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Anaerobic Co-digestion with different concentrations of whey and cow dung in biogas production

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Abstract*– In this research, the whey potential for optimization of biogas production by anaerobic co – digestion with cow dung was studied. It used a two-component multifactorial – design, where it was studied four batch type - biodigesters of 4 liters on a laboratory scale with different whey concentrations (0%, 10%, 30%, 50%) and dilute cow dung in relation 1:1.5, each one in three replications. The experimentation was performed under temperature control of 35°C±2°C; and pH control between 6.4 to 6.9 by the addition of NaOH 3N; with an average of 7% of total solids and a holding time of 35 days. Below these conditions, the 2nd biodigester with 10% of whey, it was the one which generate the largest amount of biogas with 37.9 liters in total; the 3rd biodigester, with 30% of whey generates 24.2 liters of biogas; and the 4th biodigester, with 50% of whey generates 14.5 liters. The reduction of total solids in biodigesters were 38.5%, 43.4%, 40.2% and 27.6% respectively. We used the Gompertz model, which allowed us to find high correlation grades oscillating between 86.295% y 94.268%. Therefore, it can be established, that the use of whey duplicates the biogas production until 52% and sets a limit of 30% (v/v) to obtain a good biogas production in batch-type biodigesters.

***Keywords*—Whey, cow dung, co - digestion, biogas, Gompertz model**

I. Introduction

The bioenergy is a renewable energy obtained from biomass, and its principles benefits are the reduction of greenhouse gases and waste disposal [1]. The use of non-renewable energy that are dependent of petrol is a worldwide problem, for this reason is important the use of renewable energies such as solar, wind or biomass (energy derived from biomass) [2]. Biomass is an important fuel in rural zones because it is a renewable non-fossil organic matter which can be gather and used to power a bioenergy plant [3].

The agricultural industry's waste emission is a significant contributor to environmental pollution, creating the necessity to allocate more resources towards biotechnology research to discover superior treatment methods for reuse. Considering this, anaerobic digestion emerges as a promising solution for transforming domestic or industrial organic waste into energy efficiently and effectively. [4].

Anaerobic digestion is a biological process that is developed in the absence of oxygen where a specific group of bacteria decompose the organic matter into gas products or biogas and digestate. Biogas is mainly composed of methane (CH_4), carbon dioxide (CO_2), hydrogen (H_2), hydrogen sulfide (H_2S) and ammonia (NH_3). The digestate consists of a mixture of mineral products (Ca, N, P, K) and compounds that are difficult to degrade, a liquid with fertilizing characteristics [5].

Anaerobic digestion technology has gained significance due to its valuable environmental benefits and potential to generate income for farmers. The economic viability of this technology greatly depends on several factors such as investment costs, operational expenses of biogas plants, and methane production optimization. [6].

A stable anaerobic digestion process needs balance to evade the accumulation of intermediate inhibitory compounds because it can cause a fall in pH in the middle of the process. The pH stability can be facilitated with a good balance between CO_2 and bicarbonate. Thus, a parameter that it measure is the alkalinity [7].

Livestock farming is one of the most natural resource consumers, being the grass and ground destinate for grain production to animal feeding 80% of all the agricultural land [8]. Furthermore, the use of anaerobic digestion is a promising alternative technology in respect of biomass application in energy generation, the establishment of biodigesters in rural zones is favorable to the environment and farmers [9].

Another organic waste from agriculture industries is cheese whey, which is highly polluting, it is produced in milk plants [10], also, its structure contains sugars, proteins and dissolved fats that can be transformed into methane [11].

The biogas technology establishes a sustainable alternative for organic agroindustry residues; due to the large amount and susceptibility to being converted into biogas. In addition, it has been a success from the economic and ecologic point of view; this has allowed reducing of treatment costs and waste disposal [12].

The systems of biogas generation are categorized according to the operation method, the anaerobic digestors can be classified as batch or continuous systems. Through the batch process, the raw material is added to the digester and the inoculum is added. After that, the system close and it is allowed to complete the digestion depending on the parameters [13].

The region of Cajamarca in Peru is primarily known for its cattle raising and cheese production. However, the waste generated from these industries, such as cow dung and cheese whey, cannot be disposed of directly into the environment. It is imperative to find a sustainable solution to this issue. Therefore, utilizing animal biomass as a raw material to produce electrical power and biofertilizer can serve as a promising alternative for the livestock and cheese industries. Treating cow dung and

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cheese whey as waste is not an option anymore. Rather, generating energy from these byproducts can be a viable solution to address the growing issue of waste management in the region

Anaerobic co-digestion means the digestion of two or more raw residues together in a process, which could improve the speed of this, the biodegradation, waste stabilization, digestate and raw methane production. Co-digestion can also increase the several factors that affect the digestion process such as diluting the toxic residues/inhibitors ratio and achieving a better total solids content, nutrient balance, and alkalinity.

For these reasons, the objective of this research is to study the effect of whey addition in cow dung under anaerobic conditions to find out the effectiveness in increasing biogas production and the determination of the operating variables involved in its production.

II. MATERIALS AND METHODS

Whey from the manufacture of cheese was used in the research, without any treatment; provided by a factory of matured chess in Cajamarca – Perú; which was refrigerated to 7°C until later use. The manure used was collected in the stable of the Cajamarca – Perú zone, corresponding to cattle of Jersey raze which was diluted in drinking water about 1:1.5. Before the use of these materials, they were adjusted to a 7.5 pH on average, using sodium hydroxide (NaOH) 3N. 4 different treatments (BD-1:0%, BD-2:10%, BD-3: 30%, BD-4:50%), where it used different concentrations of cheese whey in cow dung. Each treatment corresponds to a biodigester per batch, operated at a volume of 3 liters, which was studied in three repetitions each. It evaluated the effect of the concentrations of both components on the production of daily and accumulative biogas. The biodigesters were kept in heat chambers at a temperature of 35 ±2°C and kept the pH constant, adding NaOH 3N. The pH and temperature data were taken daily for each biodigester, the percentage of solids was determined at the beginning and end of the treatment, the combustion of the biogas was checked daily and the amount of methane in the biogas was determined using the Mariotte bottle method, doing each ratio in six repetitions. The data obtained using a multilevel factorial model was statistically analyzed.

III. RESULTS

A. Biogas Production

The hypothesis of this research states that cow dung has little organic matter in soluble form and many suspended solids, for this reason its biodegradability is less than 50%. On the other hand, the cheese whey has barely suspended solids and almost entirely soluble organic matter, which is why its anaerobic biodegradability is 100%. The cheese whey does not have alkalinity due to the bicarbonate, so in the experimental phase the pH was controlled in the biodigesters 1, 2, 3 and 4 with 0%, 10%. 20% and 50% of cheese whey respectively; the pH was unstable in the first 3 days for biodigesters 2, 3 and 4, since NaOH 3N was added until day 4, after that it was stable. As the days when by, the pH was corrected so that did not fall below 6.5, the pH values were within the range in which anaerobic digestion can be conducted, 6.2 to 7.8.

It was used 50, 80, 120 and 180 ml of NaOH in the biodigesters 1, 2, 3 and 4 to neutralize the organic acids that cause the pH values; it is needed a greater quantity of alkalizing, which means it must fill an excess volume in the biodigester so this can generate some drawbacks.

In figure 1, it can be observed that during the first 4 days, the amount of biogas is high, with 560mL for biodigester 1 (0% whey); 930mL for biodigester 2 (10% whey); 1200mL for biodigester 3 and 1250mL for biodigester 4 (50% whey); the reason of this values are because anaerobic digestion first phases (Hydrolysis and Acidogenesis), since in this phase is where the organic acids are generated with a high CO_2 production [8].

The maximum biogas production report are: 2650 mL, 2760 mL, 2030 mL, and 1100 mL, for biodigester 1, 2, 3 and 4, respectively. In addition, it can be clearly seen that biodigester 2 with 10% whey, exceeds the daily amount of biogas compared to other digesters. After day 15, it appears the maximum production of biogas, this tends to decrease; probably due to solid reduction and ammonia accumulation from protein digestion [14], given that in the last 2 weeks the addition of NaOH was not necessary and probably the ammonia provided the alkalinity to the medium; but since this a critic compound in the toxicity and inhibition of the medium [15], its high concentration inhibit the methanogenesis process.

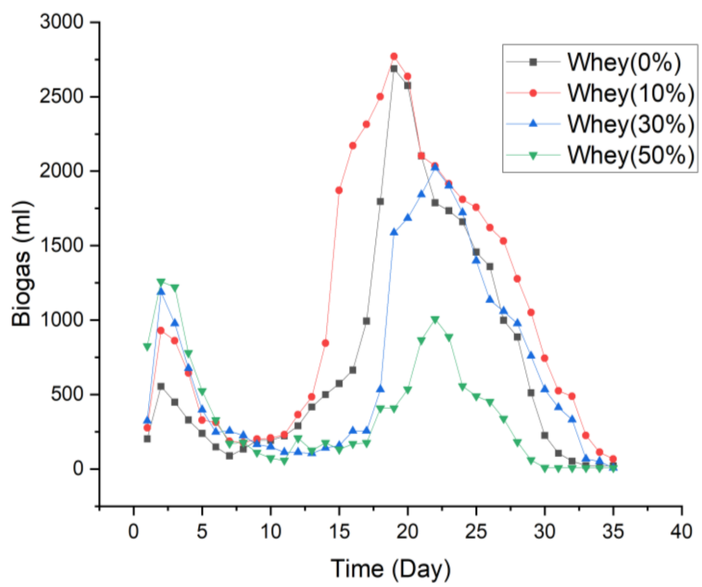

Fig. 1. Daily Biogas Production

Figure 2 shows the biogas accumulation during the process of digestion to biodigesters 1, 2, 3 and 4. In Table 2, we can see it was obtained for biodigester 1: 24916 mL, for biodigester 2:

37868 mL, for biodigester 3: 24285 mL and biodigester 4: 14455 mL. The production of biodigester 2 with 10% of whey exceeded by 52% to biodigester 1, that only have cow dung. Biodigester 3 only was surpassed by 2.5% by biodigester 1. The biogas production of biodigester 4 was not good, but it obtains biogas with a long-time adaptation.

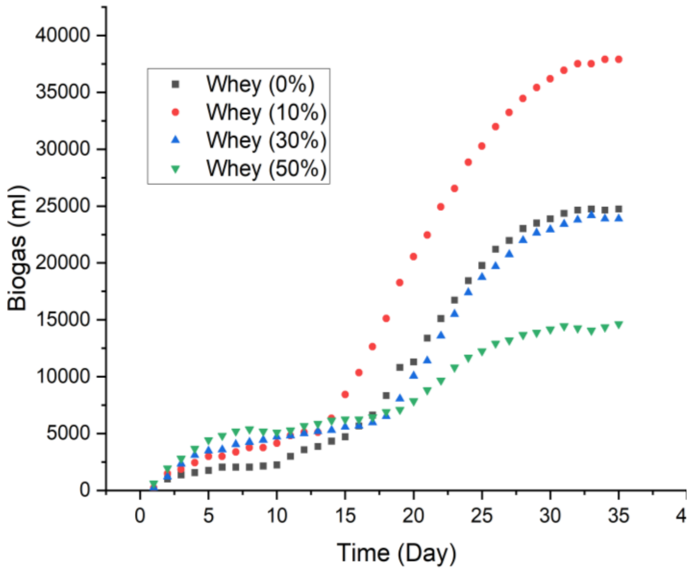

Fig. 2. Biogas accumulated production

One of the principle models to predict the kinetic analysis of biogas generation process with more detail is the modified Gompertz model [16], which is shown in equation 1:

$$ln\left(\frac{Y_n}{Y_0}\right) = a * e^{-b*e^{-c*t}} \quad (1)$$

In this equation, "Y" means the biogas accumulative production (in milliliters per solid gram) and "t" is the time of the experiment (in days). The parameters that can be estimated with these models include the potential of biogas generation "a" (in milliliters per solid gram), that relates to the substrate, the maximum rate of biogas generation "b" (milliliters per gram of dry volume per day), and delay time "c" (day). The value obtained for each treatment can be seen in Table 1.

TABLE I
GOMPERTZ MODEL INDICATORS

Gompertz model indicators	Whey (0%)	Whey (10%)	Whey (30%)	Whey (50%)
a	6.822	7.385	6.954	5.968
b	1.192	0.983	0.801	0.516
c	0.092	0.081	0.067	0.088
∑MSE	0.225	0.087	0.127	0.060
R^2	87.884	94.268	87.553	86.295

The estimation of kinetic parameters by the Gompertz model helped in the evaluation of digester performance and reactor stability. We used the modified Gompertz model to derive a correlation to biogas production from the anaerobic digestion of cow dung. This proved that the digestion process is more feasible with a concentration of 10% of whey. The model allows us to find high correlation coefficients oscillating between 86.295% and 94.268%, we can say that this model can predict the kinetic behavior of the anaerobic digestion process by considering the inhibitory behavior of methane production. mixtures that have more whey content report higher lag phase times. The untreated whey contains a high amount of complex proteins which are hard to degrade and thus lag phase times are found a bit longer.

Additionally, the biodigesters had different adaptation times, meaning that the development of methanogenic bacteria was different for each biodigester. In Table II, it shows the time that the biogas combustion was observed. This gives us the idea of biogas quality, because the biogas can only be combusted when the methane content is higher than 50% [17]. From this moment, we proceed to store the biogas in tire tubes to obtain biogas of good quality.

TABLE II
COMBUSTION TEST LOG

Treatment	Day 5	Day 7	Day 10	Day 15	Day 25	Day 32
Whey (0%)	-	-	+	+	++	++
Whey (10%)	++	++	++	++	++	++
Whey (30%)	-	+	++	++	++	++
Whey (50%)	-	-	-	+	++	++

(+): Combustion with pilot flame aid (++): Combustion without pilot flame aid. (-): Combustion is not observed.

It is observed that biodigester 2 with a concentration of 10% whey, produced a higher quality of biogas, since during the combustion test, the flame remains lit without the help of the pilot flame and its production is prolonged throughout the thirty-five days of digestion and exceeds the production levels of the other experimental mixtures. Biodigester 3, with 30% whey, generated a similar amount of biogas to biodigester 1, in which only manure was used; considering that in this digester there are less anaerobic bacteria; by the lower volume of manure in the digester.

B. *Statistical analysis*

The multifactorial analysis was performed which gave an adjustment of R^2 of 95.56% showing that the selected model adjusted to the data properly, when performing the analysis of significance time and percentage of whey, it displays significant effects on biogas production ($p<0.05$).

Figure 3 shows the standardized Pareto which displays that time had the greatest effect on biogas production, followed by the percentage of whey used. Likewise, we can see in figure 4 the graphical representation of the individual effects on the specific methanogenic productivity of the independent variables, where the main finding is that the lowest values of whey are those that obtain the greatest amount of biogas, expressing it in values of specific productivity, we can affirm that the samples of tested co-digestion was decreasing when the percentage of serum increase, obtaining specific methanogenic productivity with values between those of the substrate samples.

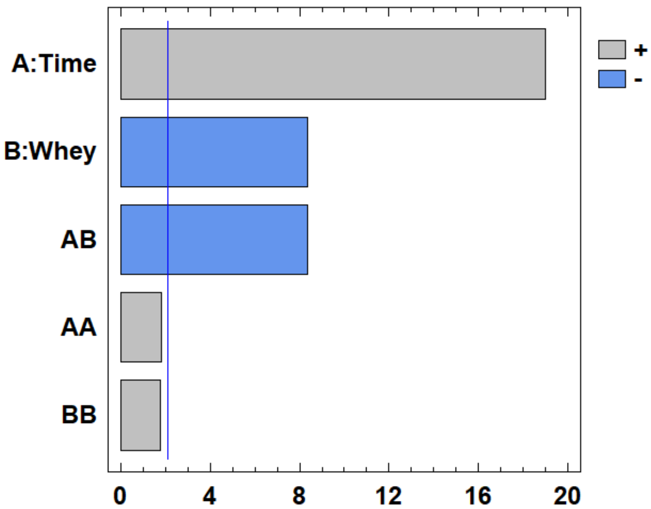

Fig. 3 Standardized Pareto for biogas production

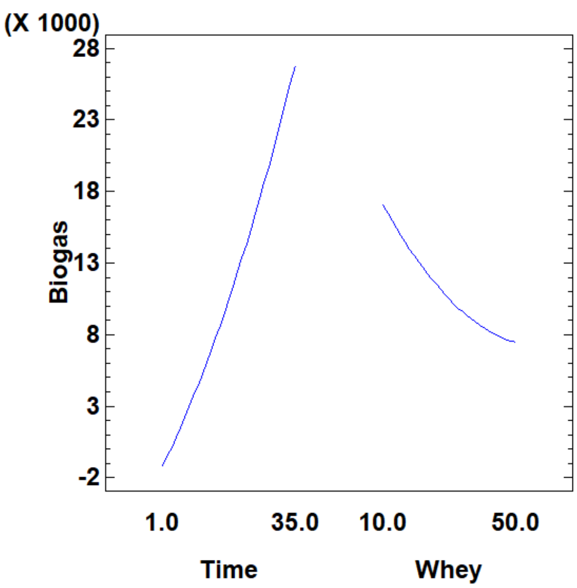

Fig. 4. Main effects for biogas production

In figure 5 is represented in 3 dimensions the interaction effect of the two independent variables in all their range of value. If the represented values in the figure are negatives, it means that the estimated value for the model is higher than the real value and if the value is positive, means that the real value is higher than the predicted value by the model. The equation of the model is shown below and has an adjust R^2 of 94.55 %.

$$B = -2424.7 + 1190.26*T - 107.791*W + 8.13355*T^2 - 22.0392*T*W + 4.41076*W^2$$

B: Biogas, T: time y W: whey

Additionally, figure 5 shows the 2D diagram, where is shown the contour lines/outlines predicted by the model in which for different values of two independent variables would obtain the same value of methanogenic specific productivity, displaying the area that would get a higher production which is represented in yellow color.

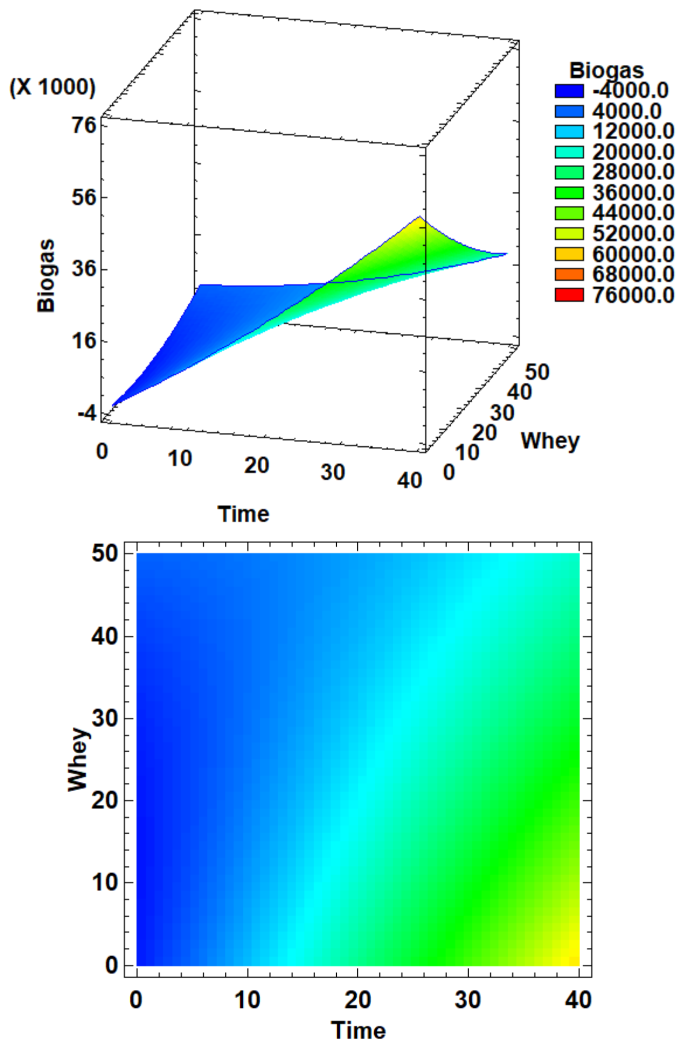

Fig. 5. Response surface for biogas production

Similarities studies indicated that is possible the use of larger amounts of whey, as in the case of Comino [18], he achieved a maximum increase of biogas of 79% with regard to the start-up phase using a high volume of whey about 60%, but with a pretreatment of the cow dung; other studies always see the substrate improvement to increase the biogas production and its quality, such as the use of sewage sludge [19], food waste [20] which together with anaerobic co-digestion turns into a well stablish technology to recovery water resources facilities to maximize methane production.

C. Solid Total Removal

The results are displayed in figure 6, where the red diagrams show pretreatment values and the blue one shows the values after co-digestion process, where is observed a solid total removal of 38.5% for biodigester 1 with 0% of whey () (Control), 43.4% for biodigester 2 with 10% of whey, 40.2% for biodigester 3 with 30% of whey and 27.6% for biodigester 4 with 50% of whey. The values of biodigesters 1 and 2 are at the minimum reported that can be allowed to have an efficient biogas production, the removal of organic material has to be between 40% and 60% of removal [21].

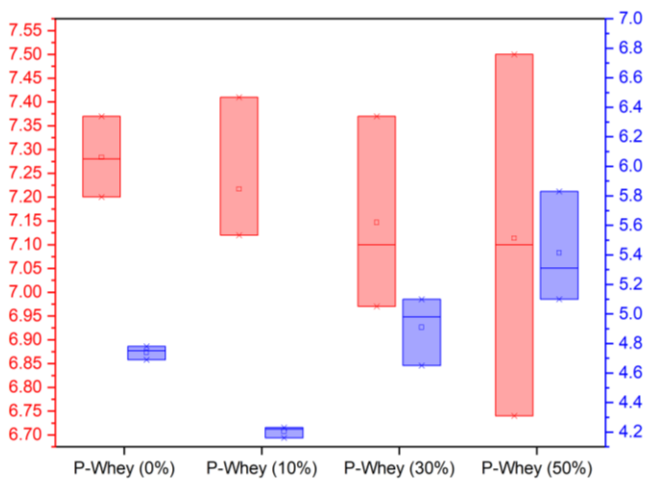

Fig. 6. Removal percentages

D. Methane percentage in biogas

The methane percentage in different biogas samples determined by the Mariotte bottle method are displayed in figure 7. Even though this method has an error rate of 10% regarding Chromatography analysis (López n.d.); we can observe a certain degree of biogas quality difference for the different whey concentrations in the biodigesters; the biodigester 2 reported 78% and the biodigester 4, 66% of methane; the biodigester 3 with 30% of whey provided us the highest methane content, with 82%; unlike biodigester 1 that only had cow dung, this provided 71% of methane; according to Torres (2008), these methane high values are due to CO_2 reduction in biogas for the use of NaOH, which reacts with the CO_2 to form Na_2CO_3, that gives alkalinity. This is why the values exceed a small proportion of those exposed by Steffen (1998), which establishes the methane percentage in biogas produced by whey at 80% and cow dung at 75%.

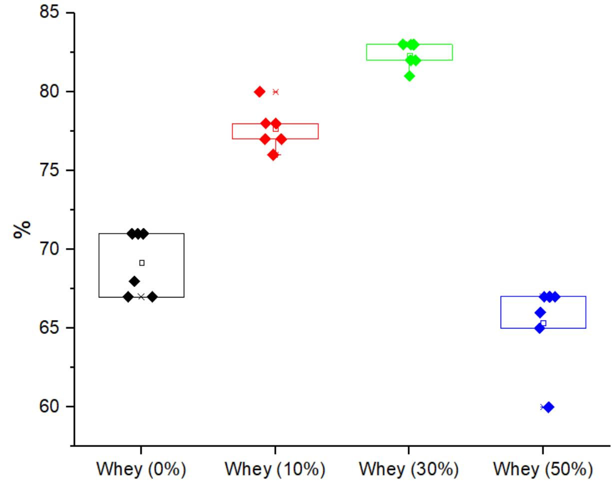

Fig. 7. Percentage of methane in different biogas samples

IV. Conclusions

The results of this research demonstrate that methane production by co-digestion of cheese whey and cow dung is possible, the data from an anaerobic digestor in a laboratory scale suggested that the addition of 10% whey has a higher biogas production potential compared to whey-free substrate, with an improvement of 52%. The anaerobic digestion of whey and cow dung cannot be conducted without pH control so it must always have the addition of some alkalizer. Experimental data from tests were well described by the modified Gompertz model. In terms of optimizing biogas production using whey and beef manure, the following operating variables are established: a maximum whey concentration of 10%, a mesophilic temperature of 35°C, a pH higher than 6.5, constant stirring, a solids concentration of 7% and a retention time of 35 days. We can conclude that in the kinetics of anaerobic digestion, manure has its limiting stage in the hydrolytic phase, this stage for the cheese serum is the methanogenic phase. Finally, we can say that this type of mixture has the energy potential typical of the co-digestion of energy crop residues and livestock. Even digestate has a valuable methane yield.

References

[1] R. H. G. de Jesus, J. T. de Souza, F. N. Puglieri, C. M. Piekarski, y A. C. de Francisco, «Biodigester location problems, its economic–environmental–social aspects and techniques: Areas yet to be explored», *Energy Reports*, vol. 7, pp. 3998-4008, nov. 2021, doi: 10.1016/j.egyr.2021.06.090.

[2] B. Singh, Z. Szamosi, Z. Siménfalvi, y M. Rosas-Casals, «Decentralized biomass for biogas production. Evaluation and potential assessment in Punjab (India)», *Energy Reports*, vol. 6, pp. 1702-1714, nov. 2020, doi: 10.1016/j.egyr.2020.06.009.

[3] Y. Chen, J. J. Cheng, y K. S. Creamer, «Inhibition of anaerobic digestion process: A review», *Bioresource Technology*, vol. 99, n.º 10, pp. 4044-4064, jul. 2008, doi: 10.1016/j.biortech.2007.01.057.

[4] N. Kalaiselvan *et al.*, «A waste to energy technology for Enrichment of biomethane generation: A review on operating parameters, types of biodigesters, solar assisted heating systems, socio economic benefits and challenges», *Chemosphere*, vol. 293, p. 133486, abr. 2022, doi: 10.1016/j.chemosphere.2021.133486.

[5] L. C. Corrales, D. M. Antolinez Romero, J. A. Bohórquez Macías, y A. M. Corredor Vargas, «Bacterias anaerobias: procesos que realizan y contribuyen a la sostenibilidad de la vida en el planeta», *Nova*, vol. 13, n.º 24, pp. 55-81, jul. 2015.

[6] D. Patowary y D. C. Baruah, «Effect of combined chemical and thermal pretreatments on biogas production from lignocellulosic biomasses», *Industrial Crops and Products*, vol. 124, pp. 735-746, nov. 2018, doi: 10.1016/j.indcrop.2018.08.055.

[7] T. Xia, H. Huang, G. Wu, E. Sun, X. Jin, y W. Tang, «The characteristic changes of rice straw fibers in anaerobic digestion and its effect on rice straw-reinforced composites», *Industrial Crops and Products*, vol. 121, pp. 73-79, oct. 2018, doi: 10.1016/j.indcrop.2018.04.004.

[8] S. A. Guares, J. D. de Lima, y G. A. Oliveira, «Techno-economic model to appraise the use of cattle manure in biodigesters in the generation of electrical energy and biofertilizer», *Biomass and Bioenergy*, vol. 150, p. 106107, jul. 2021, doi: 10.1016/j.biombioe.2021.106107.

[9] R. Alvarez y G. Lidén, «Semi-continuous co-digestion of solid slaughterhouse waste, manure, and fruit and vegetable waste», *Renewable Energy*, vol. 33, n.º 4, pp. 726-734, abr. 2008, doi: 10.1016/j.renene.2007.05.001.

[10] J. Oblitas, J. Morales, F. Rimarachin, W. Castro, y T. Chuquizuta, «Selección de factores clave en el rendimiento de biomasa de Kluyveromyces fragilis a partir de suero de queso usando un diseño Placket-Burman», ago. 2020, Accedido: 31 de agosto de 2021. [En línea]. Disponible en: http://laccei.org/LACCEI2020-VirtualEdition/meta/FP219.html

[11] J. L. Magaña-Ramírez, R. Rubio-Núñez, H. Jiménez-Islas, y M. T. Martínez-García, «Anaerobic treatment of lactic waste and goat manure», *Ingeniería e Investigación*, vol. 31, n.º 1, Art. n.º 1, ene. 2011, doi: 10.15446/ing.investig.v31n1.20530.

[12] X. Wang, X. Lu, F. Li, y G. Yang, «Effects of Temperature and Carbon-Nitrogen (C/N) Ratio on the Performance of Anaerobic Co-Digestion of Dairy Manure, Chicken Manure and Rice Straw: Focusing on Ammonia Inhibition», *PLOS ONE*, vol. 9, n.º 5, p. e97265, May 2014, doi: 10.1371/journal.pone.0097265.

[13] B.-S. Xing, S. Cao, Y. Han, X. C. Wang, J. Wen, y K. Zhang, «A comparative study of artificial cow and sheep rumen fermentation of corn straw and food waste: Batch and continuous operation», *Science of the Total Environment*, vol. 745, 2020, doi: 10.1016/j.scitotenv.2020.140731.

[14] A. Zainal, R. Harun, y S. Idrus, «Performance Monitoring of Anaerobic Digestion at Various Organic Loading Rates of Commercial Malaysian Food Waste», *Frontiers in Bioengineering and Biotechnology*, vol. 10, 2022, Accedido: 2 de enero de 2023. [En línea]. Disponible en: https://www.frontiersin.org/articles/10.3389/fbioe.2022.775676

[15] Y. Ren, C. Wang, Z. He, Y. Qin, y Y.-Y. Li, «Biogas production performance and system stability monitoring in thermophilic anaerobic co-digestion of lipids and food waste», *Bioresource Technology*, vol. 358, p. 127432, ago. 2022, doi: 10.1016/j.biortech.2022.127432.

[16] N. Ben Khedher *et al.*, «Modeling of biogas production and biodegradability of date palm fruit wastes with different moisture contents», *Journal of Cleaner Production*, vol. 375, p. 134103, nov. 2022, doi: 10.1016/j.jclepro.2022.134103.

[17] J. F. González, A. I. Parralejo, J. González, A. Álvarez, y E. Sabio, «Optimization of the production and quality of biogas in the anaerobic digestion of different types of biomass in a batch laboratory biodigester and pilot plant: Numerical modeling, kinetic study and hydrogen potential», *International Journal of Hydrogen Energy*, vol. 47, n.º 93, pp. 39386-39403, dic. 2022, doi: 10.1016/j.ijhydene.2022.09.113.

[18] E. Comino, V. A. Riggio, y M. Rosso, «Biogas production by anaerobic co-digestion of cattle slurry and cheese whey», *Bioresource Technology*, vol. 114, pp. 46-53, jun. 2012, doi: 10.1016/j.biortech.2012.02.090.

[19] K. Bella y P. Venkateswara Rao, «Anaerobic co-digestion of cheese whey and septage: Effect of substrate and inoculum on biogas production», *Journal of Environmental Management*, vol. 308, p. 114581, abr. 2022, doi: 10.1016/j.jenvman.2022.114581.

[20] T. J. Bolen *et al.*, «Feasibility assessment of biogas production from the anaerobic co-digestion of cheese whey, grease interceptor waste and pulped food waste for WRRF», *Energy*, vol. 254, p. 124144, sep. 2022, doi: 10.1016/j.energy.2022.124144.

[21] X. Flotats, E. Campos, J. Palatsi, y A. Bonmatí, «Digestión anaerobia de purines de cerdo y codigestión con residuos de la industria alimentaria», ene. 2001.