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Co-digestion and model simulations of source separated municipal organic waste with cattle manure under batch and continuously stirred tank reactors

Panagiotis Tsapekos, Panagiotis G. Kougias*, Sidhant Kuthiala and Irini Angelidaki

Department of Environmental Engineering, Technical University of Denmark, Kgs. Lyngby, DK-2800, Denmark

*Corresponding author: Panagiotis G. Kougias, Department of Environmental Engineering, Technical University of Denmark, Bld 113, 2800 Lyngby, Denmark.
E-mail address: panak@env.dtu.dk, Tel.: +45 45251454

Abstract

This study investigates the co-digestion of source separated municipal organic waste (SSMOW), pretreated using a biopulper, and cattle manure both in batch and continuous stirred tank reactors. The optimum co-digestion feeding mixture was consisted of 90% SSMOW and 10% cattle manure on organic matter basis, yielding 443 $mLCH_4/gVS$. The high performance of the co-digestion was explained by the fact that the efficient pulping pretreatment boosted the methane production from SSMOW and that the added livestock slurry provided the buffer capacity to avoid inhibition occurred by intermediates' accumulation. Moreover, batch assays focused on the effect of inoculum to substrate ratio (ISR) were performed. Results showed that the reduction of ISR had slight impact on extending the lag phase, without affecting the rest kinetic parameters. The efficiency of the co-digestion process in continuously fed reactor was comparable with the results obtained from the batch assay (i.e. <95% of the maximum expected value). Finally, the outputs from an applied mathematical model were in good agreement with the experimental data obtained from the continuous reactor operation, demonstrating that the BioModel can serve as a reliable tool to predict the process performance under real-scale conditions.

Keywords

Source separated municipal organic waste; anaerobic digestion; methane; kinetics; modeling

1. Introduction

Anaerobic digestion (AD) of source separated municipal organic waste (SSMOW) is considered as a competitive to the traditional (e.g. composting, landfilling, incineration) waste management solution as the organic matter is efficiently degraded producing bioenergy and also, biofertilizer [1,2]. In terms of bioenergy production, SSMOW can ensure high biogas yielding operation [3–5]. Specifically, the presence of soluble carbohydrates, proteins and lipids derived from the kitchen waste residues [6] settles SSMOW as a very interesting substrate for AD.

Despite the fact that SSMOW consists mainly of degradable components, non-degradable fractions (e.g. plastics) can be also found, as impurities. Thus, a well-performing separation step can increase process efficiency by initially discarding the non-degradable materials and subsequently, a suitable pretreatment method can boost the deconstruction of previously intact organic matter [7–9]. In industrial perspective, it was previously shown that the integration of two rather dissociated processes into a single and straightforward step is able to remarkably enhance the AD sustainability [10].

In this framework, pulping technology similar to the process used in paper industry can combine these two steps namely separation and pretreatment steps that are needed prior to AD of SSMOW, into a single process. A biopulper can separate the degradable organic matter and sort-out the non-degradable that can be subsequently recycled, reused or recovered [11]. In addition, the installed milling machinery assists the pretreatment of organic matter improving the biodegradability of SSMOW. In fact, a previous study demonstrated that the pretreatment of SSMOW with pulping technology, led to more than 390 $mLCH_4/gVS$ under different reactor configurations (i.e. batch assays, fed-batch and continuous stirred tank reactors (CSTR)) [5].

Notwithstanding the high bioenergy output, SSMOW is a very acidic waste, and on top of this, the AD process is prone to be inhibited at increased organic loads [1]. Thus, it is crucial to ensure high bioenergy output avoiding risks of acidification incidents and indeed, co-digestion can serve as a potential solution to such inhibition problems. More specifically, cattle slurry is able to increase the pH towards higher levels and hinder reactor's acidification due rapid volatile fatty acids (VFA) accumulation [12]. In addition, various hydrolytic and fermentative microbes which accelerate the disintegration process are already present in the livestock manure. So, the dissimilar biochemical characteristics of SSMOW and manure substrates can be combined to create a proper feedstock mixture. Furthermore, the usage of livestock slurries into the biogas sector is promoted by the policy-makers by the granted subsidies as mean to solve the manure treatment problem through AD [13]. Thus, co-digestion strategies using livestock manures are highly exploited.

However, the chemical composition of both substrates is not consistent but is strongly dependent on different parameters, which will in turn affect the final methane productivity. For instance, the major origin of SSMOW can influence positively (e.g. food residues) or negatively (e.g. green waste) the final bioenergy output [14]. On the other hand, nutritional feedstock composition, moisture content, animal species and growth stage are among the parameters that markedly affect manure's biogas productivity [15]. Hence, a universal feeding recipe for the biogas plants is not possible and thus, the optimum feedstock composition should always be independently examined within the framework of co-digestion applications.

Apart from the optimum co-digestion mixture, other kinetic parameters of the AD process are equally important and should be evaluated. For example, the achievement of a rapid and efficient disintegration of organic matter is assigned to the ratio between added feedstock and active biomass [16]. Indeed, organic overload inhibits the methanogenic community due to VFA accumulation and over-acidification [17]. Thus, kinetic parameters such as lag phase, hydrolysis and methane rate are influenced by the inoculum to substrate ratio (ISR) [18]. The imbalance between rapid hydrolysis-acidification and slow methanogenesis causes organic overload to the archaeal species which could not fully utilise the fed substrate [19]. Hence, it is crucial to secure an efficient feeding strategy to avoid toxicity that can eliminate the methanogenic activity.

Furthermore, operational parameters (e.g. reactor's configuration) play an important role towards co-digestion process optimisation. For instance, batch reactors can efficiently provide information about the duration of lag phase, maximum biogas yield, methane and hydrolysis rate. In contrast, CSTR are more appropriate to examine issues as microbiome's acclimatization at long term operation. Experiments are laborious and time consuming and therefore can only cover few experimental conditions. On the contrary, the outcome of both lab-scale reactor set-ups after data interpretation can be extremely useful as input for modeling simulations in order to expand testing at various conditions, and thereby improve the understanding of the AD system. Specifically, reliable mathematical models can reveal in advance the bottlenecks that limit the methane production (e.g. lag phase, substrates inhibition etc.) and highlight the operational conditions (e.g. hydraulic retention time, organic loading rate) that optimise process efficiency [20]. Hence, through reliable simulation outputs, the application of SSMOW for AD can be generalised in the direction of stable and high-yielding biogas production.

The aim of the present work was to provide a comprehensive research on exploitation of SSMOW as a major influent substrate for biogas digesters and to generate a dataset based on continuous reactor operation monitoring that would be used as input for mathematical modeling. Thus, mono- and co-digestion batch assays using SSMOW, pretreated using a biopulper, and cattle manure as the co-substrate were initially conducted. A subsequent batch set was performed to evaluate the kinetics of the most promising feeding mixture and to identify potential problems related to process inhibition at different ISR. Moreover, a continuously fed digester was set up to monitor and evaluate further the effect of the co-digestion process. Finally, a mathematic model (BioModel) was used to simulate the co-digestion process and validate the accuracy of the experimental work.

2. Materials and methods

2.1 Inoculum

Thermophilic inoculum was provided by a well performing lab-scale reactor fed with cattle manure. The digestate was sieved to remove the remaining organic matter and stored in thermophilic incubator for 10 days to reduce the background biogas production. The major physicochemical characteristics of the inoculum, after the degassing process, were pH: 8.36, Total Solids (TS): 26.70 ± 0.20 g/L, Volatile Solids (VS): 17.54 ± 0.22 g/L, Chemical Oxygen Demand (COD): 24.78 ± 1.19 g/L, Total Kjeldahl Nitrogen (TKN): 2.32 ± 0.09 g-TKN/L, Ammonium Nitrogen: 2.06 ± 0.10 g-NH_4^+/L and total Volatile Fatty Acids (TVFA): 0.25 ± 0.05 g/L.

2.2 Substrates

SSMOW of approximately 25% (v/v) industrial and 75% (v/v) household waste were collected from Gemidan Ecogi A/S after pulping process, as previously described [11].

In brief, municipal waste is inserted into a pulper equipped with a helical rotor. The rotor agitates to disperse the bio-degradable organic matter without damaging the non-degradable fraction. Subsequently, the two fractions are separated using a perforated plate. Cattle manure was collected from Hashøj biogas plant. The substrates were diluted with tap water to reach the same content of organic matter to prevent pumping, mixing and clogging problems in the lab scale reactors. After dilution and mixing, the substrates were stored in plastic bottles at -20°C until usage. The main chemical characteristics of the prepared substrates are presented in Table 1.

2.3 AD experiments

Biochemical Methane Potential (BMP) assays were initially performed based on Angelidaki et al. [21] in order to define the bioenergy production of the used substrates under mono- and co-digestion trials (i.e. 80:20, 60:40, 40:60 and 20:80 on VS basis). Triplicate glass reactors were used, with total and working volume of 547 and 200 mL, respectively. The inoculum represented 80% of the working volume and the organic load was 2 gVS/L. Prior to incubation, the batch reactors were flushed with pure N_2 to replace the remaining oxygen and achieve anaerobic conditions. Subsequently, they were placed in a thermophilic incubator (54 ± 1 °C). Based on the results from the first BMP test, the optimum mixing ratio of substrates in the feedstock was determined. Then, a second BMP test was set up to examine the effect of ISR on the AD of the used substrates. Specifically, batch co-digestion experiments were established at three different ISR on VS basis (i.e. 0.5, 1.5 and 3.0) keeping the amount of inoculum constant in all batches [22]. Samples for VFA determination and methane content were taken during the incubation period. For both BMP tests, daily manual stirring was conducted to avoid the creation of dead zones and monitoring of methane production was performed twice a week until cease of methane production was observed ($p < 0.05$).

Moreover, a continuously stirred tank reactor (CSTR) with 9.0 L total and 7.5 L working volume was used to examine the AD of the mixed feedstock under continuous mode operation. The reactor was initially filled with the same inoculum as the batch assays and flushed with pure N_2 to ensure anaerobic conditions. Based on the results from the BMP tests, the influent feedstock consisted of 90% SSMOW and 10% cattle manure, in terms of VS. The hydraulic retention time was set at 15 days by supplying 125 mL of feedstock four times per day using a peristaltic feeding pump. The organic loading rate of the reactor was set to 2.3 gVS/L/d. Biogas and liquid samples were taken directly from CSTR at a sequence of twice a week to measure methane concentration, pH and VFA composition. The CSTR was operated at thermophilic conditions (54 ± 1 °C) using silicone thermal jacket. The biogas volume was quantified daily with a gas meter based on water displacement principle and the bioenergy production was calculated.

2.4 Analytical methods

The standard methods for the examination of water and wastewater were followed for TS, VS, pH, COD, NH_4^+ and TKN measurements [23]. The elementary chemical composition was used to define the carbon to nitrogen ratio (C/N) of both substrates. Gas chromatography (GC-TRACE 1310) equipped with a thermal flame ionisation detector (FID) was used to determine the methane content of all biogas reactors and to quantify the VFA accumulation (GC-TRACE 1300) [5]. The content of micro- and macro- nutrients in both substrates was determined using inductively coupled plasma with optical emission spectrometry (ICP-OES). All measurements were performed in triplicate samples.

2.5 Computational methods

The modified Gompertz equation was used to describe the kinetics of the BMP tests:

$$M(t) = M_0 \times exp\left\{-exp\left[\frac{R_{max} \times e}{M_0}(\lambda - t) + 1\right]\right\}$$

where, $M(t)$ is the produced CH_4 yield over time t (mL/gVS), M_0 stands for the final CH_4 yield (mL/gVS), R_{max} is the maximum CH_4 production rate (mL/gVS/d), λ represents the lag phase (day) an e is Euler's constant (2.7183).

The co-digestion of cattle manure with SSMOW under continuous mode operation was evaluated using the extended dynamic bioconversion model (BioModel) [24]. First order kinetics was used to simulate hydrolysis and Monod kinetic was used for the rest AD steps. Moreover, inhibition of VFA to hydrolysis, acetate to acetogenesis, ammonia to methanogenesis and pH to all AD steps was examined.

2.6 Statistical analysis

Tukey post hoc test ($p < 0.05$) and one-way analysis of variance (ANOVA) was followed to determine the statistically significant variations among mono- and co-digestion samples using the software Graphpad Prism (Graphpad Software, Inc., San Diego, CA). The prediction accuracy of the regression analyses were evaluated using the coefficient of determination (R^2) and root mean square error (RMSE).

3. Results and discussion

3.1 Mono- and co-digestion of SSMOW and cattle manure

The first set of batch assays was conducted to define the maximum methane yield of SSMOW and cattle manure and to reveal the most efficient co-digestion mixture using these substrates (Fig. 1). Among different feedstocks, the usage of cattle manure as a sole substrate was associated with the lowest biomethanation potential (181 ± 6 mL/gVS). The limited biodegradability is attributed to the presence of biofibers, as a result of the animal nutrition, which are mainly composed of lignin molecules [13]. In contrast to cattle manure, the obtained methane yield using SSMOW was significantly higher (464 ± 69 mL/gVS, $p < 0.05$). The increased bioenergy production is attributed to both biomass composition (i.e. high lipid and protein content, negligible lignocellulosic biofibers) and applied pulping pretreatment before AD. Indeed, Khoshnevisan et al. [5] found that the mono-digestion of SSMOW pretreated with a biopulper led to similar results (490 mL/gVS) under mesophilic conditions and Naroznova et al. [11] found almost the same methane yield (469 mL/gVS) with the present study under thermophilic conditions.

With respect to co-digestion experiments, the higher the contribution of SSMOW in the feedstock the higher the methane production. Especially, the highest methane output was produced using 20% of cattle manure and 80% of SSMOW on VS basis in the feedstock (382 ± 16 mL/gVS). As expected, the addition of SSMOW in the feedstock boosted the biogas production. The results can be ascribed to two parameters: 1) compositional differences related to the biodegradable organic polymers with dissimilar theoretical BMP value, and 2) significant variation of co-substrates' C/N ratio (Table 1). Specifically, SSMOW contained increased amounts of lipids and soluble carbohydrates that can boost biomethanation compared to the recalcitrant cattle manure [5]. On the contrary, the high content of nitrogen into cattle manure leads to decreased C/N ratio. Thus, during co-digestion trials the markedly higher C/N of SSMOW increased the overall value. Accordingly, Zhang et al. [12] examined the co-digestion of food waste with cattle manure and concluded that the optimal C/N ratio was 15.8. The findings are in accordance with the present co-digestion experiments where a C/N ratio of 16.9 was associated with the highest methane yield. Moreover, the preference for conducting co-digestion strategies instead of using pure substrates is also induced by the micro-nutrients composition. Specifically, livestock slurries can supplement the required trace elements for high enzymatic activity that are occasionally presented in negligible concentrations in SSMOW [5]. For instance, cattle manure can serve as Mg^{2+} source to stimulate the fermentation process and additionally, decrease Na^{+} toxicity which can be detected in high levels in SSMOW depending on their origin (e.g. food residues) [12,25]. In accordance, the content of Mg^{2+} into the cattle manure (9.5 mg/gTS) was significantly higher compared to SSMOW (1.9 mg/gTS). On the hand, SSMOW had slightly higher content of Na^{+} than manure, 9.5 and 7.3 mg/gTS respectively. However, the content of Na^{+} was not high to provoke any salinity stress to the microbial cells [26]. Furthermore, the addition of livestock slurry can overcome the occasional lack of Ca^{2+} into the SSMOW (i.e. when green waste corresponds to the major fraction), which is mandatory for the growth of methanogenic archaea [27]. Nevertheless, green waste represented only a minor fraction into the used SSMOW and thus, a Ca^{2+} deficiency was not observed into the biowaste (19.7 mg/gTS) compared to manure (23.6 mg/gTS).

In order to limit the co-digestion mixtures to only four but at the same time to be able to define the maximum methane output using both substrates, a mathematical mixture design approach was followed [28]. Linear, quadratic and full cubic equations were used to fit the experimental data from the BMP tests and subsequently, R^2 and RMSE were used to evaluate the prediction accuracy (Table 2). In fact, the cubic model had the best prediction quality (i.e. highest R^2, lowest RMSE). The response optimisation using the full cubic model showed that 90% SSMOW in the feedstock mixture can lead to even higher methane production than the 20:80. While the calculated value (i.e. 10:90) was slightly lower compared to the highest BMP that was obtained at the mono-digestion of SSMOW (i.e. 0:100), these two methane yields did not differ significantly ($p > 0.05$). Hence, a mixture containing 10% of cattle manure and 90% of SSMOW was further examined, due to the relatively high methanation and the high interest with respect to the political and economic frame conditions. The selected feedstock composition was used for the second batch assay and subsequently, to the CSTR operation. Additionally, the results from the second BMP test were used to evaluate the full cubic model output.

3.2 Effect of ISR to the AD of SSMOW with cattle manure

In the second batch assay, the effect of inoculum to substrate ratio (ISR) was elucidated. The results indicated that the methane yield of the selected co-digestion mixture was not affected by the ISR as insignificant statistical differences were detected. Additionally, the average value of the recorded methane yield (443 ± 8 mL/gVS) was slightly higher but significantly meaningless ($p > 0.05$) with the predicted value (419 mL/gVS), validating the accuracy of the cubic model obtained from the first BMP test.

Based on the outcome of linear regression (i.e. high R^2, low RMSE), the modified

Gompertz equation had high prediction accuracy. Its applicability to predict similar co-digestion processes has been previously shown [29,30]; and thus, the kinetic analysis was based on the modified Gompertz model. Apart from the values of methane production, the rest kinetic parameters varied markedly upon the different inoculum to substrate content. It was demonstrated that the higher the amount of inoculum the shorter was the lag phase (Table 3 and Fig. 2). The observations are in agreement with studies examining the effect of substrate to inoculum ratio on wastes from municipalities and livestock industry [16,31]. Indeed, high load of substrate in parallel with limited content of active biomass could lead to reactor's acidification and therefore inhibition [18,22]. In the present work, the lowest pH value (i.e. 6.66) was observed during the 3rd incubation day (Fig. 3a) and was directly connected with the accumulation of TVFA (Fig. 3b) which resulted in limited methane production (Fig. 2). Acetate represented the highest portion of produced intermediates, indicating that the initial three steps of AD were efficiently conducted and only the methanogenesis was partially inhibited during the start-up period. However, on day 8 the TVFA levels of batch assays set at ISR of 0.5 were low and on the 12th day the methane production was similar with the rest ISRs. Hence, the intermediates were efficiently consumed by the methanogenic community and the initially observed accumulation did not lead to irreversible inhibition. In a recent study, the methanogenic community was clearly inhibited at low ISR in continuously fed reactors with SSMOW [5]. The inhibition was depicted by accumulation of VFA and especially acetate concentration, drop of pH, and subsequently, extension of lag phase compared to control operation. However, in the present study irreversible inhibition was not detected.

Results obtained from the second batch set showed that the decrease of ISR had only a slight impact on extending the lag phase during the co-digestion of SSMOW with cattle manure. The strong buffer capacity of livestock slurry alleviated the overload of the inoculum that otherwise can occur at low ISR [18].

3.3 Continuous mode co-digestion of SSMOW and cattle manure

CSTR operation is better to mimic the co-digestion of SSMOW with cattle manure to real conditions compared to BMP assays. At steady state conditions, the methane yield of the CSTR was relatively high (437 ± 20 mL/gVS, Fig. 4a) corresponding to 96% of the maximum expected output based on the results from the second BMP assay. Typically, the methane production of a continuous reactor reaches 70-90% of the BMP value [32], which highlights the high efficiency of the investigated system. In this context, the reactor did not face any technical challenges and after seven days of operation reached almost the maximum bioenergy production. Moreover, during the second HRT the overall process performance was already stable. During the whole experimental period, the methane content in biogas was rather constant (65.3 ± 2.3%), pH was stable (7.65 ± 0.06) and the VFA were efficiently processed by the AD microbiome and were not accumulated (Fig. 4b). Regarding the individual VFAs, acetic and propionic acids were the dominant intermediates during the whole experimental period. Nevertheless, acetic and propionic acid were always significantly lower than the suggested inhibition indicator of 2.4 and 1.8 g/L respectively [33]. In addition, the ratio between acetic to propionic acid was always higher than 1.0 g/L validating the well-performing AD process [34].

The increased performance of CSTR was in accordance with the simulation outputs, as the BioModel described efficiently both bioenergy production and biochemical parameters (Fig. 4). Indeed, the BioModel has a wide range of applicability using various organic substrates as crop residues, food waste, cheese waste, livestock slurries, wastewater sludge and SSMOW [5,35,36] and thus, it is reliably designed to simulate efficiently various co-digestion scenarios. In addition, BioModel considers also ammonia inhibition which is a major problem during the AD of either livestock slurries or SSMOW [37]. However, the used substrates were diluted with water in the present study and thus, the concentration of ammonium nitrogen was low. More specifically, the free ammonia was calculated to be less than 0.05 g/L at these conditions and on top of this, no inhibition was indicated in the simulation. In parallel, both CSTR monitoring and BioModel simulations showed that the physicochemical parameters (e.g. TVFA accumulation or pH increase), which are directly connected with ammonia problems, were within optimal range for AD process. To sum up, the overall reactor performance was good as concluded by both experimental and modeling aspects. SSMOW pretreated with biopulper can easily lead to high bioenergy output without instabilities and therefore, it should be highly considered as a primary feedstock for full-scale biogas plants.

4. Conclusions

The present study demonstrated that the anaerobic co-digestion of SSMOW with cattle manure is feasible and leads to high methane production. The kinetics of co-digestion showed that high process performance can be achieved independently from the inoculum to substrate ratio. Moreover, the mixed influent feedstock demonstrated increased biodegradation efficiency which was similar at batch assays and continuous reactor operation. Subsequently, the continuously fed reactor process was modelled using the BioModel and the results allowed close fit to the experimental measurements.

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Figure captions

Fig. 1. Methane yields of mono- and co-digestion tests of cattle manure and SSMOW

Fig. 2. Cumulative CH_4 production as a function of time during the co-digestion of SSMOW with cattle manure at different ISR.

Fig. 3. pH change (a) and TVFA accumulation (b) as a function of time during the co-digestion of SSMOW with cattle manure at different ISR.

Fig. 4. Experimental data and modelling simulations for bioenergy yield (a) and pH alteration and TVFA accumulation (b) during the co-digestion o of SSMOW with cattle manure in continuous mode operation

Tables

Table 1. Characteristics of SSMOW and cattle manure

Characteristics	**SSMOW**	**Cattle manure**
pH	4.05	7.24
TS, g/L	40.65 ± 0.64	48.25 ± 0.23
VS, g/L	35.00 ± 0.67	35.00 ± 0.04
COD, g/L	62.34 ± 1.78	56.99 ± 1.63
TKN, g/L	1.23 ± 0.04	2.46 ± 0.08
NH_4^+, g/L	0.29 ± 0.04	1.61 ± 0.08
C/N	19.01 ± 0.95	8.69 ± 0.43
TVFA, g/L	1.73 ± 0.05	6.73 ± 0.30
Acetate, g/L	1.54 ± 0.05	4.49 ± 0.29
Propionate, g/L	0.06 ± 0.00	1.19 ± 0.08
Iso-butyrate	0.01 ± 0.00	0.16 ± 0.00
Butyrate	0.11± 0.01	0.59 ± 0.02
Iso-valerate	0.01 ± 0.00	0.27 ± 0.08
Valerate	0.01 ± 0.00	0.05 ± 0.00

Table 2. Models summary statistics with BMP as response variable and VS share of SSMOW in the feedstock as regressor.

Model	**Regression equations**	R^2	***RMSE***
Linear	$BMP = 2.503 \times VS + 204.063$	0.956	18.45
Quadratic	$BMP = -0.004 \times VS^2 + 2.946 \times VS + 198.153$	0.958	17.91
Cubic	$BMP = 0.001 \times VS^3 - 0.102 \times VS^2 + 6.505 \times VS + 182.566$	0.996	5.42

Table 3. Parameters of modified Gompertz equation fitting experimental results obtained from the co-digestion of SSMOW with cattle manure at different ISR

	ISR		
Modified Gompertz equation	**0.5**	**1.5**	**3.0**
λ, days	3.11	2.63	1.95
R_{max}, mL/gVS/d	90	118	96
Measured BMP, mL/gVS	444	455	446
Predicted BMP, mL/gVS	442	452	438
Difference, %	0.5	0.7	1.9
R^2	0.999	0.999	0.999
RMSE	4.19	2.38	5.54