Treatment and energy valorization of residual glycerol in a perfectly mixed batch reactor

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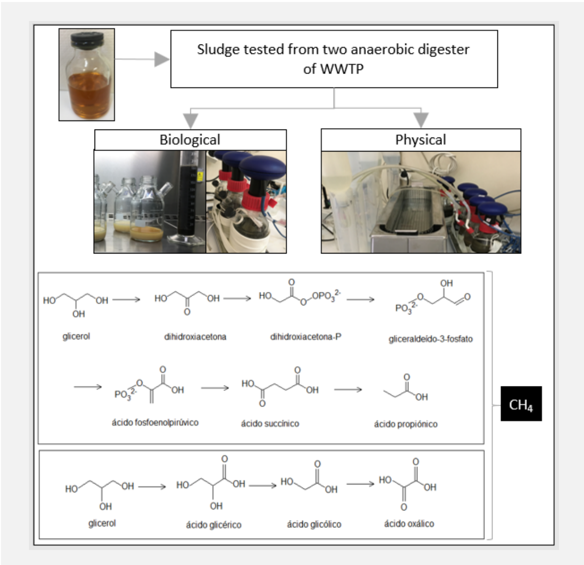

The anaerobic digestion of crude glycerol from the production of biodiesel is an alternative that has been studied for its valorization through the production of methane. The high organic load (1800 g COD.L^{-1}) of crude glycerol can cause kinetic stress, which results in an inhibition of methanogenic microorganisms. To circumvent the problem, an alternative is the prior use of ultrasonic energy and the use of microorganisms such as *Aspergillus niger* and *Escherichia coli*. Ultrasound promotes the breakdown of the cell wall and membrane, releasing intracellular material that favors biodigestion. In addition, such microorganisms can produce lipases capable of degrading other impurities contained in the residual glycerol, such as long chain fatty acids. The aim of this study is to improve the anaerobic digestion of glycerol through physical (ultrasound) and biological (*A. niger* and *E. coli*) pretreatments. The results indicate that the use of ultrasound improved methane generation by 23% for a lower concentration of glycerol (2%). There were also improvements from the use of *A. niger* in 1.7% glycerol. For the concentration of 3.2% glycerol and the use of *E. coli* in all experiments caused inhibition of methanogenic microorganisms.

Introduction

The production of biodiesel generates crude glycerol a by-product, which can be used in a wide range of industrial processes. It has purity ranging from 23% to 87% [1, 2], so depending on its use it is necessary to remove impurities, which increases the price and may become the application economically unviable [3, 4]. In addition, increased industrial production of biodiesel has been led to a surplus of glycerol, which causes a commercial devaluation and an impact on the price of biodiesel. In this context, other forms of glycerol valorization become interesting [4].

One means of valorization of the crude glycerol is its biotransformation, using microorganisms, in compounds with greater added economic value. Although glycerol is used for several industrial purposes, the impurity content of the residual glycerol limits its processing. The anaerobic digestion of glycerol to produce energy through the generation of methane is an alternative to its valorization. The high organic load of crude glycerol (1800 g^{-1} COD.L^{-1}) can cause kinetic stress that leads to the inhibition of methanogenic microorganisms [5, 6].

The use of pretreatment can improve the anaerobic digestion. Ultrasounds are used as physical pretreatment to improve the efficiency of the digestion process [7, 8]. The ultrasound helps to deagglomerate biological flakes and transforms large organic particles into smaller ones [9]. In addition, biological pretreatment using specific microorganisms such as *Aspergillus niger*, *Escherichia coli* and *Yarrowia lipolytica* [2, 10, 11] can help glycerol to follow IST metabolic pathways to produce hydrogen, ethanol, citric acid, 1,3-propanediol, among others [12, 13].

Objectives

This study had as main objective to test alternative pretreatments of the reactor feed with ultrasound and microorganisms, namely, *Aspergillus niger* and *Escherichia coli* prior to the anaerobic digestion of glycerol.

Methods

In a preliminary phase, the quality of the sludge from two different sources i.e. from the anaerobic digester of the Vila Real Wastewater Treatment Plant (AD-VR) and the anaerobic digester of the Vila do Conde WWTP (AD-VC), was evaluated for the anaerobic digestion of glycerol concentrations of 0.2%, 1.7% and 3.2%. Posteriorly the effect of pre-treatments by sonication for different times (3 to 15 h) and by the previous degradation with either *A. niger* or *E. coli* microorganisms was assessed.

Tests were performed using a continuous stirred-tank reactor in a batch operation (500 mL). The experiments were carried out with temperature control in the mesophilic regime, between 35ºC and 40ºC. A total volume of 300 mL (sludge + substrate) was employed. According to the characteristics of the glycerol, some compounds were added: potassium nitrate (C:N of 25:1), disodium phosphate (C:P of 120:1) and sodium bicarbonate (2 g.L^{-1}). The daily biogas production (% CH_4 and volume), COD (initial, after 15 days and final, after 30 days), ST and SV (after 15 days and final, after 30 days), were evaluated.

Results

Sludge Type

Sludge samples from two anaerobic reactors, one from the AD-VR WWTP and another from the AD-VC WWTP, were tested. Higher SV/ST values correspond to a greater stabilization of the sludge [14]. Tawfik and Klapwijk [15] and Al-Jamal and Mahamoud [16] indicate values of 0.66 and 0.67 as suitable for the good performance of the reactor. The SV/ST ratio was 0.75 for the AD-VC slurry and 0.65 for the AD-VR one, being the later in agreement with the literature reference values. This was evidenced by COD values. AD-VC sludge (with higher SV / ST ratio) yielded COD=0.5 g.L^{-1}, whereas AD-VR sludge (with lower SV/ST ratio) presented a COD=12.8 g.L^{-1}. According to the results shown in Figure 1, the experiments performed with 1.7% and 3.2% of glycerol originated biogas yields of 200 to 900 mL (A), however, the quality of the gas was not relevant with a variation of methane volume between 0 and 15 mL (B). In the case of the experiment with a concentration of 0.2% of glycerol, a better biogas production was obtained: the sludge from AD-VR and AD-VC allowed to obtain a methane volume of 575 mL and 294 mL, respectively (B). This indicates that the best results were obtained with the AD-VR sludge.

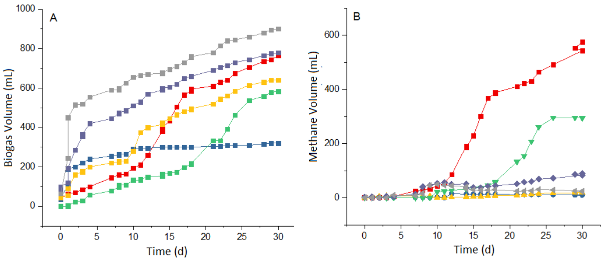

Figure 1. Volume of biogas (A) and methane (B) in the batch reactor for AD-VR and AD-VC sludges. 0.2-VR; 1.7-VR; 3.2-VR; 0.2-VC; 1.7-VC; 3.2-VC.

Pretreatment

Pretreatments provided better methane generation. The pretreatment with ultrasound (15h) for 0.2% glycerol led to an increase of about 23% of biogas with a methane content of about 77% (Figure 2-A). The exposure of microbial cells to ultrasound energy breaks the cell wall and the membrane releases intracellular material, which favors digestion [9].

For a concentration of 1.7% glycerol, the best performance was obtained after pretreatment of glycerol by the *A. niger* fungus (117 L of $CH_4.kg^{-1}$ glycerol, 76% increase) (Figure 2-B). *A. niger* produces lipase capable of catalyzing and synthesizing various dicarboxylic acids and diols, in addition, there are studies showing that the fungus can optimize the components of the biofuel synthesis of glycerol carbonate by 12% [17].

Figure 2-C shows the lowest volume of biogas generated with the lowest methane content (8.7%). Using 3.2% of glycerol leads to inhibition of methanogenic microorganisms in all the experiments. Athanasoulia et al. [18] obtained desolating results, proving the inhibition of the anaerobic digestion process when using 4% glycerol. In addition, the *E. coli* microorganism impaired the anaerobic digestion at all concentrations of glycerol.

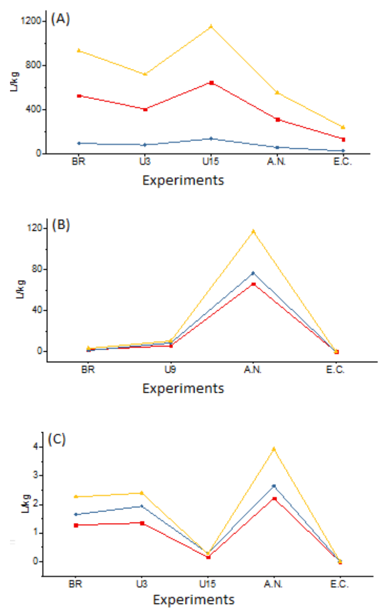

Figure 2. Volume of methane in the batch reactor for (A) 0.2% of glycerol, (B) 1.7% of glycerol and (c) 3.2% of glycerol. L $CH_4.kg^{-1}$ DOC; L $CH_4.kg^{-1}$ SV; L $CH_4.kg^{-1}$ glycerol.

Conclusion

Among the two types of sludge tested (AD-VR and AD-VC from the anaerobic digesters of Vila Real and Vila do Conde WWTPs), the AD-VR sludge allowed, in general, to obtain better results feeding the reactor with 0.2 % glycerol, i.e. a production of 937 L $CH_4.kg^{-1}$ glycerol containing 75% methane and the SV/ST=0.65.

For the batch reactor tests, the most satisfactory results were achieved with 0.2% glycerol concentration and pretreatment by ultrasounds during a period of 15 h; the obtained biogas had 77% methane, and the volume generated reached 1153 L $CH_4.kg^{-1}$ glycerol. For a concentration of 1.7% glycerol and pretreatment by *A. niger*, the content CH_4 in the resulting biogas was 65% and the production was 117 L $CH_4.kg^{-1}$ glycerol. Feeding with 3.2% glycerol resulted in an inhibition in all experimental trials.

Acknowledgements

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