

Investigating the Performance of Anaerobic Co-Digestion of Primary Sludge and Acid Whey Using a Twin Pilot Scale System

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Abstract Anaerobic Digestion (AD) has proven to be an effective method for transforming organic waste into biogas, a renewable energy source that can play a crucial role in reducing our dependence on finite fossil fuels. Despite its widespread use, mono-digestion, where a single substrate is utilized, is prone to several drawbacks, including process instability, limited feedstock options, and low biogas/methane efficiency. To overcome these limitations, anaerobic co-digestion (AcoD) has emerged as a promising alternative. In AcoD, multiple substrates are digested simultaneously, leading to a more stable process, a greater variety of feedstock options, and an increased biogas/methane yield. The co-digestion of wastewater treatment plant (WWTP) sludge and food processing wastes has proven to be a popular area of research in AcoD, offering a balanced mix of carbon, nutrients, and minerals for optimal digestion. This study investigates the performance of AcoD between primary sludge (PS) and acid whey (AW) compared to PS mono-digestion, using a twin 15 L pilot scale continuous flow complete stirring tank reactor (CSTR) system. The experiment was carried out at a mesophilic temperature (35 °C) with an increasing organic loading rate (OLR). The results suggest that AcoD of PS and AW is a promising approach as optimal process conditions were maintained while the biogas yield increased.

Keywords: Anaerobic co-digestion, AcoD, Anaerobic digestion, AD, Primary sludge, Acid whey, Biogas, Methane, waste to energy, continuous flow, BMP

1. Introduction

Anaerobic digestion (AD) technology offers numerous benefits, such as the generation of renewable energy, the reduction of greenhouse gas emissions, and the treatment of organic waste. However, mono-digestion, which utilizes a single substrate, has some limitations that can affect its efficiency and process stability (Hagos et al., 2017). It is worth noting that mono-digestion of readily biodegradable substrates can be challenging, as the accumulation of volatile fatty acids (VFAs) may hinder the performance of the process. Additionally, some substrates may be deficient in essential nutrients required for microbial growth, resulting in low biogas yield and organic removal efficiency, or even complete process failure (Schnürer, 2016). Anaerobic co-digestion (AcoD) has emerged as an alternative to mono-digestion to improve the process by using multiple substrates simultaneously. The co-digestion

of various substrates offers several advantages, including enhanced biogas production, increased process stability, and a more comprehensive utilization of resources (Nwokolo et al., 2020). In particular, the co-digestion of wastewater treatment plant (WWTP) sludge and food processing wastes has gained attention because it offers a balanced mix of carbon, nutrients, and minerals for optimal digestion conditions.

Primary sludge (PS) generated during wastewater treatment process, is rich in organic matter and nutrients, making it an ideal co-substrate. Acid whey (AW), on the other hand, is a by-product of the dairy industry that contains high amounts of lactose, making it a promising substrate for biogas production. However, due to the high content of readily biodegradable organic matter in AW and the low pH, its mono-digestion can be challenging. The accumulation of VFAs and the resulting decrease in pH can lead to process instability and reduced biogas yields. Biomethane potential (BMP) tests are widely used for measuring methane yield of a wide range of waste. This kind of tests give a very useful insight on methane potential of substrates and also is a very effective method to investigate the performance of co-digestion of two or even more waste combined on a range of ratios between the co-substrates. Furthermore, Parameters like substrate to inoculum ratio (SIR), nutrients concentration, characteristics of inoculum are pivotal for the performance of the tests (Angelidaki et al., 2009).

In a recent study of Bella and Venkateswara Rao, (2022), septic sludge and AW were tested within a range of ratios to investigate the effect of each co-substrate on biomethane yield. The higher methane yield was achieved with Septic Sludge: AW ratio of 60:40. Interestingly, the methane yield of mono-digestion of AW was 84% lower than the theoretical methane yield (TMY), suggesting that AW is not an adequate base-substrate. However, the composition of cheese whey (CW) is ideal for a secondary substrate in the mixture. Maragkaki et al., (2017, 2018) investigated the impact of co-digestion of sewage sludge with CW, using a SS-CW ratio of 90:10% (v/v). The findings showed a slight decrease in methane concentration in the biogas. Moreover, the methane yield from co-digestion was lower than that of SS mono-digestion, which could be attributed to the increased amount of carbohydrates in the final substrate. This increase in carbohydrates may have resulted in biogas with lower methane concentration compared to that from lipids and proteins.

The present study aims to investigate the effect of AW on anaerobic co-digestion with PS through two distinct experimental setups. The first experimental approach involves batch experiments utilizing a BMP unit, while the second experimental approach comprises continuous flow experiments utilizing a small twin pilot-scale anaerobic digestion system. The primary objective is to evaluate the feasibility and potential of AW as a co-substrate for anaerobic co-digestion with PS and to determine the optimal co-substrates ratio for achieving maximum biogas production and process stability.

2. Materials and methods

2.1. Substrates and inoculum

All experimental systems in this study were each inoculated with mesophilic digested sludge sourced from the Psytalia Wastewater Treatment Plant (PWTP) located in the greater Athens region, with a treatment capacity of 3,500,000 population equivalent. The PS utilized in the experiments was obtained after gravity thickening from PWTP. The AW utilized in this study was procured from a dairy industry located in the city of Patra, Greece, and derived as a byproduct of Greek yogurt production. All samples were analyzed for total and volatile solids (TS and VS), total and soluble chemical oxygen demand (COD), alkalinity according to Standard Methods (APHA – WEF – AWWA, 1992). Volatile fatty acids (VFA) of anaerobic liquid were also monitored in the continuous flow experiments. Substrates and inoculum were also analyzed for NH_4^+ , TKN, TP, $\text{PO}_4\text{-P}$ (**Table 1**).

Table 1. Physicochemical characteristics of inoculum (anaerobic sludge), primary sludge and acid whey.

Parameters	Inoculum	PS	AW
TS (%)	3.1	3.9	5.9
VS (%)	3.0	3.0	5.1
pH	7.6	6.2	4.4
Alkalinity (mg CaCO_3/L)	5003	533	-
$\text{NH}_4\text{-N}$ (g/L)	1.1	0.2	0.05
TKN (g/L)	3.60	0.73	0.49
TP (mg/L)	49.5	30	400.5
COD_T^* (g/L)	38.45	49.59	64.2
COD_s^* (mg/L)	772	3016	58950

* COD_T : Total COD, COD_s : Soluble COD

2.2. Biomethane potential system, batch experiments description

The experimental setup for measuring biogas production involved a compact BMP unit with a heated bath that maintained a constant temperature of 35 ± 1 °C (**Figure 1**). Each bottle had a working volume of 400 to 700 mL. The biogas produced was passed through a scrubber containing a 2M NaOH solution to separate CH_4 from other gases. The BMP unit control panel automatically measured and recorded the volume of CH_4 produced. The substrate to inoculum ratio (SIR) was 1:2 VS basis. At least one of the containers is used for a control test on a standard

substrate of glucose to confirm the proper response of the inoculum (anaerobic sludge) on standardized substrates. Additionally, a blind test of the inoculum is necessary by measuring the endogenous methane production, which is then subtracted from the methane production obtained in the determinations of the other substrates. The methodology used was based on the approach described by Angelidaki et al., (2009). The BMP (Biochemical Methane Potential) lasts for 30 days, and its operation is only terminated when the daily methane production for three consecutive days is <1% of the accumulated methane volume (Holliger et al., 2016). All biogas and methane values reported in the study are expressed in normal conditions.



Figure 1 Biomethane potential system

2.1. Twin Pilot scale system, continuous flow experiments

The anaerobic digestion system (**Figure 2**) consists of two fully automated 15 L complete stirring tank reactors (CSTRs) that can be operated separately. They are made of INOX and the temperature was adjusted at 35°. Each reactor is fed from a 7 L tank, which is also equipped with a stirring mechanism. The system is also equipped with a thermometer and a pH meter and a U type biogas meter. The control digester was fed with PS as mono-substrate, while the experimental digester was fed with a mixture of PS and AW. The experiment last for 32 days with an increasing OLR up to 2 gVS/L-d. Substrates were monitored throughout the experiment and VS remained at $4\% \pm 0.26$. Biogas composition was analyzed weekly with a portable multi gas meter device (GFM 406).



Figure 2 Twin CSTR anaerobic digestion system.

3. Results and discussion

3.1. Biomethane potential experiments

It is evident that the different substrates had varying methane yields, with the highest methane yield of 385.8 mL CH₄/gVS obtained from the substrate consisting of PS and AW in a 70:30 ratio. Also, PS mono-digestion exhibited a high methane yield of 383.4 mL CH₄/gVS, indicating that AW can be an effective co-substrate for anaerobic digestion with PS.

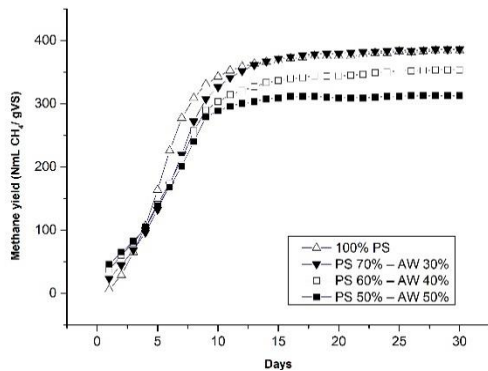


Figure 3 Cumulative methane production of the BMP tests

One effect of adding AW to the feeding mixture is an initial increase in methane production for the first three days. This effect becomes more apparent with an increase in the amount of AW added to the mixture. This is likely due to the highly readily available organic compounds present in AW, which can be easily utilized by the microorganisms involved in the process, thereby reducing the lag phase period. Based on the results of methane yield, it is suggested that despite the initial improvement in methane production due to AW, the final methane production declines with an increase in AW (**Figure 4**).

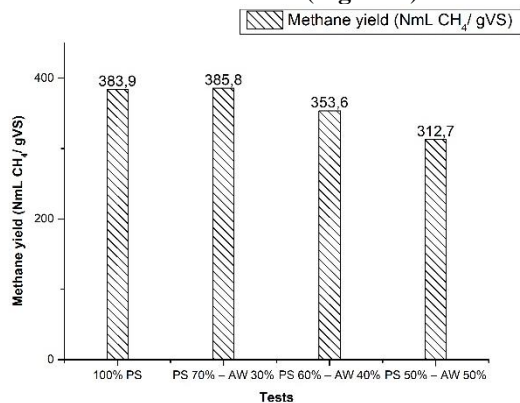


Figure 4 Methane yield of the BMP tests.

When 30% AW was added to the feeding mixture (with 70% PS), the methane yield increased slightly to 385.8 mL CH₄/gVS. However, when the proportion of AW was increased to 40%, the methane yield decreased significantly to 353.6 mL CH₄/gVS. Further increasing the proportion of AW to 50% resulted in a further decrease in methane yield to 312.7 mL CH₄/gVS.

3.1. Continuous flow anaerobic digestion experiments

Two identical CSTR anaerobic digester operated with increasing OLR for two different feedstocks: PS-AW (70-30) and PS (100). The data indicates that as the OLR increased from 1 to 2 g VS/L-d, there was a corresponding increase in biogas yield for both feedstocks. However, the

biogas yield for PS-AW (70-30) consistently outperformed that of PS (100) at all OLRs, with an average difference of 61.8 mL CH₄/gVS (**Figure 5**). The measured parameters of the anaerobic liquid from both digesters are summarized in **Σφάλμα! Το αρχείο προέλευσης της αναφοράς δεν βρέθηκε.** The percentage difference in biogas yield between the two feedstocks also increased with OLR, ranging from 4% to 20%. Similarly, the methane yield for PS-AW (70-30) was higher than that of PS (100) at all OLRs, with an average difference of 10.5 mL CH₄/gVS. The percentage difference in methane yield between the two feedstocks also increased with OLR, ranging from 3% to 11%. Although the methane content of the experimental reactor was 5% lower than that of the control reactor (62.68% and 67.85%, respectively), the higher biogas production covered the difference. These results suggest that co-digestion of PS with AW can improve biogas and methane yield compared to PS alone with the production of greater quantity of biogas. Furthermore, the data shows that the benefits of co-digestion at higher OLRs increase as the difference in biogas and methane yield between PS-AW (70-30) and PS, indicating the potential benefits of co-digestion at higher OLRs.

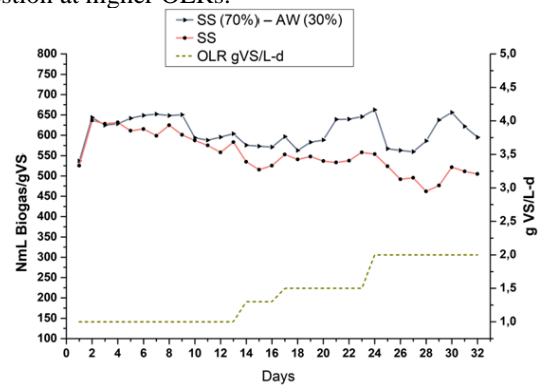


Figure 5 Methane yield of experimental and control digester for the experimental period and OLR progression.

Both digesters maintained stable pH levels, with the experimental reactor having a pH of 7.30 ± 0.02 and the control reactor having a pH of 7.27 ± 0.04 .

Table 2 Experimental and control data of anaerobic liquid.

Parameter	Experimental	Control
pH	7.30 ± 0.02	7.27 ± 0.04
TS (mg/L)	28385 ± 2049	28570 ± 1728
VS (mg/L)	18256 ± 1830	18646 ± 1453
VS (%TS)	64	65
COD _T (mg/L)	29393 ± 3466	29575 ± 2585
COD _s (mg/L)	505 ± 143	512 ± 76
Alkalinity (mg/L)	4701 ± 255	4742 ± 408
VFAs/Alk	0.2 ± 0.01	0.2 ± 0.01
NH ₄ ⁺ (mg/L)	976.71 ± 66.20	972.64 ± 72.34
Biogas yield (NmL/gVS)	607.1 ± 77.95	545.33 ± 47.44
% CH ₄	62.68 ± 4.59	67.85 ± 1.44
% CO ₂	28.23 ± 1.98	27.43 ± 2.39

The addition of AW, which is typically high in easily biodegradable organics and lactose, may have provided an easily utilized source of carbon and energy for microbial growth, contributing to the higher VS removal rate observed in the experimental treatment compared to the control treatment (Szaja & Montusiewicz, 2019). Specifically, VS removal efficiency was 55% for the experimental reactor and 52% for the control reactor. The concentration of VFAs remained low for both digesters, although this data is not shown. Both digesters maintained stable alkalinity levels, with the experimental digester having an alkalinity of 4701 ± 255 mg/L and the control digester having an alkalinity of 4742 ± 408 mg/L. Finally, the ammonium (NH_4^+) levels in both digesters were stable throughout the experiment. These results suggest that the experimental treatment was successful in maintaining the appropriate conditions for anaerobic digestion, despite the addition of a new substrate.

4. Conclusions

The results of the batch experiments showed that adding AW to the feeding mixture initially increased methane production, but with an increase in AW, the final methane

production declined. The highest methane yield was obtained from the substrate containing PS-AW in a 70:30 ratio, indicating that AW can be an effective co-substrate for anaerobic digestion with PS. In the continuous flow experiments, co-digestion of PS with AW consistently outperformed PS alone at all OLRs, with the difference in biogas and methane yield increasing with OLR. These results suggest that co-digestion of PS with AW can improve biogas and methane yield compared to PS alone, particularly at higher OLRs.

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