Heliyon 5 (2019) e02140

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.heliyon.com

The effects of temperature and duration of thermal pretreatment on the solid-state anaerobic digestion of dairy cow manure

Wilton P.A. McVoitte, O. Grant Clark *

Department of Bioresource Engineering, Faculty of Agricultural and Environmental Sciences, McGill University, 21,111 Lakeshore Road, Ste-Anne-de-Bellevue, H9X 3V9, Quebec, Canada

ARTICLE INFO

Keywords:
Bioengineering
Energy
Dairy cow manure
Thermal pretreatment
Solid-state anaerobic digestion
Lignocellulose
Central composite rotatable design
Agricultural engineering
Agricultural water management
Bioenergy
Energy sustainability
Waste treatment

ABSTRACT

Cellulosic substrates such as dairy cow manure often yield low volumes of biogas and low concentrations of methane when digested anaerobically. Thermal pretreatment of dairy cow manure was investigated to determine if pretreatment temperature and duration can be optimized to maximize biogas yield and methane concentration. A central composite rotatable design was used to select combinations of temperature and duration. Based on measured data, statistical models were generated to estimate the biogas yield and methane concentration during digestion. The highest biogas yields were from the untreated samples and samples treated at the center temperature and duration of the statistical model (125 °C, 37.5 min). The model predicted the optimum pretreatment conditions of 140 °C for 30 minutes. Under the conditions of this experiment, temperature and duration had no significant effect on the biogas yield and methane concentration. This lack of significance may indicate that thermal pretreatment may be an unnecessary step in the anaerobic digestion of dairy cow manure, which could reduce capital and operating costs for the industry.

1. Introduction

Anaerobic digestion (AD) is the biological degradation and stabilization of organic materials under anaerobic conditions (Chen et al., 2008). It is a waste treatment technology that generates renewable energy in the form of biogas and produces a semi-solid or solid output (digestate) typically used for agricultural fertilizer (Angelonidi and Smith, 2015). The methane (CH_4) in the biogas can then be burned to generate electricity and heat or upgraded as a vehicle fuel (Lymperatou et al., 2017; Passos et al., 2017).

The process is mediated by a consortium of anaerobic microbes that work synergistically (Li et al., 2011). Hydrolytic bacteria first break down complex organic polymers like carbohydrates and proteins into simpler soluble monomers (e.g. glucose and amino acids). Acidogenic bacteria then consume the products of hydrolysis, converting them into simple volatile fatty acids (VFAs) (e.g. lactic and butyric acids) and simple alcohols (e.g. methanol). Further fermentation of the organic acids and alcohols occur via acetogenesis, forming acetic acid, hydrogen (H_2) and carbon dioxide (CO_2), which are the direct substrates for CH_4 production. Methanogenic bacteria then generate CH_4, with about 70% being produced from the acetic acid and the remainder from H_2 and CO_2 (Polprasert, 2007).

Anaerobic digestion diverts organic waste from landfills, reducing negative impacts such as the production of greenhouse gases, leachate, and odor (Environment Canada, 2013; Li et al., 2011). Depending on the nature of the substrate, the digestate can be used as a nutrient-rich fertilizer (Appels et al., 2011). The digestate also contains considerable amounts of nitrogen, phosphorous and potassium, making them available for absorption by plants (Koszel and Lorencowicz, 2015). As an organic soil amendment, digestate increases the soil's carbon pool (Westphal et al., 2016). This improves plant growth and health.

Solid-state digestion operates on a substrate with a total solid content (TS) of more than 15%. It is advantageous over liquid-state digestion as it offers greater flexibility in the type of substrate, especially biomass composed mainly of lignin, cellulose, and hemicellulose. Digesting waste in this form requires reduced moving parts and lower energy input for heating and mixing (Chaikitkaew et al., 2015).

Dairy cow manure is a widely available resource that can be utilized in AD. In 2006, Canadian milk cows produced 21.6 million tonnes of manure. This represents 12% of the total manure produced by all livestock that year (Hofmann, 2006). Dairy cow manure is a common AD substrate due to the expansion of the livestock production sector and its

* Corresponding author.
E-mail address: grant.clark@mcgill.ca (O.G. Clark).

https://doi.org/10.1016/j.heliyon.2019.e02140
Received 25 December 2018; Received in revised form 11 May 2019; Accepted 18 July 2019

rich content in nutrients and microorganisms that lead to spontaneous biogas production (Lymperatou et al., 2017). Therefore, it is crucial to have a good understanding of its anaerobic biodegradability (Passos et al., 2017).

The major hindrance for the utilization of animal manure for biogas production is its low biodegradability and low CH_4 yield due to its high lignocellulosic fiber content (Andriamanohiarisoamanana et al., 2017; Nasir and Ghazi, 2015; Raju et al., 2013). Dairy cow manure consists of up to 40–50% fibrous lignocellulosic matter containing mainly plant material that has not been fully digested (Nasir and Ghazi, 2015; Rafique et al., 2010; Raju et al., 2013). Though dairy cow manure contains a high content of lignocellulose, it may not be considered truly lignocellulosic because a significant portion of the lignified material has been digested in the bovine rumen. It usually has a rather low TS concentration of 7–9% depending on the manure management system (Angelidaki and Ellegaard, 2003). In general, manure is usually high in nitrogen in the form of ammonia, posing a hindrance to AD. Manure has a low carbon-to-nitrogen (C/N) ratio, which increases the likelihood of process failure or inhibition when used as a single feedstock (Hassan et al., 2016). The manure's high water content also limits the CH_4 yield, producing 0.2–0.34 L CH_4 g^{-1} volatile solid (VS) (Angelidaki and Ellegaard, 2003; Passos et al., 2017). Conversely, substrates rich in lipids and/or easily degradable carbohydrates (e.g. used vegetable oil) can produce up to 0.65 L CH_4 g^{-1} VS (Labatut et al., 2011).

To compensate for these characteristics, pretreatment techniques have been carried out on lignocellulosic substrates to allow for more efficient degradation of the organic matter. These methods should enhance the CH_4 yield by disintegrating the cell walls of the substrate, solubilizing the hemicellulose and lignin, and allowing for more effective microbial hydrolytic activity (Nasir and Ghazi, 2015).

Thermal pretreatment is one method used to increase the biogas yield of organic biomass. It is one of the most studied pretreatment methods, having been successfully applied at an industrial scale, and one of the earliest methods recognized as having the potential to improve AD (Ariunbaatar et al., 2014; Carlsson et al., 2012). Thermal pretreatment is beneficial as the heat from the pretreatment process can be recovered and there are no added chemicals (Carlsson et al., 2012; Zhang et al., 2014).

There are gaps in the knowledge about pretreatment, specifically its application to dairy cow manure. Despite the numerous studies of pretreatment technologies at both lab and industrial scales, there are no optimum treatment combinations available for specific substrates at their specified process conditions such as TS and pH (Appels et al., 2011). Moreover, for thermal pretreatment, limited research has examined combinations of temperature and duration that enhance the biogas yield or CH_4 concentration or reduce the retention time (Ariunbaatar et al., 2014). Limited research has been conducted on the thermal pretreatment of dairy cow manure and most have been conducted on liquid manure (Passos et al., 2017). Most of the research related to the use of manure in AD has been applied to its use as an inoculum or nitrogen source or its codigestion with other biomass with low nitrogen concentrations (Appels et al., 2011). Manure has a high moisture content which acts as a solvent for dry biomass making it useful as a base substrate for codigestion (Andriamanohiarisoamanana et al., 2017). Optimization studies usually investigate codigestion or bioaugmentation to enhance AD parameters such as the C/N ratio or reduce the likelihood of inhibition, improving overall AD performance.

The objective of this research was to determine the effect of pretreatment temperature and duration on the quantity and quality of the biogas produced from the solid-state AD of dairy cow manure. The volume of biogas in liters (L) represents the quantity of biogas, while the quality of the biogas is represented by the CH_4 volume in L and the percent CH_4 ($\%CH_4$) of the total biogas generated. Response surface methodology (RSM) was employed to determine the optimal pretreatment conditions for maximum biogas yield and CH_4 volume from the dairy cow manure. After digestion, empirical models were developed to predict the biogas volume and CH_4 volume of the thermally treated manure as a function of the pretreatment temperature and duration.

2. Materials and methods

2.1. Experimental statistical design

The selected RSM, the central composite rotatable design (CCRD), was used to determine the influence of pretreatment temperature and duration on the thermal pretreatment and subsequent solid-state AD of the dairy cow manure (Bowley, 2008). The CCRD provided the experimental data and determined the relationships among the treatment factors and dependent variables by fitting full second-order polynomial models representing the response surface over a relatively broad parameter range (Khoobbakht et al., 2016).

RSM is a viable tool for establishing the optimal conditions in any given system by establishing the relationship between more than one variable and a given response (Ravindran et al., 2016). This statistical approach is useful as it requires fewer tests and less time compared to a full-factorial design (Khoobbakht et al., 2016). The CCRD is also beneficial as certain degrees of freedom are left which help create more reliable models, especially in situations where some experiments can be affected by experimental error (Rakić et al., 2014). This model is also beneficial because it offers good prediction ability.

The experimental treatment factors were the pretreatment temperature and duration. The pretreatment temperature was the oven temperature while the pretreatment duration was the length of time beginning when the manure was placed in the pre-heated oven until it was removed and placed at room temperature. The minimum and maximum values of the independent variables (temperature and duration) were first found in the literature from other similar studies (Bordeleau and Droste, 2011; Carrère et al., 2010) and the center point (0) determined from this range. The dependent variables investigated were the biogas volume, CH_4 volume, and the $\%CH_4$.

Bowley (2008), Montgomery (2005), and Khoobbakht et al. (2016) explain in further detail the five levels per treatment factor of the CCRD (Table 1) and the pairing of these five levels into specific treatment combinations. A total of 12 treatment combinations were used: four replicates of the center point, four star points, and four factorial points (Table 2). The entire experiment was replicated in two blocks to increase the statistical power giving a total of 24 experiments. One positive control and one negative control were also included for each block. The positive control contained the inoculum and raw manure (i.e. not treated). The temperature was designated as 25 °C and zero minutes indicating no pretreatment. The negative control consisted of inoculum only. No pretreatment was conducted on the inoculum, only the manure for the relevant treatment combinations.

A multiple regression analysis was carried out on the data for biogas volume, CH_4 volume and $\%CH_4$ to obtain the regression coefficients. The data was analyzed using the SAS software package (version 9.4, SAS Institute Inc., Cary, NC). The statistical models were used to predict the modeled value of the dependent variables. The polynomial equations were then fitted to the data based on the least-squares optimization technique (Bowley, 2008). Using the polynomial model, the correlation between pretreatment temperature, duration, and the various dependent

Table 1
Treatment factors and their levels for the central composite rotatable design[a].

Factor	Code	Min.	−λ	Center	+λ	Max.
		−1	−0.707	0	+0.707	+1
Temperature (°C)	T	50[b]	72	125	178	200[b]
Duration (min)	t	15[b]	22	37.5	53	60[b]

[a] The central composite rotatable design has five levels per treatment factor, namely: the center (0), the negative (–λ) and positive (+λ) factorial points, and the minimum (−1) and maximum (+1) points (Karimi et al., 2012).
[b] (Bordeleau and Droste, 2011; Carrère et al., 2010).

Table 2
Treatment combinations for the independent variables.

Experiment	1[a]	2[a]	3[b]	4[b]	5[b]	6[b]	7[d]	8[c]	9[c]	10[c]	11[c]	12[a]	13[a]
T (°C)	125	125	125	125	200	50	25	72	72	178	178	125	125
t (min)	37.5	37.5	15	60	37.5	37.5	0	22	53	22	53	37.5	37.5
Net Biogas Volume (L) Block 1	9	1	14	0	7	1	20	1	10	0	3	0	27
Net Methane Volume (L) Block 1	4	0	7	0	4	0	10	0	5	0	2	0	14
Net Biogas Volume (L) Block 2	22	22	18	4	15	3	28	3	5	5	4	5	4
Net Methane Volume (L) Block 2	12	12	9	1	8	1	14	1	2	2	1	1	1

[a] Centre points: (0,0). Generally, three to five replicates of the center treatment combination are recommended to provide precision in estimating the modeled value for the dependent variable. Four replicates of the center point were included in this experiment.
[b] Star points: These are four additional treatment combinations (0, –1; 0, +1; +1, 0; –1, 0), which are the minimum and maximum values for each factor combined with the center point of each factor. The figure formed by these points is called a star, hence they are known as star points or axial points.
[c] Factorial points: The factorial points (±λ) were first calculated using the equation found in Bowley (2008). Note for a two-factor design, λ is 0.707. Thus, the four factorial combinations are (-λ -λ; -λ +λ; +λ -λ; +λ +λ).
[d] Positive control (Not included in central composite rotatable design).

variables were obtained.

2.2. Design of digesters and water-bath

Clear standard wide-mouth soda-lime glass bottles with polyvinyl-lined caps (W216928, Wheaton, Millville, NJ) were converted to 4-L laboratory-scale digesters by piercing a 20-mm (¾-in) hole in the center of each cap using a drill press. This hole was used to accommodate a miniature through-wall fitting (8674T55, McMaster-Carr, Elmhurst, IL), which could then accommodate a 2-way right angled polyvinyl chloride (PVC) ball valve (PVC-657-4M4B-B, Cole-Parmer, Montréal, QC). Each through-wall fitting was supported by an oil resistant nitrile rubber (Buna-N) gasket (4509K12, McMaster-Carr), an O-ring (2-236/N70, R.B. Packings and Seals Inc., St-Laurent, QC) and a spacer washer (128968, Precision, Pointe-Claire, QC) with polytetrafluoroethylene (PTFE) thread seal tape (B74-280, Belanger, Pointe-Claire, QC) around the threading.

Digesters were designed for acclimatization and digestion of the inoculum and substrate. For each digester, 610 mm (2 ft) of 5 mm × 2 mm (3/16-in × 1/16-in) vinyl tubing (AGL00012, Fisherbrand, Toronto, ON) was joined with 610 mm (2 ft) of 3 mm × 2 mm (1/8-in × 1/16-in) vinyl tubing (AGL00007, Fisherbrand) via a plastic barbed tube fitting reducer (5117K59, McMaster-Carr). A flow control clamp (RK-95785-06, Cole-Parmer) was also placed on the tubing. The larger end of the vinyl tubing was attached to the digester via the PVC ball valve, while the other end was attached to a 2-L Tedlar™ Push/Pull Lock Valve (PLV) gas sampling bag (24654, Sigma-Aldrich, Oakville, ON) for gas collection. Each vessel was checked for leaks by pressurizing it with dry air, submerging it into a vessel filled with water and watching for escaping bubbles.

Two 95-L wheeled coolers (3000001054, Coleman, Montréal, QC) filled with ≈ 7-L water were used as water-baths for the digesters. Submersible aquarium heaters (T11302, Hydor Theo, Bassano del Grappa, VI) maintained the temperature between 31–38 °C. Three K-Type thermocouple probes (Omega Engineering, Laval, QC) were placed within each water-bath to monitor the temperature. A data-logger (34970A, Agilent, Loveland, CO) was connected to the thermocouples to allow for continuous data monitoring.

2.3. Inoculum and substrate

Digested biosolids were collected from the AD plant operated by the City of Saint-Hyacinthe, QC, Canada. The biosolid is sewage sludge from the city's waste water treatment plant (Ville de Saint-Hyacinthe: Technopole agroalimentaire, n.d.). The digestate was used as the inoculum. The inoculum is the starter culture that contains the microorganisms that will facilitate the digestion process. It was stored at room temperature and used within two days.

Dairy cow manure was collected from the Holstein heifers at the Macdonald Campus Dairy Unit, McGill University, Ste-Anne-de-Bellevue, QC, Canada. It was stored at room temperature and used within two days. New inoculum and manure were collected prior to each experiment.

2.4. Inoculum and substrate characterization

TS and VS tests were conducted on the inoculum and manure prior to acclimatization and digestion (APHA, 2005). These tests were conducted using an oven (1327F, VWR, Cornelius, OR), K-type thermocouple probes and balance (MS4002S/03, Mettler Toledo, Greifensee, ZH) and muffle furnace (F48025, Barnstead Thermolyne Type 48000, Dubuque, IA). These tests were done to determine the moisture and solids content of the inoculum and manure, in part to determine the mass of substrate to inoculum to be mixed on a 1:1 ratio on a TS basis.

The pH of the inoculum and manure were measured using a Soil Stik™ pH meter (2105, Spectrum Technologies Inc., Aurora, IL) with the semi-solid samples prepared in a 1:2 ratio with distilled water (Wolf, 2003).

2.5. Acclimatization of inoculum

Eight-hundred grams of inoculum was weighed into each digester. Note the inoculum was not pretreated to preserve the microorganisms for the digestion of the pretreated manure. The digester was then evacuated using bench vacuum (<10 kPa) and then purged with nitrogen gas (106717101, Praxair, Montréal, QC) under low pressure ($<7 \times 10^2$ kPa) for three-minute intervals each. This dilution technique was repeated five times to ensure an anaerobic environment. Tedlar bags were then attached to the digester.

The digesters were placed in the water-bath for two weeks to allow the microorganisms to acclimatize to the anaerobic, mesophilic conditions. This time was sufficient to minimize the background CH_4 that was produced from the inoculum when the substrate was added (Chaikitkaew et al., 2015).

2.6. Sample preparation, pretreatment, and digestion

Manure was weighed (1235 g), into a 2-L clear standard wide-mouth soda-lime glass bottle (W216927, Wheaton), the top was covered with foil, and placed in the oven at the pre-determined temperature for the selected duration (Table 2). The pretreated manure is the sample of manure heated at the pre-determined treatment temperature and duration combination. Thereafter, each sample was cooled to room temperature for five to six hours. Samples of the pretreated manure were taken

for TS, VS and pH characterization as per Section 2.4.

The mass of the digesters containing the acclimatized inoculum was noted and 1085 g of the pretreated manure was mixed with the acclimatized inoculum. The inoculated manure was the cooled pretreated manure mixed with the inoculum. Samples were then taken for TS, VS and pH analyses as per Section 2.4. The digesters were then sealed, and the air removed and displaced with nitrogen gas as explained in Section 2.5. This was carried out for each of the 24 treatment combinations, conducted in two blocks of 12. A Tedlar bag was attached to each digester and the digesters were placed in the water-bath.

One digester contained acclimatized inoculum and manure that was not pretreated (positive control) and another contained acclimatized inoculum only (negative control). Samples from these digesters were also taken for characterization (Section 2.4), purged using the dilution technique (Section 2.5), and then sealed. One positive and negative control was conducted for each block giving a total of 28 experiments.

The inoculated manure and positive and negative controls were incubated for forty days. This is the approximate retention time used in research and industry (Environment Canada, 2013).

2.7. Analytical methods

2.7.1. Gas composition

The biogas composition was measured via gas chromatography (Hewlett Packard 5890A Gas Chromatograph) with a thermal conductivity detector and a column (Alltech) in which the outer column was packed with an activated molecular sieve, and the inner column was packed with a porous polymer mixture. Helium was the carrier gas at a flow rate of 30 mL min^{-1}. The run time was six minutes at a pressure of up to 25 kPa. The detector and injector temperatures were 110–120 °C while the oven temperature was 40–43 °C.

Biogas was sampled from the collection bag in the following manner. Prior to sample analysis, the needle used for injection was flushed with ambient air. An aliquot of ambient air (0.1 mL) was injected as a quality control check. One millimeter of sample gas was withdrawn from the Tedlar bag with 0.1 mL used as the injection volume. The syringe was flushed with the sample gas prior to injecting and between samples. Each time, the volume that was withdrawn was noted and added to the total volume of biogas. Samples were taken in duplicate.

2.7.2. Biogas volume and methane volume

After determining the gas composition, the volume of biogas was measured via the water displacement method. When pressure was applied to the bag, the incoming biogas displaced the water into the measuring cylinder. The mass of the water was weighed on the balance. Using the density of water as 1.00 g mL^{-1}, the mass of water expelled was equivalent to the volume of biogas generated.

The net biogas volumes and net CH_4 volumes for each experiment and the positive control were then determined. Firstly, the cumulative biogas volumes were determined by summing the daily yields. From the gas chromatography analysis, the percent CH_4 from each gas sample was determined. This was used to determine the cumulative CH_4 volume. The biogas volume and CH_4 volume generated by the negative control was subtracted from the biogas volumes and CH_4 volumes generated by each of the other experiments and the positive control. This gave the contribution from the manure only, i.e. the net biogas volume and net CH_4 volume.

2.7.3. Carbon and nitrogen analysis

Carbon and nitrogen analysis was conducted on dried samples of the inoculum, manure and inoculated manure for the positive control. The analysis was performed by a commercial laboratory (Environex Group, Longueuil, QC).

2.8. Characterization after digestion

After the 40 days of incubation, the digesters containing the inoculated manure were weighed and samples were taken for TS, VS, and pH characterization. This characterization was used to determine the degradation efficiency.

3. Results and discussion

The TS, VS and pH characteristics of the inoculum and manure as received, i.e. prior to acclimatization and digestion, are shown in Table 3. Also shown in Table 3 are the characteristics of the pretreated manure. Based on these characteristics, it would seem that the thermal pretreatment had limited to no effect on the manure. The characteristics of the inoculated manure before and after digestion are also shown in Table 3. The reduction in the solids content after digestion is indicative of the transformation of the organic material to biogas. The C/N ratios of the inoculum, manure and inoculated manure for the positive control are also shown in Table 3. The C/N ratio of the inoculum was inconsistent between blocks, which made the inoculated manure lower than the optimum ratio of 30:1. This may have affected the metabolism of the microbes and their ability to optimally produce biogas.

In this study, pretreatment temperature and duration were investigated to determine their effect on the solid-state AD of dairy cow manure. The investigated responses are presented below.

3.1. Biogas and methane production

The response surface for total biogas volume is:

$$V_{Biogas} = -24.11 + (0.3690 \times T) - (0.00118 \times T^2) + (0.6904 \times t) - (0.00849 \times t^2) - (0.00133 \times T \times t) \quad \text{Eq. 1}$$

where V_{Biogas} is the net biogas volume (L), T is the target oven temperature, and t is the duration the manure is exposed to the treatment temperature (Table 2). The differences between the modeled and measured values (residuals) were large (up to 16.90 L) for a few of the experiments (Fig. 1a). All the other residuals were smaller, − 9.34 to +4.23 L. The lack of fit was also not significant, indicating that a higher degree polynomial regression was not required to explain the response. Neither the effect of the block nor the linear, quadratic nor interaction factors of temperature nor duration were significant (Table 4). Note that the CCRD had an unbalanced number of replicates per treatment combination. This may have resulted in a covariance between the parameter estimates leading to the model being significant despite none of the factors of the model being significant. This is not statistically inconsistent. The coefficient of determination (R^2) of the model was 43% (Table 4). As illustrated by the contour plot (Fig. 1d), biogas volume increases with temperature up to approximately 150 °C and then decreases. The biogas volume increases with duration of the pretreatment until 50 min, then decreases.

The central composite rotatable design (CCRD) had an unbalanced number of replicates per treatment combination. This may have resulted in a covariance between the parameter estimates leading to the model being significant despite none of the factors of the model being significant. This is not statistically inconsistent. CCRD also has the disadvantage of not being able to sample points at all extremes. The positive control could not be included as part of the CCRD and a significant increase or decrease of the treatment combinations in relation to the control could not be statistically determined using this approach.

In Block 1, only Experiment 13, one of the samples pretreated at the center temperature and duration (125 °C, 37.5 min) yielded more biogas (34% increase) than the positive control (Experiment 7). In Block 2, none of the pretreated samples yielded more biogas than the positive control sample (Fig. 1a).

The best-fit quadratic model for the volume of CH_4 generated by the

Table 3
Total solid, volatile solid, pH and carbon-to-nitrogen ratios of the inoculum, manure, pretreated manure and inoculated manure before digestion and the inoculated manure after digestion.

	Before Digestion								After Digestion	
	Inoculum[a]		Manure[a]		Pretreated manure[b]		Inoculated manure[b]		Inoculated manure[c]	
	Block 1	Block 2	Block 1	Block 2	Block 1	Block 2	Block 1	Block 2	Block 1	Block 2
TS (%)[d]	21.80 ± 0.01	19.74 ± 0.05	16.12 ± 0.21	14.60 ± 0.26	16.25 ± 0.01	15.94 ± 0.01	18.05 ± 0.004	16.58 ± 0.005	16.86 ± 0.004	14.65 ± 0.01
VS (%)[e]	58.89 ± 0.32	63.81 ± 0.03	84.63 ± 0.13	84.90 ± 0.03	84.83 ± 0.003	85.02 ± 0.01	72.59 ± 0.01	76.54 ± 0.01	68.20 ± 0.01	71.41 ± 1.22
pH	8.35 ± 0.07	8.07 ± 0.04	7.85 ± 0.07	8.18 ± 0.13	8.24 ± 0.36	8.11 ± 0.08	8.35 ± 0.29	8.20 ± 0.07	8.27 ± 0.09	8.46 ± 0.06
C/N	4.3	8.8	16.1	17.3	NA	NA	7.9	5.1	NA	NA

[a] The reported results are the Mean ± Standard deviation of duplicate samples.
[b] The reported results are the Mean ± Standard deviation of single samples for all treatments including the positive control.
[c] The reported results are the Mean ± Standard deviation of duplicate samples for all treatments including the positive control.
[d] Percent of total wet mass.
[e] Percent mass of total solids.

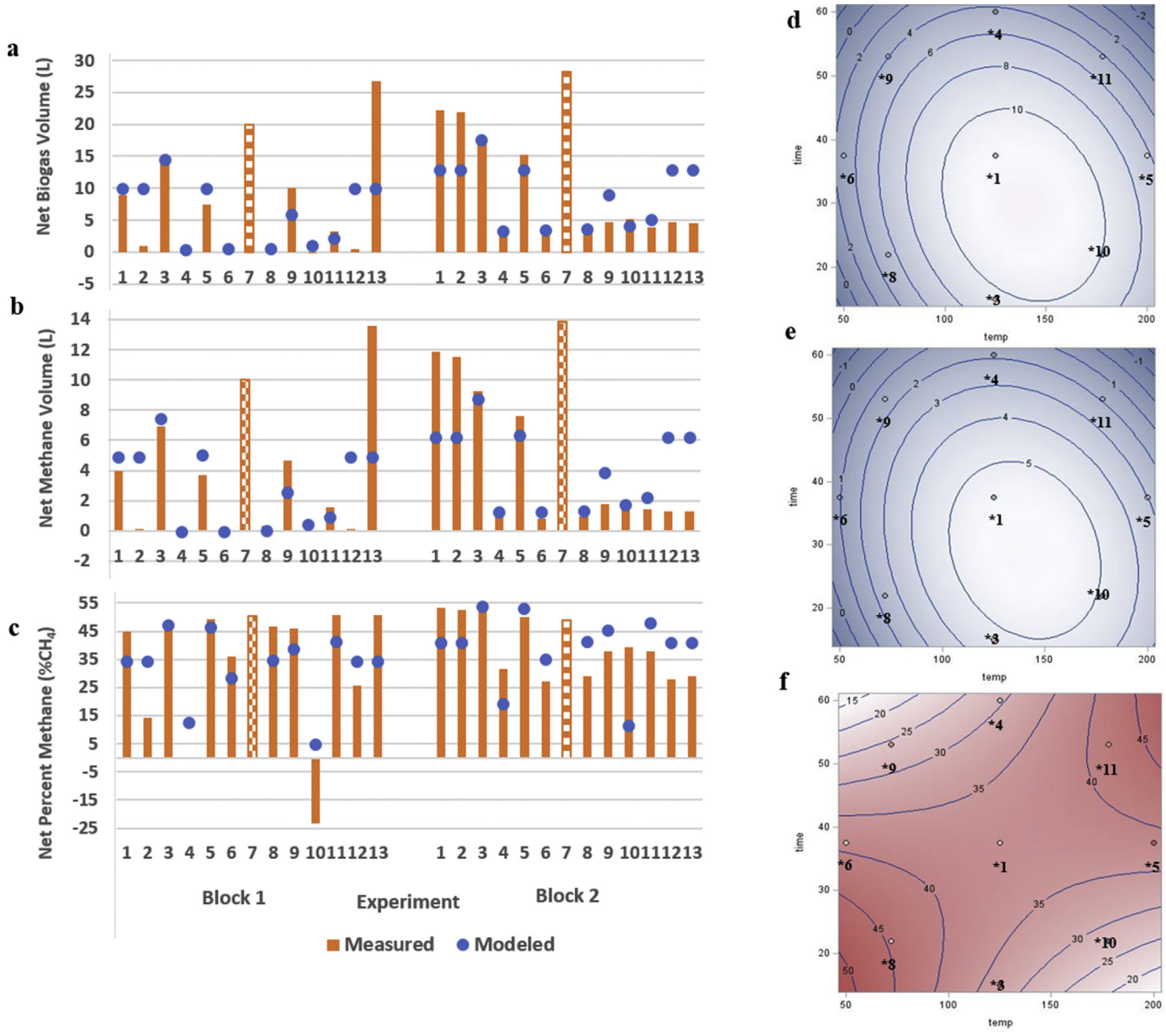

Fig. 1. a-c: Measured and modeled values for the net biogas volume, net methane volume, and the net percent methane based on response surface methodology. Treatment 7 is the positive control and was not included in the central composite rotatable design. Both blocks were analyzed together. For Fig. 1c, Block 1, Experiment 10 produced less gas than the inoculum, its net percent methane is negative. See Tables 2 and 3 for the temperature and duration combination for each treatment. Fig. 1d–f: Contour plots showing the biogas volume, which peaked at about 150 °C, 50 min; the methane volume, which peaked at about 150 °C, 40 min; and the change in percent methane, all in response to pretreatment temperature and duration. The numbers with asterisks are the numbers of the experiments.

Table 4
Estimated statistical measures for models of biogas volume, methane volume, and percent methane.

Factor	Net Biogas Volume (L)		Net Methane Volume (L)		Net Percent Methane (%CH_4)	
	F-value/Estimate	P-value	F-value/Estimate	P-value	F-value/Estimate	P-value
Block	0.94	0.3492	0.59	0.4550	0.90	0.3576
T	0.42	0.5264	0.49	0.4934	0.00	0.9626
T^2	1.90	0.1897	1.71	0.2119	0.07	0.7893
T	0.85	0.3708	0.86	0.3703	0.08	0.7842
t^2	0.99	0.3360	0.84	0.3742	0.40	0.5356
T × t	0.16	0.6918	0.12	0.7351	1.81	0.1999
Lack of fit	1.75	0.2026	1.57	0.2404	3.16	0.0581*
Model	3.47	0.0171**	2.71	0.0435**	11.82	<0.0001***
R-squared	0.429		0.400		0.477	
CV	99.728		116.309		47.708	
RMSE	7.619		4.184		17.034	
Mean	7.640		3.597		35.704	

*Statistically significant p values at α = 0.10 **α = 0.05 ***α = 0.01.

samples is:

$$V_{Methane} = -12.52 + (0.1908 \times T) - (0.00061 \times T^2) + (0.3381 \times t) - (0.00429 \times t^2) - (0.00062 \times T \times t) \quad \text{Eq. 2}$$

where $V_{Methane}$ is the net CH_4 volume (L) (Table 2). The difference between the modeled and measured values (residual) ranged from −4.84 to 8.76 (Fig. 1b). Like the biogas volume, none of the factors in this model were significant (Table 4) and the R^2 value was 40%. The contour plot (Fig. 1e) shows that the CH_4 volume peaks at about 150 °C and 40 minutes and decreases thereafter. Like the biogas volume, for Block 1, only one of the samples pretreated at the center temperature and duration (125 °C, 37.5 min) yielded more CH_4 (36% increase) than the positive control. For Block 2, none of the pretreated samples was greater than the positive control (Fig. 1b).

The literature reports other cases in which thermal pretreatment yields are like the positive controls. For example, the effect of thermal pretreatment on the biogas potential of dewatered pig manure was investigated at 25 °C–150 °C (Rafique et al., 2010). The substrate that was not treated with heat (25 °C) yielded ≈0.37 L biogas g^{-1} VS. The only samples which yielded more biogas than the positive control sample were those treated at 50 °C and 100 °C. The treatment at 100 °C yielded 0.48 ± 0.02 L biogas g^{-1} VS, a 30% increase from the raw manure sample. The other treatments at 70 °C, 110 °C, 130 °C and 150 °C all yielded less biogas than the positive control sample.

Passos et al. (2017) who also worked with dairy cow manure also found that, for almost all cases, thermal pretreatment did not significantly change the final CH_4 yields when compared to the positive controls. The treatment that yielded more CH_4 than the controls (0.29 and 0.33 L CH_4 g^{-1} VS) was conducted at 37 °C for 24 hours and generated ≈0.45 L CH_4 g^{-1} VS, a 35% increase. They concluded that exposure time (duration) had a greater effect than temperature in their experiment.

Suboptimal conditions during pretreatment could account for the lack of a significant response for both biogas yield and CH_4 volume. For instance, Rafique et al. (2010) suggest that complex organic compounds form at elevated temperatures that are very difficult to degrade. These recalcitrant compounds are released as hemicellulose and lignin solubilize. They are not only refractory but act as inhibitors and are toxic to anaerobic microbial populations. Some of these compounds include furfural and soluble phenolic compounds. Elevated temperatures can also create chemical bonds resulting in the agglomeration of the particles within the substrate. One example is the formation of melanoidines formed in the late stages of the Maillard reaction which occur in substrates containing proteins and carbohydrates. The reaction between the carbohydrates and amino acids forms a complex substrate that is difficult to biodegrade. This reaction can occur at extreme thermal treatments that exceed 150 °C or longer treatment durations at lower temperatures (<100 °C) (Ariunbaatar et al., 2014; Carlsson et al., 2012). Maillard reactions can also result in hardening and darkening of the manure during pretreatment at elevated temperatures, which results in less biodegradation and low biogas yields (Ariunbaatar et al., 2014; Rafique et al., 2010). For thermal pretreatments carried out below 100 °C it is possible that the complex molecules were not degraded but only deflocculated or dispersed (Ariunbaatar et al., 2014).

Suboptimal conditions for microbial growth during AD could also account for the lack of significant effect on the biogas yield after pretreatment. For instance, the C/N ratio of the inoculated manure was far from the ideal 30:1 ratio (Environment Canada, 2013; Li et al., 2011). The substrates in this study had C/N ratios of 5.1 and 7.9, indicating an abundance of nitrogen or protein, which leads to excessive microbial growth, production of free radicals and accumulation or overloading of ammonia inhibiting methanogenesis (Cornell Waste Management Institute, 1996).

The biogas yield in L can be converted to L of biogas per grams of VS added. For this experiment, the biogas yield was between 0.01–0.12 L g^{-1} VS_{added}. These values were low compared to what was seen in literature. The values seen in literature for cattle and dairy cow manure were ≈0.25–0.30 L g^{-1} VS (Andriamanohiarisoamanana et al., 2017; Angelidaki and Ahring, 1994). The CH_4 volume can similarly be converted to L CH_4 g^{-1} VS_{added} which, for these experiments was between 0.01–0.06 L CH_4 g^{-1} VS_{added}. The CH_4 yields reported in the literature were much higher, i.e. 0.10–0.30 L CH_4 g^{-1} VS_{added} (Andriamanohiarisoamanana et al., 2017; Labatut et al., 2011; Møller et al., 2004). Passos et al. (2017) obtained a high CH_4 yield for the positive control (0.29 and 0.33 L CH_4 g^{-1} VS_{added}) when compared to the treated samples (0.04 and 0.06 L CH_4 g^{-1} VS_{added}). They speculated that the differences in the CH_4 concentration may have been due to environmental conditions and animal feeding practices (Passos et al., 2017).

In this experiment, the digesters that produced the most biogas and CH_4 in both experimental blocks were either the positive control or replicates pretreated at the center temperature and duration (125 °C, 37.5 min) and center temperature and minimum duration (125 °C, 15 min).

Following the calculation from Montgomery (2005), the optimum temperature and duration for pretreatment were 140 °C for 30 min. This optimum temperature is similar to the one recommended by Carrère et al. (2009). In their experiments, temperatures higher than 135 °C were necessary to improve the methane potential of the total fraction of the pig manure. The best results were obtained with the highest temperature i.e. 190 °C. The context of these temperatures was determined solely on a laboratory basis. Moreover, Cano et al. (2014) conducted lab-scale hydrolysis plant experiments which consisted of a 2-L reactor fed with the substrate and heated with steam until the desired temperature, and a flash tank of 5 L where the steam explosion takes place after the hydrolysis reaction time elapsed. They determined that 170 °C for 30 minutes was the optimal conditions for the pretreatment of the manure. This heat treatment improved the AD of the manure (from 317 mL CH_4/g VS to 408

mL CH_4/g VS), producing 29% more CH_4. However, considering the results from our study, pretreatment may be an unnecessary step for the anaerobic digestion of dairy cow manure.

Pretreatment efficiency indicates the impact of the input of thermal energy before and after pretreatment and its further impact on the biogas and CH_4 production. If an energy analysis of the pretreatment and AD system was carried out, then the excess energy produced because of the pretreatment should be weighed against the extra energy required to perform the pretreatment (Carlsson et al., 2012). Combining thermal pretreatment and anaerobic digestion is advantageous as the possibility of getting the energy needed for the thermal process can be obtained from the different integration possibilities, such as recovery of heat from hot streams and the use of the biogas produced in the AD to generate steam (Pérez-Elvira et al., 2008). Limited economic studies were found for the combination of thermal pretreatment and anaerobic digestion when manure was used as the substrate. Cano et al. (2014) found that the implementation of thermal hydrolysis with steam explosion (170 °C) for cow manure yields an economic benefit of 29% (936 kW h/t) compared to conventional AD (727 kW h/t). Passos et al. (2017) conducted techno-economic analysis comparing AD without a pretreatment step, thermal alkali pretreatment followed by AD and thermal acid pretreatment followed by AD. According to the results, thermal-alkali pretreatment showed a negative total cost (−438,873 US$/year) since the surplus of energy injected to the grid (197,851 US$/year) did not offset the expenses (636,724 US$/year) related to the extra energy and the cost of the chemical agent. This result was attributed to the high temperature required for the pretreatment, which under the conditions of their experiment could not supply the required thermal energy from the biogas in the combined heat and power unit. The total income of the thermal-acid pretreatment was positive (936,770 US$/year) but it was still lower than the income obtained by the conventional AD application (1,033,506 US$/year).

The quadratic response surface model for $\%CH_4$ is presented in Eq. (3):

$$\%CH_4 = 73.30 - (0.4294 \times T) + (0.00022 \times T^2) - (0.4015 \times t) - (0.0121 \times t^2) + (0.00986 \times T \times t) \quad \text{Eq. 3}$$

The $\%CH_4$ is the percent CH_4 of the cumulative biogas volume for each experiment. As for biogas yield and CH_4 volume, none of the factors of the model were significant except the lack of fit. The R^2 value was ≈48%. The measured and modeled values are shown in Fig. 1c. Fig. 1f illustrates how the $\%CH_4$ decreases as the temperature increases for treatment durations up to 40 min. For longer durations, the $\%CH_4$ increases with temperature.

The $\%CH_4$ for the positive controls (Experiment 7) were 50.27%CH_4 for Block 1 and 48.94%CH_4 for Block 2 (Fig. 1c). The best improvement in $\%CH_4$ was seen for Experiment 13 in Block 1 and Experiments 1 and 2 in Blocks 2, all conducted at 125 °C for 37.5 min (50.89%CH_4, 52.57% CH_4, 53.33%CH_4 respectively) and Experiment 3, Block 2, 125 °C, 15 min (52.88%CH_4). This was concordant with the literature as the $\%CH_4$ of the biogas is expected to be between 55–70% (Appels et al., 2011).

4. Conclusions

No significant effect of pretreatment temperature or duration was seen on biogas volume, CH_4 volume, or $\%CH_4$. Based on previously published results from other studies, the thermal pretreatment process considered to be a promising approach to improving the yield and quality of biogas from the anaerobic digestion of substrates with high lignocellulosic content, such as cow or pig manure. Thermal energy was predicted to alter the structure of the physical, structural and chemical properties of the substrate, making the volatile component of the substrate more available to hydrolytic microbes and increasing the biogas content.

The model that was generated from our results predicted optimum pretreatment conditions of 140 °C for 30 min. The treatments for which the highest amount of biogas and CH_4 were observed were the positive control and the center temperature and duration (125 °C, 37.5 min). Only one replicate of the center temperature and duration yielded more biogas and methane than the positive control. The lack of significant effect could mean that thermal pretreatment of dairy cow manure may not be required, which would save the AD industry from investing in unnecessary infrastructure. Also, cow manure has been previously digested in the bovine gut, a process that already alters the lignocellulosic content of the substrate and may render thermal pretreatment redundant. Therefore, the best method to increase the yield from the anaerobic digestion of cow manure may be co-digestion with another appropriate biomass.

Declarations

Author contribution statement

Grant Clark: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Wilton McVoitte: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Funding statement

This work was supported by the Natural Sciences and Engineering Research Council (NSERC) of Canada, Discovery Grant RGPIN-2014-04273.

Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

References

Andriamanohiarisoamanana, F.J., Saikawa, A., Tarukawa, K., Qi, G., Pan, Z., Yamashiro, T., Iwasaki, M., Ihara, I., Nishida, T., Umetsu, K., 2017. Anaerobic co-digestion of dairy manure, meat and bone meal, and crude glycerol under mesophilic conditions: synergistic effect and kinetic studies. Energy Sustain. Develop. 40, 11–18.

Angelidaki, I., Ahring, B.K., 1994. Anaerobic thermophilic digestion of manure at different ammonia loads: effect of temperature. Water Res. 28 (3), 727–731.

Angelidaki, I., Ellegaard, L., 2003. Codigestion of manure and organic wastes in centralized biogas plants. Appl. Biochem. Biotechnol. 109 (1), 95–105.

Angelonidi, E., Smith, S.R., 2015. A comparison of wet and dry anaerobic digestion processes for the treatment of municipal solid waste and food waste. Water Environ. J. 29 (4), 549–557.

APHA, AWWA, WEF, 2005. Standard Methods for the Examination of Water and Wastewater, twenty-first ed. American Public Health Association (APHA), American Water Works Association (AWWA), Water Environment Federation (WEF), Washington, DC.

Appels, L., Lauwers, J., Degreve, J., Helsen, L., Lievens, B., Willems, K., Van Impe, J., Dewil, R., 2011. Anaerobic digestion in global bio-energy production: potential and research challenges. Renew. Sustain. Energy Rev. 15 (9), 4295–4301.

Ariunbaatar, J., Panico, A., Esposito, G., Pirozzi, F., Lens, P.N.L., 2014. Pretreatment methods to enhance anaerobic digestion of organic solid waste. Appl. Energy 123, 143–156.

Bordeleau, É.L., Droste, R.L., 2011. Comprehensive review and compilation of pretreatments for mesophilic and thermophilic anaerobic digestion. Water Sci. Technol. 63 (2), 291.

Bowley, S.R., 2008. Response surfaces. A Hitchhiker's Guide to Statistics in Plant Biology. Any Old Subject Books, Guelph, Ontario, Canada, pp. 167–180.

Cano, R., Nielfa, A., Fdz-Polanco, M., 2014. Thermal hydrolysis integration in the anaerobic digestion process of different solid wastes: energy and economic feasibility study. Bioresour. Technol. 168, 14–22.

Carlsson, M., Lagerkvist, A., Morgan-Sagastume, F., 2012. The effects of substrate pretreatment on anaerobic digestion systems: a review. Waste Manag. 32 (9), 1634–1650.

Carrère, H., Sialve, B., Bernet, N., 2009. Improving pig manure conversion into biogas by thermal and thermo-chemical pretreatments. Bioresour. Technol. 100 (15), 3690–3694.

Carrère, H., Dumas, C., Battimelli, A., Batstone, D.J., Delgenès, J.P., Steyer, J.P., Ferrer, I., 2010. Pretreatment methods to improve sludge anaerobic degradability: a review. J. Hazard Mater. 183 (1–3), 1–15.

Chaikitkaew, S., Kongjan, P., O-Thong, S., 2015. Biogas production from biomass residues of palm oil mill by solid state anaerobic digestion. Energy Procedia. 79, 838–844.

Chen, Y., Cheng, J.J., Creamer, K.S., 2008. Inhibition of anaerobic digestion process: a review. Bioresour. Technol. 99 (10), 4044–4064.

Cornell Waste Management Institute, 1996. Cornell composting: the science and engineering of composting. http://compost.css.cornell.edu/science.html. (Accessed 14 February 2017).

Environment Canada, 2013. Technical Document on Municipal Solid Waste Organics Processing. Public Works and Government Services of Canada (PWGS), Government of Canada. https://www.ec.gc.ca/gdd-mw/3E8CF6C7-F214-4BA2-A1A3-163978EE9D6E/13-047-ID-458-PDF_accessible_ANG_R2-reduced%20size.pdf. (Accessed 5 December 2017).

Hassan, M., Ding, W.M., Shi, Z.D., Zhao, S.Q., 2016. Methane enhancement through co-digestion of chicken manure and thermo-oxidative cleaved wheat straw with waste activated sludge: a C/N optimization case. Bioresour. Technol. 211, 534–541.

Hofmann, N., 2006. A geographical profile of livestock manure production in Canada. Enivironment Accounts and Statistics Division, Statistics Canada. https://www150.statcan.gc.ca/n1/pub/16-002-x/2008004/article/10751-eng.htm. (Accessed 23 March 2018).

Karimi, F., Rafiee, S., Taheri-Garavand, A., Karimi, M., 2012. Optimization of an air drying process for artemisia absinthium leaves using response surface and artificial neural network models. J. Taiwan Inst. Chem. Eng. 43 (1), 29–39.

Khoobbakht, G., Najafi, G., Karimi, M., Akram, A., 2016. Optimization of operating factors and blended levels of diesel, biodiesel and ethanol fuels to minimize exhaust emissions of diesel engine using response surface methodology. Appl. Therm. Eng. 99, 1006–1017.

Koszel, M., Lorencowicz, E., 2015. Agricultural use of biogas digestate as a replacement fertilizers. Agric. Agric. Sci. Procedia 7, 119–124.

Labatut, R.A., Angenent, L.T., Scott, N.R., 2011. Biochemical methane potential and biodegradability of complex organic substrates. Bioresour. Technol. 102 (3), 2255–2264.

Li, Y.B., Park, S.Y., Zhu, J.Y., 2011. Solid-state anaerobic digestion for methane production from organic waste. Renew. Sustain. Energy Rev. 15 (1), 821–826.

Lymperatou, A., Gavala, H.N., Skiadas, I.V., 2017. Optimization of aqueous ammonia soaking of manure fibers by response surface methodology for unlocking the methane potential of swine manure. Bioresour. Technol. 244, 509–516.

Møller, H.B., Sommer, S.G., Ahring, B.K., 2004. Methane productivity of manure, straw and solid fractions of manure. Biomass Bioenergy 26 (5), 485–495.

Montgomery, D.C., 2005. Response surface methods and designs. Design and Analysis of Experiments, sixth ed. John Wiley & Sons, Inc., NJ, USA, pp. 405–463.

Nasir, I.M., Ghazi, T.I.M., 2015. Pretreatment of lignocellulosic biomass from animal manure as a means of enhancing biogas production. Eng. Life Sci. 15 (7), 733–742.

Passos, F., Ortega, V., Donoso-Bravo, A., 2017. Thermochemical pretreatment and anaerobic digestion of dairy cow manure: experimental and economic evaluation. Bioresour. Technol. 227, 239–246.

Pérez-Elvira, S.I., Férnández-Polanco, F., Férnández-Polanco, M., 2008. Hydrothermal multivariable approach. Full-scale feasibility study. Electron. J. Biotechnol. 11 (4).

Polprasert, C., 2007. Organic Waste Recycling: Technology and Management, third ed. IWA Publishing, London, UK.

Rafique, R., Poulsen, T.G., Nizami, A.-S., Asam, Z.-u.-Z., Murphy, J.D., Kiely, G., 2010. Effect of thermal, chemical and thermo-chemical pre-treatments to enhance methane production. Energy 35 (12), 4556–4561.

Raju, C.S., Sutaryo, S., Ward, A.J., Møller, H.B., 2013. Effects of high-temperature isochoric pre-treatment on the methane yields of cattle, pig and chicken manure. Environ. Technol. 34 (2), 239–244.

Rakić, T., Kasagić-Vujanović, I., Jovanović, M., Jančić-Stojanović, B., Ivanović, D., 2014. Comparison of full factorial design, central composite design, and box-behnken design in chromatographic method development for the determination of fluconazole and its impurities. Anal. Lett. 47 (8), 1334–1347.

Ravindran, B., Wong, J.W.C., Selvam, A., Thirunavukarasu, K., Sekaran, G., 2016. Microbial biodegradation of proteinaceous tannery solid waste and production of a novel value added product – Metalloprotease. Bioresour. Technol. 217, 150–156.

Ville de Saint-Hyacinthe: Technopole agroalimentaire, n.d. Environment and residual materials: Biomethanation. Ville de Saint-Hyacinthe. http://www.ville.st-hyacinthe.qc.ca/services-aux-citoyens/biomethanisation.php. June 2, 2018.

Westphal, A., Kuecke, M., Heuer, H., 2016. Soil amendment with digestate from bio-energy fermenters for mitigating damage to beta vulgaris subspp. by heterodera schachtii. Appl. Soil Ecol. 99, 129–136.

Wolf, N., 2003. Determination of manure pH. In: Peters, J. (Ed.), The Board of Regents of the University of Wisconsin System. Cooperative Extension Publishing Operations, Madison, WI, pp. 48–49. Recommended methods of manure analysis (A3769).

Zhang, Z.K., Li, W.L., Zhang, G.Y., Xu, G.W., 2014. Impact of pretreatment on solid state anaerobic digestion of yard waste for biogas production. World J. Microbiol. Biotechnol. 30 (2), 547–554.