

**Production of biogas from crude glycerol using ultrasound as
pretreatment in a batch reactor**

Lis Geraldine Zschach

Thesis report submitted to
Escola Superior de Tecnologia e de Gestão
Instituto Politécnico de Bragança

Master degree in
Chemical Engineering

Supervisors:

Professor Doctor Ramiro José Espinheira Martins

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*To my family,
the motor of my life*

ACKNOWLEDGEMENTS

First of all, I would like to thank to God, to set my path always better than I expected. To take care of me and my beloved people.

Thanks to my supervisors for the guidance. Prof. Ph.D. Ramiro. J. E. Martins of Department of Chemical and Biological Technology, Superior School of Technology (ESTIG), Polytechnic Institute of Bragança (IPB) for the opportunity and trust given as well as all the knowledge in this area. Prof. Ph. D. Cristiane Kreutz of Environmental Department Academic, The Federal University of Technology – Paraná, Campo Mourão, Paraná, Brazil, especially for all the dedication, help, patience, support and love given, you had been a second mother for me in all this time.

This experience would not have been the same without the help, company and support of my partner and friend Mariana Ferrari Giacon. Thank you for all the time shared.

A special thanks to the Eng. Maria João Alfonso of the Chemical Process Laboratory (ESTIG-IPB), for all the help, patience and love provided.

All my gratitude to my National University of Misiones (UNaM) to allow me to have this amazing experience. Thanks to all the people involved in this agreement, specially to Prof. Ph.D. Claudia Méndez from Physicochemical Department of the Faculty of Exact, Chemical and Natural Science (FCEQyN) of the UNaM for all the support and dedication provided to this agreement. Thank you Ph.D. C. Méndez for helping me to do my firsts steps in the field of research, for all the knowledge transmitted, all the opportunities given as well as your dedication. I will always be grateful to you. Thanks to all the professors and fellows of the Laboratory of Materials of the FCEQyN for the support given in the science field and specially to the Ph.D Alicia Ares.

Thanks to my friends from the UNaM, for supporting and believing in me, I didn't feel the distance thanks to you. Thanks to the first great friends that the UNaM gave me, Belén, Marian, Marce, Mauri, David, Noe, Melanie, Ayelén and Mane.

For the company and for supporting me through the process, thanks to all my international friends of Bragança. You have been my family this all year. Brazil, Mexico, Russia, Georgia, France and Romania have pieces of my heart. Thanks specially to my closest friends for all the support and love given, Filipe, Agustina, Mariia, Andrea and Ksenia.

Finally, thanks to my all my family for the all the support and love given, thanks to push me every day to reach my objectives. All my life would not be the same without the company and unconditional love of my brothers, thank you. Thanks to my cousins and grandparents for being part of my life and giving me so much love.

Thanks to my Dad, Eng. Carlos Zschach, my inspiration.

This work was financially supported by the project UID/EQU/50020/2019 Associate Laboratory LSRE-LCM- funded by national funds through the Foundation for Science and Technology (FCT) and the Ministry of Science, Technology and Higher Education (MCTES) (PIDDAC).



ABSTRACT

Global energy supply is based on petroleum derivative. Scarcity of natural sources and environmental concerns are problems associated to the energy supply as well as its increasing demand. These problems can be solved by using more renewable sources.

Biofuels as biogas have the potential to provide energy and decrease environmental emissions. Studies made before, says that crude glycerol from biodiesel production process can be used to produce biogas by anaerobic digestion process. In order to improve biogas volume and its quality, pretreatments to the substrate are being studied.

The aim of this work is to study the effect of ultrasound, a mechanical pretreatment, in crude glycerol to produce biogas. The volume of biogas and methane produced was measured, the last to evaluate the quality of biogas, to assess the main effect of pretreatment on anaerobic digestion. They were carried out in batch reactors under mesophilic conditions (37 ° C). Different pretreatment times with ultrasound (15 and 30 minutes) of glycerol treatment were tested and compared with untreated glycerol. For biogas production, measurement was made by moving with its pressure the plunger of a syringe. Methane production by washing the biogas in sodium hydroxide (3%). Characterization tests were also performed on the inoculum and the substrate. As an inoculum, the activated sludge from septic tanks in the rural area of Bragança was used. Substrate was the crude glycerol from biodiesel production process of the Biofuels Laboratory of the School of Technology and Management of IPB.

Results shows the maximum production of biogas of 2012 ml that was produced by the crude glycerol pretreated with 30 minutes of ultrasound. It was followed by the one that had 15 minutes of pretreatment with a performance of 1356.6 ml of biogas against the untreated one that presented 1010.5 ml. Measurements of methane volume showed that the 30 minutes treatment had the maximum performance of 13184.7 ml of methane. With this time of pretreatment was performed the mesoscale, in a system with a 7 l reactor built under mesophilic conditions. A mixing and a heating system were connected to the reactor and pH adjusted. Biogas was washed in sodium hydroxide and methane production was daily measured. It achieved 16224 ml of methane. Therefore, it can be concluded that ultrasound treatment improves biogas production.

Keywords: biogas, crude glycerol, ultrasound pretreatment, anaerobic digestion.

RESUMO

O suprimento global de energia é baseado em derivados de petróleo. A escassez de fontes naturais e as preocupações ambientais são problemas associados ao suprimento de energia e à crescente demanda. Esses problemas podem ser resolvidos usando mais fontes renováveis.

Os biocombustíveis como biogás têm o potencial de fornecer energia e diminuir as emissões ambientais. Estudos feitos anteriormente afirmam que o glicerol bruto do processo de produção de biodiesel pode ser usado para produzir biogás pelo processo de digestão anaeróbica. Para melhorar o volume de biogás e sua qualidade, estão sendo estudados pré-tratamentos para o substrato.

O objetivo deste trabalho é estudar o efeito do ultrassom, um pré-tratamento mecânico, no glicerol bruto para produzir biogás. O volume de biogás e metano produzido foi medido, o último a avaliar a qualidade do biogás, para avaliar o principal efeito do pré-tratamento na digestão anaeróbica. Eles foram realizados em reatores descontínuos em condições mesofílicas (37 ° C). Diferentes tempos de pré-tratamento com ultra-som (15 e 30 minutos) de tratamento com glicerol foram testados e comparados com glicerol não tratado. Para a produção de biogás, a medição foi feita movendo com sua pressão o êmbolo de uma seringa. Produção de metano lavando o biogás em hidróxido de sódio (3%). Também foram realizados testes de caracterização no inóculo e no substrato. Como inóculo, foi utilizado o lodo ativado de fossas sépticas na área rural de Bragança. O substrato foi o glicerol bruto do processo de produção de biodiesel do Laboratório de Biocombustíveis da Escola de Tecnologia e Gestão do IPB.

Os resultados mostram a produção máxima de biogás de 2012 ml produzida pelo glicerol bruto pré-tratado com 30 minutos de ultra-som. Seguiu-se aquele que realizou 15 minutos de pré-tratamento com desempenho de 1356,6 ml de biogás contra o não tratado que apresentou 1010,5 ml. Medições do volume de metano mostraram que o tratamento de 30 minutos teve o desempenho máximo de 13184,7 ml de metano. Com esse período de pré-tratamento foi realizada a mesoescala, em um sistema com um reator de 7 l construído em condições mesofílicas. Uma mistura e um sistema de aquecimento foram conectados ao reator e o pH ajustado. O biogás foi lavado em hidróxido de sódio e a produção de metano foi medida diariamente. Atingiu 16224 ml de metano. Portanto, pode-se concluir que o tratamento com ultrassom melhora a produção de biogás.

Palavras-chave: biogás, glicerol em bruto, pré-tratamento de ultra-sons, digestão anaeróbica.

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INTRODUCTION



INTRODUCTION

Energy demand is expected to be increased as population grows. It has been growing 2% each year. Nowadays energy supply is based mainly in petroleum derivative. Studies made on this source, suggest that its use causes several environment concerns as air pollution, acid precipitation and gas emissions to the atmosphere, in addition the environmental impact caused by its exploitation [1]. In the interest of solving energy supply problems, the changes involved are to create energy savings on the demand side, to improve the efficiency of its production process and to replace petroleum derivative by renewable energy. Less than 15% of global scale of primary energy corresponds to renewable energy. The main part is hydro and wood power. Solar, wind and biomass as biofuels are a small portion [2]. Biofuels are expected to increase in percentage [3].

The biodiesel is a renewable energy source considered alternative to the Diesel. It is produced from vegetal oils, animal fats and used oils and fats. Biodiesel world production is expected to increase. By increasing its production, the quantity of its main by-product, crude glycerol, will increase [3]. For each 100 kg of biodiesel produced, 10 kg of crude glycerol are produced too [4]. The refining of crude glycerol to have application in chemical industry is expensive so others alternatives of application had been evaluated. Studies made suggest that crude glycerol can be used in an anaerobic digestion process in order to get biogas. This can represent an energy supply problem solution [4,5].

Normally, crude glycerol is used in co-digestion process to reduce its retention time and achieve a major biogas volume. For reducing this time and improve the quantity and quality of biogas, pretreatments as ultrasound are applied on the substrate that is going to be submitted to anaerobic digestion [6, 7]. The objective of pretreatments is to degrade organic matter in order to have more accessible molecules for microorganisms to carry on the anaerobic digestion. The first step of anaerobic digestion, hydrolysis, has a special attention because this is the step that causes rate-limitation on the process. This leads to longer retention times. Pretreatments could solubilize the organic matter in order to obtain less retention time [8,9].

Ultrasound is a cycle sound pressure frequency. It is a mechanical method to solubilize the organic matter by vibration. It causes the increase of the temperature and the pressure in the liquid [10]. By these properties, it has been used as a mechanical pretreatment in production of biogas [6].

Biogas produced by anaerobic digestion contains around of 55 to 65% of methane, 30 to 40% of carbon dioxide, fractions of water vapor, hydrogen sulfide and ammonia. Percentage of methane is its quality index [11]. Biogas can be used production of heat and steam, fuel uses, production of chemicals or electricity generation [12]. It can be used for heating using boilers to produce water vapor for industrial plants. Heating can be used in the process line and the remaining electricity can be sold to the national grid [11].

The aim of this work is to study the effect of ultrasound pretreatment in crude glycerol to produce biogas.

General Objective

To produce biogas using ultrasound as pretreatment from crude glycerol as substrate in an anaerobic digestion process.

Specific Objectives

- Characterize crude glycerol from biodiesel production process.
- Characterize the activated sludge used as inoculum.
- Evaluate the percentage of crude glycerol to be used in the digestion.
- Evaluate the effect of pretreatment with ultrasound in the biogas production.
- Testing the process to a mesoscale with a mixed flow reactor.

STATE OF ART



STATE OF ART

1. World's energy supply problem

As world population grows in a 2% year by year, lifestyle and energy demand rise together. The industrialized economies that contains 25% of world's population, consumes 75% of world's energy supply [1].

World's energy supplied is based on petroleum derivative as coal, oil and gas, nuclear sources and renewable energy sources (RES). Petroleum derivative is the principal source of energy nowadays and it is considered the conventional energy [2].

Actual problems in energy supply are related to the global warming and the scarcity of natural resources. Environment concerns that are related to energy conventional production are air pollution, acid precipitation, ozone depletion, deforestation, and the gaseous emissions to the atmosphere [1]. When fuel combustion takes place, exhaust gases are released to the environment. Exhaust gas emission contribute to acid rain and the global accumulation of carbon dioxide (CO₂) contributing to the greenhouse effect. Each one of them has serious consequences for human health and the environment [1].

In the last years, because of the petroleum deficit and the contamination that using petroleum-based fuels causes, man has been looking for alternative sources of energy to replace it [3]. Sustainable energy development strategies involve to have energy savings on the demand side, more efficiency in the energy production and the replacement of fossil fuels by renewable energy. RES are expected to keep on increasing as it is showed in Figure 1 [2].

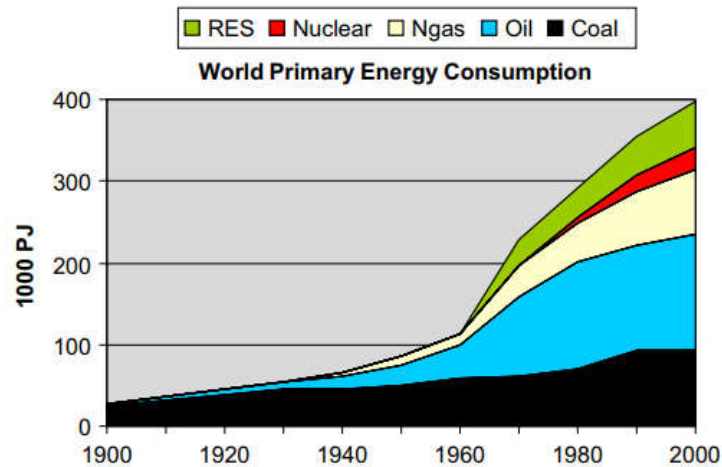


Figure 1. World primary energy consumption

Lund, H. (2007). Renewable energy strategies for sustainable development. *Energy*, 32(6), pp 912-919.

The rising use of renewable energy and the energy efficiency were important not only for reducing the climate change, but also for creating new economic opportunities. Renewable energy programs reach rural areas that are outside the conventional electricity grid. In 2014 there was a carbon stabilisation attributed to increment of the use of renewable energy and its efficiency [13].

An alternative to solve energy supplied problems are the biofuels [3]. The production of biofuels for transport presented an annual growth from 11 to 13% in 2014. They are used in electricity generation, heating or as fuel. Biofuels can be gaseous as syngas and biogas or liquid as biodiesel and bioethanol. The main biofuels produced are biodiesel and bioethanol. They are used mainly used for passenger vehicles and heavy-duty road vehicles. In European countries, the Unites States and Brazil, the fuel used in road vehicles exceeded the 20% in 2014. After liquid fuels, gaseous biofuels increased small quantities [13].

Biodiesel is considered a cleaner burning diesel replacement fuel produced from natural and renewable sources. It is an oxygenated fuel so it hence a more complete combustion [3].

According to [3] causes less formation and emission of particles showing a lower opacity of smoke compared to the conventional fuel. This biofuel works in compression-ignition or Diesel engines. Biodiesel has physical proprieties similar to the Diesel [3, 14].

2. Biodiesel

Biodiesel is a fatty acid methyl ester which is derived from triglycerides by transesterification with methanol. Is a biodegradable, non-explosive, non-flammable, renewable, and non-toxic substance. Its combustion generates low levels of greenhouse effect gases and particulate materials [3]. Is an efficient, clean and represents a natural energy alternative [14].

Biodiesel reduces emissions of solids particles less than 10 microns, carbon monoxide and sulfur oxides and dangerous pollutants. The Environmental Protection Agency (EPA) says that on vehicular emission with biodiesel, particulate matter is reduced in a 47% when using biodiesel and carbon monoxide by 48% [3].

Characterization of this biofuel is determined by its density, viscosity, high heating value, cetane number, cloud and pour points, flash and combustion points and its characteristics of distillation. Comparison between biodiesel and Diesel fuels shows that the first one has better values in aromatic content, flash point and sulfur content. Analysing the environmental factors, biodiesel has a better biodegradability [14]. Physical characteristics of biodiesel compared to Diesel are resumed in Table 1.

Table 1. Characteristics of biodiesel vs Diesel

Property	Biodiesel	No 2. Diesel
Specific gravity (kg/L)	0.87–0.89	0.84–0.86
Cetane number	46–70	47–55
Cloud point (K)	262–289	256–265
Pour point (K)	258–286	237–243
Flasf point (K)	408–423	325–350
Sulfur (wt%)	0.0000–0.0024	0.04–0.01
Ash (wt%)	0.002–0.01	0.06–0.01
Iodine number	60–135	–
Kinematic viscosity, 313 K	3.7–5.8	1.9–3.8
Higher heating value, MJ/kg	39.3–39.8	45.3–46.7

Bozbas, K. (2008). Biodiesel as an alternative motor fuel: Production and policies in the European Union. *Renewable and Sustainable Energy Reviews*, 12(2), pp 542-552.

Advantages of biodiesel are (1) reduction of engine maintenance so it prolongs engine life because comparing with Diesel has better lubricating properties; (2) safer to handle because is less toxic than Diesel and has higher flash point; (3) it is more biodegradable; (4) reduces exhaust emissions; (5) has the same performance in the engine than the Diesel and (6) has visible smoke [14].

Some disadvantages are that in cold weather this biofuel has not good performances and has a reduced energy density. When storage in prolonged periods, biodiesel degrades [14].

2.1 Biodiesel's world production

In 2014 the renewable energy continued to grow despite the increase in global energy consumption and the fall in oil prices during the second part of the year. Global energy consumption increased 1.5% due to the growing demand in the developing countries. Against this, because of the renewable energy grow, global carbon emissions associated with energy consumption stabilized in the same year [12].

In recent years, the renewable energy supplied was approximately 19.1% of the global final energy, as shown in Figure 2 [13].

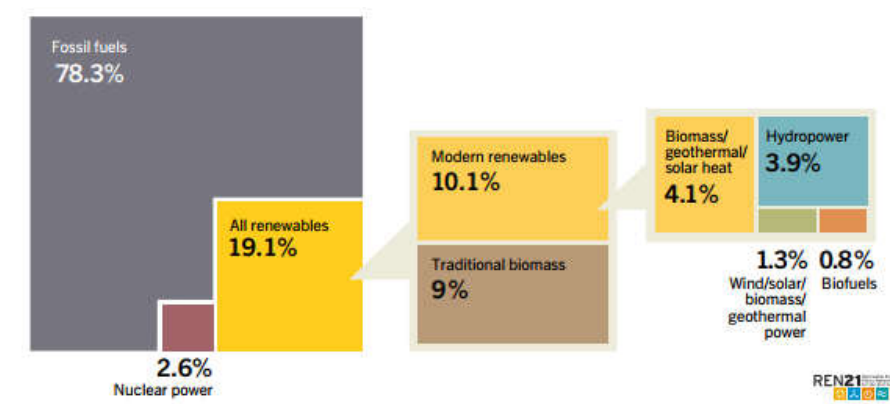


Figure 2. Estimated renewable energy of global final consumption

REN21. Renewable Energy Policy Network for the 21st Century. (2015). Annual Reporting on Renewables: Ten years of excellence.

This energy is being destined in power generation, heating and cooling, transport and energy supply of rural areas. Within this total, the biomass used for cooking and heating is approximately 9% [13].

New policies instead of market forces are encouraging biodiesel production almost in the entire world. Therefore, global biodiesel production is expected to increase as seen in Figure 3 [12,15].

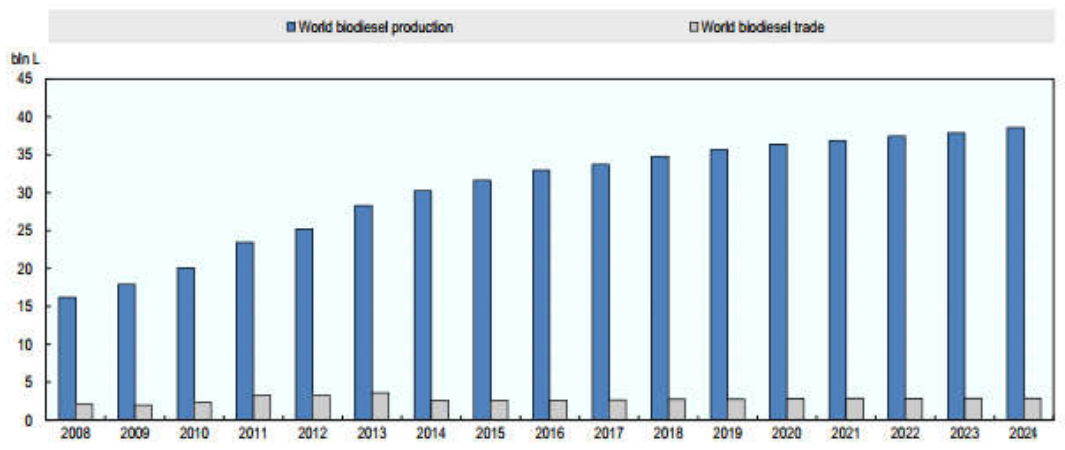


Figure 3. World biodiesel production and trade.

OECD/FAO (2015), “OECD-FAO Agricultural Outlook”, OECD Agriculture statistics (database), <http://dx.doi.org/10.1787/agr-outl-data-en>

Global biodiesel production is expected to increase in a 27% from 2014. Studies made determined that biodiesel production is expected to reach 39 billion litres (Bln L) by 2024. Also, is expected the European Union to be the major producer as shown in Figure 4 [15].

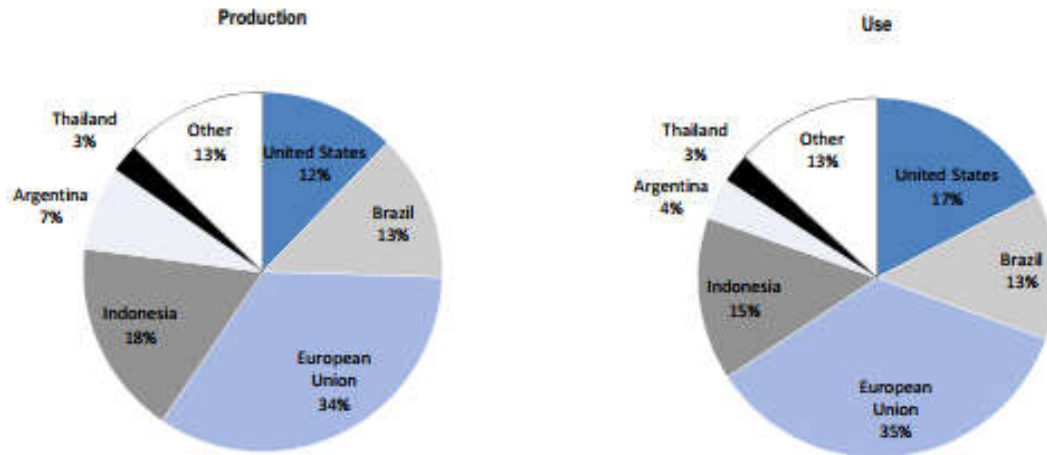


Figure 4. Biodiesel production and percentage of use in the main producing countries.

OECD/FAO (2015), “OECD-FAO Agricultural Outlook”, OECD Agriculture statistics (database), <http://dx.doi.org/10.1787/agr-outl-data-en>

The European Union, considered the number one in biofuels production, registered an increase in its production with Germany leading the region. In this region, biodiesel use is projected to increase to its highest level in 2020 with 13.6 Bln L. By 2024 expected to decline to 13.1 Bln L due lower demand [15].

The United States is supposed to lose position after the second largest biodiesel producer in the next ten years. Its production is intended to be constant at 4.8 Bln L. Canada saw a marginal increase in biofuels production [15].

Brazil expanded its production in biodiesel, as well as others Latin American countries such as Argentina, that in response of the incentives of its government jumped 28%, and Colombia. Brazil is expected to become the third largest biodiesel producer to satisfy the domestic demand. In 2024 is expected to rise up 5 Bln L. For Argentina, the domestic and international production demand must be satisfied with the increase expected in their production up to 1.4 Bln L [15].

Asian countries as China, Indonesia and Thailand increased their production by 16%, continuing being at the top of the region. It is projected that Indonesia will continue to meet its demand and be an important biodiesel exporting country. It will increase from 2 Bln L in 2012 to 5.6 Bln L in 2024. Biodiesel represented a way to add value to the palm oil produced, reducing the greenhouse gas emissions and the diesel imports in this country [15].

2.2. Production Process

2.2.1 Raw materials

Oils and fats are known as triglycerides, tri esters formed from three molecules of higher fatty acids and a molecule of propanetriol. Oils in general are triglycerides sources for the production of biodiesel. Peanut, soya bean, babassu, palm, sunflower, canola or corn oils could be used [6, 16].

Fatty acids are linear organic acids that are different in their carbon number and the amount of double bonds between their carbon atoms in the hydrophobic chain, or for the presence of some functional group in their carbon chain. The difference between oils and fats is that at room temperature, fats are solids and oils are liquids [17].

It is estimated that 70 to 95% of biodiesel production costs are due to the raw materials. The residual oil of the domestic, commercial or food industries are raw material for biodiesel. These residual oils represent low cost raw materials. Using them means cooperating with recycling, so it is friendly to the environment. In addition to these, oil extraction process is avoided, making it a technical advantage. Even though they are partially oxidized, they show similar characteristics to refined oils esters [17].

2.2.2 Production process

Many process for biodiesel fuel production have been developed. The most commonly used is the transesterification using alkaline catalysis because of its short reaction times and high levels of conversion of the triglycerides in their methyl esters [3]. The transesterification process is described in Figure 5 below.

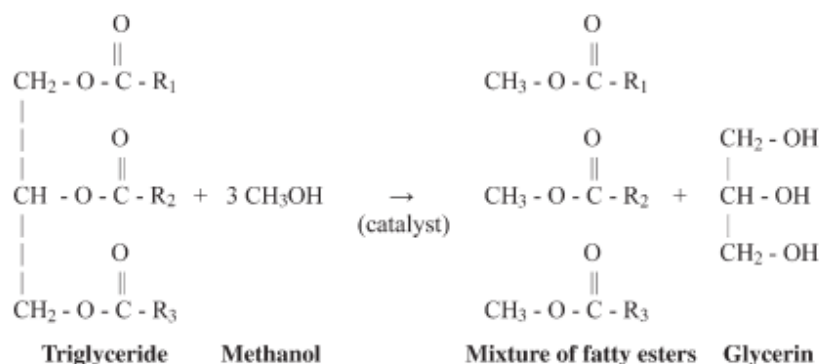


Figure 5. Transesterification process

Gerpen, J. (2005). Biodiesel processing and production. *Fuel Processing Technology*, 86(10), pp 1097-1107.

Sodium hydroxide (NaOH) or potassium hydroxide (KOH) could be used as alkaline catalyst. Methanol or ethanol and any kind of refined, crude or residual oil are the reagents of the reaction. The amount of catalyst varies from 0.5% to 1% w/w [18].

The acid catalyst is the second conventional method of biodiesel production. Sulfuric acid is commonly used in this method. It gives high yield in esters but the reaction is slow. The recovery of glycerol is difficult and the relationship between alcohol and raw material must be determined experimentally, resulting a disadvantage for the process [18].

Enzymatic transesterification using lipase is the production system that has become interesting in the recent years for industries, because the glycerol produced can be easily recovered. In addition, it has a simple purification of fatty methyl esters. The cost of lipase production is a disadvantage for this system, so the use of immobilized cells biocatalysts within biomass support particles represents a method to reduce these costs. The reaction is completed spontaneously during culture and no purification is needed [3].

The production process begins when alcohol, catalyst and oil are mixed in stirred reactor. The small plants use batch reactors; larger plants use continuous processes. In the next step, glycerol is separated from methyl esters in settling tanks or centrifuges. The methyl esters go to the step of methanol removal to be purified and methanol to be recycled [19].

To be used as a fuel, biodiesel requires several treatments to reduce its high viscosity, and high content of fatty acids that may cause gum formation, carbon deposits or polymerization thereof during the combustion process or storage [3]. Biodiesel pass through the last step of neutralization and washing before going to the dryer and being ready [19].

The most important variables in biodiesel production process are the reaction temperature, the intensity of mixture, the ratio of alcohol to vegetable oil, the type of oil used, the catalyst and amount of catalyst [18].

The principal by-product of biodiesel production process is the glycerol (known also as glycerine). 10% w/w of processed oil is converted into crude glycerol. As biodiesel production continues to grow, the amount of glycerol also. Many methods are being developed to add value to this product [20]. Glycerol has water, salts, soaps and other waste so it must be purified. After this process, it can be used in pharmaceutical, cosmetic industry, as a lubricant, solvent for varnishes and other chemical industries, and for biogas production [21]. The biodiesel production process is described in Figure 6.

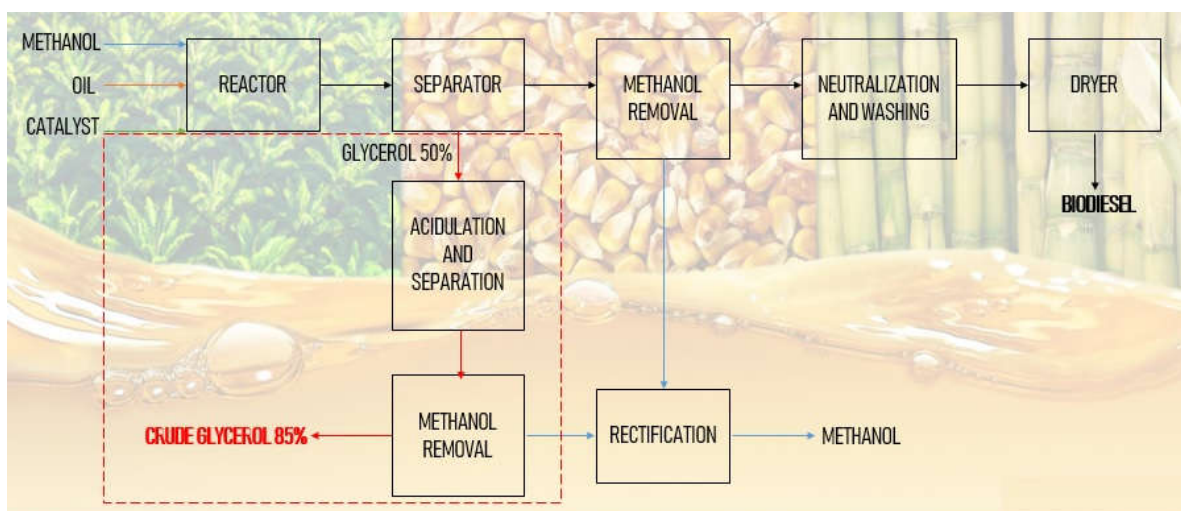


Figure 6. The biodiesel production process

3. Glycerol

Glycerol is 1,2,3-propanetriol, a colourless, odourless and viscous substance. It is considered a non-toxic alcohol. Pure glycerol with a purity over 95% can be used in chemical industry. When used in an anaerobic process, the presence of microorganisms capable of forming acetate, hydrogen and carbon dioxide is necessary. As the inoculum is used generally activated sludge [5].

The crude glycerol obtained in the biodiesel production process is usually 50% pure. The other part is basically fatty acids such as sodium and potassium soaps. Despite this, crude and pure glycerol has a similar organic matter in terms of COD, content of almost 1 kg/L. COD and others properties of crude and pure glycerol are visible in Table 2 [22].

Table 2. Properties of crude and pure glycerol.

	Crude	Pure
Appearance	Pale brown	Clear
pH	8–9	6.7
Boiling point (°C)	>130	290
Na content (mg/L)	16,939	0
K content (mg/L)	454	0
Specific gravity (@ 25 °C)	1.25	1.26
Glycerol content (%)	~ 80	≥99
Matter organic non-glycerol (%)	<20	–
COD (Kg/L)	1.14	1.05

Nghiem, L., Manassa, T., Nguyen, P., Fitzgerald, S., Dawson, M., & Vierboom, S. (2014). Co-digestion of sewage sludge and crude glycerol for on-demand biogas production. *International Biodeterioration & Biodegradation*, 95, pp 160-166.

The production of methane from glycerol anaerobic digestion has several advantages (1) involves low nutrients requirements, (2) energy savings and (3) generation of a stabilized digestate that can be used in composting as fertilizer. Glycerol can be stored in long periods of time without suffering any changes in its composition. It offers a high volume production of biogas in smaller reactor volumes, does not requires large investments and it the process is simple to operate, so it is good for local applications [4].

4. Anaerobic digestion

Anaerobic digestion is a process of decomposition in which the organic matter degrades into simpler chemical components. The whole process occurs in the absence of oxygen that leads to methane (CH_4) and carbon dioxide (CO_2) production and a smaller proportion of hydrogen sulfide (H_2S) and ammonia (NH_3) in addition to small amounts of other gases [16]. Five groups of bacteria are involved in this process working in an assembly group. Each derives energy from its biochemical reactions [23].

Basically, anaerobic digestion could be studied in four different steps: hydrolysis, acidogenesis, acetogenesis and methanogenesis (Figure 7).

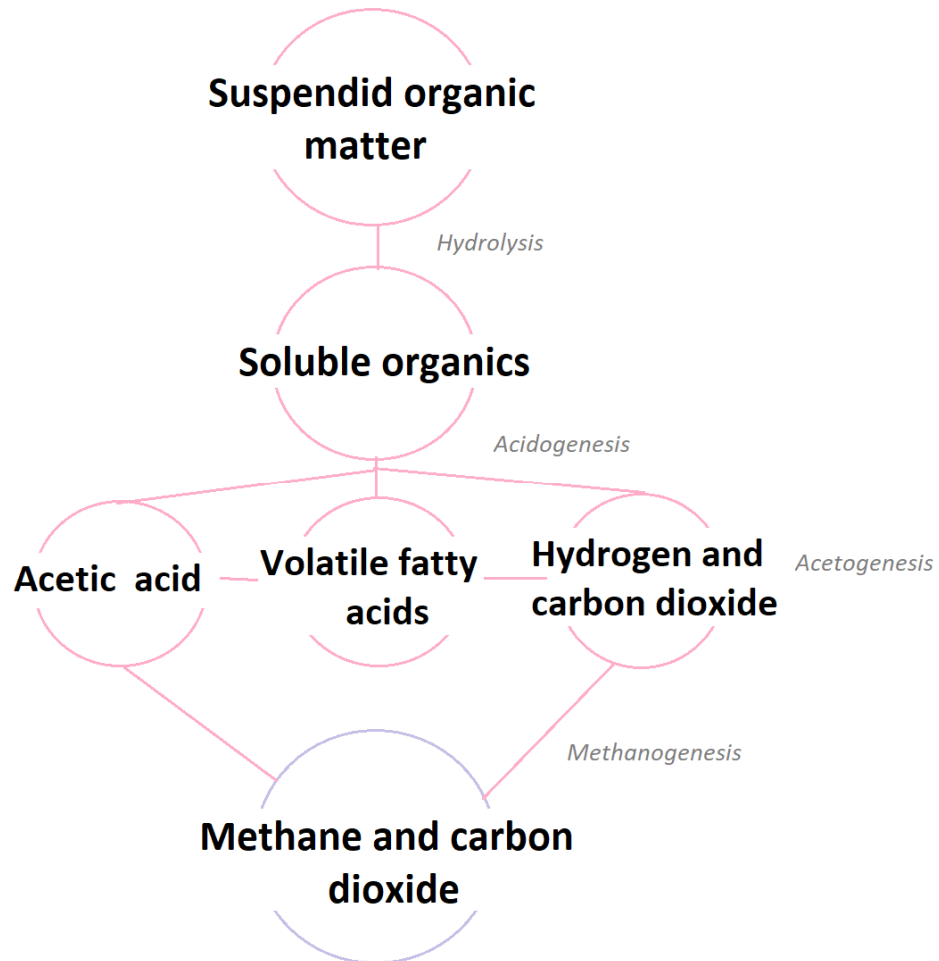


Figure 7. Steps of the anaerobic digestion process.

-Hydrolysis: in this step insoluble organic material and high molecular material are degraded. These compounds can be lipids, polysaccharides, proteins and nucleic acids [12]. By the action of bacteria, lipids are converted into fatty acids, polysaccharides into monosaccharides, proteins in amino acids and the nucleic acids in purines and pyrimidines. This stage is the most important in the production of biogas because it can become a rate limitation of the entire process. Many investigations have been carried out to find methods to enhance hydrolysis and with this, to reduce the residence time of digestion [24].

-Acidogenesis: the compounds formed in the previous step have a second split during acidogenesis. Volatile fatty acids are produced by fermentative bacteria as well as NH_3 , CO_2 , H_2S and other less important products [12].

-Acetogenesis: the higher organic acids and alcohols produced in acidogenesis are now reagents to produce essentially acetic acid along with CO_2 and hydrogen (H_2). The last gas is the one that controls the conversion of the reaction by its pressure [12].

-Methanogenesis: in the last stage of anaerobic digestion, methane-forming bacteria produce methane. These bacteria are divided in two groups, the first one splits acetate to form CH_4 and CO_2 . The second group produces CH_4 from taking H_2 as an electron donor and CO_2 as electron acceptor [12].

This process requires strict anaerobic conditions to be carried on. It depends on coordinated series of complex microbial group to transform the main organic matter into methane [12].

4.1 Control Parameters:

To obtain the major quantity of methane, some parameters must be well controlled. These parameters must be controlled from the digester during the residence time and before. Some of them are:

-Total solids content: it could be separated in three ranges according to the quantity in percentages: for a content below 10% is considered low solid, medium solid 15-20% and high solid 22-40%. It is important to determine this parameter because when it increases, the volume of digester decreases and this leads to minimize water requirements [11].

-Volatile solids: describes the biological conversion capacity of the anaerobic digestion process. Controlling the amount of volatile solids of the substrate avoids the accumulation of inhibitor substances in the digester and pH changes in organic matter [12,11].

-Temperature: influences the growth rate and metabolism of the microorganism so the population dynamic of microorganism inside the digester [12]. The selection of temperature depends on the process conditions. For mesophilic it is usually between 20-45°C and for thermophilic 50-65°C. The conditions will depend on the type of substrate and the digester to be used. The last one is the more efficient in retention time, loading rate and gas production but needs a higher temperature in the inlet so the process is more sensitive than the mesophilic. To maintain the rate of gas production, the temperature must be kept constant over time [11].

-Retention time: is the time that is needed to complete the transformation of the organic matter into methane. It depends on the temperature conditions of the digester. For mesophilic could be between 15 to 30 days and to thermophilic 12 to 14 days [11]. Other time parameters are the solids retention time (SRT) which is the average solids time inside the digester and the hydraulic retention time (HRT) which is the average liquid time in the digester. If SRT decreases, decreases the reactions extent [12].

-pH: the optimal range is 6.4 to 7.2. Lower than that can be toxic for methane-forming bacteria, that are the most sensitive group of bacteria involved in the anaerobic digestion process [11]. Fermentative bacteria are less sensitive, so they can work in a range of pH between 4.0 and 8.5. In lower values, the products are acetic and butyric acid and at higher products are acetic and propionic acids. During the anaerobic digestion, volatile fatty acids tend to reduce de pH and this is offset by the alkalinity produced by the methane-forming bacteria [12].

-Carbon to Nitrogen ratio: is the relationship between the quantity of carbon and nitrogen in the organic matter. This relationship indicates how fast nitrogen is consumed by the methane-forming bacteria. When this parameter increases, biogas production decreases. Normally it should be around 20 or 30 [11].

-Mixing: is one of the most important parameters to get attention. The mixture provides a greater area of contact between organic matter and microorganisms. In addition, prevents temperature gradients and variation in pH. An adequate mixture should be slow so as not to disturb the metabolism of decreases the but not cause scum formation [11].

4.2 General process and type of digesters

The anaerobic digestion process is generally divided into four stages: pretreatment, digestion, gas recovery and residue treatment. Depending on the substrate pretreatment is indicated. In pretreatment, the substrate is filtrated, homogenized and diluted. Then thermal, mechanical, chemical or biological pretreatment is applied on it. Normally, a heat exchanger is connected to the digestion vessel to keep the temperature constant within it [24]. The liquid is recycled for use in the initial dilution and the solids are treated for use as compost. The gas is stored in hermetic vessels until required. Depending on its subsequence use, the gas is subject to a purification process [11].

Digestion could be carried out in different types of digesters mainly separated into:

-Single-stage digester: it is a simple digester to operate that is efficient for use with sludge. The disadvantages are the high investment costs for the main reactor and for the necessary additional equipment. Commonly, a stirred tank reactor is used to ensure a good mixing system [11]

-Multi-stage digesters: represents an improvement to anaerobic digestion systems by using separated reactors for different stages. This system provides flexibility and optimization. Generally, two reactors are used. One to be carried out the hydrolysis, acidogenesis and acetogenesis and the other the methanogenesis. This allows hydrolysis to be taken care of in more detail to reduce its rate [11].

-Batch: it is a simple system where the digesters are filled once with the fresh substrate and allowed to complete the anaerobic digestion process. When the process is completed, the substrate is collected. There are three types of batch systems (Figure 8): single step, sequential system and the up flow anaerobic sludge blanket (UASB) reactor [11].

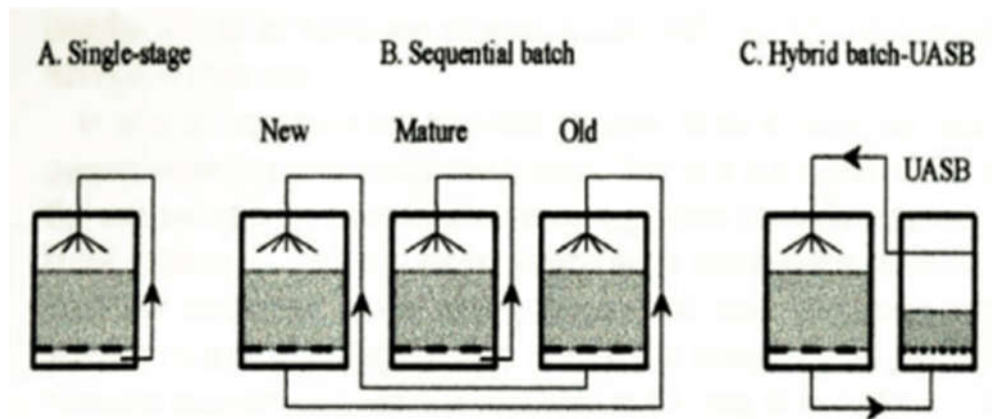


Figure 8. Types of batch reactors.

Vandevivere, P., De Baere, L., Verstraete, W. (1999) unpublished manuscript

The single-stage batch system has recirculation of the leachate at the upper part of the digester. Before feeding the substrate to the digester, it is mixed with the inoculum. When the biogas production stops, the reactor is empty. The sequential batch process involved two or more reactors where in the first part of the digestion process occurs in the first reactor and the methanogenesis in the second. The pH buffering agents are re-circulated to the first reactor so the pH continues into the convenient ranges. UASB reactors are similar to the sequential ones, but the second reactor is an upflow anaerobic sludge blanket reactor [24].

Batch reactors are technically simple, non-expensive and robust. The disadvantages are settlement of the material inside the digester and the complications that this can cause to the whole process [24].

5. Pretreatment

The slow degradation of the activated sludge is a disadvantage for anaerobic digestion processes and is the same to the glycerol. This leads to high retention times that imply significant space requirements for the reactors in which the digestion is getting through. During the hydrolysis, the combination of particle solubilisation and biological decomposition of organic polymers causes rate-limitation [8, 9].

Part of the organic matter is enclosed in bacterial flocs that do not allow microorganisms to degrade it [25]. This can be enhanced thus pretreatments to the substrate and the inoculum, in the studied case, sewage sludge [26].

Biological, thermal, chemical or mechanical pretreatments can be used. They led to rupture of the cell wall of the organic matter and the bacteria membrane, resulting cell lysis. All organic substances that were inside of the cell can be easily hydrolysed by decreasing the time of this step. As consequence, the retention time of the entire process decreases. [8,27]

-Biological: in this pretreatment different methods are used to disintegrate the organic matter with enzymatic reactions or by autolytic processes [10]. It consists of a wide range of processes that involves in-process and prior to anaerobic digestion sludge destruction. A process that combines a higher stage on thermophilic or hyper-thermophilic condition known as temperature phased anaerobic digestion is commonly used.

Thermophilic conditions increase the rate of disintegration of organic solids by increasing the hydrolytic activity [12]. Other methods that can be used are the composting or micro-aeration that produces superior hydrolytic enzymes [6].

-Thermal: are considered thermal pretreatments those that work using temperatures of 60 to 200°C, usually applied to the sludge by heat exchangers or by steam injections. This increase in temperature of sludge allows the degradation of the sludge gel structure so the water is released and the sludge is partially solubilized [27]. The tests performed showed an increment in the solubilisation of sludge as temperatures increased. For temperatures higher than 200°C there were formed compounds harder to dewater [6].

-Chemical: this type of pretreatments uses chemical reagents produce the solubilisation of the substrate by breaking down the cell walls. Commonly, organic matter to increasing methane production is usually made by ozonation, hydrogen peroxide, Fenton's reagent, acids or alkalis [10]. This pretreatment could be perform with a combination of others pretreatments to improve results [6].

-Mechanical: this pretreatment uses physical effect to break up bacterial cell walls causing solubilization and increasing the surface area. The particle size is inversely proportional to the rate in which the microorganism degrades the substrate [16]. Pressure, translational or rotational energy as high pressure homogenizer, lysate centrifuge, high performance pulse technique, microwave and ultrasound could be used [10]. Simpler methods such as the use of a stirred ball mill or shear gap homogenisation that shows better results when immobilized microorganism is used [6].

5.1 Ultrasonic system

The ultrasonic system is composed by a power supply, a converter that is a transducer, a booster and a horn as represented in the Figure 9, below [6].

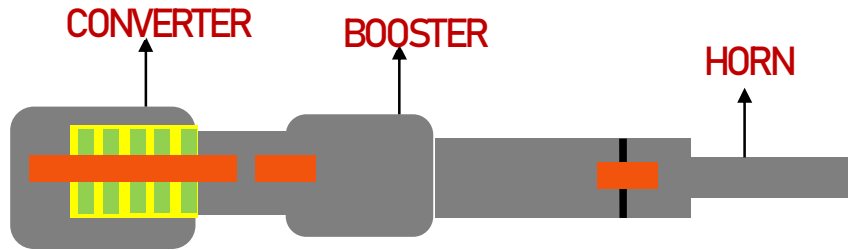


Figure 9. Ultrasonic system.

The power supply provides the system with electrical energy that achieves the converter to be converted into mechanical energy. This mechanical energy will be converted into ultrasonic frequencies and transmitted to the booster, where the amplitude of the frequencies increases. The horn is the last piece that works as an acoustic tool that transfers those waves as vibratory energy to the media that will be treated [6].

Moreover, ultrasonic waves could be treated as sound waves based in their similarity. Depending on the frequency, the ultrasound is divided in categories. Those used in pretreatments are called power ultrasound and range from 20-100 kHz. Working at these frequencies allows to solubilize the matter by the mechanical vibration [28].

Ultrasonic decay is affected by substrate characteristics such as TS content, temperature, pH, particle size and the ultrasound condition as time, intensity, frequency, amplitude and input power [6].

Previous studies showed that ultrasonic treatment time has the most significant effect on the substrate disintegration, as well as the combination of time and high ultrasonic power [6].

5.2 Ultrasound pretreatment application in anaerobic digestion process

The ultrasound was tested as an accelerator of anaerobic digestion by [29] with a frequency of 31 kHz focus in the reduction of residence time. The volatile solids reduction was 45.8% for untreated sludge and 50.3% for treated one. Biogas production was significantly improved.

[30] studied this pretreatment as a method to increase the soluble chemical oxygen demand (SCOD) in sewage sludge for digestion using a frequency of 22 to 40 kHz comparing different ultrasound times. The longest treatment time, 10 minutes, presented the highest SCOD. It was 4200 mg/l, higher than the 2100 mg/l presented by the 2.5 minutes of treatment. Untreated samples had a SCOD of 620 mg/l. Biogas production also increased with pretreatment time. A volume of 34 ml was reached by the 10 minutes treatment, 7 ml for the 1 minutes treatment and 2 ml for the untreated sample.

Other authors studied the improvement of anaerobic digestion through the application of this pretreatment, as [31] that discovered that with a continuous system and a 24 kHz ultrasonic treatment, the application of ultrasound has a clear effect on disintegration and solubilisation of the sludge and obtained a 40% of increment in biogas production.

On the other hand, studies of the ultrasound treatment with a co-digestion with crude glycerol were made by [7]. The experiments were performed with a mixture of cattle manure, food waste and crude glycerine (glycerol) to see the increment of methane production. The results obtained of the substrate treated were 70% compared to untreated.

Finally, [32] evaluated the biogas production with a co-digestion of cattle manure with crude glycerine obtained from biodiesel production using ultrasound as pretreatment. The applied frequency was of 20kHz for 4 min. Compared to the untreated substrate, biogas production increased by 800%.

6. Biogas

6.1 Biogas composition

As produced by digestion, biogas is a clean and environmentally friendly fuel [12]. It contains 55 to 65% of methane (CH₄), 30 to 40% of carbon dioxide (CO₂), water vapour fractions, hydrogen sulfide (H₂S) and ammonia (NH₃) [9]. There are traces of hydrogen (H₂), carbon monoxide (CO), and saturated oxygen (O₂). The composition of biogas is similar to landfill gas, but this last one contains significant amounts of halogenated compounds [11]. Composition of biogas is summarized in Table 3.

Table 3. Composition and parameters from different gas sources.

Parameter	Unit	Landfill gas	Digestion biogas
Lower heating value	MJ/N m ³	16	23
	KWh/N m ³	4.4	6.5
Density	MJ/kg	12.3	20.2
	kg/N m ³	1.3	1.2
Methane number		> 130	> 135
Methane (and variation)	vol%	45 (30-65)	63 (53-70)
Higher hydrocarbons	vol%	0	0
Hydrogen	vol%	0-3	0
Carbon monoxide	vol%	0	0
Carbon dioxide (and variation)	vol%	40 (15-50)	47 (30-50)
Nitrogen (and variation)	vol%	15 (5-40)	0.2 (-)
Oxygen (and variation)	vol%	1 (0-5)	0 (-)
Hydrogen sulphide (and variation)	ppm	< 100 (0-500)	< 1000 (0-10 ⁴)
Ammonia	ppm	5	< 100
Total chlorine (as Cl ⁻)	mg/N m ³	20-200	0-5

Appels, L., Baeyens, J., Degrève, J., & Dewil, R. (2008). Principles and potential of the anaerobic digestion of waste-activated sludge. *Progress In Energy And Combustion Science*, 34(6), pp 755-781.

Calorific value for landfill gas is 36.14 MJ/m³ and for biogas is 21.48 MJ/m³. Despite this, the composition of biogas depends on glycerol and the sludge used as raw material, so it can vary [11]. The heating value of biogas is determined by the CH₄ content. The higher heating value can be reached when 1 N m³ of biogas is combusted and the water vapour formed within the combustion is condensed [12].

The methane number describes the gas resistance to shocks when used in a combustion engine. Biogas contains several sulfur compounds that are very corrosive in presence of H₂O. H₂S is reactive with many metals and this is reinforced by concentration and pressure, by the presence of H₂O and high temperatures. High concentrations of ammonia cause problems in combustion engines when producing NO_x, so it must be controlled. All the biogas plants must have filters to reduce the amount of particles in the gas and the content of water droplets or scum [12].

6.2 Biogas utilization

According to biogas utilization, the treatment will need an improvement. There are four basic utilizations for it, heat and steam production, electricity generation, fuel use or chemicals production [12].

Biogas can be used for heat using boilers to produce water vapor for industrial plants. In this case, a treatment to remove H₂S is suitable to avoid corrosion problems. For the combined units of heat and power, H₂S content must be even less than in the boilers. Heating is used in the process and the remaining electricity can be sold to the national grid [11].

For the use of biogas as a fuel, it must be improved to have a higher calorific value, constant quality and not have particles that can cause damage to the mechanical parts of the engine. As for the previous applications, there should be no danger of corrosion caused by H₂S. As a fuel, it can be destined for fuel cells. Fuel cells are power generation systems to produce DC electricity as long as the fuel is supplied [11].

MATERIALS AND METHODS



MATERIALS AND METHODS

1. Inoculum and substrate

Anaerobic sewage sludge

The anaerobic sewage sludge that was used as inoculum on the experiments was taken from septic tanks of Bragança rural area. After collection, it was stored at refrigerator at 4°C until it was needed for the tests. Feasibility tests and sludge characterization were performed to ensure that we will have enough biomass to carry out the process. Sewage sludge is shown in Figure 10.



Figure 10. Sewage sludge used as inoculum.

Crude glycerol

The glycerol used in the experiments to obtain biogas was taken from the Laboratory of Biofuels, Superior School of Technology and Management (ESTIG) of the Polytechnic Institute of Bragança (IPB). This crude glycerol is the principal by-product of biodiesel production that is produced in the laboratory. It is shown in Figure 11.



Figure 11. Crude glycerol used as substrate.

Inoculum and substrate characterization

For substrate and inoculum characterization the following parameters were studied: pH, Total Solids (TS), Fixed Solids (FS), Volatile Solids (VS), Total Suspended Solids (TSS), Fixed Suspended Solids (FSS), Volatile Suspended Solids (VSS), Alkalinity, Volatile Acids (VA), Chemical Oxygen Demand (COD), Total Nitrogen by Kjeldahl method (TNK). The analytical methods used are described in Table 4.

Table 4. Analytical methods used in the determination of the characterization parameters.

Parameter	Analytical method of reference
pH (Soresen scale)	Digital meter with pH electrode, Hanna model EDGE according to the method 4500 H+ B of Standard Methods [33]
TS (mg/l)	2540 D of Standard Methods [33].
FS (mg/l)	2540 E of Standard Methods [33].
VS (mg/l)	2540 E of Standard Methods [33].
TSS (mg/l)	2540 C of Standard Methods [33].
FSS (mg/l)	2540 G of Standard Methods [33].

VSS (mg/l)	2540 G of Standard Methods [33].
Alkalinity (mg/l)	2320 B of Standard Methods [33].
VA (mg/L)	[34]
COD (mg/l)	Colorimetric Method 5220 D of Standard Methods [33].
NTK (mg/l)	4500-Norg Macro-Kjeldahl Method of Standard Methods [33].
AN (mg/L)	Direct Nesslerization Method 4500-NH ₃ Method of Standard Methods [33].

Ultrasound pretreatment

Ultrasound pretreatment was performed in an ultrasound-H bath of J.P. Selecta of 9 liters (Figure 12). The ultrasound equipment works with a frequency of 50 kHz and 900 W of power. 15 and 30 minutes of pretreatment time were selected for testing crude glycerol.



Figure 12. Ultrasound bath.

2. Batch Reactor at laboratory scale

Biogas measurement

In order to evaluate biogas production, anaerobic digestion was performed in 250 ml batch reactors system under mesophilic conditions (37°C). They were perfectly closed in order to maintain anaerobic conditions. To keep the temperature of the reactors constant, they were placed inside a hot bath and controlled with a digital thermometer.

The reactors were filled with 200 ml of a mixture between the substrate and the inoculum. The proportion of substrate was 1%. Each reactor had a rubber in its stopper that allows to measure biogas volume by moving with gas pressure the plunger of a syringe. The volume produced by each reactor was daily measure directly from the syringe. The system used to measure biogas production is described in Figure 13.

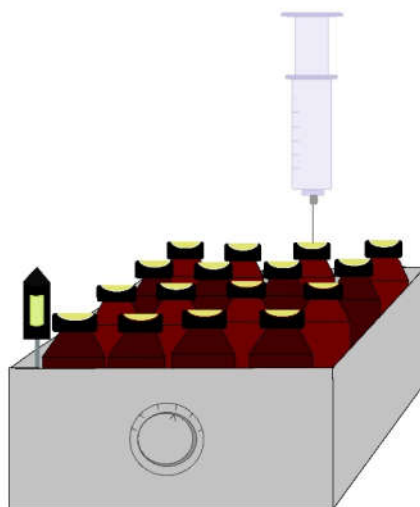


Figure 13. System made to measure biogas production at laboratory scale.

Different times of ultrasound (US) pretreatment were tested of glycerol treatment and compared with glycerol untreated. After a literature review, times of 15 and 30 minutes of ultrasound pretreatment were established. Control tests for the inoculum were also performed to make the discount of biogas production of sewage sludge.

Tests were performed by triplicated as it is shown in the Table 5.

Table 5. Reactors used for biogas production and their content.

Reactor	Content of crude glycerol
C1-C2-C3	Control test without glycerol
1.1-1.2-1.3	Glycerol without pretreatment
3.1-3.2-3.3	Glycerol with US treatment of 15 minutes
4.1-4.2-4.3	Glycerol with US treatment of 30 minutes

This test was carried out for 23 days. This time was determined when the biogas production dropped below the 1% of its accumulated production value. The experimental apparatus is shown in the Figure 14. Each material used and its functions is described in Table 6.

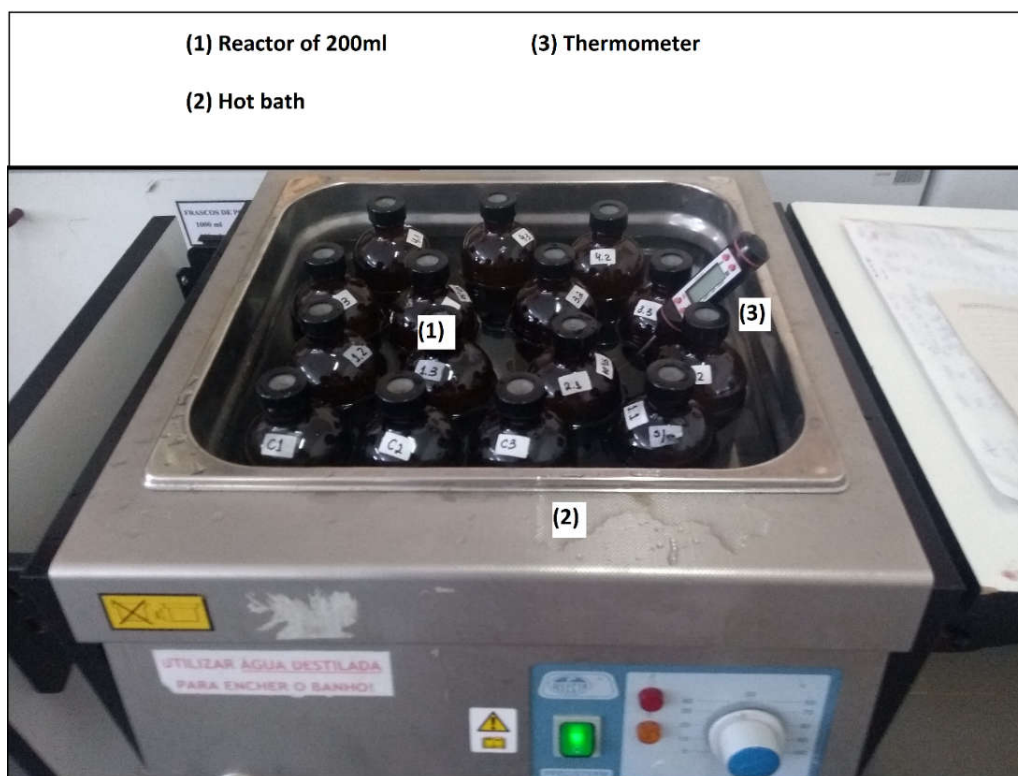






Figure 14. Biogas production system at laboratory scale.

Table 6. Materials of the biogas production system at laboratory scale and their function.

Name of material		Function
Reactors of 200ml		To hold the digestion process and keep anaerobic conditions.
Thermometer		To measure bath temperature
Hot bath		To keep the reactors under mesophilic temperature conditions
Syringe		To measure by pressure the volume of biogas

Methane measurement

On the another hand, to evaluate the quality of biogas, methane production tests were performed. Anaerobic digestion was performed in a 500 ml batch reactors system under mesophilic conditions (37°C). To maintain the temperature, they were placed inside a hot bath. Reactors were perfectly closed in order to keep the anaerobic conditions. The volume of methane produced was measured by alkaline displacement columns.

The reactors were filled with 400 ml of a mixture between the substrate and the inoculum. The proportion used was the same than in biogas production tests, 1% of substrate in the inoculum. Reactor were connected by a tube to an alkaline trap that is the main element of the displacement column measurement system.

When biogas is produced, it goes to the top of the reactor due to density difference and is conducted through a tube to the alkaline trap that is a 3% sodium hydroxide reservoir. This alkaline solution is saturated with carbon dioxide, so the others gases, mainly methane, produce the displacement of the same volume of the alkaline column. Therefore, the solution displaced by methane passes to the next bottle where it can be measured by the volumetric analysis. The system used is described in Figure 15.

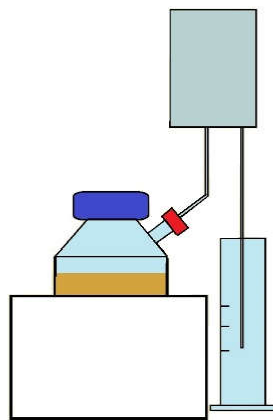


Figure 15. Methane production system for laboratory scale.

As the previous test, different pretreatment times with ultrasound (US) were tested in glycerol and compared with glycerol untreated. 15 and 30 minutes of pretreatment time. Control tests for the inoculum were performed to discount the methane production from sewage sludge to have the results for crude glycerol in order to compare the effect of the ultrasound system in the methane production.

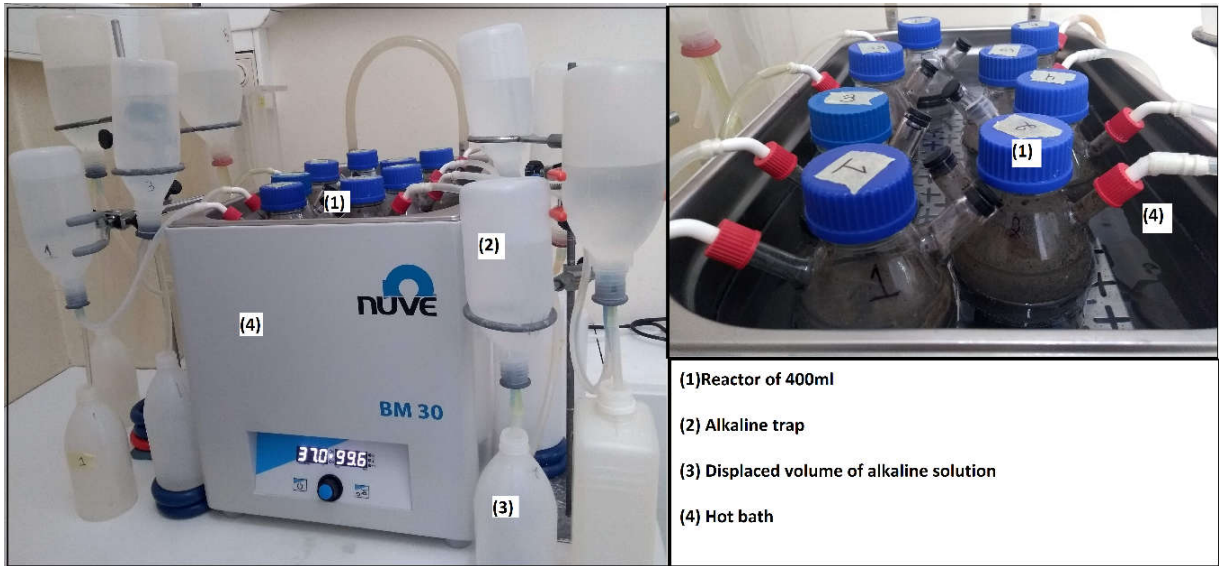
Methane production was measured volumetrically from the bottles with displaced alkaline solution. Tests were performed by duplicated as it is shown in the Table 7.

Table 7. Reactors used for methane production process and their content.

Reactor	Content of crude glycerol
1 and 2	Glycerol with US treatment of 15 minutes
3 and 4	Glycerol with US treatment of 30 minutes
5 and 6	Glycerol without pretreatment
7 and 8	Control test without glycerol

This test was carried out for 32 days. This time was determined when the methane production dropped below the 1% of its accumulated production value.


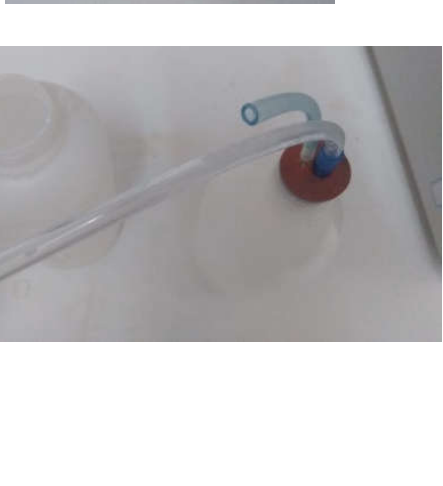
The system built to achieve the objective is shown in the Figure 16. Each material used and its functions it is described in Table 8.



(1) Reactor of 400ml
 (2) Alkaline trap
 (3) Displaced volume of alkaline solution
 (4) Hot bath

Figure 16. Methane production system at laboratory scale.

Table 8. Materials of the methane production system and their function

Material	Function
Reactor of 200ml 	To hold the digestion process and keep anaerobic conditions.
Carbon dioxide trap and displaced volume of alkaline solution 	To absorb carbon dioxide with the alkaline solution and displace the volume of solution equivalent to the volume of methane produced.

Hot bath



To keep the reactors
under mesophilic
temperature
conditions

3. Batch Reactor at mesoscale

The mesoscale of methane production was tested in a batch reactor of 7 l under mesophilic conditions (37°C). To maintain the temperature, a heating system was connected to the reactor. The distilled water was heated with a serpentine inside the main water tank. Then the hot water was plumped to the outside part of the reactor where tubes were formed a circle around the reactor.

The pH was daily controlled and adjusted to 7.2 to ensure that microorganisms can degrade the organic matter. To control it, it was necessary to take samples of the content of the reactor and to be measured in a pH meter. It was daily controlled. The pH was adjusted by adding 40% sodium hydroxide solution.

The test was made with the best results obtain in the laboratory scale testes. When analysing their results, it was concluded that the 30 minutes ultrasound pretreatment achieved the highest amount of methane and biogas productions. Thus, reactor was filled with 6 l of a mixture between the substrate (crude glycerol with the 30 minutes ultrasound pretreatment) and the inoculum. The proportion of substrate was the same as in the previous tests, of 1%.

The volume of methane produced was measured by alkaline displacement columns. The reactor was connected by a tube to the alkaline trap. The solution displaced by methane went to the reservoir where it was measured by the volumetric analysis.

A mixing system was needed to keep the reactor content homogenous. A mixing machine with an umbrella type stirrer of two pieces was used. It worked at 40 rpm.

The mixing and the heating system, the reactor and the alkaline displacement columns are part of the system where the digestion was carried out. It is described in the design below.

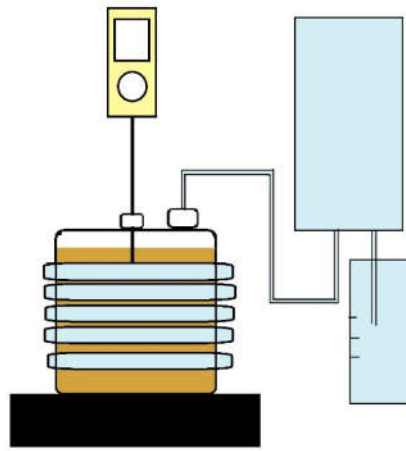


Figure 17. Mesoscale methane system.

The test of mesoscale methane production system was carried out for 22 days. The time was determined when the methane production fell below the 1% of its accumulated production value. Methane production was daily measured.

The system is described in Figure 18 and each of its materials and function in Table 9.

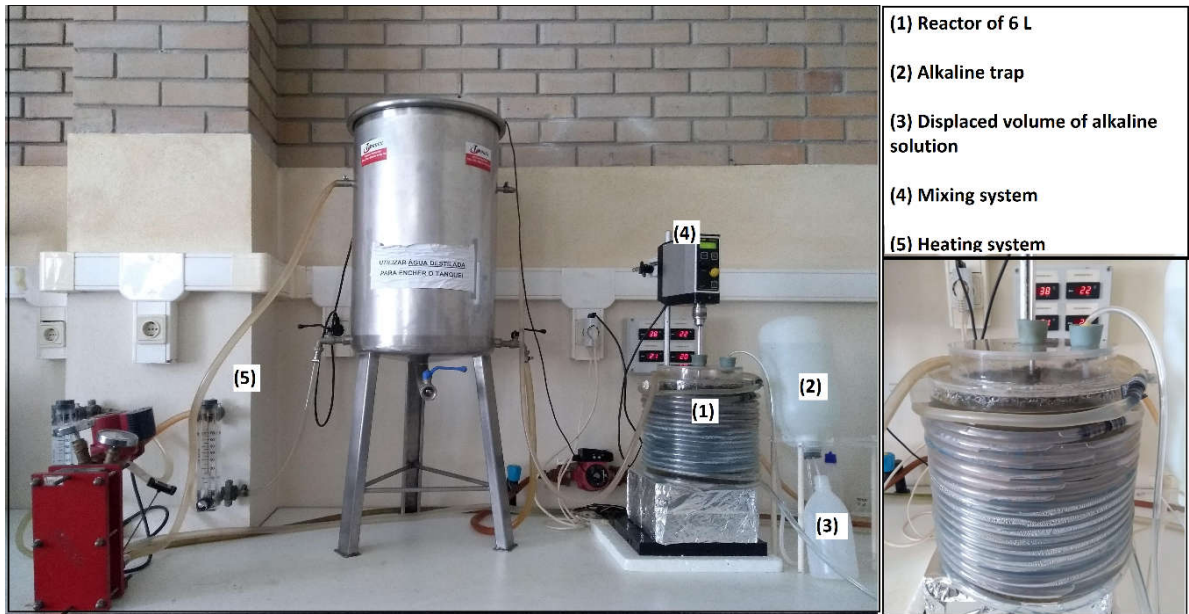

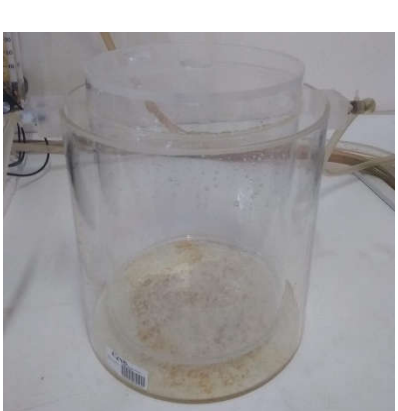


Figure 18. Methane production system at mesoscale.

Table 9. Materials used for mesoscale methane production process and their functions.

Name of material	Function
Heating system 	To keep the reactors under mesophilic temperature conditions
Reactor of 6 L 	To hold the digestion process and keep anaerobic conditions.

Mixing system



To mix the content of reactor to keep homogenous conditions of pH, temperature and composition.

Carbon dioxide trap



To absorb carbon dioxide with the alkaline solution and displace the volume of solution equivalent to the volume of methane produced.

pH meter



To measure the pH of the reactor content

RESULTS AND DISCUSSION



RESULTS AND DISCUSSION

1. Substrate and inoculum characterization

Anaerobic sewage sludge from septic tanks of the rural area of Bragança and crude glycerol from biodiesel production process of the Laboratory of Biofuels of ESTIG were tested. The characterization tests were performed in accordance with the procedures described in Table 4. The characterization tests results are described in Table 10 and 11.

Table 10. Characterization of activated sludge

Parameter	Value
pH	7.2
Conductivity ($\mu\text{S}/\text{cm}$)	11.3
TS (g/l)	11.0
FS (g/l)	3.5
VS (g/l)	7.5
TSS (g/l)	20.6
FSS (g/l)	3.1
VSS (g/l)	7.5
Alkalinity	0.033
COD (g/l)	40.1
AV (mg/l)	0.2
NTK (g/l)	1.3
C/N	30.7

Table 11. Characterization of glycerol

Parameter	Value
pH	8.4
COD (g/l)	976.4
TS (g/l)	755.7

For mesoscale methane production, the pH had to be adjusted at 7.3 because when anaerobic digestion is carried out, this parameter changes step by step. A pH below 5 could stop functioning of bacteria, mainly methanogenic bacteria that are the most sensitive to pH and temperature conditions. To adjust it was necessary to add 40% solution sodium hydroxide.

According to [35], the relationship VS/TS is a parameter that can indicate the microorganisms proportion in the sludge. For a good performance, they agree that this parameter should be around 0.66 or 0.67. VS/TS relationship in the sewage sludge from septic tanks is 0.68, so it indicates that is a good sludge to be used as inoculum.

2. Batch Reactor at laboratory scale

Biogas measurement system

1. COD and solids determination

For the system tested, COD, TS, FS and VS determinations were made to evaluate the content of each reactor after the digestion process that helps to make conclusions about the pretreatment effect and to select the best conditions of working. As tests were made in triplicated, the final values are an average of both reactors values.

For the COD, the reactors that had glycerol without pretreatment had a value of 22.4 g/l, while with 15 and 30 minutes of ultrasound pretreatment the values were of 25.6 and 39.1 g/l, respectively. As analysed before, with pretreatment time increases COD values.

The results are represented as a line at Figure 19. Solids determined are shown in bars in the same Figure.

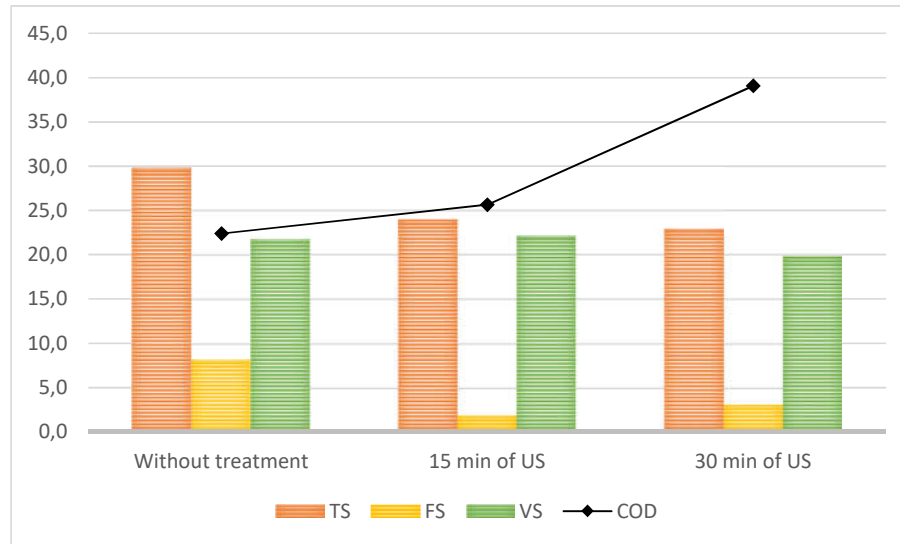


Figure 19. COD and solids determination for biogas production at laboratory scale

The total solids (TS) in the reactor that had glycerol without treatment was of 29.8 g/ml. In the reactors that had 15 and 30 minutes of pretreatment, were of 24.0 and 19.8 g/ml. From these values it can be concluded that volatile solids were decreasing with pretreatment time. According to [35] volatile solids (VS) can give an estimation about the amount of substrate that can be turned into methane, so it is expected to decrease after anaerobic digestion process.

2. Biogas production

Daily biogas production is summarized in Figure 20. Each curve represents the accumulated volume of methane produced by the treatment. As the test was made in triplicated, values are an average of reactors. To represent the effect of the pretreatment in the glycerol, the biogas production values of the control test that had only sewage sludge in the reactors, were discount from the values of the test with glycerol.

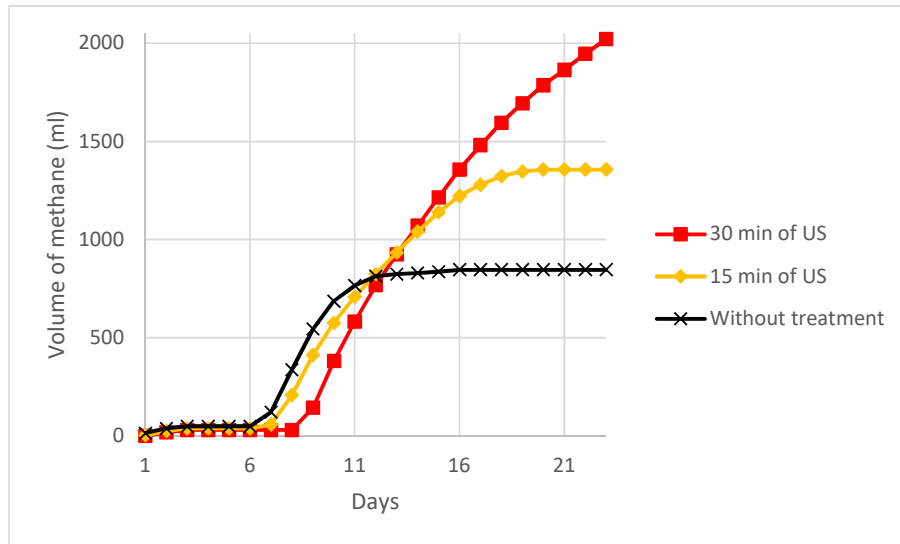


Figure 20. Daily biogas production at laboratory scale.

The results show that the maximum biogas production was achieved by the reactors that had glycerol with 30 minutes of ultrasound pretreatment and was 2021 ml. Then, the reactors that had glycerol with 15 minutes of pretreatment showed a better performance with a final value of 1356.6 ml of biogas than the 1010.5 ml of the reactors that did not have treated glycerol.

Compared with the results of the experiences carried out by [37], the maximum biogas production was obtained for 30 minutes of ultrasound pretreatment as well, for a co-digestion of maize straw and dairy manure. It concludes that ultrasound pretreatment changed the physical and chemical properties of the substrate.

According to the tests performed by [30], biogas production also increased with the pretreatment time. A volume of 34 ml was reached by the treatment of 10 minutes, 7 ml for the treatment of 2.5 minutes and 2 ml for the untreated sample. They were made on sewage sludge. Analysing these scenarios, it can be concluded that ultrasound pretreatment enhanced the biogas production.

Methane measurement system

1. COD and solids determination

COD, TS, FS and VS determinations were made to evaluate the content of each reactor after the digestion process. As tests were made in duplicated, the final values are an average of both reactors values.

For the COD, the reactors that had glycerol without pretreatment, presented a value of 29.11 g/l, meanwhile with 15 and 30 minutes of ultrasound pretreatment the values were of 43.5 and 45.09 g/l, respectively. As was analysed before, with pretreatment time increases COD values. Results are represented as a line at Figure 21. Solids determined are shown in bars in the same figure.

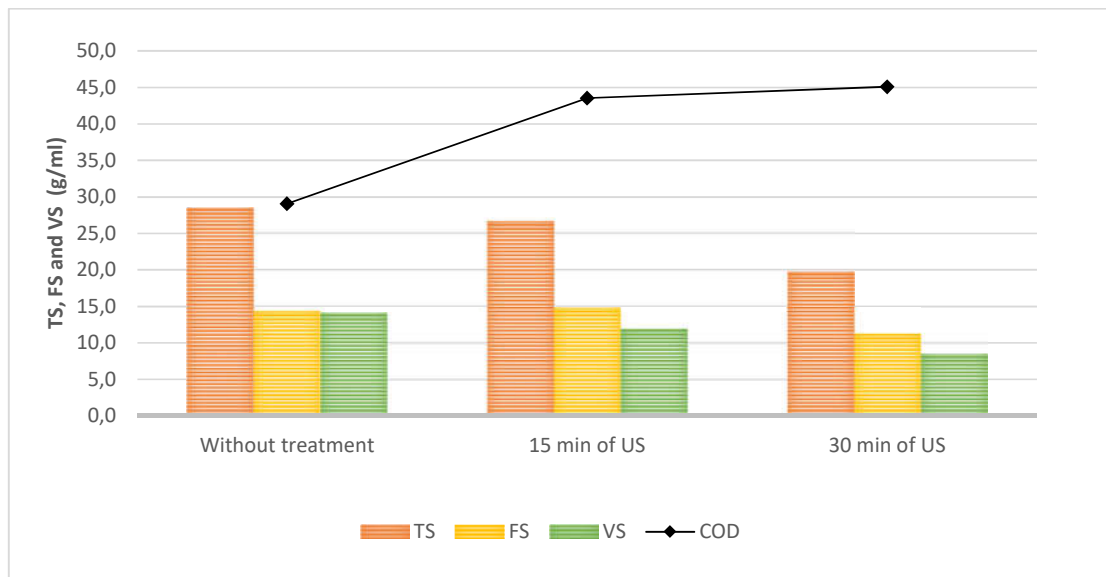


Figure 21. COD and solids determination for methane production at laboratory scale

As the pretreatment time in glycerol increases for each reactor, TS, FS and VS decreases. Total solids (TS) in the reactor that had glycerol without treatment was of 28.4 g/ml and in the reactors that had 15 and 30 minutes of pretreatment was of 26.6 and 19.7 g/ml.

2. Methane production

Daily methane production is resume in Figure 22. Each curve represents the accumulated volume of methane produced by treatment.

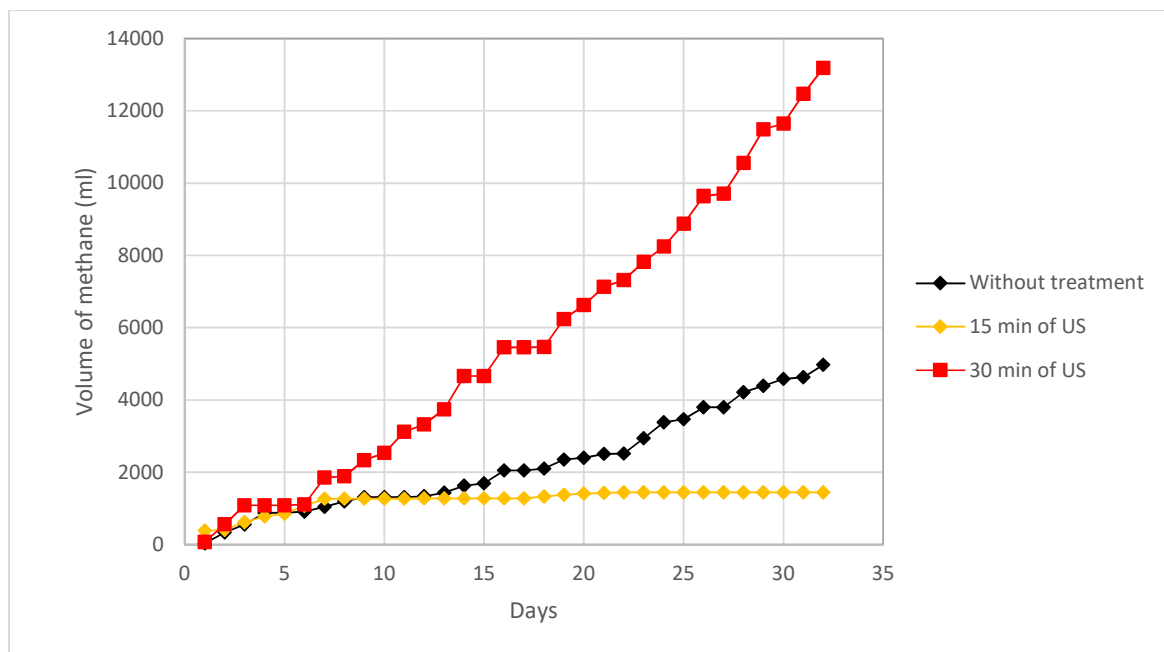


Figure 22. Methane production at mesoscale

As the test was made on duplicated, values are an average of both reactors. To represent the effect, the pretreatment in glycerol, methane production values of the control test that had only sewage sludge in the reactors, were discount from the values of the test with glycerol.

The results show that the maximum methane production was reached by the reactors that had glycerol with 30 minutes of ultrasound pretreatment and it was 13184.7 ml. Then, the reactors that had glycerol without treatment showed a better performance with a final value of 4966.9 ml of methane than the 1439.6 ml of the 15 minutes of pretreatment.

According to the studies made by [7], the ultrasound pretreatment in a co-digestion of cattle manure and crude glycerol (3%) improved the methane production by 90% than in untreated one.

In both cases, 30 minutes of ultrasound pretreatment had the maximum methane production, so the pretreatment improved production.

Comparing these results with the biogas production performed previously, it can be suggested that for a 30 minutes of ultrasound treatment, the production of biogas and methane is improved. On the another hand, in addition to the fact that in a15 minutes of ultrasound treatment, biogas volume is higher than the untreated one. The quality of biogas determined by methane percentage cannot be ensured. The amount of biogas could be related to carbon dioxide or other gases formed in the tested performed.

Finally, from the results found in this phase of the work, can be conclude that ultrasound pretreatment enhanced biogas production and had the best performance for 30 minutes of pretreatment.

3. Batch Reactor at mesoscale

1. COD and solids determination

First, COD, TS, FS and VS determinations were made to evaluate the content of the 7 l reactor before and after the digestion process, to compare the results and make conclusions about the physical and chemical effects of ultrasound pretreatment in the anaerobic digestion of crude glycerol.

In COD determinations, before anaerobic digestion began, the reactor had a value of 142.0 g/l. After finishing the process, this value increased up to 207.0 g/l. Since the substrate was in a disintegration process, the amount of COD should decrease.

The variation of solids content in the methane production are shown in Figure 23. As mentioned above, volatile solids (VS) can give an estimation about the amount of substrate that can be converted to methane, so it is expected that decreases after anaerobic digestion process. It should represent a good amount of solids before the digestion, so that a good methane production can be expected.

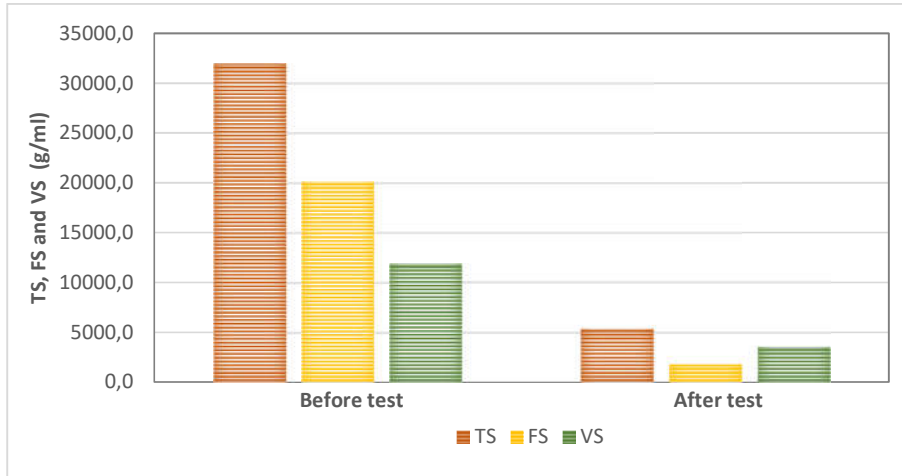


Figure 23. Content of solids for mesoscale methane production.

Before the anaerobic digestion, the substrate and inoculum presented a value of TS of 32.0 g/l, FS of 20.1 g/l and VS of 11.9 g/l. After the digestion, these values become TS of 5.3 g/l, FS of 1.77 g/l and VS of 3.53 g/l. The results show a decrease in solids content, in order of 83.7, 91.1 and 70.5% respectively, after the anaerobic digestion.

According with these results, the colour differences between the content of the reactor before and after the digestion can be observed in Figure 24. After anaerobic digestion, the content became darker and more homogenous than before.



Figure 24. Content of reactor before and after anaerobic digestion.

2. Methane production

Daily methane production is resume in Figure 25. The curve represents the accumulated volume of methane produced by the mesoscale reactor (7 l), with 30 minutes of ultrasound pretreatment. The total volume obtained for the test was of 16224 ml of methane.

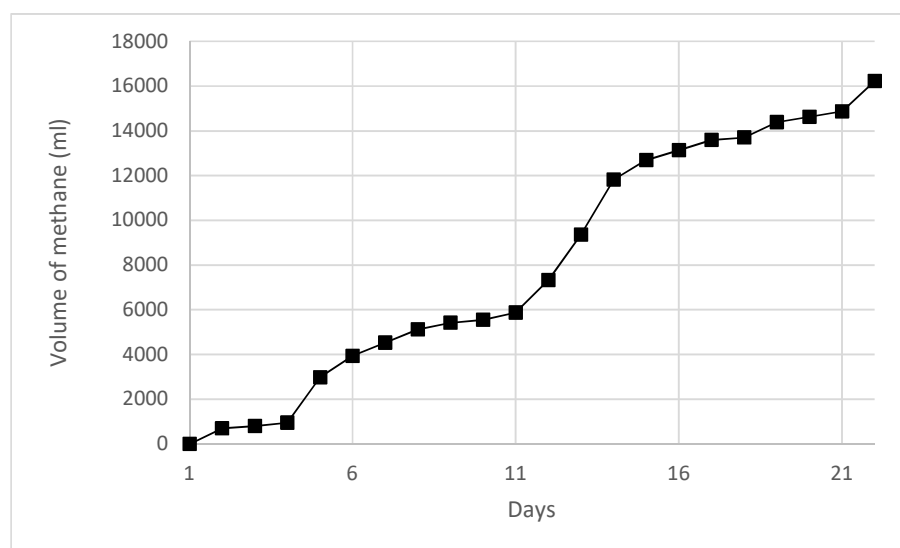


Figure 25. Methane production for mesoscale test.

For the test made in a 400 ml batch reactor, the results obtained after 32 days of operation for the sewage sludge with 1% of glycerol with a 30 minutes of ultrasound pretreatment was 13184.7 ml of methane. It was made under mesophilic conditions for 32 days. The mesoscale tests, were performed with 6000 ml (6 l) of the same proportion, 1% of crude glycerol under mesophilic conditions too but with a continue mixing and a controlled pH for 22 days. These two controlled parameters helped to have a better anaerobic digestion as was mentioned in [11]. The results in mesoscale tests were 123% higher than in laboratory scale.

If the volume of methane values in both tests are compared in the same period of time, for the laboratory scale test was obtained 5,455.5 ml of methane until day 22. This value is 297% lower than the 16224 ml obtained at mesoscale at day 22 too. Therefore, it can be concluded that the mesoscale test was performed successfully. The control of variables like mixing and pH in addition to temperature and anaerobic conditions, could be another effect to evaluate in a future research.

CONCLUSIONS



CONCLUSIONS

- The characterizations tests performed on the substrate and crude glycerol show that they had good properties to carry out the experiences.
- For biogas production, the reactors that had glycerol without pretreatment presented a value of 22.4 g/l, while with 15 and 30 minutes of ultrasound pretreatment the values were of 25.6 and 39.1 g/l of COD.
- In the case of the determining quality of biogas through the determination of methane, the behaviour of organic matter was the same. The reactors that had glycerol without pretreatment presented a value COD of 29.11 g/l, while with 15 and 30 minutes of ultrasound pretreatment the values were of 43.5 and 45.09 g/l.
- The increase in COD after ultrasound pretreatment may suggest that this mechanical method can disintegrate the organic matter and leave the molecules of crude glycerol more accessible to the microorganisms.
- The results of biogas production test show that the maximum biogas production was 2021.0 ml for the reactors that had glycerol with 30 minutes of ultrasound pretreatment. Then the reactors that had glycerol with 15 minutes of pretreatment showed a performance of 1,356.6 ml of biogas than the 1010.5 ml of the reactors that have not got glycerol treated.
- In the determination of methane production, the maximum value was reached by the reactors that had glycerol with 30 minutes of ultrasound pretreatment, 13184.7 ml. After this result, the reactors that had glycerol without treatment reached 4966.9 ml leaving the 15 minutes of pretreatment in the last place with 439.6 ml of methane.
- Thus, the comparison of results obtained in biogas and methane production, can be suggest that ultrasound pretreatment improves biogas production. By the volume of methane produced can be suggest that for a longer pretreatment time, the quality of biogas improves.

Can be conclude that, in this work, the pretreatment ultrasound with 30 minutes, enhanced biogas production and had the best performance in both on the bench scale and on the mesoscale.

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