Effect of agitation on the process of bi methanization of sludge from low-temperature wastewater treatment plants

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Wastewater treatment is a serious environmental problem, especially in developing countries such as Morocco. Their discharge into the natural environment has negative impacts on water resources and the environment. They are therefore purified to reduce the concentration of the main pollutants. Often biological remediation is used when wastewater contains biodegradable organic pollutants. This generally leads to treated water that can be discharged into the environment reused, or used as wastewater sludge. In the case of domestic or municipal water, this sludge has a fermentable character, which can have negative impacts on the environment especially when it is discharged directly into the environment. This sludge is subjected to various aerobic or anaerobic treatments to reduce the fermentable fraction. Anaerobic treatment seems more attractive because it gives the way to eliminate organic matter, produce biogas and manure compost both in one time. In this work, an experimental study was conducted on the ambient temperature anaerobic digestion of sewage sludge in two reactors, one operating without agitation and the other with agitation, to evaluate the impact of the latter on the biological process.

Key words: Bio-methanization, ambient temperature, agitation, methanogenic potential, sludge, wastewater treatment plant, COD, pH.

INTRODUCTION

Some solid wastes have a fermentable organic fraction which is an environmental problem. Their injection in the
environment could be a real threat. Nevertheless, this organic character can be used wisely, by valorizing its potential in value-added products when subjected to anaerobic biological treatment, which, in addition to reducing negative effects (emission of bad odors, methane, mercaptans, etc.), makes it possible to produce biogas and sludge mineralization for soil amendment. This solid waste includes sludge from wastewater treatment plants. They are often disposed of in landfills or dumped into the natural environment. In Morocco, their quantity is estimated at 435,600 tonnes/year (Afilal, 2013), and have a fermentable organic fraction between 50 and 70% (Eva, 2004); this is a good source for anaerobic biogas production, consisting mainly of methane 60 to 75% (Eva, 2004), the equivalent of 128,000 tonnes of CH₄ per year.

The anaerobic biological biomethanization treatment proceeds in four stages (Figure 1). The first step is the hydrolysis of organic matter, for example proteins, fats, cellulose and starch. These polymers such as amino acids, fatty acids and simple sugars are divided into monomers.

The second step is acidogenesis, during which the hydrolytes are oxidized to organic acids (for example lactates), alcohol (ethanol) or volatile fatty acids (propionate, butyrate and valerate).

In the third step, the resulting acidogenesis compounds are transformed into methane precursor products: acetic acid, carbon dioxide (CO₂) and hydrogen (H₂).

The fourth step biomethanization, which is done by two ways. One, it called hydrogenotrophic using hydrogenotrophic bacteria that draw their needs from the couple H₂/CO₂. These bacteria get their energy from the reduction of carbon dioxide by hydrogen to produce methane according to the following reaction.

\[ 4H_2 + CO_2 \rightarrow CH_4 + 2H_2O \]  (Reaction 1)

The other way called acetoclast where the strict anaerobic bacteria known as acetates, which extract their needs from the acetates; use acetate as their only carbon source (Reaction 2).

\[ CH_3COOH \rightarrow CH_4 + CO_2 \]  (Reaction 2)

The anaerobic process is a complicated method controlled by physico-chemical conditions such as the nature of the substrate, temperature, pH, agitation, etc. for instance, psychrophilic digestion is optimal between in a temperature range around 6 and 15°C, while mesophilic digestion in a range between 30 and 35°C; for thermophilic anaerobic digestion, it is favored and most effective at temperatures above 45°C because the reaction is accelerated by heat. However, anaerobic digestion is often used in mesophilic conditions, a compromise between performance and energy costs due to heating and especially because of its greater stability (Laskri, 2016).

The anaerobic fermentation takes place in a pH range between 5.5 and 8 for all phases. For the acidogenesis phase, it is between 5.5 and 6.5 (Souza et al., 2012), for acetogenesis, it is near neutrality. For methanogens, the pH range is between 6 and 8 (Gourdon, 2002).

Agitation plays a significant function in the anaerobic metabolic process. It ensures a good mixture, which improves the contact between the purifying bacteria and the sludge (Borole et al., 2006). Many studies have shown the importance of agitation on the fermentation process; in the first hand Johan Lindmark (Lindmark, 2014) evaluated the effect of mixing by comparing three regimes of mixing: 150 rpm, 25 rpm and continuous mixing; The results prove that the biogas production is better with a speed of 25 rpm which reaches 240 Nml, while the speed of 150 rpm gives a lower production of biogas; the author found that strong agitation inhibits the anaerobic fermentation process.

In the second hand, Hajji and Rhachi (2016) studied the influence of agitation on biogas production by comparing two reactors with and without agitation; he found a biogas production rate that reaches a value of about 0.61 m³ for a reactor of 40 rpm, then in the reactor without agitation the final volume of the final biogas is reduced up to 62%, most of these methanisation plants operate at temperatures of 37 or 55°C, however Ukondalemba et al. (2016) studied an anaerobic digestion with recirculation (type of agitation) of juice in the hydrolysis phase and acidogens at an ambient temperature of 25°C; results showed that 83% of the COD is transformed into biogas.

This study aimed at studying the biomethanization of sludge from a wastewater treatment plant at ambient temperature. The objective is to evaluate the effect of agitation (agitated versus no agitated reactors) on the anaerobic biological process by comparing two identical reactors.

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**Figure 1.** Diagram of the trophic chain of methanogenesis and its different stages (Moletta, 1993).
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MATERIALS AND METHODS

Origin and characterization of the substrate

The sludge samples collected from sludge wastewater treatment plant of the National Office of Water and Electricity (ONNEE) of Bourereg in Rabat-Morocco. This station receives discharges at an average rate = 86.4 m³/d, and a peak flow rate = 259.2 m³/d; these discharges have the following characteristics: COD = 15.1 Kg/d, BOD₅ = 9.0 Kg/d, mean TSS = 16.2 Kg/d. It is a station that produces between 7 to 10 Kg/d of fresh liquid sludge in the open air on drying beds and then stocked.

Before their use, the dry sludge (Plate 1) was crushed and sieved to obtain a homogeneous powder with a grain size of less than 2 mm. Masses of 5 g of these sludge are added to 125 ml of distilled water. The mixture was then agitated every 15 min and then rested every 15 min for 1 h (Hamdani, 2008).

The mixture was recovered to determine the following parameters:

1) The hydrogen potential (pH) is measured by using a multi-parameter probe.
2) The suspended matter has a centrifugation method.
3) The electrical conductivity.
4) Dissolved oxygen.

Organic matter

The chemical oxygen demand (COD) was determined according to the standard method (AFNOR T-101) (Kalloum et al., 2007).

Metal measurement: ICP-AES

Batch digester operation

The experimental system (Plate 2) used for the anaerobic digestion of sludge consists of two opaque plastic tanks each with a volume of 30 L. Each tank was filled with 1,350 kg of dry sludge at a rate of 50 g/L. The tests were conducted at an average ambient temperature of 18°C for 40 days. In the reactor, agitation is ensured by using a mechanically controlled agitation motor with a speed of 40 rpm (Kalloum et al., 2007).

The follow-up was made by daily samples of 50 ml from the reactors. The test was run during the month of February.
RESULTS AND DISCUSSION

Physical and chemical characteristics of sludge

Table 1 presents the characteristics of the sludge from the Bouregreg wastewater treatment plant. These results show that the sludge has a COD of 10.89 g/l, and contains a number of metals of significant concentrations, in this case Ca (71720 mg/L), Fe (15470 mg/L), Mg (8808 mg/L), Al (7125 mg/L) and to a lesser extent K (1301 mg/L), Na (1087 mg/L) and Zn (1652 mg/L). The organic matter content is 63%. The typology of this sludge is similar to that of domestic or municipal wastewater treatment plants (Choo-Kun, 2015; Mehrez et al., 2017).
The temperature variation

Figure 2 represents the evolution of temperature. The temperature data shows a same trend between the two samples (agitated reactor and unagitated reactor). The values vary between 17 and 24°C. However, it is noted that temperature was higher in the agitated reactor.

The pH variation

Figure 3 represents the evolution of the pH; the profile shows two phases: the first probably corresponding to the hydrolysis and acetogenesis phase; the pH decreased from 6.82 to 5.8 in the unagitated bioreactor and from 6.82 to 6.2 in the agitated bioreactor (Zhai et al., 2015; Kalloum et al., 2007); the second corresponding to the biomethanization phase; the pH increased from 5.8 to 6.5 in the unagitated bioreactor and from 6.2 to 7.5 in the agitated bioreactor. The agitated bioreactor had good pH conditions. This can be explained by a better use of volatile fatty acids (VFAs) by heterotrophic bacteria following their contact with the substrate enhanced by agitation process, as was highlighted by many authors.
It can also be explained by the effects of agitation in favour of the emission of gases formed during the various stages of anaerobic fermentation, including in particular acidic materials: CO$_2$, volatile fatty acids, H$_2$S, etc. The partial hydrogen pressure is particularly important in the process. Excessively high hydrogen content prevents the conversion of intermediate products from being converted. As a result, organic acids accumulate and prevent the formation of methane (Wandrey and Alivasidis, 1983); hydrogen sulphide had an inhibiting effect on methane formation. The inhibition thresholds encountered in methanogenic bacteria vary according to the type of substrate and physicochemical conditions and ranged from 50 to 1000 mg/L.

The variation of COD

Figure 4 represents the evolution of COD; the COD decreased for both bioreactors. This decrease is relatively more significant for the agitated bioreactor. The abatement rates achieved after 40 days are 60 and 81% respectively for the unagitated and agitated bioreactors. This difference is probably due to agitation, which on the one hand allows the organic matter and the purifying microorganisms present in solution to be mixed, as pointed out Haoqin Zhou and Zhiyou (Zhou and Wen, 2019), and on the other hand to facilitate the liberation and disengagement that can remain trapped in the solid raw material Haoqin Zhou and Zhiyou (Zhou and Wen, 2019), such as hydrogen formed during the acetogenesis step and H$_2$S which can be generated by sulfate-reducing bacteria, both inhibitors of bacterial activity and paricular for methanogenic bacteria with respect to hydrogen).

The work done by Kalloum (Kalloum et al., 2013) by using manual agitation showed a significant reduction in the COD of sludge to conclude that anaerobic digestion is an efficient method for the reduction of organic pollution and that most of the organic matter that is present in the digester is biodegradable. Pinho et al. (2004) confirmed that agitation rate plays an important role in the solubilization of suspended organic matter As well as the acceleration of the degradation of the COD in suspension. The same results were reported by Mehrez (Mehrez et al., 2017) and Ahmed et al. (2016).

Variation of dissolved oxygen

The Figure 5 represents the evolution of the dissolved oxygen; the concentration values reflect the correct performance of anaerobic digestion in both bioreactors. By the third day, this concentration decreases below the value required for anaerobic conditions. It then reaches an average concentration of 0.3 mg/l. This value is comparable to what is being reported by other authors (Botheju and Bakke, 2011).

Variation in conductivity

Figure 6 represents the evolution of The Electrical conductivity; the data showed an increase throughout the
anaerobic fermentation process. This could be due to the appearance of small species of high ionic mobility and/or mineralization of the environment. This is more pronounced in the case of the agitated bioreactor. (El Hafiane and El Hamouri, 2002) reported that conductivity increases from the inlet to the outlet of the reactor, indicating progressive mineralization of the medium.

Conclusion
The comparative psychrophilic anaerobic fermentation tests between two unagitated and agitated bioreactors have shown conclusive results regarding the role of agitation. Indeed, the monitoring parameters: pH, COD, temperature, conductivity highlighted the effect of agitation. The anaerobic fermentation processes are to the advantage of the agitated reactor. The agitation promotes the homogenization of the reactor and a good mixing between the substrate and the purifying bacteria. It also makes it possible to break the layer of solid products that can float on the surface of the solution and thus prevent the escape of certain gases such as AGV, CO$_2$, H$_2$, H$_2$S, which by their presence also make the medium acid and/or inhibit the activity of certain bacteria including methanogenic bacteria.
CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

REFERENCES

Affial ME (2013). Potentiel des déchets organiques et valorisation énergétique au Maroc. Bulletin d’Information de l’Académie Hassan II, Sciences et Techniques. http://www.academie.hassan2.sciences.ma/pdf/bulletin14.pdf

Ahmed T Khelafi M, Kaidi K (2016). Production De Biogaz A Partir Du Dechet De La Pomme De Terre. p. 6, mai 2016. Conference PAPER: JET 2016 à Hamammet en Tunisie, At Hôtel Laico Hammanet Tunisie.

Borole AP, Klasson KT, Ridenour W, Holland J, Karim K, Al-Dahhan MH (2006). Methane production in a 100-L upflow bioreactor by anaerobic digestion of farm waste. Applied Biochemistry and Biotechnology 131(1-3):887-896.

Botheju D, Bakke R (2011). Oxygen Effects in Anaerobic Digestion – A Review.pdf. The Open Waste Management Journal 4:1-19.

Choo-Kun M (2015). Intégration de la méthanisation des boues dans une filière alternative de traitement des eaux usées basée sur le procédé A/B: Vers la station d’épuration à énergie positive. p. 171. Ingénierie de l'environnement. INSA de Lyon, 2015. Français. (NNT : 2015ISAL0142). (tel-01417804).

Eva (2004). Valorisation des boues de la station d’épuration en biogaz. http://hmf.enseeht.fr/travaux/CD0405/beiere/4/html/binome3/biogaz.htm (consulté le juill. 18, 2019).

El Halfane F, El Hamouri B (2002). Performances d’un système anaérobie à deux phases dans l’épuration des eaux usées domestiques sous climat méditerranéen. Revue Marocaine des Sciences Agronomiques et Vétérinaires 22(3):133-141.

Gourdon R (2002). Aide à la définition des déchets dits biodégradables, fermentation, méthanisables, compostables. p. 153-200. https://record-net.org/storage/etudes/00-0118-1A/rapport/Rapport_record00-0118_1A.pdf 2.

Haji A, Rhachi M (2016). Effet de l’agitation sur la digestion anaérobie des déchets ménagers et assimilés en régime mésophile Effect of agitation on anaerobic digestion of household and similar waste under mesophilic regime. p. 8 Journal of Materials and Environmental Science 7:11:4136-4143: ISSN : 2028-2508.https://www.materenvironics.com/Document/voi7/voi7_N11/443-JMES-2344-Haji.pdf

Hamdani I (2008). Gestion des boues des stations d’épuration au Maroc : Quantification, caractérisation et options de traitement et de valorisation. p. 116. https://www.susana.org/en/knowledge-hub/resources-and-publications/library/details/2006, juill. 2008.

Kalloum S, Khelafi M, Djiafri M, Tahri A, Touzi A (2007). Etude de l’influence du pH sur la production du biogaz à partir des déchets ménagers. Revue des Energies Renouvelables. 10(4):539-543.

Kalloum S, Iddou A, Khelafi M, Touzi A (2013). Utilisation du procédé de la digestion anaérobie pour le traitement des boues des stations d’épuration des eaux usées. Les Energies Renouvelables 16(4):611-618.

Laskri N (2016). Dépôt des déchets riches en matière organique (boues de station d’épuration et déchets d’abattoir) Par digestion anaérobie. http://biblio.univ-annaba.dz/wp-content/uploads/2017/04/These-Laskri-Nabila.pdf

Lindmark (2014). The effects of different mixing intensities during anaerobic digestion of the organic fraction of municipal solid waste. Waste Management 34(8):139-1397. doi: 10.1016/j.wasman.2014.04.006. https://www.sciencedirect.com/science/article/abs/pii/S0956053X14010469

Mehrez I, Kalloum S, Khellfi O (2017). Study of biogas production from lagooning sludge by anaerobic digestion. p. 6. Journal of Materials, Processes and Environment May edition 5:1.

Monou M, Kythreotou N, Fatta D, Smith SR (2009). Rapid screening procedure to optimise the anaerobic codigestion of industrial biowastes and agricultural livestock wastes in Cyprus. Waste management 29(2):712-720.

Pinho SC, Ratusznei SM, Rodrigues JA, Foresti E, Zaiat M (2014). Influence of the agitation rate on the treatment of partially soluble wastewater in anaerobic sequencing batch biofilm reactor. Water Research 48(9):2699-2705.

Souza MA, Chaguri MP, Castelini FR, Lucas Junior JD, Vidotti RM (2012). Anaerobic bio-digestion of concentrate obtained in the process of ultra filtration of effluents from tlapia processing unit. Revista Brasileira de Zootecnia 41(2):242-248.

Ukondalemba LM, Aleke AL, Ngahane EL, Anki DME, Vasel J (2016). Valorization of Organic Household Waste and Septic Tank Sludge By Anaerobic Digestion. International Journal of Innovation and Scientific Research 20(2):272-281. http://www.ijissr-journals.org/abstract.php?article=IJISR-15-245-05.

Wandrey C, Aivasidis A (1983). Zur reaktionstechnik der anaeroben fermentierung. Chemie Ingenieur Technik 55(7):516-521.

Zhai N, Zhang T, Yin D, Yang G, Wang X, Ren G, Feng Y (2015). Effect of initial pH on anaerobic co-digestion of kitchen waste and cow manure. Waste Management 38:128-31.