OID90/30	28.2
Soluble COD Removal (%)	S	8.6
	T	19.9
	MBR	38.4
	OID	27.3
	T127/60	47.9
	MBR127/60	51.6
	OID127/60	58.9
	T90/30	29.5
	MBR90/30	44.5
	OID90/30	47.6

When performances of different samples are compared, removal of TS were similar for all three types of sludge. On the other hand, reduction of VS for OID is lower than the reduction of VS for Tatlar and MBR samples.

Table 4-16. *Produced methane amount for BMP assays*

Sample Code	BMP Methane Volume (mL)
S	8.1
T	159.8
MBR	195.2
OID	84.9
T127/60	359.6
MBR127/60	292.0
OID127/60	212.7
T90/30	251.7
MBR90/30	219.7
OID90/30	177.0

OID samples, containing industrial sludge, has less removal rates and methane percent than other samples. COD removal percentage reactor is the highest in the sludge samples pretreated 60 minutes at 127◦C, as expected. For methane generation, pretreated 60 minutes at 127◦C samples has higher amount than the others which is expected result with respect to both preliminary experiments and initial characterization.

When all the results are evaluated, it is seen that municipal sludge pretreated 60 minutes at 127◦C has the highest methane production potential and it is followed by MBR127/60, T90/30 and MBR90/30 samples, respectively. On the other hand, methane production potential of OID sludge is the lowest due to toxic characteristics of the OID sludge.

CHAPTER 5

CONCLUSION

The main objective of this study was to enhance biogas production of biological sludge by thermal hydrolysis pretreatment. According to COD/BOD ratio, biodegradable fraction of wastewaters was determined. In Tatlar and METU MBR WWTP influent, the biodegradable fraction of wastewater was high; therefore, biological treatment and anaerobic digestion processes were applicable. As expected, high anaerobic digestion performance for Tatlar and MBR sludges were observed, in BMP tests. On the other hand, the non-biodegradable fraction of OID wastewater was high. Thus physical and chemical treatment processes were applied in WWTP. In addition, if anaerobic digestion system were present in OID, it would not be as effective as the anaerobic systems in Tatlar and MBR. On the other hand, Tatlar WWTP sludge sample was typical domestic sludge which has higher anaerobic digestion performance. Due to longer solid retention time with respect to typical domestic WWTP, MBR sludge had less initial stability with respect to Tatlar WWTP. Therefore, performance of methane generation was slightly lower that Tatlar WWTP. On the other hand, OID sludge possibly contained toxic compounds due to containing various sectoral wastes in it. Thus, methane generation was lower than the others. A performance comparison for biogas and methane generation from different types of sludge which were thermally pretreated at different temperatures and durations, was also conducted. Initially different temperatures and exposure time were used for thermal hydrolysis then these results were used to determine the optimal thermal pretreatment conditions for the sludge samples. Three different types of sludge collected from Tatlar Wastewater Treatment Plant, Middle East Technical University MBR wastewater treatment plant, and OID Treatment Plant.

Temperature and time screening results showed that thermal hydrolysis application at 127^{o}C for 60 minutes gave the highest increase in soluble and total COD values for all sludge types. At 127^{o}C for 60 minutes thermal hydrolysis setup soluble COD concentration of domestic WAS increased up to 264% and solubilisation was improved by 24%. Soluble COD concentration of MBR sample was increased up 9% and solubilisation was improved by 34%. Soluble COD concentration of the industrial sludge was increased by 2%, and the solubility by 19% respectively. The results of the research showed that soluble COD improvement was the highest for domestic sludge and lowest for the industrial type sludge.

After BMP assay set-up with pretreated and non-pretreated sludge samples reduction in TS, TSS, VS, VSS, CST and soluble COD values was observed. For best condition (pretreated 60 minutes at 127^{o}C), TS removal for T127/60, MBR127/60 and OID127/60 were 17.4%, 14.7% and 19%; TSS removal for T127/60, MBR127/60 and OID127/60 were 15.8%, 13.5% and 16.5%, respectively. VS removal for T127/60, MBR127/60 and OID127/60 were 48%, 44.8% and 29.4%, respectively. VSS removal for T127/60, MBR127/60 and OID127/60 were 46.2%, 41.6% and 22.1%, respectively. Thermal hydrolysis pretreatment before anaerobic digestion improved also biogas production. For non-pretreated Tatlar samples, generated biogas amount was 316.86 mL; whereas generated biogas amount of T127/60 and T90/30 were 609 mL and 438.43 mL, respectively. For non-pretreated MBR samples, generated biogas amount was 365.04 mL; compared to the generated biogas amount of MBR127/60 and MBR90/30 were 498.01 mL and 389.43 mL, respectively. For non-pretreated OID samples, generated biogas amount was 170.59 mL; whereas generated biogas amount of OID127/60 and OID90/30 were 368.31 mL and 324.10 mL, respectively.

When normalized methane generation (mL CH4/gr VS reduction) is examined, it is clearly seen that thermal hydrolysis has positive impact on all sludges under all pretreatment conditions. For Tatlar sludge the improvement of normalized methane production was 60% and 120% for T90/30 and T127/60 pretreatment conditions, respectively compared to untreated sludge. On the other hand, this improvement was 19% and 38% for MBR sludge. Finally, normalized methane production improved by 135% and 155% for OID sludge pretreated under 90/30 and 127/60 pretreatment conditions, respectively compared to un-pretreated sludge. These results showed that thermal hydrolysis has a significant impact on all sludges, even on the difficult to degrade OID sludge. Therefore, thermal hydrolysis can be used to enhance the reactor performance after determination of optimum hydrolysis temperature and time for different kinds of sludges.

Thermal hydrolysis applied to three different sludge types did not change the pH values significantly before and after anaerobic digestion. Thermal hydrolysis changed the dewaterability of sludge. Sludge samples pretreated at 127^{o}C for 60 minutes gave the highest CST, for MBR and industrial sludge.

CHAPTER 6

RECOMMENDATIONS FOR FUTURE WORK

In this thesis thermal hydrolysis is applied as a pretreatment process to three selected sludge types to enhance the biogas production. During the study, temperature and exposure time was selected as the parameters to control the effectiveness of the thermal hydrolysis on sludge minimization and biogas production.

Our results show that sludge type is a determining parameter on the biogas and methane production after thermal hydrolysis. Therefore, a study with different types of sludge is recommended.

Pressure may have an impact on the performance of thermal hydrolysis of sludge therefore, effect of pressure used for thermal hydrolysis application and its impact on biogas production must investigated in future studies.

In this study, the C/N ratio was not examined. If the carbon amount is less and the nitrogen amount is high, it can cause a toxic effect on sludge. Therefore, the C/N ratio to optimize biogas production after thermal hydrolysis can be investigated.

After thermal pretreatment, pathogenic microorganism amount in digested sludge was not analyzed in this study. After conducting thermal hydrolysis sterilization of sludge was anticipated. Therefore, pathogenic activity in the sludge types after thermal hydrolysis can be investigated in further studies. If pathogenic activity meets the standards, sludge after thermal hydrolysis possibly utilized for land application, which will extinguish the disposal and transportation expense of digested sludge into landfills.

REFERENCES

Abe, N., Tang, Y., Iwamura, M., Morimura, S., & Kida, K. (2013). Pretreatment followed by anaerobic digestion of secondary sludge for reduction of sewage sludge volume. Water Science and Technology, 67(11), 2527-2533. doi: 10.2166/wst.2013.154

Abelleira-Pereira, J., Pérez-Elvira, S., Sánchez-Oneto, J., de la Cruz, R., Portela, J., & Nebot, E. (2015). Enhancement of methane production in mesophilic anaerobic digestion of secondary sewage sludge by advanced thermal hydrolysis pretreatment. Water Research, 71, 330-340. doi: 10.1016/j.watres.2014.12.027

Abiero, Z., & Owili, D. (2016). Suitability of Sewage Sludge Ash as A Filler Material In Asphalt Concrete. Journal Of Multidisciplinary Engineering Science Studies, 2(3), 322-331. Retrieved from http://www.jmess.org/wp-content/uploads/2016/03/JMESSP13420062.pdf

Albright, W., Benson, C., Gee, G., Abichou, T., McDonald, E., Tyler, S., & Rock, S. (2006). Field Performance of a Compacted Clay Landfill Final Cover at a Humid Site. Journal of Geotechnical And Geoenvironmental Engineering, 132(11), 1393-1403. doi: 10.1061/(asce)1090-0241(2006)132:11(1393)

APHA, AWWA, & WEF. (2005). Standard Methods for the Examination of Water and Wastewater (21st ed.). Washington, DC.: American Public Health Association, American Water Works Association, Water Environment Federation.

Apul, O.G., & Sanin, F. D. (2010). Examination of sludge minimization potential and associated costs by ultrasonic pretreatment. Katı Atık ve Çevre.

Ariunbaatar, J., Panico, A., Esposito, G., Pirozzi, F., & Lens, P. (2014). Pretreatment methods to enhance anaerobic digestion of organic solid waste. Applied Energy, 123, 143-156. doi: 10.1016/j.apenergy.2014.02.035

Aski Genel Müdürlüğü. Ankara Merkezi Atıksu Arıtma Tesisi. Retrieved June 12, 2019, from http://www.aski.gov.tr/TR/ICERIKDETAY/Tatlar-Atiksu-Aritma-Tesisi/30/22

Aslan, Y. (2015). Membran Biyoreaktör (MBR) Atıksu Arıtma Sistemleri. Çevre Portal. Retrieved June 12, 2019, from https://www.cevreportal.com/membran-biyoreaktor-mbr-atiksu-aritma-sistemi/

Barber, W. (2016). Thermal hydrolysis for sewage treatment: A critical review. Water Research, 104, 53-71. doi: 10.1016/j.watres.2016.07.069

Barjenbruch, M., & Kopplow, O. (2003). Enzymatic, mechanical and thermal pre-treatment of surplus sludge. Advances in Environmental Research, 7(3), 715-720. doi: 10.1016/s1093-0191(02)00032-1

Baysal Arıtma ve Mühendislik (2017). İleri Arıtma-MBR(Membran Biyoreaktör) Sistemleri. Retrieved June 12, 2019, from http://www.baysalaritma.com/faaliyet-alani/ileri-aritma-mbr-membran-biyoreaktor-sistemleri

Bjerg, P., Albrechtsen, H., Kjeldsen, P., Christensen, T., & Cozzarelli, I. (2003). The Groundwater Geochemistry of Waste Disposal Facilities. Treatise On Geochemistry, 9, 579-612. doi: 10.1016/b0-08-043751-6/09057-5

Bolzonella, D., Pavan, P., Battistoni, P., & Cecchi, F. (2005). Mesophilic anaerobic digestion of waste activated sludge: influence of the solid retention time in the wastewater treatment process. Process Biochemistry, 40(3-4), 1453-1460. doi: 10.1016/j.procbio.2004.06.036

Bougrier, C., Battimelli, A., Delgenes, J., & Carrere, H. (2007). Combined Ozone Pretreatment and Anaerobic Digestion for the Reduction of Biological Sludge Production in Wastewater Treatment. Ozone: Science & Engineering, 29(3), 201-206. doi: 10.1080/01919510701296754

Bougrier, C., Delgenès, J., & Carrère, H. (2006). Combination of Thermal Treatments and Anaerobic Digestion to Reduce Sewage Sludge Quantity and

Improve Biogas Yield. Process Safety and Environmental Protection, 84(4), 280-284. doi: 10.1205/psep.05162

Bougrier, C., Delgenès, J., & Carrère, H. (2007). Impacts of thermal pre-treatments on the semi-continuous anaerobic digestion of waste activated sludge. Biochemical Engineering Journal, 34(1), 20-27. doi: 10.1016/j.bej.2006.11.013

Carrère, H., Bougrier, C., Castets, D., & Delgenès, J. (2008). Impact of initial biodegradability on sludge anaerobic digestion enhancement by thermal pretreatment. Journal of Environmental Science and Health, Part A, 43(13), 1551-1555. doi: 10.1080/10934520802293735

Carrère, H., Dumas, C., Battimelli, A., Batstone, D., Delgenès, J., Steyer, J., & Ferrer, I. (2010). Pretreatment methods to improve sludge anaerobic degradability: A review. Journal of Hazardous Materials, 183(1-3), 1-15. doi: 10.1016/j.jhazmat.2010.06.129

Cesaro, A., & Belgiorno, V. (2014). ChemInform Abstract: Pretreatment Methods to Improve Anaerobic Biodegradability of Organic Municipal Solid Waste Fractions. Cheminform, 240(15), 24–37. doi: 10.1002/chin.201415287

Chen, L., Neibling, H. (2014). Anaerobic Digestion Basics. University of Idaho Extension, 1-6 p.

Chu, L., Yan, S., Xing, X., Sun, X., & Jurcik, B. (2009). Progress and perspectives of sludge ozonation as a powerful pretreatment method for minimization of excess sludge production. Water Research, 43(7), 1811-1822. doi: 10.1016/j.watres.2009.02.012

Da Silva, C., Astals, S., Peces, M., Campos, J., & Guerrero, L. (2018). Biochemical methane potential (BMP) tests: Reducing test time by early parameter estimation. Waste Management, 71, 19-24. doi: 10.1016/j.wasman.2017.10.009

de Figueirêdo Lopes Lucena, L., Thomé Juca, J., Soares, J., & Portela, M. (2014). Potential Uses of Sewage Sludge in Highway Construction. Journal of Materials In Civil Engineering, 26(9), 04014051. doi: 10.1061/(asce)mt.1943-5533.0000937

Doğan, I., & Sanin, F. (2009). Alkaline solubilization and microwave irradiation as a combined sludge disintegration and minimization method. Water Research, 43(8), 2139-2148. doi: 10.1016/j.watres.2009.02.023

Elliott, A., & Mahmood, T. (2007). Pretreatment technologies for advancing anaerobic digestion of pulp and paper biotreatment residues. Water Research, 41(19), 4273-4286. doi: 10.1016/j.watres.2007.06.017

EPA. (1994). Land Application of Sewage Sludge; A Guide for Land Appliers on the Requirements of the Federal Standards for the Use or Disposal of Sewage Sludge, 40 CFR Part 503, 2-3 p.

Eshtiaghi, N., Markis, F., Yap, S., Baudez, J., & Slatter, P. (2013). Rheological characterisation of municipal sludge: A review. Water Research, 47(15), 5493-5510. doi: 10.1016/j.watres.2013.07.001

Eskişehir Organize Sanayi Bölgesi. Tarihçe. Retrieved June 12, 2019, from http://www.eosb.org.tr/hakkimizda/

Eurostat. (n.d.). Sewage sludge production and disposal from urban wastewater (in dry substance (d.s)) Retrieved April 15, 2019, from https://ec.europa.eu/eurostat/tgm/refreshTableAction.do?tab=table&plugin=1&pcode=ten00030&language=en

Everett, J.G. (1972). Dewatering of wastewater sludge by heat treatment. J. Water Pollution Control Federation, 44(1), 92-100.

Farno, E., Baudez, J., Parthasarathy, R., & Eshtiaghi, N. (2014). Rheological characterisation of thermally-treated anaerobic digested sludge: Impact of temperature and thermal history. Water Research, 56, 156-161. doi: 10.1016/j.watres.2014.02.048

Farno, E., Baudez, J., Parthasarathy, R., & Eshtiaghi, N. (2015). Impact of temperature and duration of thermal treatment on different concentrations of anaerobic digested sludge: Kinetic similarity of organic matter solubilisation and sludge rheology. Chemical Engineering Journal, 273, 534-542. doi: 10.1016/j.cej.2015.03.097

Fytili, D., Zabaniotou, A. (2008). Utilization of sewage sludge in EU application of old and new methods—A review. Renewable and Sustainable Energy Reviews, 12(1), 116-140.

Gavala, H., Yenal, U., Skiadas, I., Westermann, P., & Ahring, B. (2003). Mesophilic and thermophilic anaerobic digestion of primary and secondary sludge. Effect of pre-treatment at elevated temperature. Water Research, 37(19), 4561-4572. doi: 10.1016/s0043-1354(03)00401-9

Goel, R., Tokutomi, T., Yasui, H., & Noike, T. (2003). Optimal process configuration for anaerobic digestion with ozonation. Water Science and Technology, 48(4), 85-96. doi: 10.2166/wst.2003.0228

González-Fernández, C., & García-Encina, P. (2009). Impact of substrate to inoculum ratio in anaerobic digestion of swine slurry. Biomass and Bioenergy, 33(8), 1065-1069. doi: 10.1016/j.biombioe.2009.03.008

Gurjar, B.R., Tyagi, V.K. (2017). Sludge Management, CFC Press, 5-27 p.

Haug, R., Stuckey, D., Gossett, J., & McCarty, P. (1978). Effect of Thermal Pretreatment on Digestibility and Dewaterability of Organic Sludges. Journal (Water Pollution Control Federation),50(1), 73-85. Retrieved from http://www.jstor.org/stable/25039508

Higgins, M., Beightol, S., Mandahar, U., Suzuki, R., Xiao, S., & Lu, H. et al. (2017). Pretreatment of a primary and secondary sludge blend at different thermal hydrolysis temperatures: Impacts on anaerobic digestion, dewatering and filtrate characteristics. Water Research, 122, 557-569. doi: 10.1016/j.watres.2017.06.016

Hiraoka, M., Takeda, N., Sakai, S., & Yasuda, A. (1985). Highly Efficient Anaerobic Digestion with Thermal Pretreatment. Water Science and Technology, 17(4-5), 529-539. doi: 10.2166/wst.1985.0157

HMC Europe (2003). Operating Manual, 16 p.

Huang, X., Dong, W., Wang, H., & Feng, Y. (2018). Role of acid/alkali-treatment in primary sludge anaerobic fermentation: Insights into microbial community structure, functional shifts and metabolic output by high-throughput sequencing. Bioresource Technology, 249, 943-952. doi: 10.1016/j.biortech.2017.10.104

Jiang, D., Ni, G., & Ma, G. (2010). Reuse of Municipal Wastewater Sludge for Construction Material. Advanced Materials Research, 156-157, 939-942. doi: 10.4028/www.scientific.net/amr.156-157.939

Khiari, B., Marias, F., Zagrouba, F., & Vaxelaire, J. (2004). Analytical study of the pyrolysis process in a wastewater treatment pilot station. Desalination, 167, 39-47. doi: 10.1016/j.desal.2004.06.111

Kim, J., Park, C., Kim, T., Lee, M., Kim, S., Kim, S., & Lee, J. (2003). Effects of various pretreatments for enhanced anaerobic digestion with waste activated sludge. Journal of Bioscience And Bioengineering, 95(3), 271-275. doi: 10.1016/s1389-1723(03)80028-2

Kor-Bicakci, G., & Eskicioglu, C. (2019). Recent developments on thermal municipal sludge pretreatment technologies for enhanced anaerobic digestion. Renewable and Sustainable Energy Reviews, 110, 423-443. doi: 10.1016/j.rser.2019.05.002

Köksoy, G. T., & Sanin, F. D. (2010). Effect of digester F/M ratio on gas production and sludge minimization of ultrasonically treated sludge. Water Science and Technology, 62(7), 1510-1517.https://doi.org/10.2166/wst.2010.447

Kuan-Yeow, S., Joo-Hwa, T. and Duu-Jong, L. (2018). Sustainable Sludge Management:Resource Recovery for Construction Applications.

Lamb, D., Heading, S., Bolan, N., & Naidu, R. (2012). Use of Biosolids for Phytocapping of Landfill Soil. Water, Air, & Soil Pollution, 223(5), 2695-2705. doi: 10.1007/s11270-011-1060-x

Leitão, R., van Haandel, A., Zeeman, G., & Lettinga, G. (2006). The effects of operational and environmental variations on anaerobic wastewater treatment systems: A review. Bioresource Technology, 97(9), 1105-1118. doi: 10.1016/j.biortech.2004.12.007

Liao, X., Li, H., Zhang, Y., Liu, C., & Chen, Q. (2016). Accelerated high-solids anaerobic digestion of sewage sludge using low-temperature thermal pretreatment. International Biodeterioration & Biodegradation, 106, 141-149. doi: 10.1016/j.ibiod.2015.10.023

Liew, A., Idris, A., Samad, A., Wong, C., Jaafar, M., & Baki, A. (2004). Reusability of sewage sludge in clay bricks. Journal of Material Cycles and Waste Management, 6(1), 41-47. doi: 10.1007/s10163-003-0105-7

Liu, X., Wang, W., Gao, X., Zhou, Y., Shen, R. (2012). Effect of thermal pretreatment on the physical and chemical properties of municipal biomass waste. Waste Management, 32(2) 249-255. doi:10.1016/j.wasman.2011.09.027

Lui, D., Liu, D., Zeng, R. J., & Angelidaki, I. (2006). Hydrogen and methane production from household solid waste in the two-stage fermentation process. Water Research, 40(11), 2230-2236. https://doi.org/10.1016/j.watres.2006.03.029

Lundin, M., Olofsson, M., Pettersson, G., & Zetterlund, H. (2004). Environmental and economic assessment of sewage sludge handling options. Resources, Conservation and Recycling, 41(4), 255-278. doi: 10.1016/j.resconrec.2003.10.006

Malerius, O., Werther, J. (2003). Modelling the adsorption of mercury in the flue gas of sewage sludge incineration. Chemical Engineering Journal, 91 (1-3), 197-205. https://doi.org/10.1016/j.cej.2003.08.018

Menendez, J.A., Inguanzo, M., Pis J.J. (2002) Microwave-induced pyrolysis of sewage sludge. Water Research, 36(13), 3261-3264. doi:10.1016/S0043-1354(02)00017-9

Mihelcic, J.R., Zimmerman, J.B. (2010) Environmental Engineering Fundamentals, Sustainability, Design. John Wiley & Sons, Inc.

Mills, N. (2015). Unlocking the Full Energy Potential of Sewage Sludge. 1-37 p.

Minimi, G., Di Bartolo Zuccarello, R., Lotito, V., Spinosa, L., Di Pinto, A.C. (1997). A design model of sewage sludge incineration plants with energy recovery. Water Science & Technology, 36(11), 211-218. https://doi.org/10.1016/S0273-1223(97)00685-9

Ministry of Environment and Urbanization. (2013). Municipal/Urban Treatment Sludge Management Project, 1514-1522 p.

Morosini, C., Conti, F., Torretta, V., Rada, E. C., Passamani, G., Schiavon, M., Cioca L. I., Ragazzi, M. (2016). Biochemical methane potential assays to test the biogas production from the anaerobic digestion of sewage sludge and other organic matrices. WIT Transactions on Ecology and The Environment, 205, 235-243. doi: 10.2495/EQ160221

Müller, J. (2001). Prospects and problems of sludge pre-treatment processes. Water Science and Technology, 44(10), 121-128. doi: 10.2166/wst.2001.0598

Müller, J.A., Winter, A. and Stünkmann, G. (2004). Investigation and Assessment of Sludge Pre-treatment Processes. Water Science Technology, 49 (10), 97-104. doi: https://doi.org/10.2166/wst.2004.0618

Oosterhuis, M., Ringoot, D., Hendriks, A., Roeleveld, P. (2014). Thermal hydrolysis of waste activated sludge at Hengelo Wastewater Treatment Plant, The Netherlands. Water Science & Technology, 70 (1), 1-7. https://doi.org/10.2166/wst.2014.107

Orta Doğu Teknik Üniversitesi. (2013). ODTÜ/ODTÜ Teknokent Membran Arıtma Tesisi Rio +20'de Türkiye'yi Temsil Edecek! Retrieved June 12, 2019, from ODTÜ/ODTÜ Teknokent Membran Arıtma Tesisi Rio +20'de Türkiye'yi Temsil Edecek!. (2019). Retrieved from https://www.metu.edu.tr/tr/duyuru/odtuodtu-teknokent-membran-aritma-tesisi-rio-20de-turkiyeyi-temsil-edecek

Perez-Elvire,S.I., Nieto Diez, P., Fdz-Polanco, F. (2006). Sludge Minimisation Technologies. Environmental Science and Biotechnology, 5(4), 375-398. https://doi.org/10.1007/s11157-005-5728-9

Phothilangka, P., Schoen, M.A., Wett, B. (2008). Benefits and drawbacks of thermal pre hydrolysis for operational performance of wastewater treatment plants. Water Science and Technology, 58(8), 1547-1553

Pontoni, L., Dalessandro, G., Dantonio, G., Fabbricino, M., Piozzi, F., Esposito., G. (2015). Effect of Anaerobic Digestion on Rheological Parameters and Dewaterability of Aerobic Sludges from MBR and Conventional Activated Sludge Plants. Chemical Engineering Transactions, 43, 2311-2316. https://doi.org/10.3303/CET1543386

Pontoni, L., Papirio, S., Dalessandro, G., Caniani, D., Gori, R., Mannina, G., Capodici, M., Nicosia, S., Fabbricino, M., Piozzi, F., Esposito., G. (2018). Dewaterability of CAS and MBR Sludge: Effect of Biological Stability and EPS Composition. Journal of Environmental Engineering, 144(1), 1-9. doi: 10.1061/(ASCE)EE.1943-7870.0001299.

Sanin, F.D., Clarkson, W.W., Vesilind, P.A., (2011). Sludge Engineering: The Treatment and Disposal of Wastewater Sludges.

Sapkaite, I., Barrado, E., Fdz-Polanco, F., Perez-Elvira, S.I. (2017). Optimization of a thermal hydrolysis process for sludge pre-treatment. Journal of Environmental Management, 192, 25-30. https://doi.org/10.1016/j.jenvman.2017.01.043

Sato, Y., Oyamada, T., & Hanehara, S. (n.d.). Applicability of sewage sludge ash (SSA) for paving materials: A study on using SSA as filler for asphalt mixture and base course material. Third International Conference on Sustainable Construction Materials and Technologies. Retrieved April 15, 2019, from http://www.claisse.info/2013%20papers/data/e283.pdf

Seshadri, B., Bolan, N.S., Thangarajan, R., Jena, U., Das, K.C., Wang, H., Naidu, R. (2016). Chapter 5- Biomass Energy from Revegation of Landfill Sites. Bioremediation and Bioeconomy. doi: 10.1016/B978-0-12-802830-8.00005-8

Singh, R.P., Agrawal, M. (2008). Potential benefits and risks of land application of sewage sludge. Waste Management, 28(2), 347-358. https://doi.org/10.1016/j.wasman.2006.12.010

Sperling, M. (2017). Biological Wastewater Treatment Series Volume One: Wastewater Characteristics, Treatment and Disposal, IWA Publishing, 40 p.

Stuckey, D.C., McCarty, P.L. (1984). The effect of thermal pretreatment on the anaerobic biodegradability and toxicity of waste activated sludge. Water Research, 18(11), 1343-1353. https://doi.org/10.1016/0043-1354(84)90002-2

Su ve Çevre. (2007). Odtü Yeni Biyomembran Arıtma Tesisi. Retrieved June 12, 2019, from http://www.suvecevre.com/yayin/531/odtu-yeni-biyomembran-aritma tesisi_15912.html#.XPQMlIgzaUm

Şahinkaya, S. (2018). Termokimyasal Ön İşlem İle Atık Aktif Çamurun Parçalanması. Uludağ Üniversitesi Mühendislik Fakültesi Dergisi. 23 (2). doi: 10.17482/uumfd.319836

Takashima, M. (2008). Examination on process configurations incorporating thermal treatment for anaerobic digestion of sewage sludge. Journal of Environmental Engineering, 134(7). doi:10.1061/(ASCE)0733-9372(2008)134:7(543)

Tanaka, S., Kobayashi, T., Kamiyama, K, ve Bildan, M.L. (1997) Effects of thermochemical pre-treatment on anaerobic digestion of waste activated sludge, Water Science Technology, 35(8), 209-215. https://doi.org/10.2166/wst.1997.0315

Tchobanoglous, G., Stensel, H., Tsuchihashi, R., & Burton, F. (2014). Wastewater engineering: Treatment and resource recovery. New York: McGraw-Hill.

Valo, A., Carrere H., and Delgenes, J.P. (2004). Thermal, chemical and thermochemical pre-treatment of waste activated sludge for anaerobic digestion. Journal of Chemical Technology and Biotechnology, 79(11). https://doi.org/10.1002/jctb.1106

Van Lier, J.B., Tilche, A., Ahring, B.K., Acarie, H., Moletta, R., Dohanyos, M., Hulshoff Pol, L.W., Lens P., Verstraete, W. (2001). New perspectives in anaerobic digestion. Water Science Technology, 43(1), 1-18. https://doi.org/10.2166/wst.2001.0001

Vlyssides A.G., Karlis P.K. (2004). Thermal-alkaline solubilizatiob of waste activated sludge as a pre-treatment stage for anaerobic digestion. Bioresource Technolog, 91(2), 201-6. doi:10.1016/S0960-8524(03)00176-7

Weemaes, M., Grootaerd, H., Simoens, F., Verstraete, W. (2000). Anaerobic digestion of ozonized biosolids, Water Research, 34(8), 2330-2336. https://doi.org/10.1016/S0043-1354(99)00373-5

WEF, & ASCE/EWRI. (2009). Design of Municipal Wastewater Treatment Plants, Volume 3: Solids Processing and Management. Virginia: WEF Press.

WEF. (1996). Operation of Municipal Wastewater Treatment Plants (Manual of Practice, No. 1). WEF Press.

Weng, C.-H., Lin, D.-F., & Chiang, P.-C. (2003). Utilization of sludge as brick materials. Advances in Environmental Research, 7(3), 679-685. https://doi.org/10.1016/S1093-0191(02)00037-0

Wilson, C.A., Murthy, S.N., Novak, J.T. (2008). Digestibility Study of Wastewater Sludge Treated by Thermal Hydrolysis. Residuals and Biosolids.

Wilson, C.A., Novak, J.T. (2009). Hydrolysis of macromolecular components of primary and secondary wastewater sludge by thermal hydrolytic pretreatment. Water Research, 43(18), 4489-4498. doi:10.1016/j.watres.2009.07.022

Worrell, W.A. & Vesilind, P.A. (2012) Solid waste engineering. (2nd ed.). Stanford: Cengage Learning

Xue, Y., Liu, H., Chen, S., Dichtl, N., Dai, X., Li, N. (2015). Effects of thermal hydrolysis on organic matter solubilization and anaerobic digestion of high solid sludge. Chemical Engineering Journal, 264, 174-180. https://doi.org/10.1016/j.cej.2014.11.005

Yeom, I.T. Lee, K.R. Lee, Y.H. Ahn, K.H. Lee, S.H. (2002). Effects of ozone treatment on the biodegradability of sludge from municipal wastewater treatment plants, Water Science Technology, 46(4-5), 42-425. https://doi.org/10.2166/wst.2002.0641

APPENDICES

A. Calibration of Gas Chromatography

To calibrate the GC, two gas samples which percentages are known measured in GC

Table A-1. *Nitrogen-Methane mixture measurement*

Gas Sample 1	Percentage	Measurement										Avg
		1	2	3	4	5	6	7	8	9	10	
Nitrogen	20%	19.2	19.0	20.6	20.5	19.3	20.1	20.6	19.9	20.5	20.5	20.1
Methane	25%	17.6	16.6	17.6	17.5	17.5	17.6	17.3	17.3	16.7	17.6	17.4

Table A-2. *Nitrogen-Carbon Dioxide and Methane mixture measurement*

Gas Sample 2	Percentage	Measurement										Avg
		1	2	3	4	5	6	7	8	9	10	
Nitrogen	10%	13.1	12.1	13.1	12.1	18.5	13.0	13.0	12.4	13.0	13.3	13.3
Carbon Dioxide	25%	33.9	33.4	33.0	33.2	33.9	33.0	32.9	33.3	33.0	32.9	33.2
Methane	65%	52.9	54.6	53.9	54.6	47.7	54.0	54.1	54.3	54.0	53.8	53.4

For methane real percentages and average of measured percentages showed in Table A-3

Table A-3. *Real and average of measured percentages of methane*

Gas Sample	Unit	Real percentage	Average of measured percentages
1	%	25	17.3
2	%	65	53.3

After average of measured percentages were determined for two gas samples, graph was plotted with both real percentages and average of measured percentages.

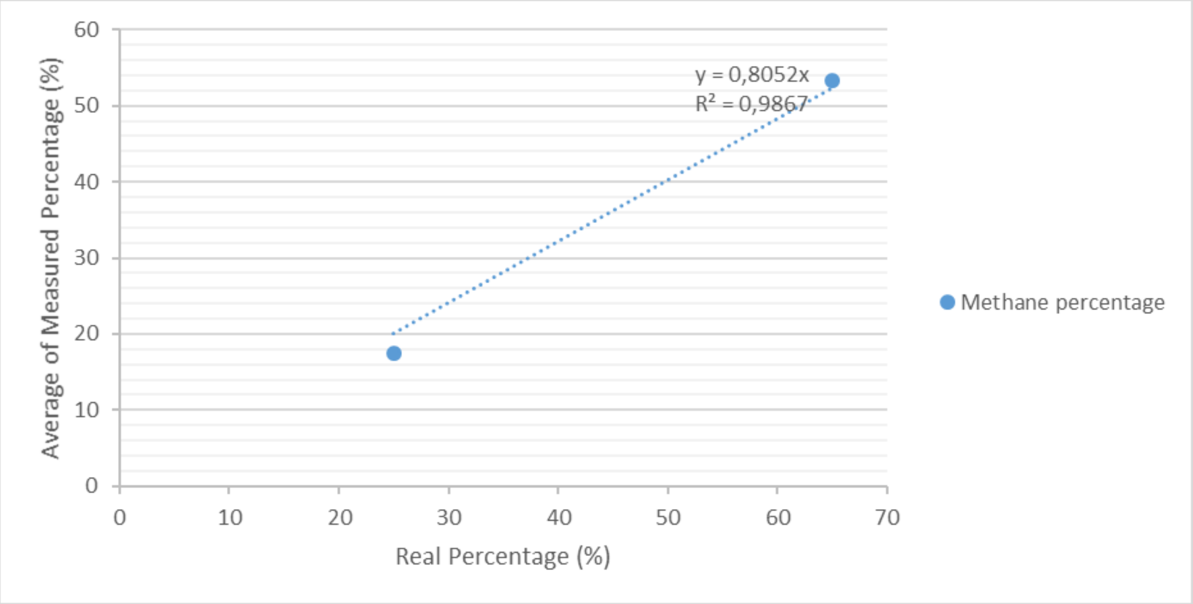


Figure A-1. Calibration curve for methane

With calibration curve, measured methane percentages calibrated.