



Anaerobic co-digestion of livestock and vegetable processing wastes: Fibre degradation and digestate stability

Beatriz Molinuevo-Salces^{a,*}, Xiomar Gómez^b, Antonio Morán^b, Mari Cruz García-González^a

^a Agricultural Technological Institute of Castilla y Leon, Ctra. Burgos, km. 119, 47071 Valladolid, Spain

^b Chemical Engineering Department, University of Leon, IRENA, Avda. de Portugal 41, 24071 León, Spain

ARTICLE INFO

Article history:

Received 24 July 2012

Accepted 21 February 2013

Available online 26 March 2013

Keywords:

Swine manure
Vegetable wastes
Anaerobic digestion
Lignin
Thermal analyses

ABSTRACT

Anaerobic digestion of livestock wastes (swine manure (SM) and poultry litter (PL)) and vegetable processing wastes (VPW) mixtures was evaluated in terms of methane yield, volatile solids removal and lignocellulosic material degradation. Batch experiments were performed with 2% VS (volatile solids) to ensure complete conversion of TVFAs (total volatile fatty acids) and to avoid ammonia inhibition. Experimental methane yields obtained for the mixtures resulted in higher values than those obtained from the sum of the methane yields from the individual components. VPW addition to livestock wastes before anaerobic digestion also resulted in improved VS elimination. In SM-VPW co-digestions, CH₄ yield increased from 111 to 244 mL CH₄ g VS_{added}⁻¹, and the percentage of VS removed increased from 50% to 86%. For PL-VPW co-digestions, the corresponding values were increased from 158 to 223 mL CH₄ g VS_{added}⁻¹ and from 70% to 92% VS removed. Hemicelluloses and more than 50% of cellulose were degraded during anaerobic digestion. Thermal analyses indicated that the stabilization of the wastes during anaerobic digestion resulted in significantly less energy being released by digestate samples than fresh samples.

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1. Introduction

One of the common problems associated with waste treatment by anaerobic digestion is inhibition of the process due to accumulation of intermediate products or by-products. As repeatedly described for manure anaerobic digestions, nutrient imbalances may cause high concentrations of N-NH₃, leading to process failures (Angelidaki and Ahring, 1993). The optimum carbon–nitrogen (C/N) ratio for anaerobic digestion is in the range of 20–25 (Yen and Brune, 2007). Livestock wastes (swine manure (SM) and poultry litter (PL)) have high nitrogen contents resulting in low C/N ratios. By contrast, vegetable processing wastes (VPW) have high C/N ratios, which make them a suitable co-substrate for anaerobic treatment with manures. However, the organic content of VPW is readily hydrolysed to total volatile fatty acids (TVFAs), which may limit anaerobic degradation if they build-up rapidly during digestion. Massive production of TVFAs during the acidogenesis stage may lead to process acidification, which can inhibit the methanogenic activity of microorganisms (Bouallagui et al., 2005; Molinuevo et al., 2008). Several studies of co-digestion of livestock wastes with VPW have been described in an attempt to obtain a

more balanced C/N ratio (Callaghan et al., 2002; Habiba et al., 2009). Livestock residues provide nitrogen for cell growth and carbon degradation (Fricke et al., 2007), and the high buffering capacity of these residues (González-Fernández et al., 2008) may avoid pH drops due to TVFAs build-up. Consequently, mixing VPW and livestock residues can provide a balanced C/N ratio, thereby preventing toxic concentrations of N-NH₃ during the process and improving the biodegradability of the waste resulting in a more stable digestion process.

The biodegradability of wastes depends, among other things, on the presence of lignocellulosic complex structures. Lignocellulosic materials include hemicelluloses, cellulose and lignin. Hemicelluloses and cellulose are complex polysaccharides present in plant cell walls. Hemicelluloses are easily hydrolysable due to their amorphous structure, which is more vulnerable than cellulose or lignin structures to enzymatic attack (Ghosh and Henry, 1985). Cellulose has a simple structure and few different enzymes are necessary to digest it. Cellulose solubilisation is dependent on the inoculum source, biomass concentration and cellulose availability (Jensen et al., 2009). Anaerobic digestion of cellulose may be hindered by cell wall components, including lignin in particular. Lignin molecules reduce cellulose bioavailability by reducing the surface area available to enzymatic penetration and activity (Haug, 1993). Lignin is a recalcitrant compound and its degradation is a limiting step in waste anaerobic digestion (Robbins et al., 1979; Pavlostathis and Giraldo-Gomez, 1991). In addition, lignin

* Corresponding author. Tel.: +34 983 317 388.

E-mail addresses: beamol6@hotmail.com, gargonmi@itacyl.es (B. Molinuevo-Salces).

degradation products, such as phenolic acids and aldehydes, have been reported to be toxic to methanogenic microorganisms (Chen et al., 2008).

Stabilisation of wastes by appropriate anaerobic digestion processes results in an odour-free product with reduced putrefaction potential. Various analytical techniques have been used to study the degree of stabilisation attained during biological processes, although assessing the result of the biological degradation is not an easy task. The thermal analysis (TA) has been demonstrated to be a useful tool for evaluating the stability of biological products, including anaerobic digestates (Gómez et al., 2007) and composts (Dell'Abate et al., 1998). TA assesses the changes in the properties of a material with changing temperature. This technique can be used to determine the combustible fraction of organic matter and therefore the energy potential of a substrate. Several studies have addressed fibre degradation during anaerobic digestion of plant-derived substrates (Tong et al., 1990; Chanakya et al., 2009). However, little is known about fibre degradation during anaerobic digestion of livestock wastes, and these wastes indeed often contain recalcitrant organic fibre (lignin) due to the presence of straw bedding material.

The objective of this study was to evaluate lignocellulosic complex degradation during the anaerobic co-digestion of various mixtures of livestock and vegetable wastes. The stability of digestates was also studied to assess process efficiencies. TA was used for evaluating conversion during anaerobic digestion in batch tests.

2. Methods

2.1. Substrates and sludge characteristics

Three different substrates were used: SM, PL and VPW. SM was obtained from a pig farm in Avila (Spain), with 2600 finishing pigs with an annual manure production of 8750 m³. PL was collected from a poultry farm in Palencia (Spain) with 86,000 laying hens and an annual manure production of 5850 m³. VPW were from a vegetable-processing factory in Segovia (Spain); they were composed of green pea, maize, carrot and leek residues and were ground to particles of about 1 mm diameter in a fruit mill. The anaerobic sludge (AS) used as inoculum was obtained from the anaerobic digester of the municipal wastewater treatment plant (WWTP) of Valladolid (Spain). All the substrates were homogenized and stored at 4 °C before use. The chemical compositions of substrates and inoculum are reported in Table 1.

2.2. Experimental set-up

Batch assays were run with a volatile solid (VS) concentration in the substrate of 2%. This VS concentration was selected to avoid

Table 1
Chemical characterisation of substrates (VPW, SM, PL) and anaerobic sludge (AS).

Parameters	VPW	SM	PL	AS
pH	4.4	7.7	9.2	7.4
VS (g L ⁻¹)	134.7 (1.6)	17.1 (0.0)	201.3 (0.8)	5.6 (0.1)
TS (g L ⁻¹)	143.6 (1.9)	25.2 (0.2)	305.2 (13.5)	12.3 (0.1)
CODs (g L ⁻¹)	70.9 (1.7)	5.1 (0.1)	12.4 (0.9)	11.7 (3.5)
CODt (g L ⁻¹)	224.1 (49.4)	29.8 (0.3)	n.d.	17.5 (0.6)
TKN (g L ⁻¹)	3.5 (0.1)	4.1 (0.1)	13.8 (1.4)	1.0 (0.0)
N-NH ₄ ⁺ (g L ⁻¹)	0.9 (0.2)	2.8 (0.0)	12.8 (0.7)	0.6 (0.0)
C ³ :N	38	4	15	6
Hemicellulose (% TS)	7.8	2	4.4	n.d.
Cellulose (% TS)	17.4	22	19	n.d.
Lignin (% TS)	4.4	9.8	4.2	n.d.

n.d.: not determined.

^a Corrected values taking into account TVFAs lost (Vedrenne et al., 2008).

Table 2

Proportion of the various mixtures (C1–C9) used for batch digestion tests. Percentages are expressed in dry weights (DW).

	% VPW (DW)	% SM (DW)	% PL (DW)
C1	0	100	0
C2	25	75	0
C3	50	50	0
C4	75	25	0
C5	100	0	0
C6	0	0	100
C7	25	0	75
C8	50	0	50
C9	75	0	25

TVFAs- and N-NH₃-mediated inhibition, because the main objective of this study was to evaluate lignocellulose degradation. The VS concentration was chosen according to Molinuevo-Salces et al. (2010), who studied the effect of substrate concentration on methane yield and VS reduction when co-digesting VPW-SM and VPW-PL under batch conditions. Different mixtures of livestock waste and VPW were prepared. These mixtures were denoted as C1–C9, as shown in Table 2. C1–C4 were for SM-VPW co-digestion tests, C6–C9 for PL-VPW co-digestion tests and C5 for VPW digestion tests.

The anaerobic assays were conducted in 500 mL bottles containing 100 mL of inoculum and 100 mL of substrate. Blanks containing 100 mL of inoculum and 100 mL of distilled water were also run to determine the endogenous methane production of the anaerobic sludge. A solution of KHCO₃ at 14 g L⁻¹ was used to adjust the pH to 7.5 at the beginning of the experiment (Cirne et al., 2007). The bottles were closed with a septum and flushed with N₂ to remove oxygen. They then were incubated in a thermostatic shaker at 100 rpm and 35 ± 2 °C for up to 80 days. Production of biogas was monitored by measuring the overpressure in the bottle headspace at time intervals depending on the production of biogas (Colleran et al., 1992).

2.3. Analyses

Analyses consisted of assaying total solids (TS), VS, total and soluble chemical oxygen demand (CODt and CODs), total Kjeldahl nitrogen (TKN) and ammonium nitrogen (N-NH₄⁺). Standard methods were used for all analyses (APHA, 2005). Free ammonia concentrations at the end of the tests were calculated in accordance with Hansen et al. (1998).

Fresh (pre-digestion) and digested samples from SM-VPW (C3), VPW (C5) and PL-VPW (C8) assays were collected for analysis of lignin, acid and neutral detergent fibres (ADL, ADF, and NDF, respectively). These analyses were performed by the method of Van Soest and Wine (1967) using a ANKOM 2000I fibre analyser. Cellulose and hemicellulose contents were determined by subtraction: ADF-ADL and NDF-ADF, respectively. Total carbon was calculated by the addition of VS and TVFAs values at the beginning of the assay. TVFAs volatilization of volatile acids during drying for the determination of VS was taken into account in these calculations (Vedrenne et al., 2008).

Biogas composition was analysed using a gas chromatograph (Varian CP 3800 GC) with a thermal conductivity detector, provided by a CP-Molsieve5A column (15 m × 0.53 mm × 15 μm) followed by a CP-Porabond Q column (25 m × 0.53 mm × 10 μm). Hydrogen (13.6 mL min⁻¹) was used as the carrier gas. The injection port temperature was set at 150 °C and the detector temperature was 175 °C. TVFAs was analysed using a gas chromatograph (Varian CP 3800 GC) equipped with a Nukol capillary column (30 m × 0.25 mm × 0.25 μm) and a flame ionization detector. The carrier gas was helium and the temperature of the injector was

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