BIOMASS AND BIOENERGY 56 (2013) 545–554

Available online at www.sciencedirect.com

SciVerse ScienceDirect

http://www.elsevier.com/locate/biombioe

# Anaerobic digestion of maize and cellulose under thermophilic and mesophilic conditions – A comparative study

*Katarzyna Golkowska**, *Manfred Greger*

*University of Luxembourg, Faculty of Science, Technology, and Communication, 6, rue Richard Coudenhove-Kalergi, 1359 Luxembourg, Luxembourg*

ARTICLE INFO

*Article history:*
Received 12 September 2011
Received in revised form
18 April 2013
Accepted 28 May 2013
Available online 4 July 2013

*Keywords:*
Anaerobic digestion
Cellulose
Maize silage
Thermophilic
Mesophilic
Batch

ABSTRACT

The aim of this study was to advance in understanding of digestion process of energy crops. Cellulose and maize silage were fermented in batch mode at mesophilic (38 °C) and thermophilic (55 °C) conditions and corresponding organic loads of 5.5 ± 0.2 kgVS/m$^3$, 11.2 ± 0.3 kgVS/m$^3$ and 16.7 ± 0.4 kgVS/m$^3$.

For both substrates more stable and faster digestion took place at 38 °C. Due to complex structure maize degradation was characterized by varying digestion rate and longer total digestion time resulting form breakdown of hard-degradable fractions. The digestion retard at increased OLRs of cellulose and lower degradation level obtained for all cellulose series confirm a higher overloading potential for systems dealing with single-component-substrates but also the enhanced sensitivity of such systems to any inconvenient digestion conditions.

Based on observed patterns of volatile fatty acids and oxidation-reduction potential, different fermentation mechanisms can be concluded for cellulose and maize, but also for different temperature modes. Conversion of maize at highly reductive conditions with increased concentrations of butyric acid was accompanied by much higher activity of hydrogenotrophic methanogens than for cellulose digestion.

Two factors showed a strong potential to influence test results: an insufficient VS content of inoculum, which caused reduced biogas yields, and a high natural biodiversity of maize silage, resulting in higher biogas yields than calculated based on the maize composition.



## 1. Introduction

The global energy demand is forecasted to grow by more than one-third until 2035. However the energy demand of OECD and thus of most European countries is barely anticipated to rise. In the coming years a shift from oil, coal (and nuclear) towards natural gas and renewable energy sources is expected on the European energy market [1]. Such development has been promoted by the Renewables Directive of the EU [2] and in some countries (e.g. Germany or Switzerland) additionally triggered by the Fukushima Daiichi nuclear disaster from 2011. For that reason the enhanced political and financial

*Abbreviations:* DS, dry solids; $DS_K$, corrected dry solids; $DS_N$, uncorrected dry solids; FM, fresh mass; HAc, acetic acid; iso-HBu, iso-butyric acid; n-HBu, n-butyric acid; HPr, propionic acid; iso-HVa, iso-valeric acid; n-HVa, n-valeric acid; OL, organic load; OLR, organic loading rate; ORP, oxidation-reduction potential; sGP, specific biogas production; sGPR, specific biogas production rate; VFA, volatile fatty acids; VS, volatile solids; $VS_K$, corrected volatile solids; $VS_N$, uncorrected volatile solids.

* *Corresponding author*. Present address: Public Research Centre Henri Tudor, Resource Centre for Environmental Technologies, 6A, avenue des Hauts-Fourneaux, L-4362 Esch-sur-Alzette, Luxembourg. Tel.: +352 42 59 91 4433; fax: +352 42 59 91 555.

E-mail addresses: katarzyna.golkowska@tudor.lu (K. Golkowska), manfred.greger@uni.lu (M. Greger).

0961-9534/$ – see front matter 

http://dx.doi.org/10.1016/j.biombioe.2013.05.029

support for renewable energy and in particular for biogas industry has been provided in many European countries. As a result the annual primary biogas production in the EU increased from 8.4 Mtoe in 2009 to 10.1 Mtoe in 2012 [3,4]. In particular the fermentation and co-fermentation of energy crops has developed rapidly in recent time. Use of agricultural feedstock and waste allows constructing more compact and therefore more efficient installations and improving performance of the biogas plants digesting mainly manure. The new energy approach shifts the main focus of the agricultural biogas plants from disposing of manure to energy production. This change has an additional social dimension: a traditional farmer can become energy producer and has a possibility to profit from the new developments on the energy market.

The predominant number of biogas plants nowadays is run mesophilic (36–38 °C) [5], while thermophilic digestion mode (55 °C) is widespread mainly in Scandinavian countries. Most of the findings related to anaerobic digestion are based on the experiments conducted with waste water (e.g. [6–9]), manure (e.g. [10–12]) and solid waste (e.g. [13–15]) or refer to co-digestion of these substrates (e.g. [16–19]). Though, not many papers have been published on mono-digestion of energy crops by now [20–28]. Similar to the industrial trends, mesophilic fermentation was better described in former publications, while thermophilic digestion has been mainly a subject of more recent studies. However, hardly any publications comparing experiments conducted in the similar way under thermophilic and mesophilic conditions can be found.

Originally it was assumed that the degradation pathways and the performance of process parameters for all types of anaerobic digestion must be similar to that observed for digestion of waste water and manure, independent of the substrate, temperature mode or the source of the inoculum. The methane was believed to be mainly produced via propionic acid (HPr) and acetic acid (HAc) [29–33]. The last findings reveal that this is not always the case [24,34,35]. Many aspects of anaerobic digestion of energy crops, such as impact of micronutrients, the exact microbiological composition of bacterial biocenosis, or degradation pathways, have not been researched by now and together with advanced modeling constitute the core of the current research.

The aim of this study was to advance in understanding of digestion process of energy crops. Since most of the experiments concentrate on one operating temperature, no studies comparing performance of the bacterial biocenosis for different temperatures and (OLs) loads under similar experimental conditions can be found. This study investigating mono-fermentation of a model substrate (cellulose) and agricultural substrate (maize silage) under mesophilic and thermophilic temperature conditions should help to close this research gap.

## 2. Material and methods

### 2.1. *Inoculum*

Inoculum for thermophilic (55 °C) tests was obtained from continuously operated 50-l thermophilic plug-flow research fermenter fed with maize and grass silage mix at organic loading rate (OLR) of 1.5 kgVS/($m^3$ d). The uptime of the reactor before inoculum sampling amounted to one year. Mesophilic inoculum (38 °C) was provided by an industrial biogas plant from Beckerich (Luxembourg). This unit consists of three CSTRs with the working volume of 1500 $m^3$ each, which are under operation since 2004. The reactors are run at OLR of 2.5 kgVS/($m^3$ d) and fed with manure (25,000 tonnes/a), maize silage (5400 tonnes/a), grass silage (1000 tonnes/a) as well as corn whole-crop-silage (800 tonnes/a).

Both inocula were prepared following the guidelines for the fermentation of organic matter [36]. Before the beginning of the first experiments both inocula were adapted to cellulose or maize respectively. The adaptation process was conducted in the similar way as the later experiments. The inoculum was placed in fermenters fed with maize or cellulose at OLs of 4–5 kgVS/$m^3$. After digestion period of *ca.* 2 weeks (or shorter if the biogas production ceased) the feeding process was repeated and the digestion continued for 2–6 weeks to remove the residual degradable components. The inocula from all thermophilic and mesophilic reactors were remixed (separately for each temperature mode) to receive homogenous start conditions and filtered through a kitchen strainer (2 mm mesh size). The homogenous filtrate was used to inoculate all reactors of the same experimental series. The subsequent batch series (with increasing OLs) were performed with the inoculum retrieved from the previous experiment to achieve better bacterial adaptation to increased OLs. The step including inoculum homogenization and filtration was repeated before every new OL was investigated. The volatile solids (VS) content of inoculum ranged between 1.75 and 2.71% of fresh mass (FM) and 54–60% of dry solids (DS), except for thermophilic test with maize at 5.7 kgVS/$m^3$, in which the VS reached 0.59% of FM and 59% of DS. The substrate to inoculum VS ratios measured for all the experimental series are given in Table 1.

The initial pH of the inoculum in different experimental series, together with the final pH values reached after the digestion, are summarized in Table 1. For all trials the pH values ranged from neutral to slightly alkali conditions (6.98–8.60).

### 2.2. *Substrate*

Microcrystalline cellulose powder of pharmaceutical grade was purchased from Euro OTC Pharma GmbH (Bönen, Germany). For maize degradation commercial maize silages of two harvests were applied: MZ I – for thermophilic batches, MZ II – for mesophilic batches. The ensilaged maize was cut into ~5 mm fibers, stored frozen and defrosted at low temperatures (4 °C) for 24 h before charging of the fermenters. Both silages were characterized by Van Soest and Weende analysis [37,38]. Further calculation of corrected DS content ($DS_K$) of the substrate was done according to [39]. The DS content was increased by the amount of volatile compounds lost through volatilization during DS determination according to Eq. (1), where: $DS_N$–measured DS value and $DS_K$–corrected DS value.

$$DS_K[\%] = 2.22 + 0.960\ DS_N[\%] \quad (1)$$

The compositions of cellulose and maize silages corrected according to Eq. (1) are presented in Table 2. Lignin was considered as a non-degradable component of maize. Therefore the total theoretical biogas yield was calculated according to [40] based on the substrate composition but excluding the