Fuel 193 (2017) 15–21

Contents lists available at ScienceDirect

Fuel

journal homepage: www.elsevier.com/locate/fuel

Full Length Article

Evaluation of the effect of glycerol supplementation on the anaerobic digestion of real municipal solid waste in batch mode

S. Zahedi [a,b], R. Solera [a,*], J.L. García-Morales [a], H. Ennouri [c], D. Sales [a]

[a] Department of Environmental Technologies, University of Cadiz, Faculty of Marine and Environmental Sciences (CASEM), Pol, Río San Pedro s/n, 11510 Puerto Real (Cádiz), Spain
[b] Catalan Institute for Water Research (ICRA), Scientific and Technological Park of the University of Girona, 17003, Spain
[c] Université de Carthage, LR: LETMi, INSAT, B.P. 676, 1080 Tunis, Tunisia

ARTICLE INFO

Article history:
Received 29 October 2016
Received in revised form 5 December 2016
Accepted 8 December 2016

Keywords:
Methane production
Real municipal solid waste
Batch mode
Methanogenic activity
Glycerol

ABSTRACT

The effect of low glycerol supplementation (<1% v/v) in the thermophilic anaerobic digestion (AD) of real municipal solid waste (RMSW) on methane production (MP) is not known. This study explores the effect of five different glycerol supplementations (0%, 0.1%, 0.25%, 0.5% and 1%) on effluent characteristics, anaerobic consortia and MP. Specifically, by adding 0.25% v/v crude glycerol to the feed, the methane production rate increased by 48% (from 7.40 ± 1.17 l CH_4/l to 11.01 ± 1.66 l CH_4/l), in line with the increase in total volatile solids (TVS) removal (from 65 ± 7% to 81 ± 7) and methanogenic activity (from $110 * 10^{-12} \pm 17 * 10^{-12}$ l CH_4/cell to $156 * 10^{-12} \pm 24 * 10^{-12}$ l CH_4/cell). Extra glycerol added to the feed (0.5% or 1%) was shown to be non-feasible for thermophilic AD of RMSW, as inhibition of methanogenesis was observed. Fluorescent in situ hybridization (FISH) studies showed that the percentages of *Eubacteria*, *Archaea*, H_2-utilising methanogens (HUM) and acetate-utilising methanogens (AUM) were stable within the 94.0–97.0%, 6.0–3.0%, 1.8–3.6% and 0.6–2.8% ranges, respectively.

1. Introduction

Municipal solid waste (MSW) generation is increasing significantly in current society. Almost 1.9 billion tons of MSW are generated worldwide every year, which means about 218 kg/person/year [1]. Anaerobic digestion (AD) is an attractive treatment strategy for generating energy from putrescible MSW and for mitigating the problem of disposal [1–3]. It also has a reduced environmental impact, especially with respect to the greenhouse effect. Due to the advantages of AD, many research studies have sought to optimize the AD of MSW, including the interesting option of the co-digestion process, which increases the load of biodegradable organic matter and produces a higher biogas yield [4–8]. Studies on the AD of MSW have shown that the C/N ratio of this waste presents average values of 10:1, below the optimum for anaerobic digestion (25:1) [9], while methane production (MP) is reduced due to the washing up of microorganisms and not to overloading [10,11]. Therefore, an increase in the loading rate of the AD process via the addition of readily biodegradable organic substances, such as glycerol, a major by-product of biodiesel production, could be an ideal strategy [5]. Biodiesel constitutes a promising alternative as a renewable fuel and its production capacity has been well developed in recent years [12,13]. The increased production of biodiesel supposes an increase in crude glycerol. The global biodiesel market is expected to reach 37 billion gallons by 2016, with an average annual growth rate of 42%. This means about 4 billion gallons of crude glycerol will be produced each year. A recent study [7] has demonstrated the effectiveness of glycerol supplementation (1% v/v) on the hydrogen production steps in thermophilic-dry dark fermentation of real MSW (RMSW) in batch mode. However, no previous studies have been published on the effect of glycerol addition on the biodegradation of RMSW and subsequent methane generation or on the effect on the microbial groups involved in the digestion process, which have varying sensitivities to environmental changes, especially acetogenic bacteria and methanogenic *Archaea*.

The aim of this study was to investigate the effect of different glycerol supplementations (0%, 0.1%, 0.25%, 0.5% and 1%) on the AD of RMSW. This residue is a known substrate for methane production (MP) [9,10,14,15]. For this purpose, anaerobic batch reactors were used to determine the anaerobic biodegradation and methane generation potential.

The effect of operational parameter modifications on dissolved chemical oxygen demand (COD_D), volatiles fatty acids (VFA), total volatile solids (TVS), ammonia, alkalinity, pH, microbiological population and MP were considered to study the effect of glycerol on

* Corresponding author.
E-mail address: rosario.solera@uca.es (R. Solera).

http://dx.doi.org/10.1016/j.fuel.2016.12.024
0016-2361/

Nomenclature

AD	anaerobic digestion
AUM	acetate-utilising methanogens
COD	chemical oxygen demand
FISH	fluorescence in situ hybridization
GP	gas production
HUM	H_2-utilising methanogens
RMSW	real municipal solid waste
MP	methane production
MSW	municipal solid waste
OFMSW	organic fraction municipal solid waste
OLR	organic loading rate
SMP	specific methane production
TS	total solids
TVS	total volatile solids
VFA	volatile fatty acids

the anaerobic batch processing of RMSW. The anaerobic consortia was analysed by fluorescent in situ hybridization (FISH), employing different oligonucleotide probes.

2. Materials and methods

2.1. Inoculum and substrate

Anaerobic digester effluent from single-phase dry-thermophilic AD of RMSW was used as inoculum. The RMSW and glycerol used to feed the reactors came from an industrial trommel (30 mm) located at the Las Calandrias MSW treatment plant (Jerez de la Frontera, Cádiz-Spain) and the Abengoa Bionergy biofuel company (San Roque, Cádiz-Spain), respectively.

The RMSW was stored in 25 kg drums at 4 °C to avoid anaerobic degradation by the microorganisms present in the solid waste itself. The characterisation of the substrates used in the tests is shown in Tables 1 and 2.

Table 1
Physical-chemical and microbiological characterisation of the RMSW used in the trials.

Parameter	Average	Range
COD_D (g/l)	170(10)	160–180
TVFA (g acetic/l)	5(1)	4–6
TVS (g/kg)	225(10)	215–235
C:N (Organic matter)	13(3)	10–16
Total population (10^7 cells/ml)	36(7)	29–43
EU (%)	88(2)	86–90
Archaea (%)	12(2)	10–14
AUM (%)	7(1)	6–8
HUM (%)	5(1)	4–6

Average values are shown with standard deviations in parentheses.

Table 2
Characterisation of the glycerol used in the trials.

Parameter	Average	Range
COD_D (g/l)	1400(100)	1300–1500
TVFA (g acetic/l)	<0.1	<0.1
TVS (g/kg)	820(10)	810–830
H_2O	12(1)	11–13
K	<0.1	–
Na	5.5(0.5)	5.0–6.0

Average values are shown with standard deviations in parentheses.

2.2. Experimental reactors

To investigate the effect of different glycerol supplementations on the AD of RMSW, five different amounts of glycerol were tested (0%, 0.1%, 0.25%, 0.5% and 1%). The batch system used was made up of twelve reactors (2 for each condition + 2 blanks (inoculum without waste)), each with a total volume of 2 l and a useful volume of 1.7 l. Each reactor was equipped with an independent agitation system capable of maintaining an uniform moisture content and of redistributing the soluble substrate and microorganisms, as well as electric control of temperature. Ten digesters were initially loaded with a mixture of inoculum (700 g) and substrate (351 g RMSW in 1 l of tap water), resulting in a final concentration of 40% v/v inoculum, which is considered optimum for biogas production and substrate acclimatization [16]. Different amounts of glycerol were then added to the reactors to give glycerol percentages in the 0% v/v (Test 1), 0.1% v/v (Test 2), 0.25% v/v (Test 3), 0.5% v/v (Test 4) and 1% v/v (Test 5) range (two for each test). The two blank experiments were conducted in order to determine the amount of endogenous methane production of the inoculum. This amount was accordingly subtracted from all the experimental results. The reactors were monitored until reaching a MP plateau, which represented a monitoring period of at least 25 days.

2.3. Analytical methods

The analytical methods employed in this study can be grouped into two categories: the physical-chemical parameters controlling the process of degradability, and methods for quantifying the microbial population in the reactors.

2.3.1. Physical-chemical analysis

For process monitoring and control of the reactors, the following analytical determinations were performed: COD_D, pH, TVS, VFA, ammonia and alkalinity. All analyses were carried out in

Table 3
Oligonucleotide probes used in the study.

Probes	Probe sequences (from5′ a 3′)	Target	Formamide (%)	Time (h)	T (°C)	Reference
EUB338	GCTGCCTCCCGTAGGAGT	*Eubacteria*	20	1.5	46	Zahedi et al. [10,11]
S-D-Bact-0338-a-S-18	ACTCCTACGGGAGGCAGC	None (negative control)	20	1.5	46	Zahedi et al. [10,11]
ARC915	GTGCTCCCCCGCCAATTCCT	*Archaebacteria*	35	1.5	46	Zahedi et al. [10,11]
MB1174	TACCGTCGTCCACTCCTTCCTC	HUM	35	1.5	46	Zahedi et al. [10,11]
MX825	TCGCACCGTGGCCGACACCTAGC	AUM	20	1.5	46	Zahedi et al. [10,11]