CHAPTER 1

INTRODUCTION

1.1 BACKGROUND

In most wastewater treatment process, either municipal wastewater or industrial wastewater, activated sludge process is always been used. The usage of activated sludge to treat wastewater is very efficient, particularly in removal of organic matters and nutrients. However, wastewater treatment process using activated sludge will produce large amount of waste sludge which is not favourable to environment. Basically, the waste sludge has the high content of pathogen and hazardous materials. Therefore, waste sludge must be treated by reducing its volume reducing its impact towards the environment before being disposed. This process is called as sludge stabilization. Anaerobic digestions process has been proposed as the efficient method for treating wastewater sludge (Appels et al., 2008). Basically, anaerobic digestion is a sequence of biological processes where microbes breaking down the biodegradable materials in the condition where oxygen is absence. In anaerobic digestion, there are three stages before end product which is biogas is produce. The first stage is hydrolysis and fermentation, then will be continue to the second stage, which is acetogenesis and dehydrogenation. Third stage of the anaerobic process is, methanogenesis, where in this stage methane gas will be produce. The idea of anaerobic digestion is illustrate in the figure 1 below.

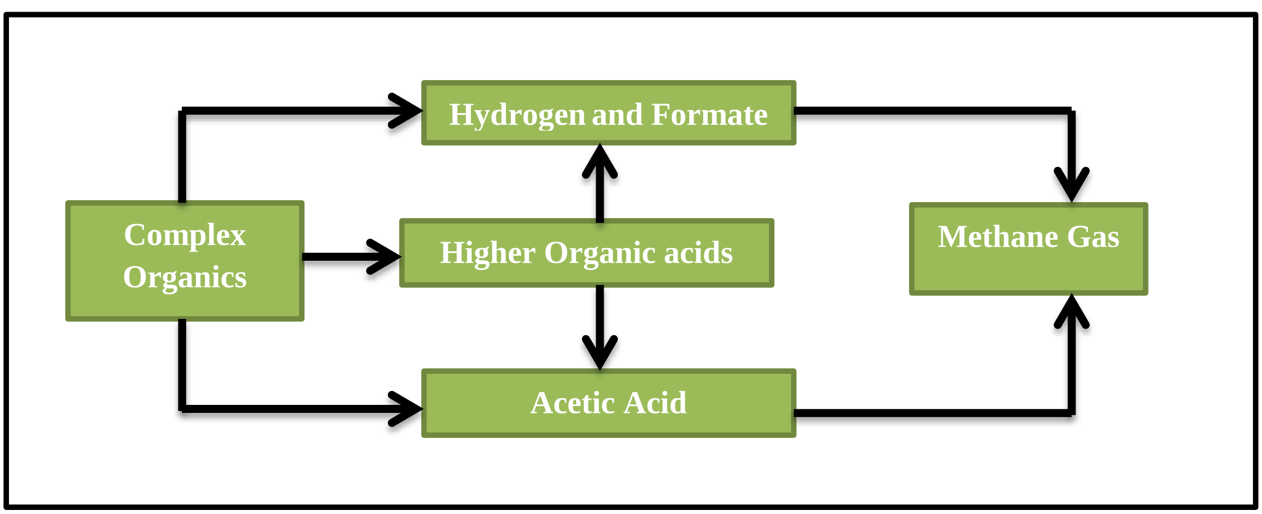

Figure 1: Anaerobic Process Flow

The benefits of anaerobic digestion process in treating wastewater sludge including volume reduction, odour removal, pathogens reduction and most significantly is the biogas production from this process which is methane that can be used as a potential source of energy (Maria et al., 2014). The production of methane gas is the main focus in this research. Optimum condition for anaerobic digestion process was studied and determined, so that the highest amount of biogas was produced and at the same time treat the waste sludge.

Anaerobic digestion for waste sludge may facing low production of biogas and biodegradability, hence having high retention times (Bolzonella et al., 2005). Therefore, pre-treatment of waste sludge is a suggested to increase the biodegradability of the waste sludge.

Some example of pre-treatment is physical pre-treatment, bio-chemical pre-treatment, acidic or alkaline process. In this research, physical pre-treatment of waste sludge was studied. The type of physical pre-treatment used in this research was the ultrasonic pre-treatment. Sonicator was used to sonicate and liquefy the waste sludge before being digested. Sonication of the waste sludge may increase the biodegradability and the biogas production (Mohammed et al., 2008).

Therefore, the high possibility of practicing ultrasonic pre-treatment of sludge in enhancing biogas production was studied further. To assess the anaerobic digestion of sludge and ultrasonic pre-treatment, the waste municipal sludge from Sewage treatment plant in Universiti Teknologi PETRONAS (UTP), Tronoh is used as the sample of study. In order to study about the waste sludge, the sludge from the Return Activated Sludge (RAS) is collected. The Return Activated Sludge can be assume having the same property in terms of its concentration as compare to the waste sludge, as both waste sludge and Return Activated Sludge were come from the secondary clarifier. Thus, it is acceptable to take the Return Activated Sludge as the sample in the research.

Basically, Sewage treatment plant of Universiti Teknologi PETRONAS (UTP) is an extended aeration activated sludge system that consists of an inlet, primary screen, equalization tank, pumping station, secondary screens, grit chamber, grease chamber, two aeration tanks in parallel, two secondary clarifier in parallel, chlorine contact tank, sludge thickener, sludge holding tank, sludge sand drying beds, dewatering facility and a control room. At the beginning of its operation, the influent coming into the sewage treatment plant was only from the new academic complex. However, the facility is now receiving full organic load and hydraulic load with the decommissioning of the north and south oxidation ponds in August 2004 and October 2004, respectively. It served all the student villages, cafeterias, old University Sains Malaysia (USM) buildings and the new academic complex. Figure 2 below briefly explain the system of sewage treatment plant in Universiti Teknologi PETRONAS.

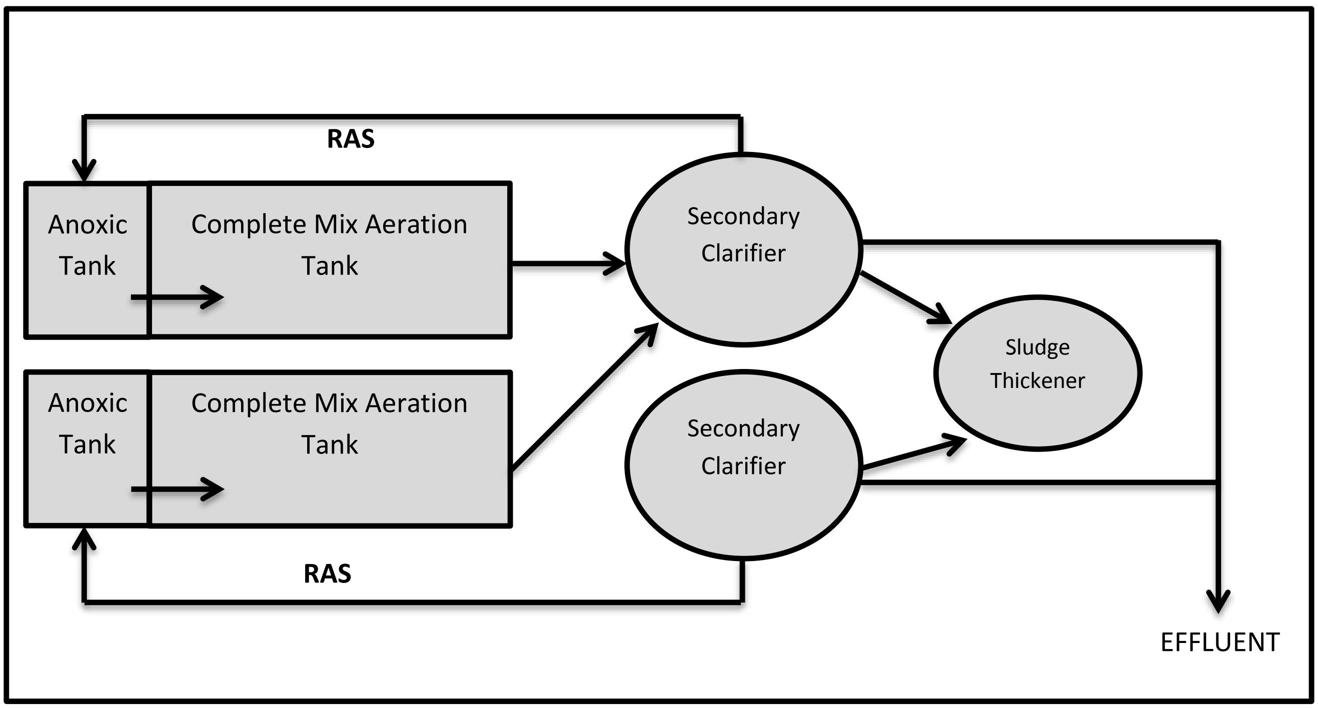

Figure 2: Diagrammatic flow of UTP Sewage Treatment Plant Process

1.2 PROBLEM STATEMENT

Waste sewage sludge is not favorable to environment when disposed without any treatment. Waste sewage sludge may contain high amount of pathogens and toxic. When the waste sludge is dispose to the land, it may contaminate the soil, emit odour and generate polluting lixiviate. If the waste sludge is consider to be dispose to the landfill, the cost impose is high. Therefore, treating the waste sludge with anaerobic digestion process is a good method in reducing waste sludge volume and lowering the biodegradable content. Plus, anaerobic digestion process will help in supplying new source of energy which is methane gas that can be used in heating or electricity generation. The biogas is the end product in the anaerobic digestion.

The biogas is composed of methane gas; carbon dioxide and a very small volume of other trace gases such hydrogen, nitrogen and carbon monoxide. Since anaerobic treatment process of sludge manages to produce methane gas which can be used as new energy source, research of the anaerobic digestion process shall be study in maximizing the biogas production. Some factors are recognize in affecting the anaerobic digestion, for instance temperature, pH, alkalinity, and nutrients.

As for the temperature, there are two temperature ranges that have been identified that favorable for anaerobic digestion, which are Thermophilic (49 – 57 °C) and Mesophilic (30 – 38 °C). Therefore, optimum condition for anaerobic process to take place need to be determines in order to collect the high amount of biogas gas. Besides that, the usage of ultrasonic pretreatment mechanism needs to be study in its effect on biogas production through anaerobic digestion.

1.3 Objectives and Scope of Study

The objectives of this project are:

i. To determine the effect of ultrasonic pre-treatment process towards the organic matter solubility, biodegradability and methane production.
ii. To determine the best condition, either mesophilic or thermophilic digestion with consideration of ultrasonic pre-treatment in producing biogas.

The scopes of study in this project are:

i. Anaerobic digestion process (Mesophilic and Thermophilic).
ii. Physical pre-treatment of waste sludge using sonicator.
iii. Assessment of biogas production from anaerobic digestion

CHAPTER 2

LITERATURE REVIEW

Anaerobic digestion process is an efficient method in treating waste sludge and at the same time producing methane gas. However, the methane gas production in anaerobic digestion process is very dependent on many factors, for instance, temperature, pH and rate of feeding. Anaerobic digestion can be divide into four steps which are hydrolysis, acidification, acetogenesis and methanogenesis. Biological process like anaerobic digestion have its own advantages compare to non-biological process, in the way it use the microorganism to react and treat the wastewater, which consume less energy and less harm to the environment. Anaerobic digestion is more reliable for treating high moisture content or semi-organic materials (Dhamodharan et al., 2014).

There are two types of anaerobic digestion, which are mesophilic digestion and thermophilic digestion. Mesophilic digestion take place at the range of temperature of 30°C – 38 °C, whereas thermophilic digestion take place at the range of temperature of 49°C – 57 °C. Thermophilic temperature often being suggested as better in breaking down of organics matter, and consequently producing methane gas. However, at 27 days SRT, mesophilic digestion will have higher rate of methane production (Rubia et al., 2002). Besides that, pre-treatment of waste sludge also can affect the production of biogas and methane gas. Different pre-treatments can be utilized for solid waste such as physical such as sonication, thermal, chemical, ozonation and biological pre-treatments (Dhamodharan et al., 2014).

The sludge pre-treatment can help in reducing the necessary retention time of sludge for digestion, the final volume of the sludge and also increase the production of methane (Valo et al., 2004). Using physical pre-treatment, ultrasound or sonication is the most potential method for increasing the solubility of organic matter, hence enhancing the anaerobic digestion yield (Cesaro et al., 2011). The sonication is work based on monolithic cavitations, with physical and chemical effects to the wastewater. The physical effects are occur by the collapse of cavitational bubbles, and consequently elevate alteration in the chemical nature through the formation of free radicals (Cesaro et al., 2011). These effects will lead to the destruction of microbiological cells and the oxidation of toxic chemical compounds. Using sonication, COD solubility and anaerobic biodegradability of sewage sludge can be significantly improve (Kim et al., 2003). Kim et al. found that ultrasonic pre-treated sludge can produce 34% higher methane volume as compared to untreated sludge.

The biogas production for pre-treated sludge is higher than untreated sludge because the Ultrasonic pre-treatment process has increase the solubility of organic matter (Cesaro et al., 2011). Braguglia et al (2010) in his research has found that, pre-treated sludge producing 26% more biogas volume than untreated sludge, the difference is due to heterogeneity of Organic Fraction of Municipal Solid Waste. According to Bragulia et al (2010), the proposed duration for the sonication is 1 to 150 minutes with the power values range from 0.2 to 9.0 kilo Watt. To prevent loss of volatile compounds, the pretreated sludge will be cooled down right after the ultrasonic pre-treatment process. In Bragulia et al research, Total volatile solids, Total Nitrogen (TN), and Total Organic Carbon (TOC) of the pre-treated sludge were assess in order to study the effect of ultrasonic pre-treatment. Figure 3 shows the results of the sludge characteristics in terms of Total Volatile solids, Total Nitrogen and TOC in research of Bragulia:

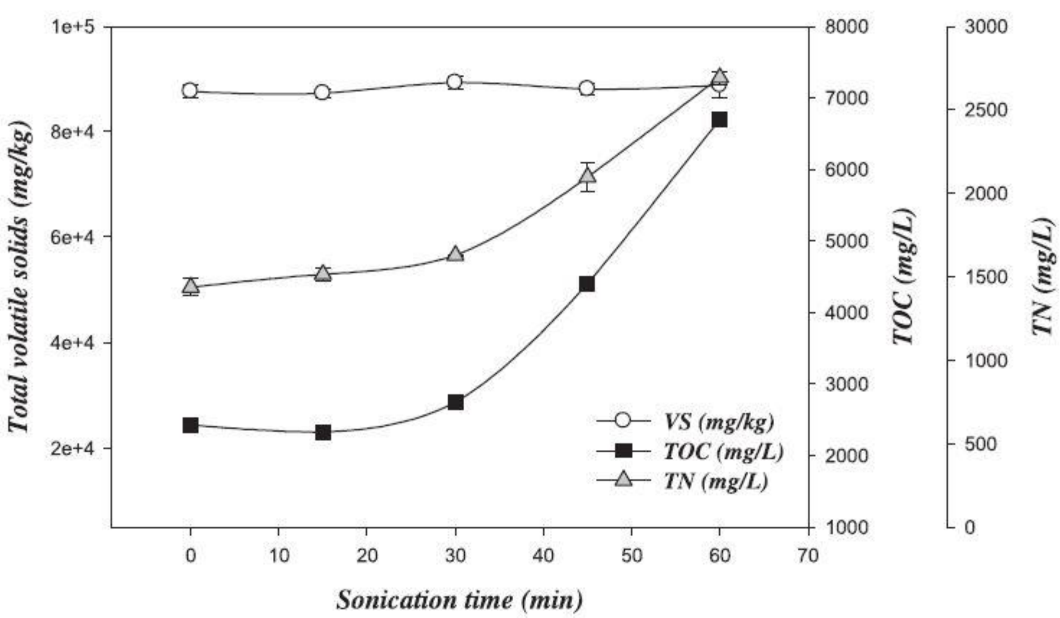

Figure 3: Variation in Total Volatile Solids, TOC, TN with sonication times

Based on the graph obtain in Bragulia et. al research, the variation of time in sonication can cause difference effects towards the pre-treated sludge. It is found that, the longer sonication time will cause the sludge to have higher Total Organic Carbon content as compared to shorter sonication time of sludge. It is also found that, the Total Nitrogen will increase as the sonication time of the sludge increase. However, the Total Volatile Solids will have not much difference when the sonication time is varies. Bragulia also research in how the sonicated sludge will affect the methane yield after the sludge is digested anaerobically. The results of anaerobic digestion from Bragulia research is shown in the Table 1 below:

Operational variables of the biomethanization of raw sewage sludge and pre-treated sewage sludge for loads of 1.0 g VS/L

	Raw sewage sludge	Sonicated sewage sludge (45 min)
Methane yield coefficient (mL_{STP}/g VS)	88 ± 4	172 ± 10
pH	7.91 ± 0.03	8.20 ± 0.06
Alkalinity (mg $CaCO_3$/kg)	6460 ± 310	5795 ± 35
VFA/Alk ratio (eq acetic acid/eq $CaCO_3$)	0.07 ± 0.02	0.22 ± 0.01
OLR (kg VS/$m^3 \cdot d$)	2.2 ± 0.4	0.9 ± 0.1

Table 1: Comparison between unsonicated sludge and sonicated sludge, adapted from Bragulia et al. (2010)

Besides that, Neis et al. (2001) also reported the enhancement of organic matter removal and increment of biogas production from anaerobic digestion of ultrasonic pre-treated sludge. While, Lafitte –Troque and Forster (2002) concluded that ultrasonic pre-treatment of sludge will bring more impact on thermophilic digestion than mesophilic digestion. High efficiency of thermophilic will slightly improve when it is combined with ultrasonic pre-treatment in biogas production (Benabdallah et al., 2006).

In other hand, different frequency of sonication of the sludge can affect the anaerobic biodegradation and biogas production. Low-frequencies of ultrasonic between 20-kHz is most effective in pre-treating the sludge (Dhamodharan et al., 2014). Bougrier et al., (2006) has proved that low-frequencies of ultrasonic is more efficient in degrading the excess sludge, where this mechanical affects facilitate particle solubilisation, thus increasing the readily digestible organic matter.

According to Benabdallah et al. (2006), ultrasonic pre-treatment will cause disintegration of Chemical oxygen demand (COD) and solubilization of organic matter which refer as volatile solids solubilization. At specific energy of greater than 11,000 kJoule/kg TS, Benabdallah found that 40% organic matter solubilization was reached. The results of assessment of pre-treated sludge by Benabdallah is shown in Figure 4:

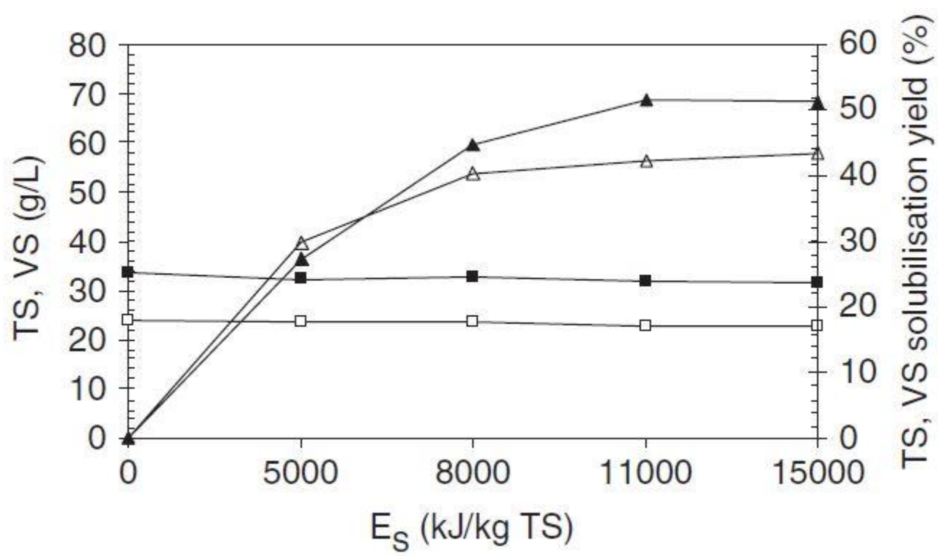

Figure 4: **Effect of ultrasonic pretreatment on TS (■) and VS (□) concentration and TS (▲) and VS (△) solubilization yield at E_s doses tested.**

In terms of anaerobic digestion, Benabdallah found that biogas production can be increase by using sonication pre-treatment. Besides that, Benabdallah also found tha Mesophilic digestion will perform better in remocing COD and producing biogas as compared to the Thermophilic digestion. The results of the research is shown in Figure 5 below:

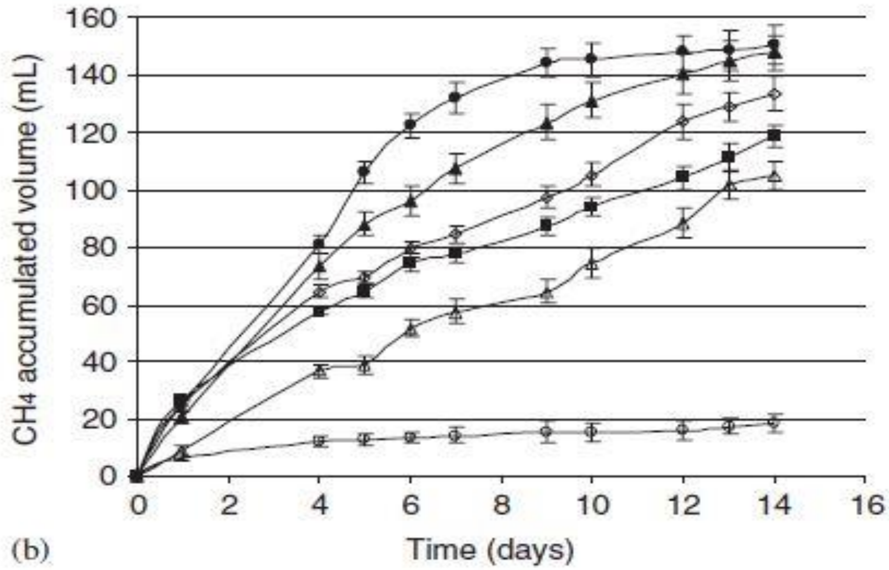

Figure 5 – **Methane accumulated volume in the biodegradability test of feed sludge (622 mg COD) under (a) mesophilic and (b) thermophilic conditions. ((○) Blank; (△) $E_s = 0$ kJ/kg TS; (■) $E_s = 5000$ kJ/kg TS; (◇) $E_s = 8000$ kJ/kg TS; (▲) $E_s = 11{,}000$ kJ/kg TS; (●) $E_s = 15{,}000$ kJ/kg TS).**

Table 2 Improvement (%) in biogas production, COD removal and specific biogas production (SBP) when using pretreated sludge in the mesophilic and the thermophilic digestion

	Mesophilic	Thermophilic
Biogas production	30.88±3.27	16.44±1.76
COD removal	18.32±2.61	6.69±1.13
SBP[a]	16.24±1.82	9.45±0.91

[a] SBP calculated as the biogas production per unit of VS aggregate.

CAHPTER 3

METHODOLOGY

3.1 Overview

The research was conducted at Biological Laboratory located inside the Sewage Treatment Plant (STP), Universiti Teknologi PETRONAS. Since the lab situated inside the Sewage Treatment Plant, the sludge sample collection from the sewage treatment plant is easier. The sludge sample that was used in the experiment is collected from Return Activated Sludge (RAS) of the sewage treatment plant. The Return Activated Sludge (RAS) was collected before it is return into the anoxic tank. The picture of the location for sludge sample collection can be view in Figure 6 below:

Figure 6: Return Activated Sludge at Anoxic Tank

From the Return Activated Sludge (RAS), 5 liters of sludge sample was collected and stored in the container. The sludge was carried to the biological lab for testing. The raw sludge sample was tested for Chemical Oxygen Demand (COD). The Soluble COD (SCOD) and Total COD (TCOD) was tested and recorded. After the SCOD and TCOD for the raw sludge sample was tested, the raw sludge was undergo sonication process.

During preliminary study, the optimum sonication parameters need to be determined. Three same sonicator model VCX 750, manufacture by Sonics were used for sonication process. The sonicators have the frequency of 20 kHz and Power of 750 Watt. The sonicator have the ability to perform sonication process at three different intensity or amplitude, which are 20% amplitude, 30% amplitude and 40% amplitude. In order to determine the optimum amplitude for the sonication process, which subsequently will aid the anaerobic digestion, preliminary test was conducted.

To determine which sonication amplitude will bring the greater impact on the sludge solubilization and biodegradability, sonication of sludge was conducted under three different amplitude 20%, 30% and 40%. 1 liter of sludge was used in the sonication. The sonication time for three of the sonicator is fixed for 2 hours. Therefore, the raw sludge were undergo sonication process for 120 minutes. The increase in TCOD of the sonicated sludge was determine by finding the percentage of difference for TCOD between the sonicated sludge and the raw sludge before sonication. Highest increment in TCOD represents the highest COD solubilization, and it will be assume as the most optimum for sonication.

After the optimum amplitude for sonication was determine, the optimum time for sonication process was tested. The sonication at 40% amplitude were done for 15 minutes. The volume of sludge sample fixed at 1 liter. The percentage of TCOD increase and percentage of COD solubilization of the sonicated sludge was determine for that particular sonication process. After that, the same process is repeated for another sonication time. The second test was sonication process for 30 minutes. The sonication process was kept repeated, until the optimum time of sonication is found. The optimum sonication time was determined when the percentage of TCOD increament and percentage of COD solubilization is stable and not increase anymore.

After the optimum sonication time was determine, that parameter was used in the sonication process to sonicate the sludge before digested in anaerobic digester. For anaerobic digestion, the anaerobic digester model TR 37 which is available at the lab was used. Four reactors were used in order to study the difference in biogas production. In each reactor, 2 liters of the sludge sample was fed. Two reactors were fed with 2 liters of raw sludge sample, whereas another 2 reactors were fed with 2 liters of sonicated sludge sample. The sonicated sludge sample was undergo sonication process for the optimum time that have been found out earlier, and at optimum intensity of sonication.

For the raw sludge sample, one of the reactors was undergo Mesophilic digestion, where the temperature in that reactor was set at 35°C (within Mesophilic range). While, another raw sludge sample undergo Thermophilic digestion, where the temperature of 55°C was maintain in the reactors. The Thermophilic digestion range was between 49°C – 57 °C. The procedure for Mesophilic and Thermophilic condition were repeated for the sonicated sludge in other reactors.

The biogas generate daily will be collected using water displacement method into a gas collection tank. The volume of biogas generate daily from each reactor was recorded, and the accumulate biogas production also recorded. To ensure the gas collected only contain of methane gas, Sodium Hydroxide (NaOH) of 25 g/L was added into the water that fill the gas collection tank in the beginning. This is to ensure, when the biogas displace the water in the tank, all the Carbon dioxide (CO_2) in the biogas content were absorbed into the Sodium Hydroxide solution. Thus, only methane gas shall contain in the gas collection tank.

The existence of any other trace gas in the gas collection tank were omitted as the volume of other gases is too small and not significant. The gas collection from each gas collection tank of the reactors were recorded after 7 days. The SCOD of the treated sludge after 7 days was tested, to find the COD removal through the anaerobic digestion.

The flow of the research work can be illustrate as in the flow chart below.

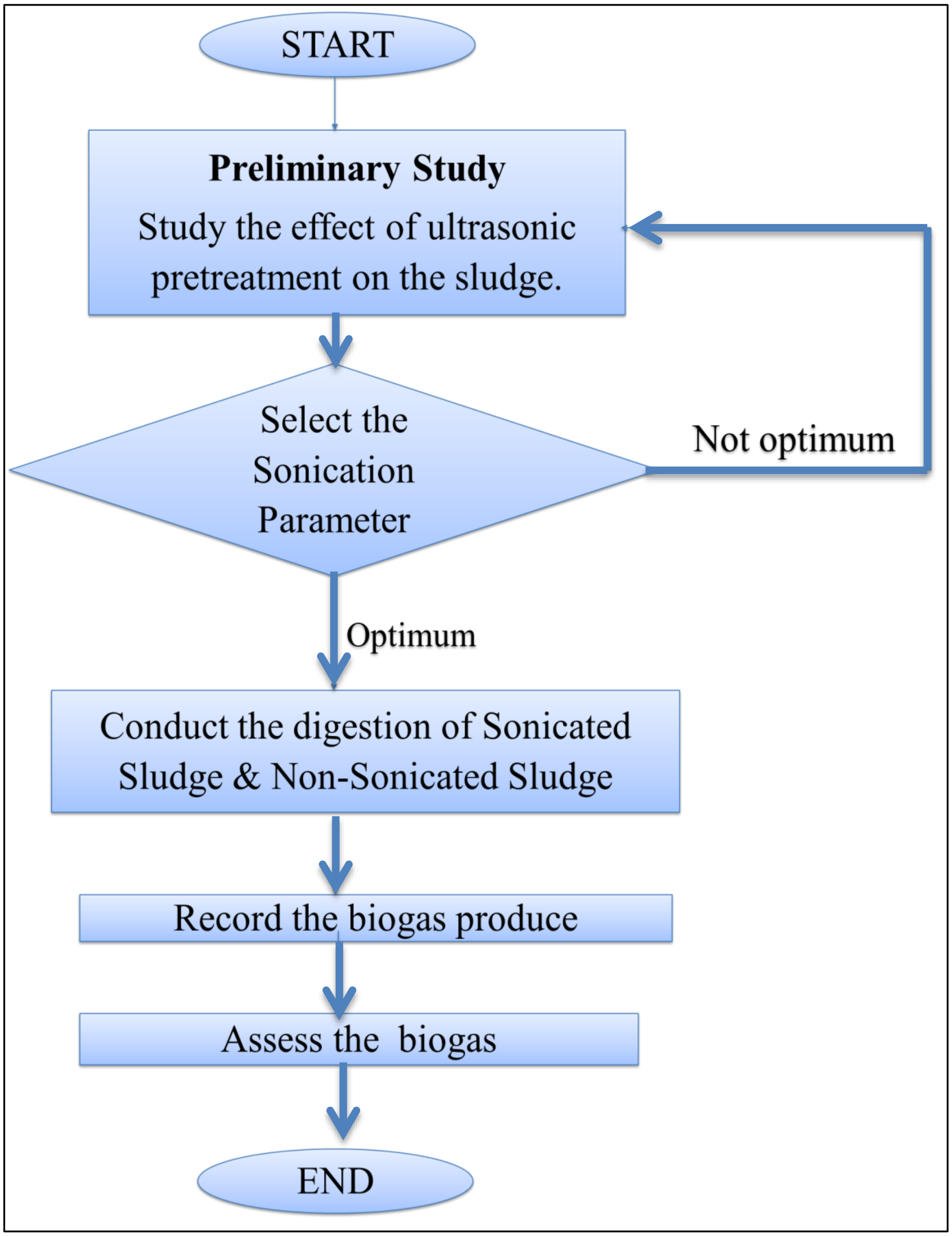

Figure 7: Flow Chart for Research Methodology

3.2 Ultrasonic Pre-treatment

The sludge was sonicated using Ultrasonic Processor VCX 750, manufacture by Sonics. The sonicator operate at the fixed frequency of 20 kHz and power of 750 Watt. However, the sonication process can be varied on its amplitude. There are three amplitude level available, which are, 20%, 30% and 40%. There are probes that act as mechanical transformer to convey the ultrasonic vibration from the converter to the sample. The probes for the sonication is adjustable, where the types of probes can be select based on its diameters of tips and number of tips. There are single tip probes, double-horn probe, eight elements probes, and et cetera. However, throughout this experiment, single probes was selected in conducting the sonication process. The probes of part number 630-0209 was selected. The probes having the tip diameter of 1 inches which equivalent to 25 mm. This type of probes able to sonicate the sample batch up to 1 liter. At the UTP biological lab, three units of sonicators are available. The sonication process that has been carried out in this experiment used three of the sonicator at one time. For example, when sonication test for parameter 40% amplitude and 15 minutes sonication time, all three sonicator should conduct the same sonication process in order to ensure the accuracy and precision in the result. The sonicator at the Biological Lab can be view in the picture below.

Figure 8: Sonicator in Biological Lab, Universiti Teknologi PETRONAS.

3.3 Chemical Oxygen Demand (COD)

The Chemical Oxygen Demand (COD) value indicates the amount of oxygen which is needed for the oxidation of all organic substances in the sludge. COD also indirectly measure the amount of organic matter in the sludge. The Total COD (TCOD) in a batch of sludge sample is compose from Soluble COD (COD$_S$) and Particulate COD (COD$_P$) . The Soluble COD is the COD that was measure form soluble fraction of the sludge, while the Particulate COD is the COD measure from the particulate fraction of the sludge.

To determine the Total COD (TCOD) of the sludge sample, the sludge was stirred vigorously to ensure the homogeneity in the sludge compound, before 1 mL of the raw sludge was suck using pipette. The 1 mL of raw sludge was placed in 250 mL volumetric flask for dilution process. 1:250 dilution factor was used in finding the Total COD. Then, the volumetric flask was filled with distilled water until reach the dilution marks on the volumetric flask. The picture of dilution process can be view in figure 9 below.

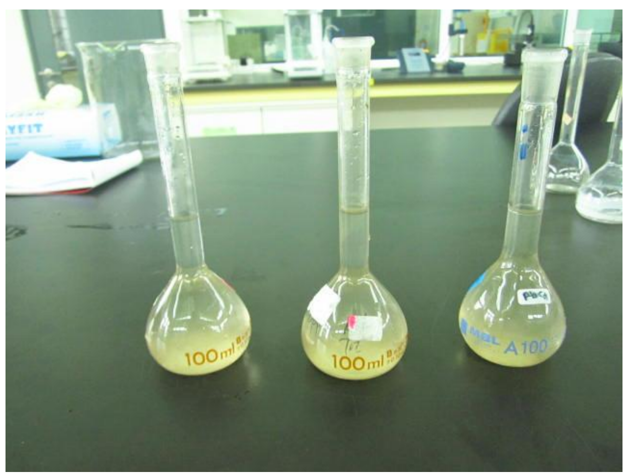

Figure 9: Dilution process using Volumetric Flask

EPA method 410.4 was used to test the COD in this experiment. In this method, Potassium Dichromate acts as the oxidizing agent in determining how much oxygen id used for the oxidization of that particular compound. 2 mL of the diluted sludge earlier was put into the High Range COD vials. A blank also was prepared, using 2 mL of distilled water which also put into another COD vials. This blank sample was used to calibrate the spectrophotometer in recognizing zero oxidization.

After the samples were put into the COD vials, the vials were be placed in the heater. The samples were digested at temperature of 150 °C for 2 hours. After the sample cooled down, then the vials that contained the samples was put in the Spectrophotometer. The reading by the spectrophotometer represent the COD value for the diluted sample in unit of mg/L. To know the Total COD (TCOD) of the sludge sample, COD value read by the Spectrophotometer need to be multiply by the dilution factor, which is 250.

To determine the Soluble COD (COD_S) of the sludge sample, the supernatant of the sludge was took and diluted with the dilution factor of 1:2. The supernatant of the sludge can be collected after the solid fraction in the sludge was settled, leave sludge into two layers, where solid fraction layer at the bottom, and supernatant layer at the top. Then, the supernatant was collected and placed into another container. The supernatant was stir vigorously to ensure uniformity in the solution. Then, 50 mL of the supernatant was put into 100 mL Volumetric flask to do the dilution of 1:2.

After the supernatant was diluted, it was filter using filter paper to remove any solid particles in the solution. This to ensure the COD reading from this solution is only come from the soluble fraction, not exert by the solid fraction. After the supernatant was filtered, 2 mL of the filtered supernatant was put into Low Range COD vials for COD test. The procedure is same as previous COD test.

After the sonication, COD was anticipate to solubilize. The COD Solubilization (S_{COD}) represents the transfer of COD from the particulate fraction of the sludge to the soluble fraction of the sludge. To calculate the COD solubilization, difference between Soluble COD (COD_s) of sludge after sonication and the initial Soluble COD (COD_{so}) of sludge, in relation of the initial Particulate COD (COD_{PO}). The formula used :

$$\boxed{COD\ solubilization\ (\%) = \frac{(COD_S - COD_{SO})}{COD_{PO}} \times 100}$$

3.4 Anaerobic Digestion

The digester TR 37 was used to conduct anaerobic digestion of waste activated sludge. TR 37 is the convertible digester that can perform aerobic or anaerobic digestion. In this research, TR 37 was used anaerobically to digest the waste sludge. TR 37 has 6 reactors. The unit consists of mainly reactor, influent tank, effluent tank, feeding pump (designed for sludge), jacket heater, aeration system, stirrer motor complete with variable speed controller, pH and DO probe. These components are mounted within a specially designed chemical resistance phenolic resin and stainless steel supporting framework. However, TR 37 digester was having problem with the feeding pump, therefore, feeding was done manually. The cap of the reactors was dismantled, and then 2 Liter of sludge was putted into each the reactor. The pictures of TR 37 can be observed in Figure 10 below:

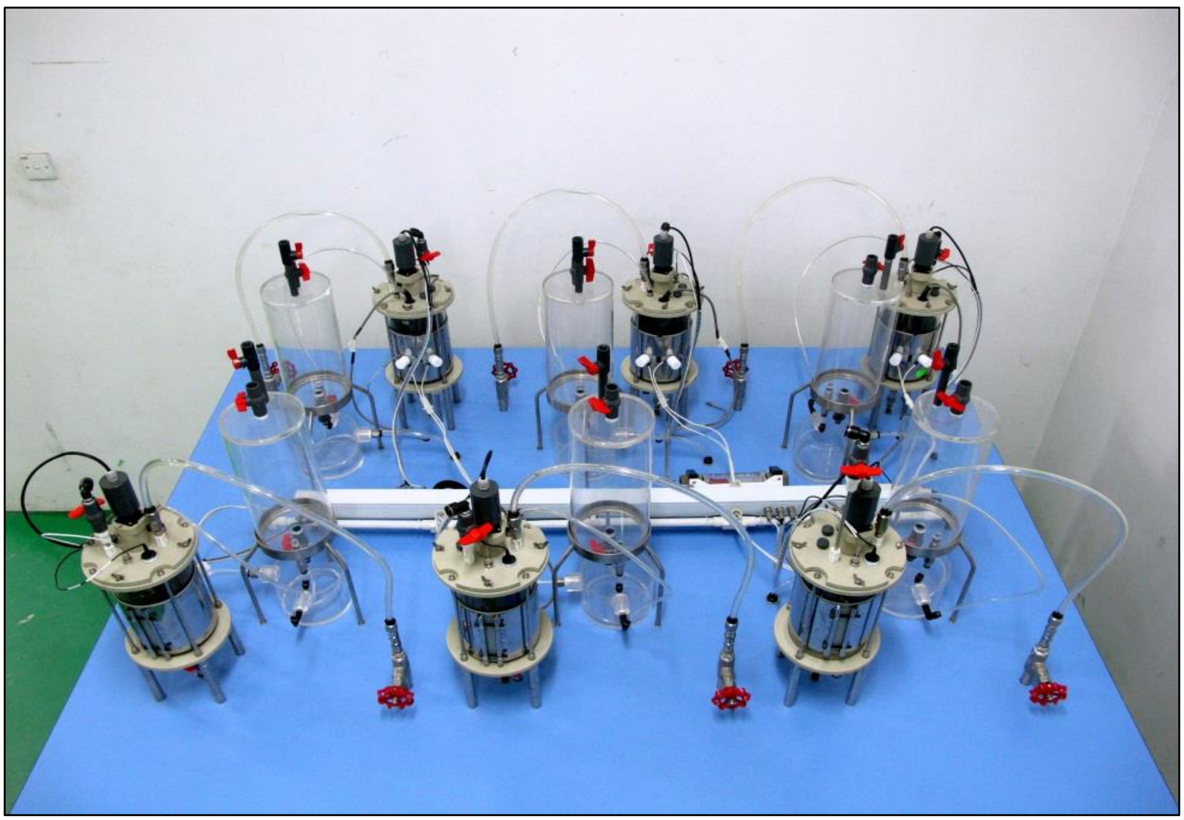

Figure 10: TR 37 Digester in Biological Lab, Universiti Teknologi PETRONAS.

From those six reactors, only four reactors were used for this experiment. Two reactors will using Mesophilic digestion, whereas another two reactors will using Thermophilic digestion. For the two reactors which using Mesopihlic digestion, the temperature was set at fixed point of 35°C, same as thermophilic where a fixed temperature was fixed at temperature of 55°C. The purpose of fixing the temperature for each digestion is to ensure the anaerobic digestion take place in the same environment, either in Mesophilic or Thernophilic. Besides that, the sludge inside the reactors is continuously stirred rapidly. The real time temperature inside the reactors and revolution rate of the stirrer can be observed and control through the digital meter.

Figure 11: Meter showing the Temperature inside the reactors

Throughout the anaerobic digestion process that occur in the digester, biogas was produce. The biogas produce in the enclosed reactors was transfer to the gas collection tank. The gas was capture using the water displacement method. The reactors system was ensure to be enclosed and air tight, so that without any exposure to the atmosphere, the level of the water inside the collection tank was remain at the same level. When gas is released from the reactor, additional pressure is created inside the tank and thus forced the water level to decrease from its initial level. The volume of water displaced by the additional pressure is released through valve that connects the reactors to the gas collection tank. The difference in water level is directly proportional to the amount of gas collected. However, to ensure carbon dioxide is not mixed with methane gas in the collection tank, the water that was used to filled the collection tank was dissolved with 0.1 M Sodium Hydroxide, NaOH. Carbon dioxide gas was absorbed by the Sodium Hydoxide solution, leaving only methane gas and other small volume of trace gas in the collection tank. The gas collection tank can be view as in the figure 12 below:

Figure 12 Gas Collection Tank (Fill with NaOH solution)

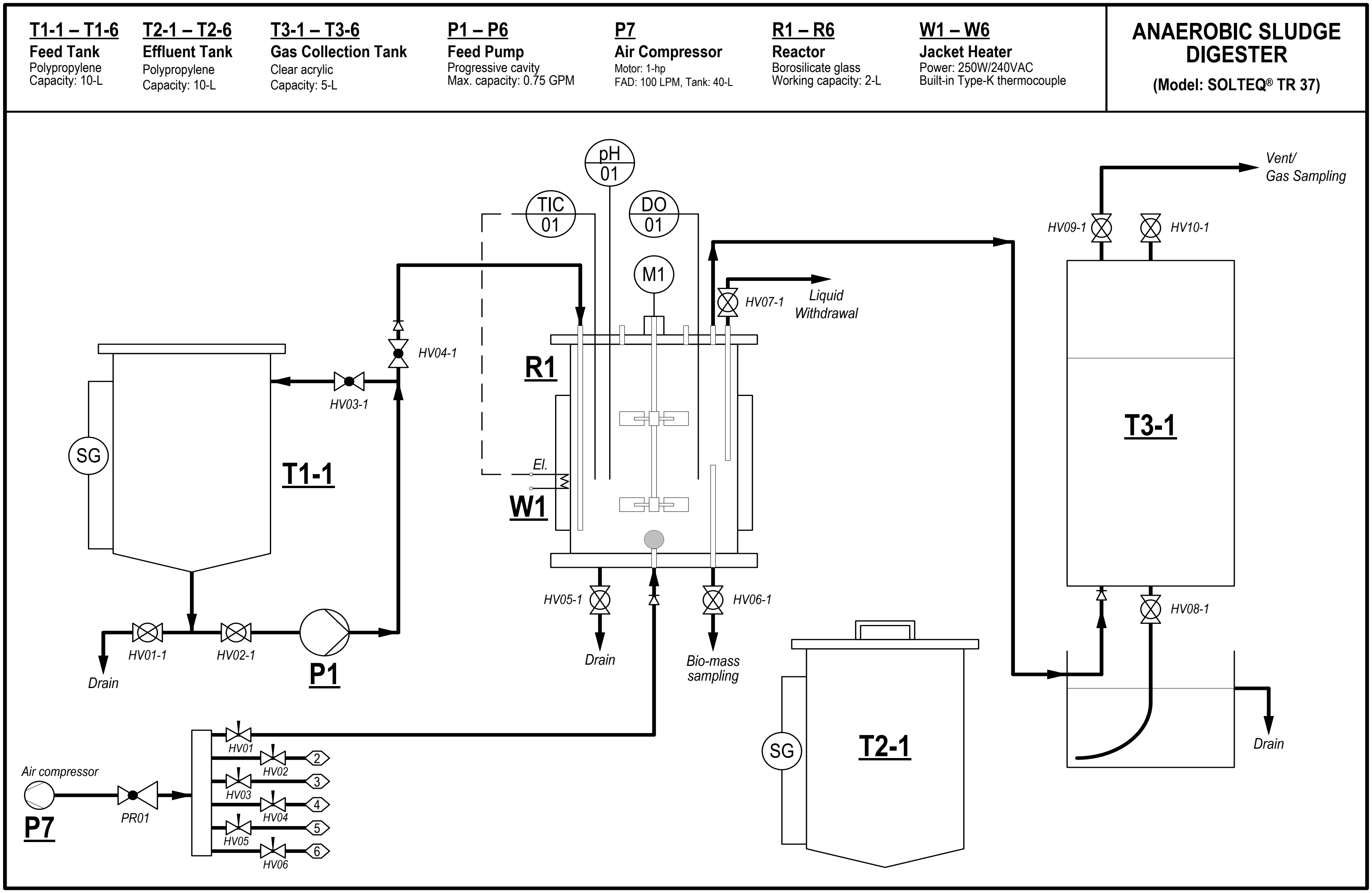

Figure 13: TR37 Process & Instrumentation Diagram

3.5 COD Removal

After the sludge undergo anaerobic digestion for 7 days, the Total COD (TCOD) was measured once again. The purpose of measuring the Total COD (TCOD) after 7 days of anaerobic digestion is to determine the COD removal through the anaerobic process. The COD removal percentage is calculate by finding the difference between the initial Total COD (TCOD) of the before entering the digester, with the final Total COD (TCOD) of the sludge after 7 days of anaerobic digestion. The formula for the COD removal:

$$\boxed{COD\ removal\ (\%) = \frac{(initial\ TCOD\ - Final\ TCOD\ )}{initial\ TCOD} \times 100}$$

To collect the sludge sample from the reactors after 7 days of digestion, valve HV-06 x was used. The sludge was discharge through the valve, and then the sludge was tested for Total COD. The Valve HV-06 x used can be view in figure 14 below.

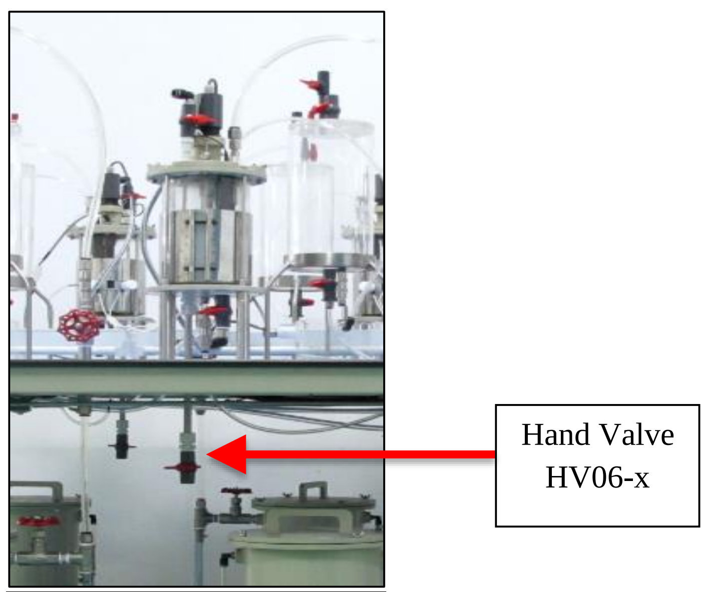

Figure 14: Valve HV-06 for Sludge Discharge from Reactors

3.6 Gant Chart

Activities	November 2014	December 2014	January 2015	February 2015	March 2015	April 2015
Proposal Submission						
Order Experiment Materials						
Preliminary Studies						
Determination of Optimum Sonication Parameters					Milestone: Optimum Sonication	
Start Anaerobic Digestion						
Collection of the biogas						Milestone: Collect biogas
Result analysis						
Report write up						Milestone: Finish Report

CHAPTER 4

RESULTS AND DISCUSSION

4.1 Preliminary Results (Optimum Sonication Amplitude Determination)

In this section, the result for the determination of the optimum amplitude that needs to be used for the sonication process to pre-treat the sludge will be show. Three sonication amplitude percentages have been tested. The sonication amplitudes percentages that were used 20%, 30% and 40%. The sonication process on the sludge sample was monitored on its capability in increasing the Total COD (TCOD) of the sludge. The result of the preliminary study on the determination of the optimum amplitude for sonication process can be view in the figure 15 below.

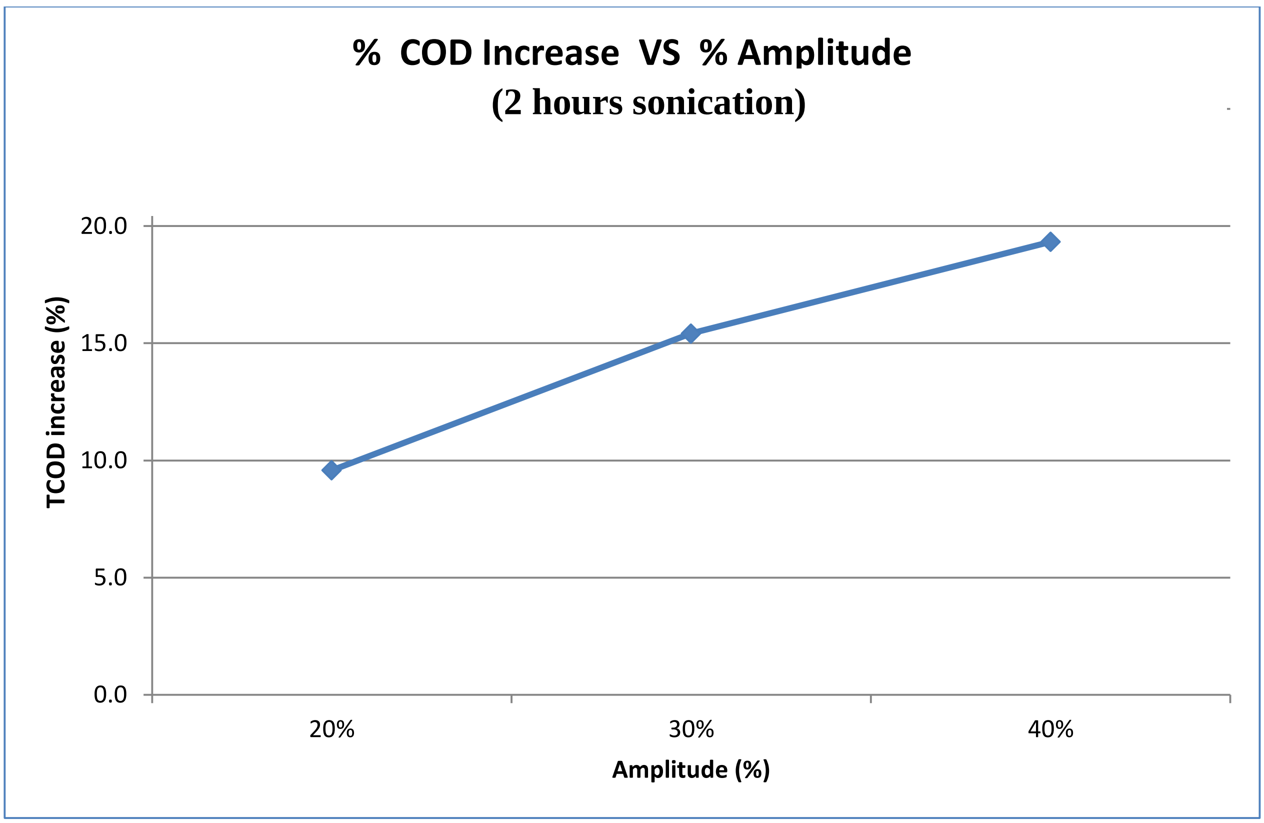

Figure 15: Plot of Percentage of COD increase VS Amplitude (%)

Based on the bar chart for TCOD increase (%) versus Sonication Amplitude (%), it is clearly shown that, as the intensity of the sonication increase, the percentage of increment of TCOD in sludge is increase. From the bar chart, 20% sonication amplitude giving 9.6% of TCOD increase in the sludge, while 30% sonication amplitude increase the TCOD in sludge to 15.4%. The highest sonication amplitude that can be perform by the Ultrasonic equipment in the lab, which is 40% sonication amplitude showing the highest impact on TCOD increase in sludge. The TCOD in sludge is increase to19.3% after the sonication process using 40% amplitude sonication. Therefore, 40% amplitude was selected as the best amplitude for this experiment when using Ultrasonic Processor VCX 750. As for reminder, all other parameters for this particular test in determining amplitude was fixed. The frequency for the sonication was fixed at 20 kHz, Power of 750 Watt, and 2 hours sonication time.

4.2 Preliminary Results (Optimum Sonication Time Determination)

In this section, the result in determining the best sonication time will be discussed. To perform the sonication process for determining the optimum sonication time, the sonication amplitude was fixed. Based on the result from previous preliminary test, 40% sonication amplitude was found out as the best amplitude in performing sonication as it will bring the highest impact on the TCOD increment in sludge. The higher TCOD increase represent more COD solubilization occur in that sonication process. It shows that the sonication succeed to enhance the COD solubilization in the sludge, and subsequently increase the biodegradability of the sludge which is important in helping the anaerobic digestion.

To assess the impact of the sonication time towards the sludge in this preliminary test, the increase in TCOD of the sludge, and the increase in COD Solubilization (S_{COD}) will be be monitor. The COD Solubilization can be calculate using the formula provided in the Section 3.3 Methodology section. The result of the test can be view in the by the plot in figure 16 and figure 17.

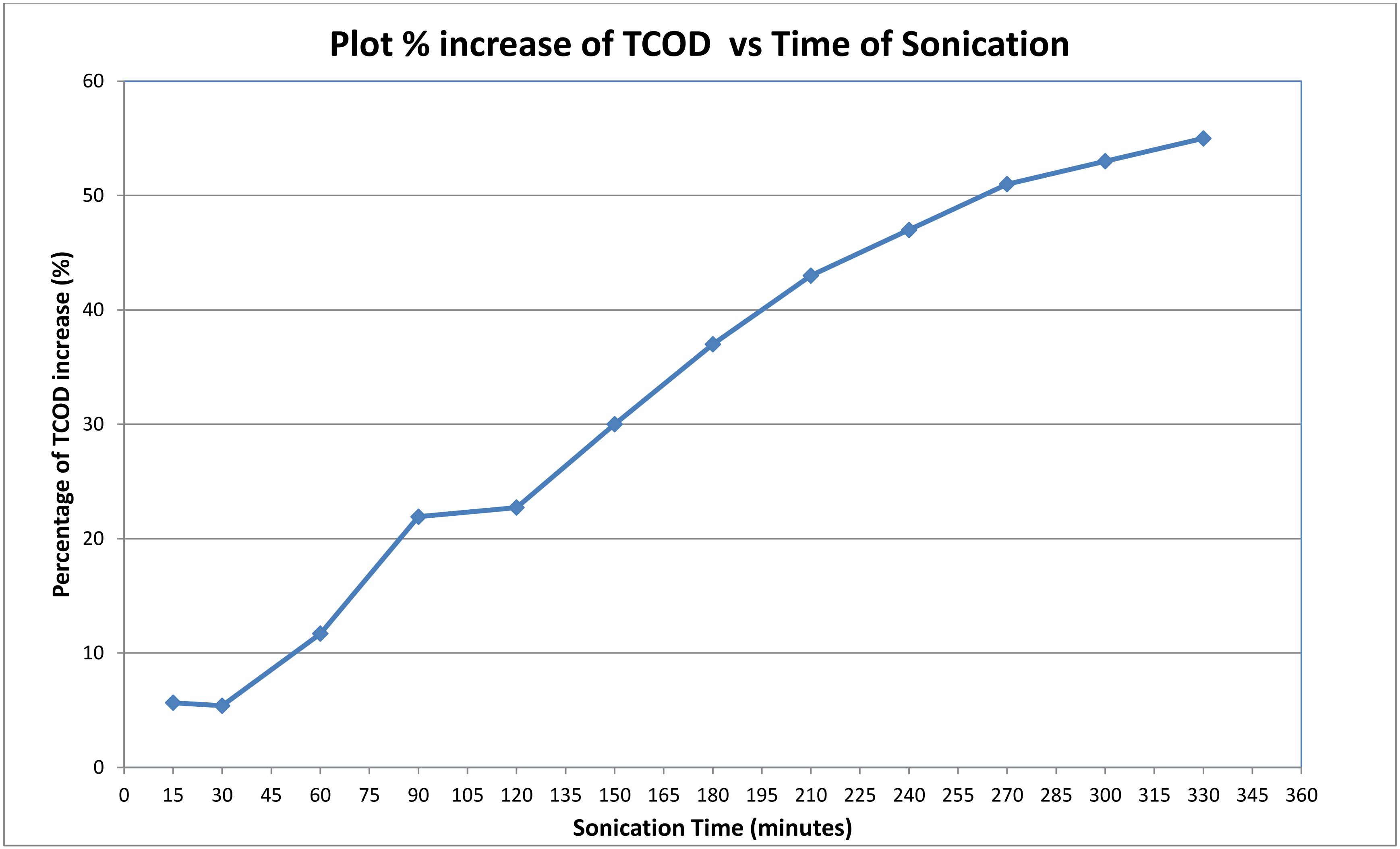

Figure 16: Plot for Percentage of TCOD increase VS Time of Sonication

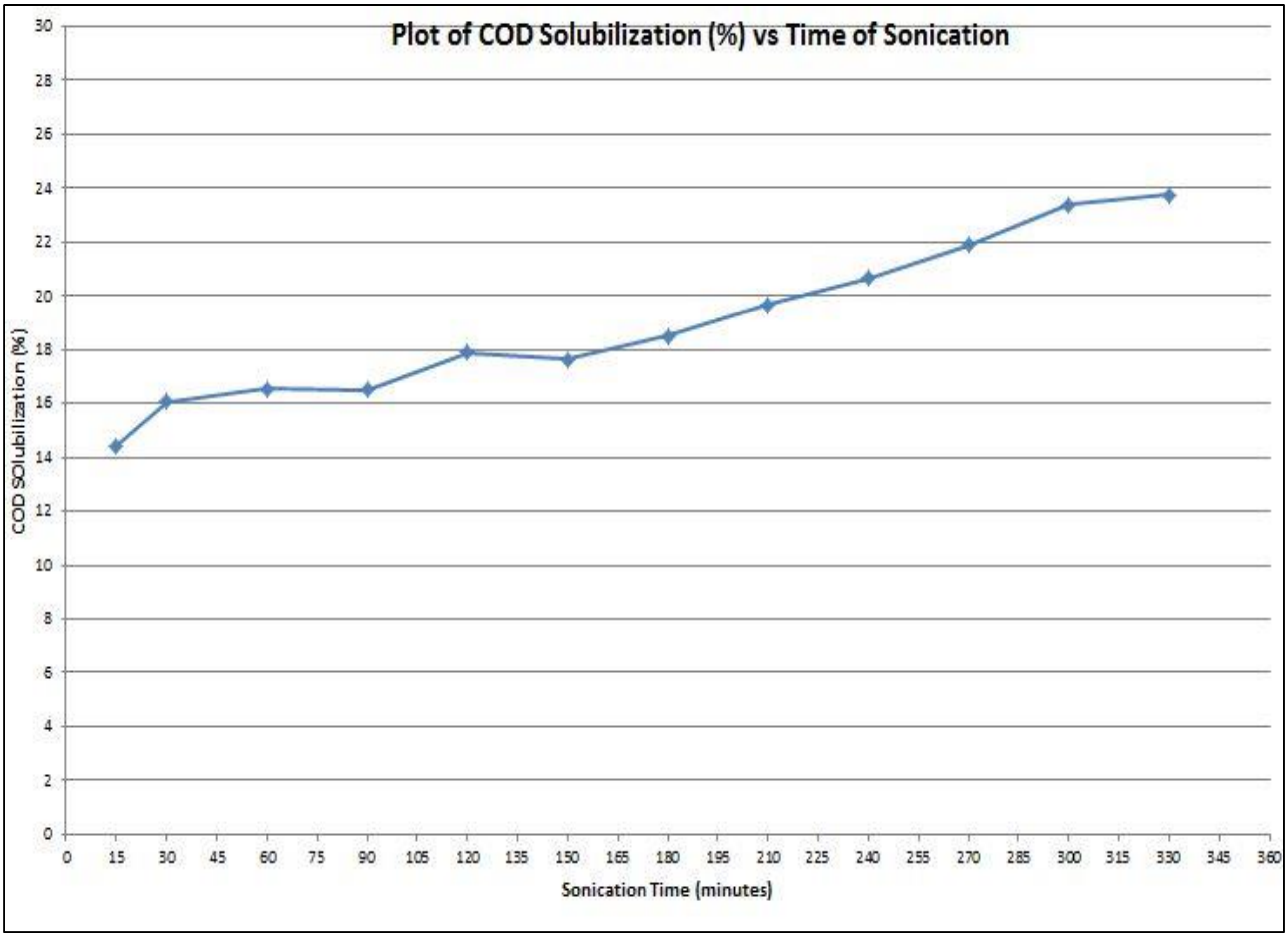

Figure 17: Plot for Percentage of COD Solubilization VS Time of Sonication

Based on both plot that was obtained from the experiment, it is clearly shows that the higher the sonication time, the greater impact it will bring to the sludge in terms of its TCOD increase and the COD Solubilization. From the plot, the trend for the increment in TCOD percentage and also COD solubilization start to stabilize at sonication time of 300 minutes to 330 minutes. The slow increment trend for the TCOD increase and COD Solubilization is expected when the time of sonication increase. However, for this experiment, due to the time constraint, sonication time of 330 minutes is selected as the best sonication time for sonicating the sludge, but the optimum time may be higher than 330 minutes. Further test need to be conducted in order to find the exact optimum time for the sonication process.

Based on the plots, the increase in TCOD between 15 minutes sonication time and 30 minutes sonication time show not much increment. While, in the COD Solubilization plot, it shows that small increase in COD solubilization percentage between 15 minutes to 30 minutes of sonication time. However, after 30 minutes of sonication, as the time of sonication increase, the TCOD is keep increasing almost uniformly, until the sonication time of 300 minutes is reach, then the trend of TCOD increase has started to slow down.

The reason for the TCOD not increase significantly when the sludge was sonicate between 15 minutes sonication time and 30 minutes sonication time, is due to the impact of the sonication to the sludge flocs. When the time of sonication is short, the solid particle in the sludge do not have enough time to break much. As compare to the longer sonication time, the sonication process is able to break the solid flocs further. When the solid flocs is break, the more organic matter is release and react, thus increasing more oxygen demand. This has lead to the increase in Total COD (TCOD).

Besides floc breakage, the sonication also able to break the cell membrane of the individual cell of organic matter. As reported by Thiem et al, sonication at higher intensity will break the cell membrane and cell lysis will occur. The cell lysis also contribute in the incrase of COD in the sludge. This mean, floc breakage and cell lysis has occur in this experiment, and subsequently increase the TCOD of the sludge.

The COD solubilization also keep increasing, as COD solubilization represents the transfer of COD from the particulate fraction of the sludge to the soluble fraction of the sludge. This means, more COD from the particulate has dissolve in the soluble fraction after the sonication, cause by the solid flocs breakage and cell lysis.

The optimum parameters of the sonication is use in next stage of research, in finding the effect of it in biogas production through anaerobic digestion. The sludge was pre-treated using the sonication of 40% amplitude, and sonicate for 330 minutes, before the sludge is digested in the anaerobic digester.

4.3 Biogas Production

In this section, the result of biogas produce from anaerobic digestion is discussed. Four reactors have been fed with the sludge that each of the reactors having the different variables. The biogas produce was captured in the collection tank. The production of biogas was observed daily, until 7 days of anaerobic digestion. The result for the biogas production can be view in the table below.

Table 3: Cumulative Volume of Biogas Produce

	Accumulated Biogas Produce (Liter)						
Days	1	2	3	4	5	6	7
(Mesophilic) Non-sonicated	0	0.05	0.1	0.15	0.25	0.3	0.4
(Thermophilic) Non-sonicated	0.05	0.1	0.15	0.25	0.4	0.55	0.75
(Mesophilic) Sonicated	0.05	0.1	0.2	0.3	0.5	0.7	0.95
(Thermophilic) Sonicated	0.05	0.15	0.3	0.45	0.65	0.85	1.1

Based on the table above, it shows the production of the biogas for each reactor. The accumulated biogas after 7 days is vary based on the parameters set for each reactor. From the table, the biogas production after 7 days is highest for Thermophilic digestion of sonicated sludge, with the Total volume of biogas produce of 1.1 Liter. Whereas, the Mesophilic digestion of the non-sonicated sludge shows the lower biogas production with only 0.4 Liter of biogas produce after 7 days of the digestion. The trend of the biogas production for each case is plotted on the graph in figure 18 the next page.

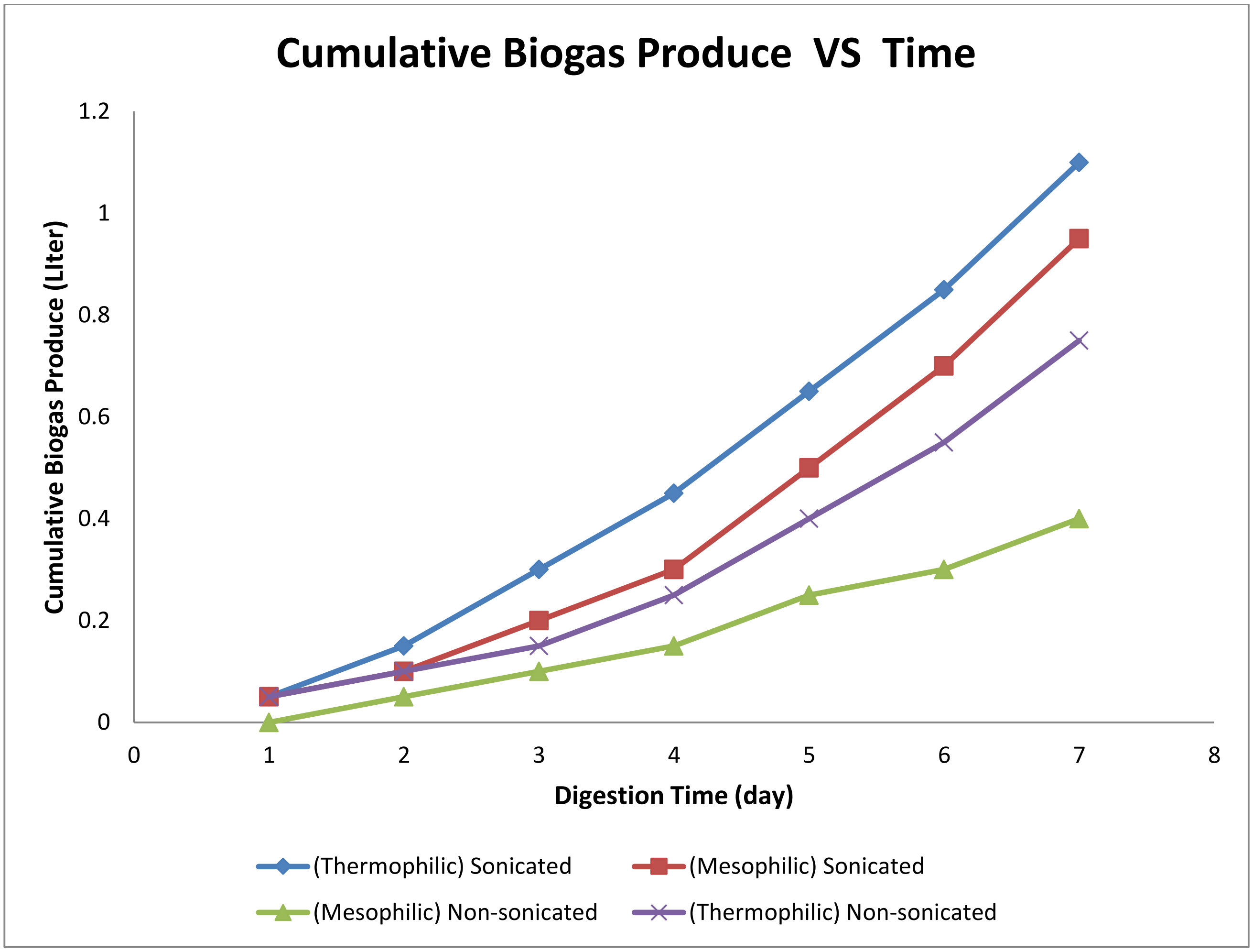

Figure 18: Plot of Cumulative Biogas Produce VS Time

Based on the plot above, it is clearly show that the biogas production is keep increasing from day 1 of the digestion, until day 7 of the digestion. As of day 7, Thermophilc digestion in both sonicated and non-sonicated sludge have show the greter biogas production when compare the Mesophilic digestion. However, Thermophilic digestion of sonicated sludge has produce higher biogas volume when compare to the Thermophilic digestion of Non-sonicated sludge.

In terms of trend in rate of biogas production, overall rate for each digestion is almost uniform. It is also found out that, Sonication do not significantly affect the rate of biogas production in

Thermophilic. However, in Mesophilic, the rate of Biogas production is increase slightly when the sludge is sonicate.

4.4 COD Removal

This section will discuss the result of COD removal from the digestion in each reactors. The result of the percentage of COD removal can be view in the table and bar chart below.

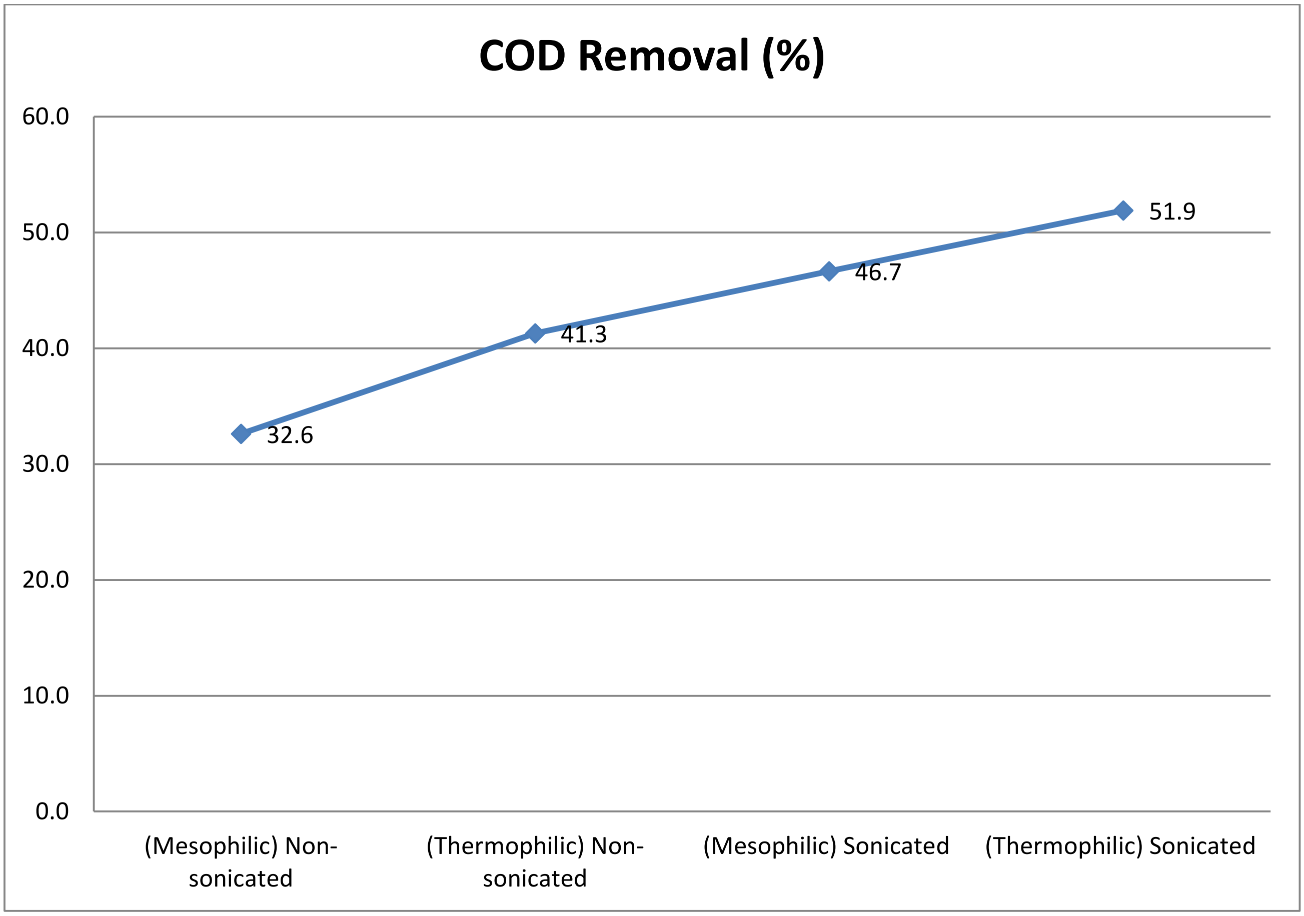

Figure 19: Percentage of COD Removal for each Digester

COD removal is found highest in Thermophilic digestion of sonicated sludge and lowest in mesophilic digestion of non-sonicated sludge. Sonicated sludge has undergo sonication that increase the biodegradability of the sludge, hence increase the efficiency of the COD removal. The COD is much easier to be degraded as the sludge itself have become more liquefy due to the sonication process. Therefore, the efficiency of the anaerobic digestion on sonicated sludge is higher. While, in terms of temperature, Thermophilic show the higher efficiency when compare to the Mesophilic, as higher heat has promote the anaerobic bacteria activity.

CHAPTER 5

RECOMMENDATIONS

In conducting this research, some limitations have been faced by the author. The limitation in the experimental work could bring impact on the result output. In future research, the author has some recommendations that are suggested, in obtaining more comprehensive result for this research topic.

First recommendation for the research is the consideration of the frequency for ultrasonic pre-treatment. In this research, the author facing the limitation in setting the frequency for the sonication process, as the Ultrasonic equipment provided in UTP Biological Lab have no ability in setting the frequency. The frequency for sonication process is fixed at 20 kHz in this experiment. The frequency shall be varied in future research so that more comprehensive result could be obtained.

Second recommendation for the research is the usage of the sonication probe. In this research, only Probe type 630-0209 was selected. The probes having the tip diameter of 1 inches which equivalent to 25 mm. As stated in the user manual of the Ultrasonic equipment, Probes with smaller tip diameters produce greater intensity of cavitation, but the energy released is restricted to a narrower, more concentrated field. Conversely, probes with larger tip diameters produce less intensity, but the energy is released over a greater area. The larger the tip diameter, the larger the volume that can be processed, but at lower intensity. Therefore, it can be understand that, different types of probes will give the different impact towards the result output. Study in types probes relation need to be studied in future research.

CHAPTER 6

CONCLUSION

As the conclusion for the research, ultrasonic pre-treatment has been found to increase the biodegradability of the raw sludge through it sonication process. The sonication process will break the solid flocs in the sludge and also break the cell membrane of some individual cell in the sludge. When the solid fraction is break and become soluble, the COD of the sludge is increase. This outcome also has been reported in the research by Thiem et al.

When the sludge biodegradability increase, anaerobic digestion can be perform more efficiently, thus increasing the biogas production. In this research, sludge that has been undergo ultrasonic pre-treatment has produce more biogas than the untreated sludge. For Thermophilic digestion, the sonicated sludge has produce 1.1 liter of biogas, while the non-sonicated sludge only produce 0.75 liter of biogas. Whereas, in Mesophilic digestion, the biogas produce for the sonicated sludge is 0.95 liter, while the non-sonicated sludge only produce 0.45liter of biogas.

When comparing between Thermophilic and Mesophilic, it is found that Thermophilic digestion will produce higher Total gas volume, compare to the Mesophilic digestion. Besides that, COD removal also higher in the Thermophilic digestion as compare to the Mesophilic digestion.

In conclusion, Thermophilic digestion with ultrasonic pre-treatment perform better in biogas production from degradation of the waste sludge.

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APPENDIX

Raw Data from lab experiment.

Result of Different Sonication Amplitude on TCOD increase

Sonication Amplitude (%)	**20 %**	**30 %**	**40 %**
Initial TCOD of the Raw sludge (mg/L)	12,000	12,000	12,000
Final TCOD of Sonicated Sludge (mg/L)	13,150	13,850	14,320
TCOD Difference (mg/L)	1,150	1,850	2,320
% of TCOD increase	9.6 %	15.4 %	19.3 %

Result for the preliminary result in determining optimum Sonication Time

Sonication Time (minutes)	TCOD increase (%)	COD Solubilization (%)
15	6	14
30	5	16
60	12	17
90	22	17
120	23	18
150	30	18
180	37	19
210	43	20
240	47	21
270	51	22
300	53	23
330	55	24

COD REMOVAL

Type if Digestion	TCOD before Digestion	TCOD Final	COD REMOVAL (%)
(Mesophilic) Non-sonicated	11500	7750	32.6
(Thermophilic) Non-sonicated	11500	6750	41.3
(Mesophilic) Sonicated	17500	9333	46.7
(Thermophilic) Sonicated	17500	8417	51.9

40% Sonication for 15 minutes (Sludge 5 February 2015 / 10.30 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	55	13750	13750
		2	56	14000	
		3	54	13500	
	Supernatant	1	17	17	19
		2	19.3	19.3	
		3	20.5	20.5	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	65	16250	15250
		2	60	15000	
		3	58	14500	
	Supernatant (Dilution 1:2)	1	1015	2030	2031
		2	1019	2038	
		3	1013	2026	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	58	14500	13583
		2	51	12750	
		3	54	13500	
	Supernatant (Dilution 1:2)	1	984	1968	1952
		2	979	1958	
		3	965	1930	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	61	15250	14750
		2	57	14250	
		3	59	14750	
	Supernatant (Dilution 1:2)	1	1009	2018	2024
		2	1012	2024	
		3	1015	2030	

	TCOD	**SCOD**	**ssCOD**
Raw Non-Sonicated	13750	19	13731
Sonicated	14528	2002	12525
		%	
Difference TCOD	777.7777778	6	
% COD Solubilisation	14		

40% Sonication for 30 minutes (Sludge 6 February 2015 / 9.30 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	45	11250	11333
		2	45	11250	
		3	46	11500	
	Supernatant	1	18	18	19
		2	19	19	
		3	18.7	18.7	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	47	11750	11333
		2	45	11250	
		3	44	11000	
	Supernatant (Dilution 1:2)	1	915	1830	1829
		2	914	1828	
		3	915	1830	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	51	12750	12250
		2	49	12250	
		3	47	11750	
	Supernatant (Dilution 1:2)	1	920	1840	1836
		2	917	1834	
		3	917.4	1834.8	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	50	12500	12250
		2	49	12250	
		3	48	12000	
	Supernatant (Dilution 1:2)	1	925	1850	1846
		2	921	1842	
		3	923	1846	

	TCOD	**SCOD**	ssCOD
Non-Sonicated	11333.33333	18.56666667	11314.76667
Sonicated	11944.44444	1837.2	10107.24444
		%	
Difference TCOD	611.1111111	5	
% COD Solubilisation	16		

40% Sonication for 60 minutes (Sludge 19 February 2015 / 9.30 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	47	11750	11900
		2	47.5	11875	
		3	48.3	12075	
	Supernatant	1	17	17	18.3333333
		2	19	19	
		3	19	19	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	53	13250	13500
		2	55	13750	
		3	54	13500	
	Supernatant (Dilution 1:2)	1	989	1978	1987.33333
		2	997	1994	
		3	995	1990	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	51.7	12925	12975
		2	51	12750	
		3	53	13250	
	Supernatant (Dilution 1:2)	1	984	1968	1965.33333
		2	979	1958	
		3	985	1970	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	53	13250	13400
		2	55	13750	
		3	52.8	13200	
	Supernatant (Dilution 1:2)	1	995	1990	1998.66667
		2	998	1996	
		3	1005	2010	

	TCOD	**SCOD**	ssCOD
Non-Sonicated	11900	18.33333333	11881.66667
Sonicated	13291.66667	1983.777778	11307.88889
		%	
Difference tcod	1391.666667	12	
% COD Solubilisation	17		

40% Sonication for 90 minutes (Sludge 20 February 2015 / 10.00 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	48	12000	12166.6667
		2	49	12250	
		3	49	12250	
	Supernatant	1	18	18	18.3333333
		2	19	19	
		3	18	18	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	57	14250	14583.3333
		2	60	15000	
		3	58	14500	
	Supernatant (Dilution 1:2)	1	1004	2008	2020
		2	1012	2024	
		3	1014	2028	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	58	14500	14833.3333
		2	61	15250	
		3	59	14750	
	Supernatant (Dilution 1:2)	1	1009	2018	2017.33333
		2	1006	2012	
		3	1011	2022	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	60.5	15125	15083.3333
		2	61	15250	
		3	59.5	14875	
	Supernatant (Dilution 1:2)	1	1018	2036	2036.66667
		2	1021	2042	
		3	1016	2032	

	TCOD	**SCOD**	**ssCOD**
Non-Sonicated	12166.66667	18.33333333	12148.33333
Sonicated	14833.33333	2024.666667	12808.66667
		%	
Difference TCOD	2666.666667	22	
% COD Solubilisation	17		

40% Sonication for 120 minutes (Sludge 27 February 2015 / 9.00 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	45	11250	11250
		2	46	11500	
		3	44	11000	
	Supernatant	1	16	16	15
		2	14	14	
		3	15	15	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	55	13750	14000
		2	57	14250	
		3	56	14000	
	Supernatant (Dilution 1:2)	1	1021	2042	2037.33333
		2	1019	2038	
		3	1016	2032	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	56	14000	13750
		2	54	13500	
		3	55	13750	
	Supernatant (Dilution 1:2)	1	1010	2020	2015.33333
		2	1004	2008	
		3	1009	2018	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	55	13750	13667
		2	57	14250	
		3	52	13000	
	Supernatant (Dilution 1:2)	1	1008	2016	2023.33333
		2	1011	2022	
		3	1016	2032	

	TCOD	**SCOD**	**ssCOD**
Non-Sonicated	11250	15	11235
Sonicated	13805.55556	2025.333333	11780.22222
		%	
Difference TCOD	2555.555556	23	
% COD Solubilisation	18		

40% Sonication for 150 minutes (Sludge 19 March 2015 / 9.00 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	47	11750	11750
		2	48	12000	
		3	46	11500	
	Supernatant	1	17	17	18
		2	19	19	
		3	18	18	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	62	15500	15083.3333
		2	60	15000	
		3	59	14750	
	Supernatant (Dilution 1:2)	1	1043	2086	2085
		2	1041	2082	
		3	1044	2088	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	62	15500	15250
		2	61	15250	
		3	60	15000	
	Supernatant (Dilution 1:2)	1	1042	2084	2081
		2	1039	2078	
		3	1041	2082	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	61	15250	15500
		2	62	15500	
		3	63	15750	
	Supernatant (Dilution 1:2)	1	1043	2086	2087
		2	1045	2090	
		3	1043	2086	

	TCOD	**SCOD**	**ssCOD**
Raw Non-Sonicated	11750	18	11732
Sonicated	15278	2085	13193

		%
Difference TCOD	3527.777778	30

% COD Solubilisation	18

40% Sonication for 180 minutes (Sludge 19 March 2015 / 1.00 pm)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	50	12500	12583
		2	50	12500	
		3	51	12750	
	Supernatant	1	21	21	20
		2	20	20	
		3	20	20	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	69	17250	17167
		2	70	17500	
		3	67	16750	
	Supernatant (Dilution 1:2)	1	1171	2342	2342
		2	1172	2344	
		3	1170	2340	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	70	17500	17417
		2	71	17750	
		3	68	17000	
	Supernatant (Dilution 1:2)	1	1174	2348	2349
		2	1174	2348	
		3	1175	2350	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	69	17250	17250
		2	70	17500	
		3	68	17000	
	Supernatant (Dilution 1:2)	1	1173	2346	2345
		2	1172	2344	
		3	1173	2346	

	TCOD	**SCOD**	ssCOD
Non-Sonicated	12583.33333	20.33333333	12563
Sonicated	17277.77778	2345.333333	14932.44444

		%
Difference TCOD	4694.444444	37

% COD Solubilisation	19

40% Sonication for 210 minutes (Sludge 20 March 2015 / 8.30 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	45	11250	11500
		2	47	11750	
		3	46	11500	
	Supernatant	1	16	16	16.6666667
		2	17	17	
		3	17	17	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	65	16250	16500
		2	66	16500	
		3	67	16750	
	Supernatant (Dilution 1:2)	1	1136	2272	2272
		2	1135	2270	
		3	1137	2274	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	65	16250	16333.3333
		2	66	16500	
		3	65	16250	
	Supernatant (Dilution 1:2)	1	1138	2276	2276.66667
		2	1138	2276	
		3	1139	2278	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	66	16500	16583.3333
		2	68	17000	
		3	65	16250	
	Supernatant (Dilution 1:2)	1	1137	2274	2272.66667
		2	1137	2274	
		3	1135	2270	

	TCOD	**SCOD**	ssCOD
Non-Sonicated	11500	16.66666667	11483.33333
Sonicated	16472.22222	2273.777778	14198.44444

		%
Difference tcod	4972.222222	43

% COD Solubilisation	20

40% Sonication for 240 minutes (Sludge 21 March 2015 / 9.00 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor	Average
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	50	12500	12416.6667
		2	49	12250	
		3	50	12500	
	Supernatant	1	15	15	14.6666667
		2	14	14	
		3	15	15	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	73	18250	18250
		2	72	18000	
		3	74	18500	
	Supernatant (Dilution 1:2)	1	1284	2568	2572.66667
		2	1287	2574	
		3	1288	2576	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	74	18500	18500
		2	75	18750	
		3	73	18250	
	Supernatant (Dilution 1:2)	1	1289	2578	2577.33333
		2	1289	2578	
		3	1288	2576	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	73	18250	18083.3333
		2	72	18000	
		3	72	18000	
	Supernatant (Dilution 1:2)	1	1287	2574	2575.33333
		2	1287	2574	
		3	1289	2578	

	TCOD	**SCOD**	**ssCOD**
Non-Sonicated	12416.66667	14.66666667	12402
Sonicated	18277.77778	2575.111111	15702.66667

		%
Difference TCOD	5861.111111	47

% COD Solubilisation	21

40% Sonication for 270 minutes (Sludge 21 March 2015 / 2.00 pm)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	51	12750	12833
		2	51	12750	
		3	52	13000	
	Supernatant	1	19	19	18.6666667
		2	18	18	
		3	19	19	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	77	19250	19167
		2	77	19250	
		3	76	19000	
	Supernatant (Dilution 1:2)	1	1411	2822	2821.33333
		2	1411	2822	
		3	1410	2820	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	77	19250	19500
		2	79	19750	
		3	78	19500	
	Supernatant (Dilution 1:2)	1	1409	2818	2821.33333
		2	1411	2822	
		3	1412	2824	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	78	19500	19417
		2	78	19500	
		3	77	19250	
	Supernatant (Dilution 1:2)	1	1412	2824	2822
		2	1411	2822	
		3	1410	2820	

	TCOD	**SCOD**	**ssCOD**
Non-Sonicated	12833.33333	18.66666667	12814.66667
Sonicated	19361.11111	2821.555556	16539.55556

		%
Difference TCOD	6527.777778	51

% COD Solubilisation	22

40% Sonication for 300 minutes (Sludge 23 March 2015 / 8.30 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	44	11000	11000
		2	44	11000	
		3	44	11000	
	Supernatant	1	13	13	13.6666667
		2	14	14	
		3	14	14	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	67	16750	17000
		2	68	17000	
		3	69	17250	
	Supernatant (Dilution 1:2)	1	1290	2580	2580.66667
		2	1291	2582	
		3	1290	2580	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	68	17000	16833
		2	67	16750	
		3	67	16750	
	Supernatant (Dilution 1:2)	1	1290	2580	2582.66667
		2	1292	2584	
		3	1292	2584	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	66	16500	16583
		2	67	16750	
		3	66	16500	
	Supernatant (Dilution 1:2)	1	1292	2584	2583.33333
		2	1292	2584	
		3	1291	2582	

	TCOD	**SCOD**	**ssCOD**
Non-Sonicated	11000	13.66666667	10986.33333
Sonicated	16805.55556	2582.222222	14223.33333

		%
Difference TCOD	5805.555556	53

% COD Solubilisation	23

40% Sonication for 330 minutes (Sludge 24 March 2015 / 11.00 am)					
		Trial	COD Read (mg/L)	COD * Dilution Factor (mg/L)	Average (mg/L)
Non-Sonicated (50 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	46	11500	11500
		2	46	11500	
		3	46	11500	
	Supernatant	1	15	15	14.6666667
		2	14	14	
		3	15	15	
1st Trial (113 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	71	17750	17833
		2	71	17750	
		3	72	18000	
	Supernatant (Dilution 1:2)	1	1371	2742	2742.66667
		2	1372	2744	
		3	1371	2742	
2nd Trial (109 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	71	17750	17833
		2	72	18000	
		3	71	17750	
	Supernatant (Dilution 1:2)	1	1371	2742	2742
		2	1370	2740	
		3	1372	2744	
3rd Trial (118 minutes Settling Time)	**Particulate** (Dilution 1:250)	1	71	17750	17667
		2	70	17500	
		3	71	17750	
	Supernatant (Dilution 1:2)	1	1373	2746	2743.33333
		2	1371	2742	
		3	1371	2742	

	TCOD	**SCOD**	**ssCOD**
Non-Sonicated	11500	14.66666667	11485.33333
Sonicated	17777.77778	2742.666667	15035.11111

		%
Difference TCOD	6277.777778	55

% COD Solubilisation	24