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Production of Biogas from Olive Mill Waste Waters Treated by Cow Manure

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Abstract

The olive mill waste waters (OMWW) generated from olive oil extraction is a major environmental concern since they are characterized by their role as a pollutant (high organic and mineral matters) and their pH acid. The aim of this study was to valorize (OMWW) by anaerobic fermentation in the presence of cow manure in order to produce biogas and reduce their toxic load. Many tests were carried out by fermenting (OMWW) with polyphenols in the presence of cow manure in thermophile temperatures. The performance of this treatment was valuated through measurements of biogas production and by the determination of different parameters of fermented media (pH, volume of the biogas and polyphenols).

Keywords

Olive Mill Waste Waters, Anaerobic Fermentation, Cow Manure, Biogas, Polyphenols

1. Introduction

Olive oil production represents a considerable share of the Tunisian economy. Olive oil is assumed to have a healthy image during its consumption due to its oleic acid content, which may prevent some human diseases [1]. Ironically, by-products of olive oil production such as olive mill waste (OMW) pose a serious environmental risk, especially in the Mediterranean. OMW is composed of many complex substances that are not easily degradable (e.g. polyphenols, tannins, etc.).

The disposal of 30 million·m^3 of olive mill wastewaters (OMW) every year is a major environmental problem in the Mediterranean countries. OMW has significant pollutant properties, especially due to the high concentration of phenols and polyphenols which are toxic and may inhibit biological treatment [2]. Given the bactericide nature of these chemical structures when found at high concentrations, the classic biological treatment cannot be applied and alternative wastewater treatments have to be taken into consideration. In order to find convenient solutions for spreading this by-product into agricultural lands without harmful environmental effects, many studies were conducted in several olive oil-producing countries. Most of these [3] [4] [5] [6] [7] studies were focused on the pretreatment of OMW using different microorganisms and different aerobic or traditional anaerobic processes [8] [9]. Few studies have been also performed to identify the OMW toxicity sources. Reference [10] [11] [12] [13] Indeed, using durum wheat germinability as a biotest, it has been suggested that phenols are the main phytotoxic compound of OMW [14].

In this study, the feasibility of using some rabbit manure in order to enhance the methane production of OMW was investigated. For this purpose, biochemical methane assay was carried out for OMW mixed with varying amounts of other substrates such as cow manure in the serum bottles, respectively. It was demonstrated that anaerobic digestion of OMW with rabbit manure significantly enhanced the biodegrability of OMW which was too low if it was digested alone. Over 90% increase in biogas production was obtained when digesting OMW with cow manure. It was demonstrated that the biodegrability of OMW could be significantly enhanced by anaerobic digestion and thereby integrated management of OMW using anaerobic degradation could be proposed as an economically viable and ecologically acceptable solution for the safe disposal of OMW.

2. Materials and Methods

2.1. Characterization of OMW

The OMW used in this study was obtained from a discontinuous olive oil processing plant located in Sousse-Tunisia, according to a process of extraction and pression then centrifugation the OMW was stored in a refrigerator for a year at 4˚C for other uses.

2.2. Characterization of Cow Manure

The cow manure used was given from a farm located in Monastir-Tunisia. It's an under-product of manure, which has an intrinsic value as an organic improvement; these products are composed of the animal extretas associated with carbonic composition (straw, sawdust).

At first, these products are sieved and divided into samples of 4 g. They are mixed with 10 ml of distilled water in tight bottles. Finally, the bottles are incubated at 37˚C to start anaerobic fermentation.

2.3. Chemical Products

Ethyl acetate, sodium bicarbonate, sodium hydroxide (NaOH), ethanol, sulfuric acid (H_2SO_4).

2.4. Casting Off (Figure 1)

1) Solution of (sodium bicarbonate + acetic acid); production of CO_2 that used to escape gas from mixture in the bottle n˚3.

2) Double boiler.

3) Bottle in which we put the mixture to ferment.

4) Solution of sulfuric acid 0.1 N to catch NH_3.

5) Solution of (NaOH 5M + lime water) to catch CO_2 and H_2S.

6) Burette that leads to detecting the volume of recovered gas.

7) Empty pump.

8) Tap.

In the bottle n˚3:

There is a mixture of 100 ml of cow manure + 700 ml of OMW + sodium bicarbonate. We add the sodium bicarbonate to adjust pH between 7 and 8 [15] [16]. We paddle OMW by N_2 to have anaerobic middle. We put the mixture in the double boiler at the thermophilic zone [55˚C - 65˚C]. This bottle had a knuckle that connects the sample to outgoing pipes. One is connected to an Erlenmeyer that contains a mixture of bicarbonate and acetic acid (A solution that produces the CO_2). This mixture is used when we start the experience to provide anaerobic conditions (during 5 minutes). The second pipe is used to collect the gas produced during the anaerobic fermentation. These paddle into the solution of NaOH (5M) and $Ca(OH)_2$ to catch the CO_2, H_2S and NH_3 produced during the anaerobic fermentation. The other gas (O_2, H_2, CH_4) are recovered into the tight and upset burette.

Finally, the pump used at the beginning of the experience, helps to aspire the gas from the burette until the solution n˚5 (NaOH + CH_3COOH) reaches the upper of the graduated burette extremity. We let the mixture ferment in the bottle n˚3 for 10 hours.

The experience was finished when the production of the gas becomes too weak (<1 ml/1 hour).

Figure 1. Apparatus used during the study.

During the fermentation, we measure every hour, the volume of the gas released in the burette. These volumes need us to calculate the number of biogas mole's obtained.

A sample was taken from the fermentation middle, at the beginning and at the final of the fermentation on which the experimental test was carried out: determination of pH, the volume of the gas given off and the polyphenols content.

2.5. Extraction of Phenolic Compounds from OMW

The procedure to obtain the phenolic extract was as follows. The samples were acidified with HCl (pH 2.0) and extracted three times with ethyl acetate (v/v) at ambient temperature.

The three organic fractions were combined and dried with anhydrous Na_2SO_4 for 30 - 40 rain.

The extract was concentrated to dryness in a rotavapour and redissolved with a methanol/water mixture (60/40). For the determination of total phenol content [17] the method used the Folin-Ciocalteau's phenol reagent (from Merck, Darmstadt) involving the successive addition of 5 ml sodium carbonate (200 $g \cdot l^{-1}$) and 2.5 ml Folin-Ciocalteau phenol reagent to a 50.0 ml sample. After 60.0 min at 20˚C the absorbance was measured at 725 nm against a distilled water and reagent blank [18].

2.6. UV Analysis of OMW Extract

Spectrophotometric measurements were performed with a UV-1650 PC/vis spectrophotometer, using Cary WinUV "version 2.21" software (varian), on crude extracts after dilution (1:100) with (methanol/water).

Phenolics were identified on the basis of their absorbance and their concentration in comparison with standard curve. When necessary, co-injection and elution with standards were used to insure about the identity of the compounds.

2.7. GC Analysis

Gas samples were taken with a syringe from the headspace of the serum bottle and analysed by gas chromatography with a SHIMADZU instrument chromatograph equipped with a flame ionization detector. The flame ionization detector was fitted with a 30 cm stainless steel column. N_2 was used as carrier gas at 28 ml/min with H: and air flows of 25 and 30 ml/min, respectively. The oven, injector and detector temperatures were 200˚C.

2.8. Measurement of pH

The measurement was carried out with a pH meter (METTLER DELTA 340), after diving the electrod into homogenous solution.

3. Results and Discussion

Figure 2-4 summarize the variation of the biogas volume according to time;

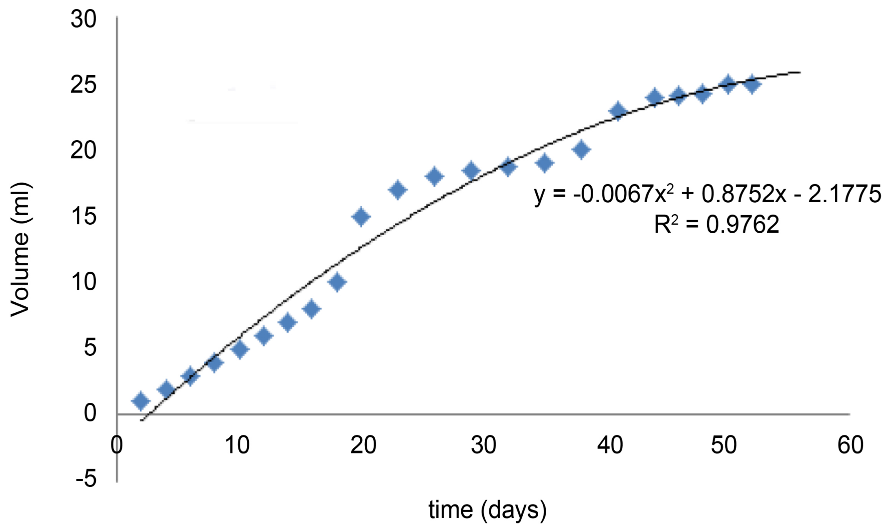

Figure 2. Variation of the biogas volume according to time.

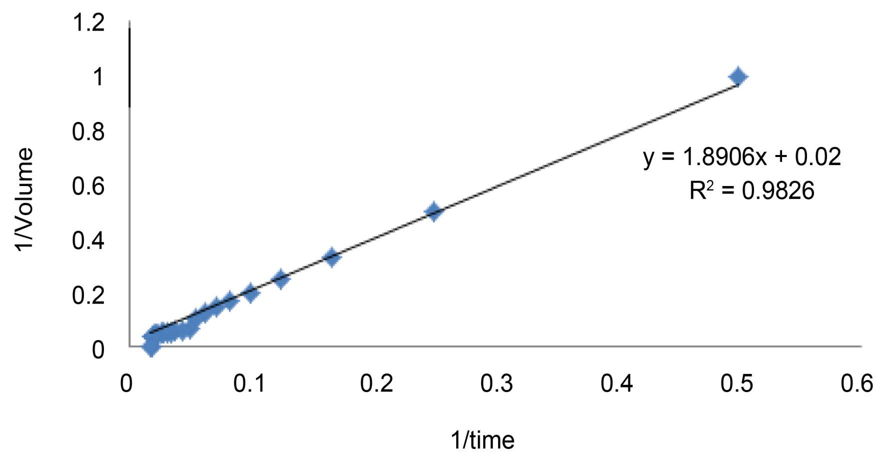

Figure 3. Variation of (1/Volume) according to (1/time).

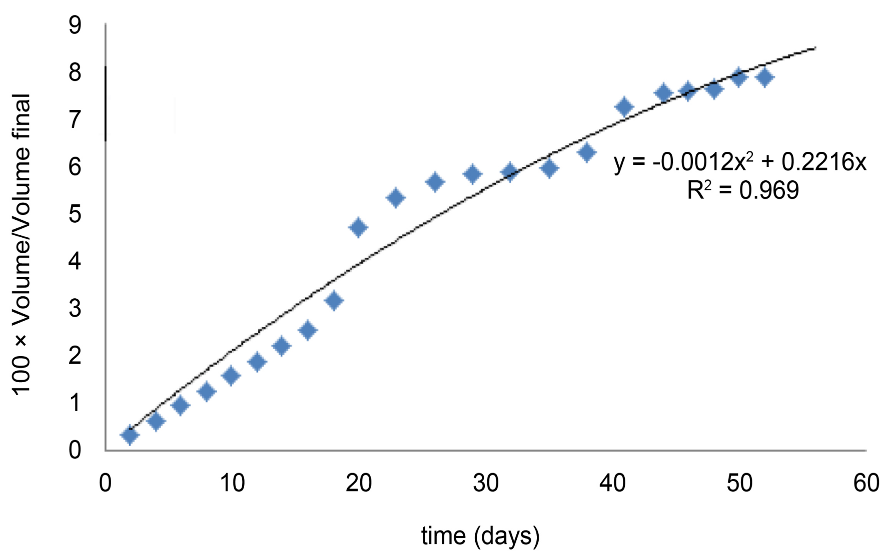

Figure 4. Variation of the yield according to time.

they show that the production of biogas started from the first moment.

3.1. Influence of the Cow Manure in OMW Fermentation

This biogas production is due to the nutriment's nature and microorganisms present in the cow manure dissolved in OMW.

Then, to provide anaerobic digestion, we must regulate the pH between 7 and 8 [19]. We added sodium bicarbonate. The addition of the basis product hasn't any effect on the production of the biogas [16] but the ion of HCO_3^- leads to the neutralization of the free organic acid and to accumulate the volatile fatty acids [20].

$$\text{Proteins} \rightarrow NH_3 + H_2O \rightarrow NH_4^+ + OH^-$$

$$CO_2 + OH \rightarrow HCO_3^-$$

3.2. Chromatographic Analysis

The chromatographic analysis of gas proves the presence of methane; the following chromatogram shows the pic of methane result of anaerobic digestion of OMW by cow manure (**Figure 5** & **Figure 6**).

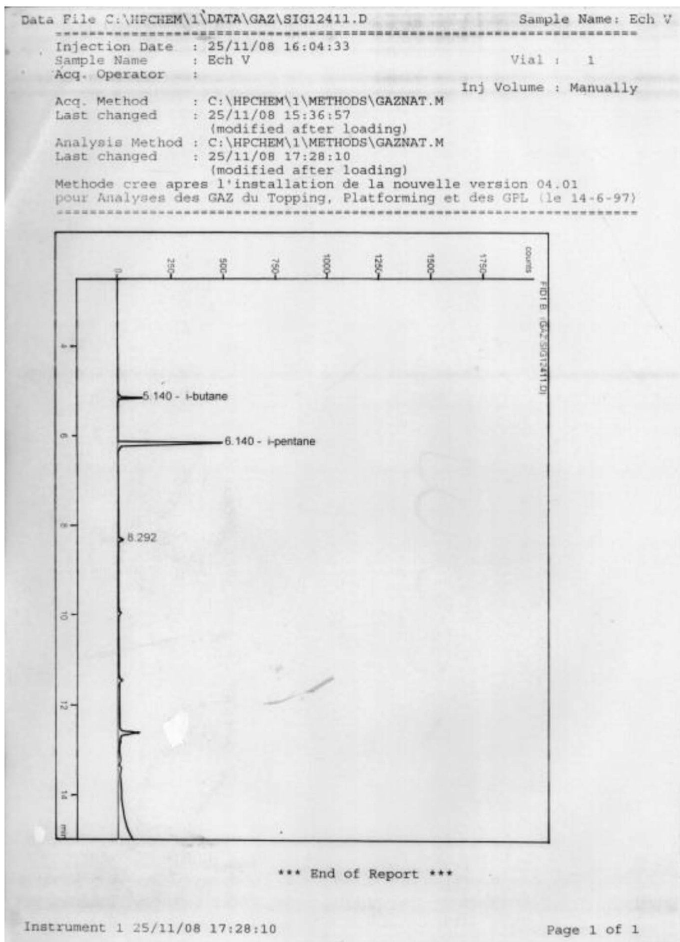

Figure 5. Original chromatogram.

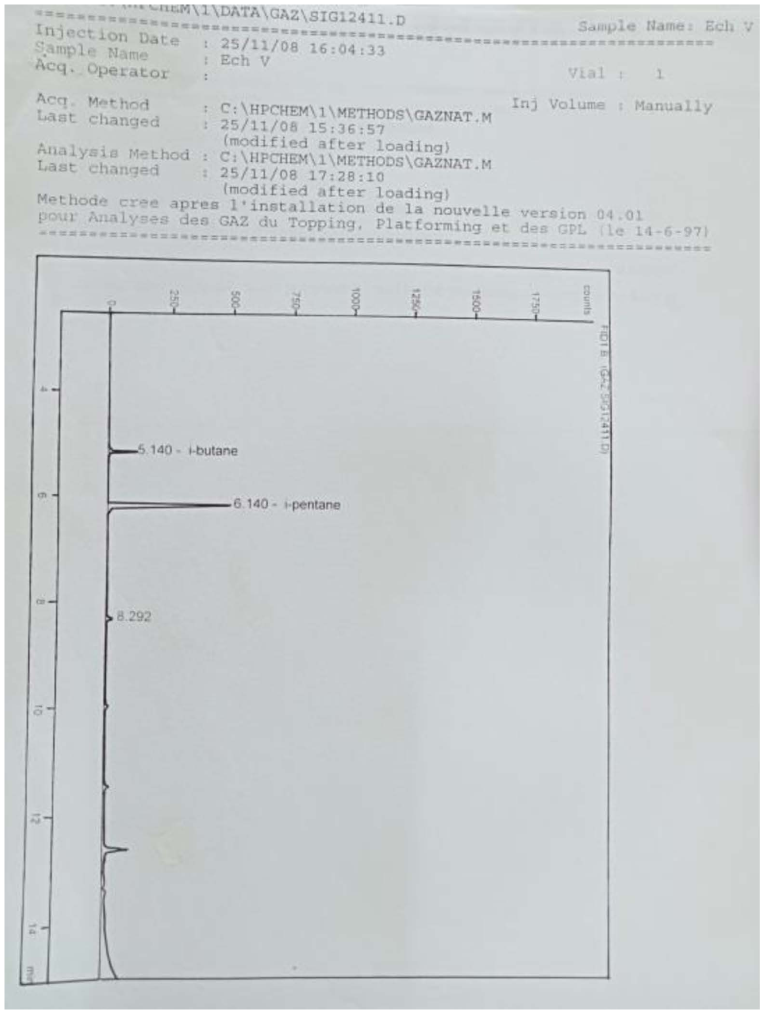

Figure 6. Figure caption.

3.3. Influence of Polyphenols

The OMW contains easily fermentable products like sugar and glucose, and also products hard to be degradable like fatty acids and polyphenols [21]; these polyphenols have inhibitor effect on the anaerobic digestion [22] but in the presence of methanogenic activities, they can be partially degradabled at pH near to 8.5. Then the biogas production varies according to the nature of manure (methanogenic activity) and the nature of OMW.

3.4. UV Analysis

We carried out the UV visible spectrophotometric analysis to pure OMW under a wave length of 725 nm. We found that the polyphenol concentration is about 1751.2 ppm.

We did the same analysis with a sample of OMW treated with cow manure. We found that the polyphenol concentration is about 0 ppm.

This informs us that the fermentation by cow manure damaged about 100% of the total phenol.

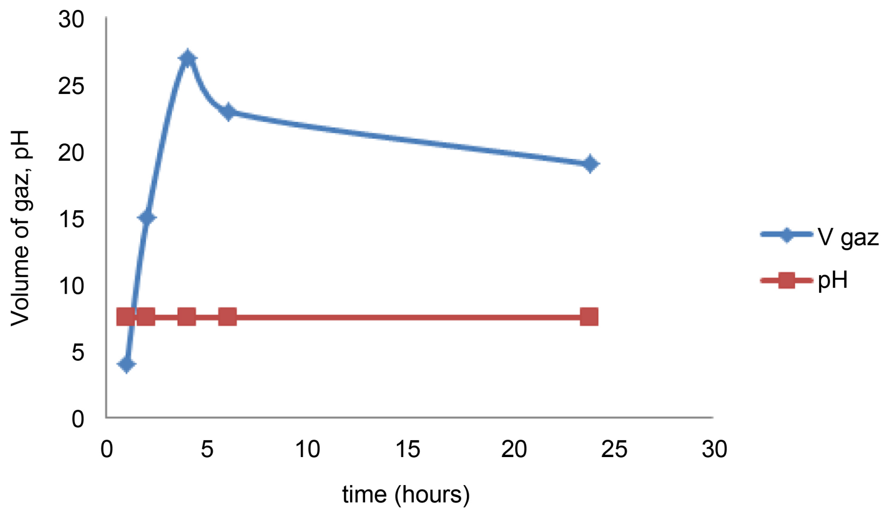

Figure 7. Variation of the volume of the gaz and pH according to time.

3.5. Influence of Incubation Time

We varied the incubation time four times (1, 20, 40, and 60) hours; we followed the variation of the production of the gas according to incubation time.

We observed that the volume of the gas increases between 15 and 21 hours, it reaches 15 ml after 20 hours; on the contrary, the value of pH stays constant (Figure 7).

4. Conclusions

This study reveals the importance of OMW anaerobic fermentation treated by cow manure.

This minimizes his undesirable effect on the environment by his degradation capacity under the action of biomass bacteria activity, the harmful organic material (decreasing the rate of polyphenols), and the production of energy in the form of biogas. This gas is butane.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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