Stabilization of thermophilic and mesophilic anaerobic digestion of sugar beet by-products and co-digestion with cow manure in semi-continuous stirred tank reactors

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Abstract

The aim of this study was to compare the single digestion of sugar beet by-products with the co-digestion of this waste using cow manure as co-substrate. The semi-continuous anaerobic processes were carried out working at a hydraulic retention time of 30 days and an 8% of total solids concentration in the feeding. The average methane yields reached in the single digestion tests were 30 mL CH_4/gVS $_{added}$ in mesophilic reactor (35ºC) and 110 mL CH_4/gVS$_{added}$ for the thermophilic (55ºC) one. However, co-digestion with cow manure leads to an increase in the average methane yield for both temperatures. Indeed, 178.6 mL CH_4/gVS $_{added}$ for mesophilic and 182.44 mL CH_4/gVS$_{added}$ for thermophilic process were obtained when co-digestion was applied. The low methane yields obtained for single digestion of SBB were accompanied by the accumulation of volatile fatty acids in the system. In fact, acetic acid was the most extensively accumulated volatile fatty acid throughout the process and, hence, an inhibition of the acetoclastic methanogenic archaea population must be occurring.

Keywords: Anaerobic digestion, Sugar beet, Cow manure, Thermophilic, Mesophilic.

1. Introduction

Sugar beet is an energy crop classified as plant biomass from the agri-food sector. One of the possibilities for the production of bioenergy from lignocellulosic biomass - such as sugar beet byproducts (SBB) - is the production of biogas by the process of anaerobic digestion (AD) [1]–[3]. The production of biogas requires that the different microbiological phases involved in the AD process are perfectly coordinated since an imbalance between them could lead to instability of the whole process. Several limitations have been reported in literature for the bioconversion of agri-food by-products [2], [4]. In this sense, the lignocellulose content of these residues is non-easily biodegradable and, in addition, these wastes are normally deficient in nitrogen with respect to their high carbohydrate content. Therefore, limitations in the performance of the anaerobic digestion process are due to the characteristics and complexity of this type of material. The application of different pretreatments and modification of process variables, such as temperature, are mainly used to increase the specific methane productivity in the AD of lignocellulosic biomass [4]–[6].

The global rate of the anaerobic process is affected by the working temperature. Indeed, the temperature affects the activity and growth rate of the microorganisms, the solubility of the gases and the type of microorganisms involved in each stage of the process. Moreover, small oscillations of temperature can produce great distortions of the process, causing imbalance between the rates of production and consumption of a particular compound, as volatile fatty acids for example [7].

Co-digestion with cow manure can improve the digestion of lignocellulosic residues by balancing the nutrient content in the system and adding microbial population. Thus, cow manure provides nitrogen, alkalinity and active microorganisms, adapted to the degradation of vegetable fibers [3], [8]–[11]. Likewise, lignocellulosic wastes supply carbon, stabilizing the process by mitigating the negative effects of inhibition by excess nitrogen in the system [12], [13].

The present study has been conducted to analyze the start-up and stabilization stages of the AD of sugar beet by-products and its co-digestion with cow manure (CM). Moreover, in both single digestion and co-digestion processes, mesophilic and thermophilic temperatures have been studied

2. Material and methods

2.1 Feedstock

Sugar beet by-products (SBB) used in this study came from the sugar factory (AB Sugar^{TM}) located in Jerez de la Frontera (Cadiz), Spain. The used SBB consisted of pellets of exhausted sugar beet cossettes. CM was collected from an agricultural facility at El Puerto de Santa Maria (Cadiz), Spain. Fresh CM was collected periodically and stored at 4 °C. For the start-up of the co-digestion tests, anaerobic sludge, coming from the WWTP "El Torno", Chiclana de la Frontera (Cadiz), Spain, has been used as inoculum.

2.2 Experimental procedures

The experimental system consisted of four semi-continuous reactors, built in stainless steel, with a working volume of 10 L.. The produced biogas was daily collected and stored in Tedlar® gas bags. The mixing was warranted by a centred vertical axis with stirring blades and the working temperature was controlled in each reactor by water circulation, coming from a thermostatic recirculation bath, through the reactors jackets. Therefore, four semi-continuous reactors, operating at 30 days-HRT, have been used in this study: SD-M; SD-T; CD-M and CD-T.

The Table 1. shows the applied conditions in each reactor.

	Temperature (ºC)	
Single Digestion (SD)	35	55
Co-digestion (CD)	35	55

2.3 Analytical methods

2.3.1 Characterization of the substrate and effluent

For the monitoring and control of the process, the samples were characterized in terms of pH, alkalinity, total solids (TS), volatile solids (VS) and volatile fatty acids (VFAs). All analytical determinations were performed according to the Standard Methods from APHA-AWWA-WPCF [14]. For the pH measurement, the potentiometric technique was used (WPFC-4500-H^+). The alkalinity was determined by the volumetric titration technique(WPCF-2330 method). TS and VS were analyzed by the gravimetric technique (WPFC-2540-B and WPFC-2540-E). On the other hand, for the VFAs analysis, the method described by Alvarez-Gallego was used [15]. Samples were filtrated twice (first 0.47 mm glass microfiber filter and second 0.22 micron Teflon filter) and then analysed by using a gas chromatograph (Shimadzu GC-2010) equipped with a flame ionization detector (FID) and Nukol® filled capillary column (diameter 0.25 µm and 30 m in length). A standard mixture of acetic, propionic, isobutyric, butyric, isovaleric, valeric, isocaproic, caproic and heptanoic acids was used for the calibration of the system (SIGMA-ALDRICH). The total acidity (TVFA) has been calculated as the weighted sum (molecular weight basis) of all individual volatile fatty acids found in the sample. The feed added to the reactors was maintained at a TS content of 8.0% [1]. The mixture used in Aco-D tests was 25:75 (SBB: CM)[16], based on the weight percent of each substrate (Table 2).

The volume of biogas produced was measured daily by using a high precision wet drum-type gas meter (Ritter^{TM} TG5). The gas composition, in terms of CH_4, CO_2, H_2 and O_2, was determined by using a gas analyzer (EMERSON^{TM} X-STREAM).

Table 2. Characteristics of raw substrates (sugar beet and cow manure) and the reactor feeds

	Raw substrates		Reactor Feed for single digestion	Reactor Feed for co-digestion
	SBB	**CM**	SBB	SBB-CM (25:75)

pH	5.6	8.2	5.7	6.1
% TS	85.85	19.62	7.49	7.34
% VS (dry basis)	90.01	71.81	91.04	80.08
sCOD (g O_2/Kg)	31.55	11.30	13.85	12.83
TVFA (gHAc/L)	4.06	6.11	3.21	2.68

3. Results and discussion

3.1 Anaerobic digestion of sugar beet by-product

The hydraulic retention time was 30 days in all the reactors (organic loading rate of 2.46 gVS/$L_{reactor}$·day). The pH was the key parameter to ensure optimal growth of microorganisms in the system. According to Montañés, et al., [17] acetogenic and methanogenic microorganisms require higher pH and their optimum activities are found in the range 7.5-8.5. In this sense, the optimum pH range depends on the temperature, with higher values for thermophilic than mesophilic processes. Therefore, a daily correction of the pH was performed by the addition of alkali (K_2CO_3) maintaining pH values 7.25 in the mesophilic reactor and 7.93 in the thermophilic one, which were found to offer better stability to the two systems. The chemical agent additions were 15 g/day and 20 g/day, respectively (Fig 1). Thus, stable operating conditions were obtained for mesophilic and thermophilic reactors, despite the fact that there was a significant accumulation of VFAs in both cases (Fig. 2). The SBBs used in the work led to low biogas production due to a very high acetic acid accumulation[18] (Fig. 3-4). The inhibition of the AD process by long chain fatty acids (LCFA) [19]–[21] or by propionic acid accumulation have been extensively reported. Indeed, the ratio of propionic to acetic acid has been used as an inhibition indicator of the AD process [1], [5], [18], [19], [22]. However, accumulation of acetic acid in the system is a non-typical behavior of AD process and must be related to inhibition of the acetoclastic methanogens. However, the biogas production remained stable in the reactors although the partial inhibition of methanogenesis occurred, limiting the conversion of acetic acid into CH_4. The average daily methane productions were 0.07 $LCH_4/L_{reactor}$·d and 0.26 $LCH_4/L_{reactor}$·d for mesophilic and thermophilic reactors, respectively. The methane yields obtained were 30 mL CH_4/gVS_{added} for the mesophilic treatment and 110 $mLCH_4/gVS_{added}$ for the thermophilic process (Fig. 5). For the same HRT, the productivity of CH_4 during the start-up process was higher in thermophilic process than in mesophilic one. This may be due to this, that the SMP obtained in this work were lower than that reported by other authors [1], [23], [24]. Suhartini et al., [23] have carried out a study to compare mesophilic and thermophilic anaerobic digestion of sugar beet pulp with a weekly supplementation with trace elements). The study was conducted in semi-continuous reactors operating at OLR of 4 gVS/L·day and 30 days-HTR. The authors obtained specific methane productions of 292 and 345 $mLCH_4$/gVS-day in mesophilic and thermophilic processes, respectively.

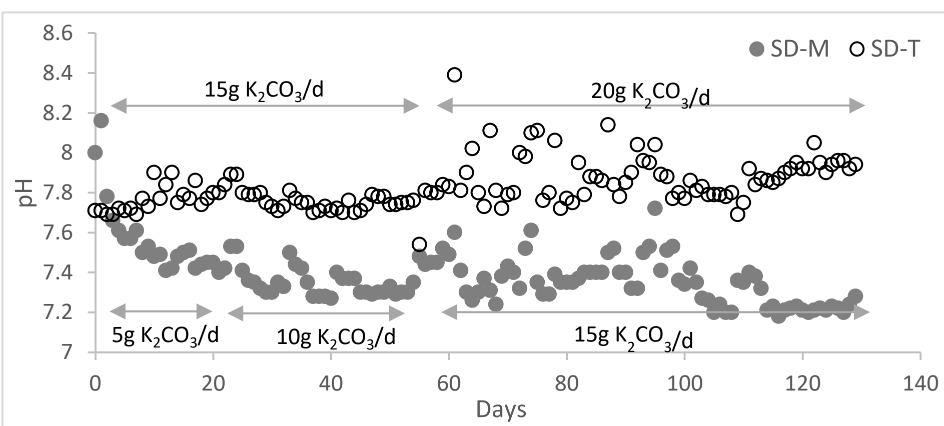

Fig.1 Comparison of pH for SD-M and SD-T

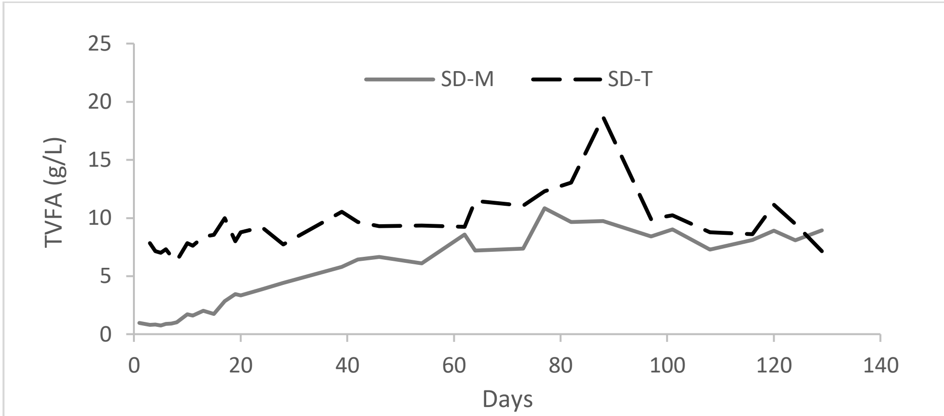

Fig.2 Comparison of TVFA concentrations for SD-M and SD-T

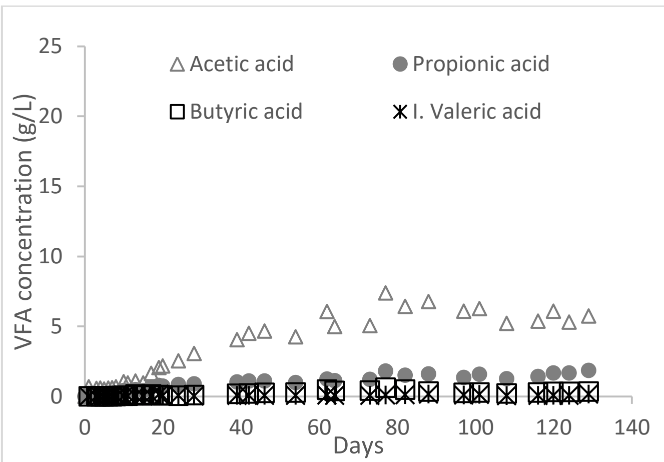

Fig.3 Individual VFAs concentrations in SD-M

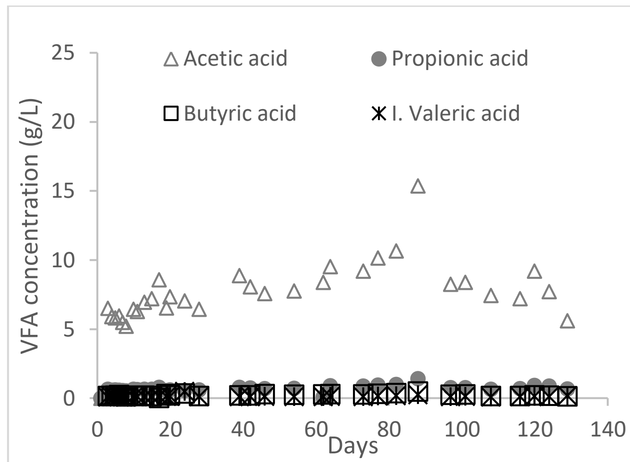

Fig.4 Individual VFAs concentrations in SD-T

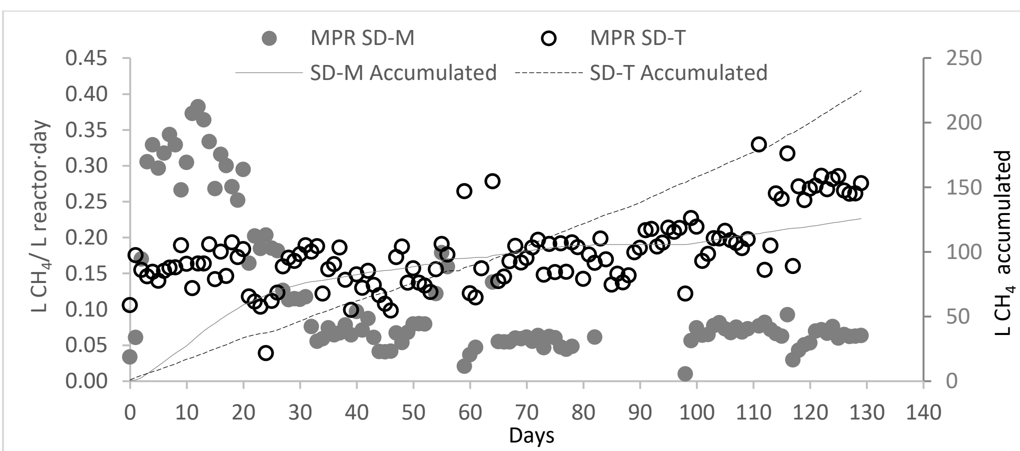

Fig.5 Daily and accumulated methane productions for SD-M and SD-T

Stable biogas productions have been achieved, in both mesophilic and thermophilic conditions, for the treatment of sugar beet by-products, although a very high HRT (30 days) was required because of the accumulation of VFA. It must be taken into account that SBB used in this work present a low alkalinity as a consequence of a change in the production process of the sugar by the supplying factory (exhausted pulp without molasses and a different drying process) and hence, it was necessary a daily alkali addition in order to maintain the stability of the pH.

3.2 Co-digestion of SBB with cow manure

In the co-digestion tests, a better pH stability was observed for both temperature ranges when compared to single AD experiments. In fact, an average addition of the alkali of 2 g/d was required in the mesophilic reactor and 5 g/d in the thermophilic reactor. The average daily methane productions were 0.35 $LCH_4/L_{reactor}$·day and 0.36 $LCH_4/L_{reactor}$·day for mesophilic and thermophilic reactors, respectively (Fig.6). The methane yields obtained were 178.62 mL CH_4/gVS_{added} for the mesophilic reactor and 182.44 $mLCH_4/gVS_{added}$ for the thermophilic one (Table 2). Compared to the single AD process, it was observed a clear decrease in the total acidity of the system, leading to an increase in the specific productivity of CH_4 of 80% and 28% for mesophilic and thermophilic processes, respectively. For the individual AD of sugar beet by-products, in mesophilic and thermophilic ranges, TVFA concentrations of 8.09 gHAc/L and 9.09 gHAc/L were obtained, respectively. However, in co-digestion assays, the maximum value of the total acidity obtained was 1.01 gHAc/L in the thermophilic range (Fig.7). Acetic and propionic acids are the dominant VFA in co-digestion. As can be seen in Fig. 8 and 9, the acetic acid production is lower than in single AD. This fact is indicative of the stabilizing effect of cow manure in the co-digestion process. Other authors have reported specific methane productivities of 242.59 ml/gVS_{added} for the co-digestion of sugar beet by-products with cow manure, working at 20 day-HTR in the mesophilic temperature range [1]. Cheng Fang et al. [25], have obtained a SMP of 260 mL/gVS_{added} with an OLR of 2,95 $gVS/L_{reactor}$·d and using 15:85 ratio (SBP:CM). The specific characteristics of the SBB used in this study seem to be behind of the low methane productivities obtained in this study. In the co-digestion of dairy manure (DM) with three crop straw residues (SRs) for CH_4 production, Li et al.[26], have reported low yields of methane (less than 100 mL/d) for a 9:1 ratio (SRs : DM) working in mesophilic conditions and maintaining a TS content of 8%.

Table 2. Comparison of the main results obtained in the mesophilic and thermophilic individual AD of SBB and Aco-D with cow manure

	Mesophilic		**Thermophilic**	
	AD	**Aco-D**	**AD**	**Aco-D**
HRT (days)	30	30	30	30
OLR (gVS/Lreactor·day)	2.46	2.00	2.46	2.00
pH	7.25	7.52	7.93	7.97
MPR ($LCH_4/L_{reactor}$·day)	0.07	0.35	0.26	0.36
SMP ($mLCH_4$/gVSadded)	30.0	178.6	110.0	182.4
TVFA (gHAc/L)	8.09	0.19	9.09	1.02

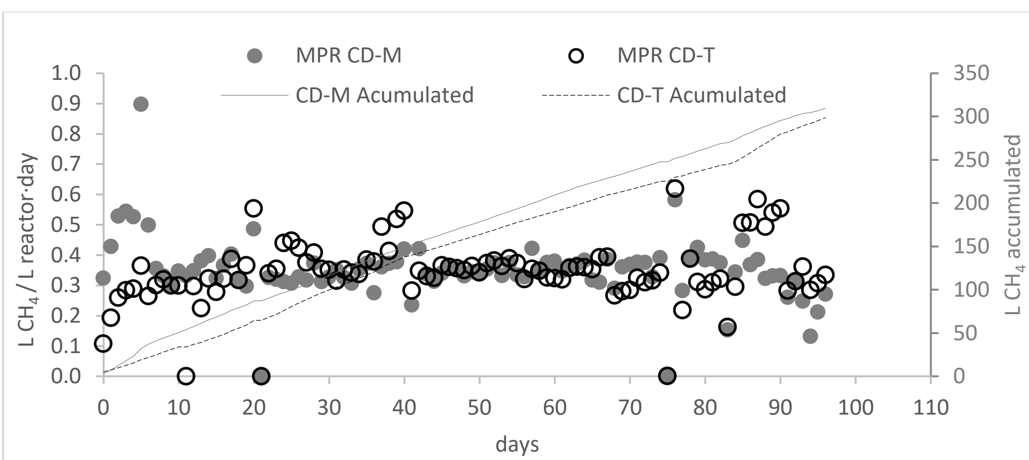

Fig.6 Comparison of daily and accumulated methane productions for CD-M and CD-T

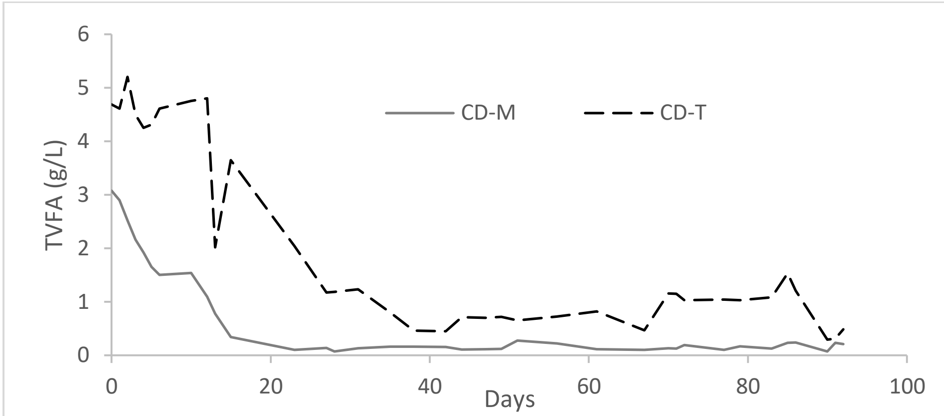

Fig.7 Comparison of TVFA for CD-M and CD-T

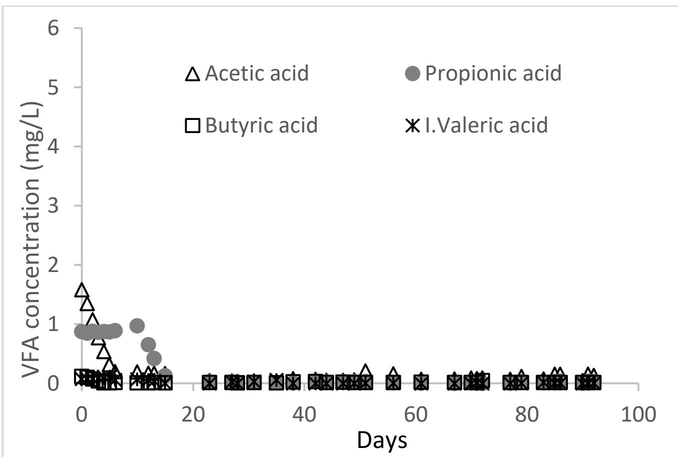

Fig.8 Individual VFA concentrations in CD-M

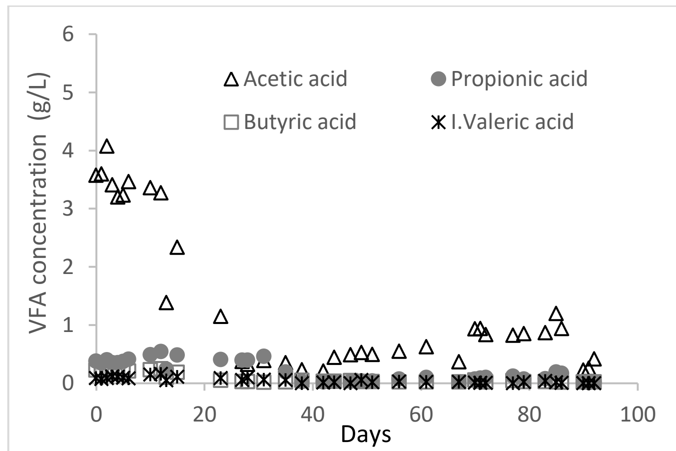

Fig.9 Individual VFA concentrations in CD-T

4. Conclusions

The comparison of the AD of SBB and its co-digestion with CM performed in the two temperature ranges (thermophilic and mesophilic) indicates that the productivity of methane is higher in thermophilic process than in the mesophilic one. Moreover, it can be observed that co-digestion process increases the methane production rate due to the stabilization of the process. The addition of cow manure have supplied alkalinity, nitrogen sources and active microorganisms, improving the degradation of the VFAs and avoiding the inhibition of acetoclastic methanogenic archaea.

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