A simple method to evaluate the short-term biogas yield in anaerobic codigestion of WAS and organic wastes

D. Scaglione, S. Caffaz, E. Ficara, F. Malpei and C. Lubello

ABSTRACT

The present study was aimed at setting and applying a procedure to measure the anaerobic degradability of different organic substrates by short-term tests (2–7 days) carried out at lab-scale with a low food to biomass (F/M) ratio. All tests were carried out using an acclimated sludge taken from a pilot-plant anaerobic digester (200 L). Trials were performed with a manometric system.

The experimental reliability of the device in measuring the anaerobic degradability was assessed by several preliminary tests carried out using acetate and glucose as reference substrates. The average conversion to methane was 99% for acetate and of 83% for glucose. The results of tests in triplicate showed the high repeatability of the method with an average coefficient of variation lower than 2%.

Then, the lab-scale procedure was applied to study the short-term anaerobic degradability of complex organic substrates: thickened waste activated sludge, two kinds of organic fraction of municipal solid waste (a kitchen waste and a fruit and vegetable waste collected at the wholesale market of Florence), olive mill wastewater and freshly harvested grass. Results indicated that organic fraction of municipal solid waste, olive mill wastewater and grass were characterized by a much higher anaerobic degradability if compared to the thickened activated sludge, well in agreement with literature data.

Key words | anaerobic biodegradability, anaerobic codigestion, batch tests, biogas yield, organic wastes, waste activate sludge

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INTRODUCTION

In Italy, anaerobic digesters for sludge stabilisation are often under-loaded and their performance in term of biogas production, is poor. Co-digestion with suitable organic substrates, such as the source-sorted organic fraction of municipal solid wastes, food wastes, farm wastes, septage and slaughterhouse wastewaters, had been proven to improve biogas production and several successful full scale experiences are now operating in Italy.

In the Florence region, available candidate co-substrates are the organic fraction of municipal solid wastes and olive mill wastewaters. The OFMSW collected in Florence (about 42,000 t/y) is currently composted; however, high treatment costs as well as the limited marketability of the resulting compost suggest investigating alternative solutions. According to literature data, the biogas yield of OFMSW ranges between 0.20 and 0.50 Nl_{CH_4}/gVS_{fed} (Alatriste-Mondragón *et al.* 2006; Castillo *et al.* 2006; Hartmann & Ahring 2006; Davidsson *et al.* 2007; Zhang *et al.* 2007), with a methane content in the biogas varying from 55 to 73%. Food market wastes and fruit and vegetable wastes could reach a biogas yield as high as 0.70 Nl_{biogas}/gVS_{fed} (Hartmann & Ahring 2006). Literature

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studies on anaerobic digestion on OMW reported methane yield values ranging between 0.13 and 0.24 Nl_{CH4}/g_{CODfed} (Borja *et al.* 2003; Rincon *et al.* 2006). Less common but with high potential, is the degradation of grass coming from household gardening and landscape management. In previous studies on several grass species, biogas yield of 0.65–0.86 m^3/kgSV (Mähnert *et al.* 2005) and 0.28–0.55 m^3/kgSV (Prochnow *et al.* 2005) were obtained.

Tests to assess the biogas yield or the biomethanisation potential (BMP) consist in following the anaerobic biodegradation of the organic substrate under study by means of the assessment of biodegradation-related parameters. Proposed methods are based on the monitoring of the rate of either substrate consumption or product formation. However, the majority of established methods relay on measuring biogas production. This is done by liquid displacement at constant pressure (volumetric systems), by pressure measurement at constant volume (manometric system) or by gas chromatography. Other studied methods focusing on product formation are based on set-point titration (Rozzi *et al.* 2001) and calorimetry (Jolicoeur *et al.* 1998). Different standardized methods for anaerobic biodegradability testing have been proposed (Müller *et al.* 2004) and various commercial equipments are now available on the market (Figure 1).

Figure 1 | Picture of instrumental equipment.

Because of the availability of simple, automated and cheap laboratory equipments for pressure monitoring, manometric methods appear especially convenient and has been proposed to evaluate anaerobic sludge characteristics (Fdz-Polanco *et al.* 2005) and chemicals toxicity versus anaerobic bacteria (Fdz-Polanco *et al.* 2006).

The main objective of this study was to assess the feasibility to implement short term batch tests (4–7 days) to get a rough estimation of the anaerobic digestibility of candidate organic substrates in terms of short-term biogas production and methane potential (stBMP). To assess the biogas production a commercial manometric system was applied (the OxiTop Control® system), whose reliability was preliminary assessed.

MATERIALS AND METHODS

Trials were performed with the manometric OxiTop Control® system. This is a manometric device consisting of a glass bottle (1,140 ml) provided with a pressure transducer located in a measuring head Two lateral openings, sealed by a rubber septum and by a teflon airtight valve, are used for substrate injections and for biogas discharge, respectively. During anaerobic tests, bottles are kept in an incubator at 35°C and mixed by a magnetic stirrer; the overpressure due to biogas accumulation in the headspace is automatically registered by the measuring heads. Taking into account the headspace volume and the incubation temperature, the ideal gas equation of state allows the calculation of cumulative biogas volumes from overpressure data. At the end of each test, the methane and CO_2 content in the headspace were assessed by means of gas-chromatographic analysis (Agilent 3,000 micro gas-chromatograph).

All tests were carried out using an acclimated sludge taken from a pilot-scale anaerobic digester (200 L) that was operated in a fed-batch mode (Caffaz *et al.* 2005). During the start-up period, the pilot-plant was fed with thickened activated sludge; later two co-digestion phases were started.

Each vessel was filled with 100–300 ml of anaerobic sludge, so that a sufficient headspace volume was provided not to exceed the maximum pressure value of 1,350 hPa. The volume of the organic substrate was generally negligible (1–20 ml). Before starting, the vessel headspace was flushed

with N_2. The initial headspace pressure was set at the atmospheric value by discharging the overpressure in a water bottle. A phosphate buffer maintained the pH value within the optimal range (6.8–7.2) during all trials. A control test with the sludge inoculum only was always performed. The net biogas production was obtained by difference between the biogas measured in the vessel with added substrate and the control one. Maximum and average gas production rates (GPR), biogas and methane yields, and mass balances were calculated for each test.

Analytical measurements before and after every batch test were carried out in order to assess the process efficiency and calculate the mass balance. N-NH_4^+, total COD, soluble COD were measured by Hach-Lange spectrophotometric methods, TS and VS were measured according to the *Standard Methods* (APHA *et al.* 2005). The efficacy of CO_2 absorption on NaOH pellets located in the bottle headspace was tested; however, it was proved not to be reliable since CO_2 gascromatographic determinations evidenced the presence of a residual CO_2 content at the end of the trials probably caused by slow absorbtion dynamics. For this reason, CO_2 traps were not applied.

During each test, the ratio between the amount of COD removed and VS removed (ΔCOD/ΔVS) was calculated; the ΔCOD was obtained both by analytical measures and by considering the COD transferred to the gas phase as methane.

The experimental reliability of the device in measuring the anaerobic degradability was assessed by several preliminary tests carried out using acetate (CH_3COONa) and glucose as substrates with a F/M value of 0.01–0.05 mgCOD/mgVS. The concentration of the solutions used for the injection were of 200 mgCOD/mL.

Then, the lab-scale procedure was applied to study the short-term anaerobic degradability of complex organic substrates: thickened waste activated sludge (TAS), two kinds of the OFMSW (a kitchen waste (KW) and a fruit and vegetable waste (FVW), collected at a wholesale market of Florence), olive mill wastewater (OMW) and freshly harvested grass (FG). Before testing, the OFMSW samples were shredded and homogenized by a kitchen mechanical mixer. The OMW came from one of the largest three-phase mill of Italy, located in Quarrata (Pistoia); OMS samples were pre-treated by means of centrifugation (4,000 rpm for 10 min). The FG came from household gardening (cut in May–June) and belong to the common kind of grass *Festuca*. Organic substrate characteristics are summarised in Table 1.

The liquid phase of the kitchen waste was obtained by means of centrifugation (15 minutes, 3,000 rpm) of the shredded waste (C-KW).

RESULTS AND DISCUSSION

Validation tests

To evaluate the repeatability of the method 7 tests in triplicate were conducted with similar samples of sludge inoculum. The average coefficient of variation was of 1.8% ± 0.8% (GPR) demonstrating the good repeatability.

Then, a set of experiments were conducted aimed at verifying the reliability of the measuring system in assessing biogas production and, from the latter, the biodegradability of the tested organic substrate under anaerobic conditions, or 'biomethanization'. As test substrates, simple and rapidly biodegradable molecules (glucose and acetate) were selected for which reference values for their average biomethanization were available. A typical cumulated biogas production over time during such tests is depicted in Figure 2. After approximately 24 h of endogenous

Table 1 | Characteristics of the experimented organic substrates

Substrate		COD [g/l]	TS [%]	VS/TS [%]	No of samples
OFMSW	KW		27.8–31.7	93.9–96.1	7
	FVW		8.5–15.8	83.6–89.6	6
	C-KW	96			2
OMW	Centrifuged	52.0–91.2	3.1–4.7	89.0–91.0	4
FG			31.5–34	82.1–89.0	5
TAS			2.6–4.9	61.9–70.9	26

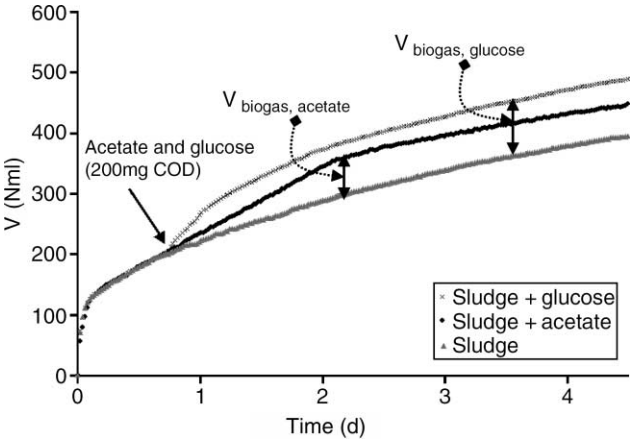

Figure 2 | Biogas production measured after acetate and glucose addition in a lab-scale test with a low F/M ratio.

degradation, substrate was injected, resulting in a clear change in the rate of biogas production. Once the biodegradation process appeared to be completed, a sample of biogas was analysed for its methane partial pressure (p_{CH4}). The fraction of the added COD (COD_{added}) converted into methane was computed from the net volume of biogas (V_{biogas}), according to the following equation:

$$\text{Biomethanization [\%]} = \frac{V_{biogas}[NL_{biogas}] \cdot p_{CH_4}[NL_{CH_4}/NL_{biogas}]}{COD_{added}[g] \cdot 0.35[NL_{CH_4}/gCOD]} \cdot 100$$

Results of these tests are summarised in Table 2. The average biomethanization was found to be 99% ± 8% for acetate and of 83% ± 0.7% for glucose. As for acetate, its full biomethanization is well in agreement with the low biomass growth yield of acetoclastic methanogens (e.g. 5% $gCOD_{biomass}/gCOD_{substrate}$, as assumed by the ADM1 model; Batstone *et al.* 2002). As for glucose, it has to be previously converted into acetate and hydrogen to be finally available for methanogens; therefore, a higher fraction of its COD is taken for growth by the numerous bacteria groups involved. The average biomethanisation measured in this experimentation is in accordance with what expected from stoichiometric reactions (Zoetemeyer *et al.* 1982). The tested experimental procedure was found to be well repeatable, as indicated by the very low coefficient of variation (8% and 0.7% for acetate and glucose, respectively).

Table 2 | Experimental results of reliability tests

Substrate	COD_{added} [mg]	p_{CH_4}	Biomethanization [%] (mean ± standard deviation)	No of repetitions
Acetate	200	0.76 ± 0.02	99 ± 8	5
glucose	200	0.57 ± 0.005	83 ± 0.6	5

Moreover, a moving window linear regression allowed the biogas production rate (BPR) to be computed from the cumulated biogas production data. The specific gas production (sBPR) was calculated by referring the BPR to the volatile solid content of the sludge sample.

When acetate is degraded, a constant biogas composition can be reasonably assumed, allowing the estimation of the specific methanogenic activity of the sludge (SMA). Typical trends of the net SMA (subtracting methane endogenous production) during the degradation of acetate and glucose are depicted in Figure 3.

The average SMA under endogenous conditions was found to be 5 $NmL_{CH4}/g_{VS}/d$, while it increased to 15 $NmL_{CH4}/g_{VS}/d$ during acetate degradation. Both SMA values are quite low as compared to literature values (e.g. 28–35 $NmL_{CH4}/g_{VSS}/d$ as typical range for SMA of digested sludges fed on agro-wastes; Raposo *et al.* 2006). This may be due to the low acetate concentration (0.25–0.5 gCOD/L) if compared with the one normally adopted in SMA tests (1–2 gCOD/L; Colleran & Pender 2002) and to the low and poorly degradable organic load of the pilot anaerobic digester from which the inoculum was taken. As depicted in Figure 4, experimental net SMA data could be well fitted by a Haldane model:

$$SMA = SMA_{max} \frac{[COD]}{[COD] + k_S + [COD]^2/k_I}$$

with:

$SMA_{max} = 220\ NmL_{CH4}/g_{VS}/d$;
$k_S = 2.9$ gCOD/L;
$k_I = 0.14$ gCOD/L.

As for glucose, a simple model interpretation is not possible, for the simultaneous occurrence of various processes. A comprehensive model, such as the well known ADM1 (Batstone *et al.* 2002) has to be applied to this purpose, that is beyond the scope of this paper.

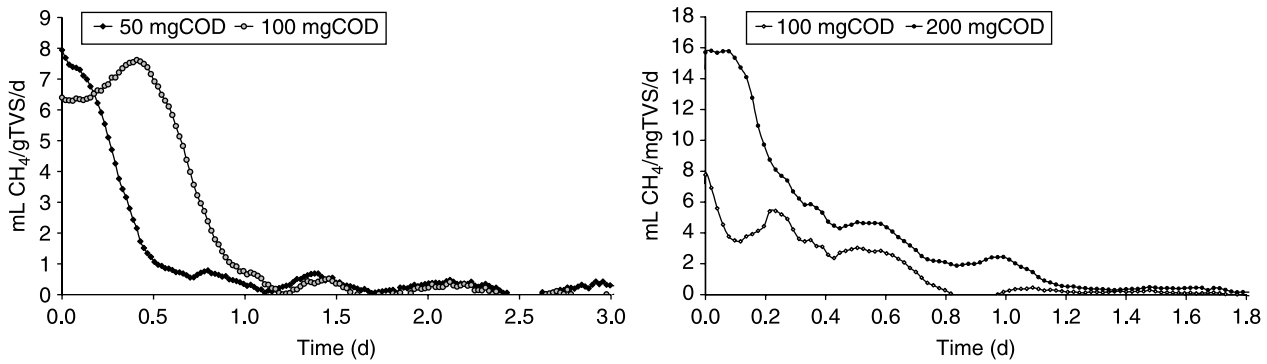

Figure 3 | Net SMA trends in acetate (on the left) and glucose (on the right) methanization tests.

However, the most relevant conclusion that can be drawn from these results is the reliability and reproducibility of the experimental procedures, which suggests the feasibility to extent this method to the study of more complex substrates.

Application to complex organic wastes

Figure 5 shows the specific biogas production (NLbiogas/gVSfed) obtained for some organic feedstocks used in this experimental study. Biogas yields for OFMSW were well in accordance with values obtained under laboratory (Einola *et al.* 2001), and full scale conditions (Edelmann *et al.* 2000; Bolzonella *et al.* 2006). Results indicated that OFMSW, OMW and grass were characterized by a higher anaerobic degradability if compared to the thickened sludge (TAS). Codigestion with a mix of TAS and OFMSW (fractions in terms of TVS, Figure 5), increased the biogas yield up to 0.4 NL/gVS (Table 3).

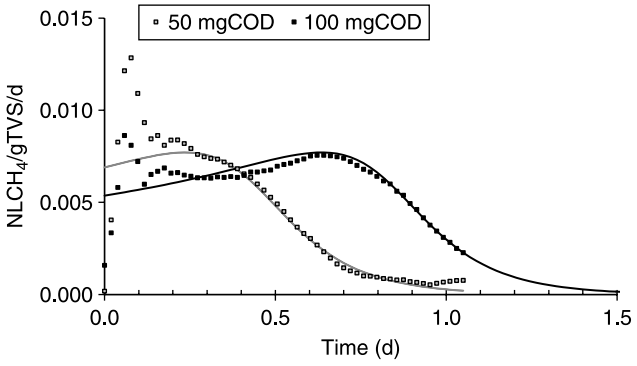

Figure 4 | Experimental SMA on acetate fitted by a Haldane kinetics model.

Biodegradability tests on fresh grass samples were performed at increasing F/M and thus at increasing initial substrate concentration; the corresponding net biogas production is depicted in Figure 6 (on the left). Short term biogas yield resulted to be $0.37 \pm 0.07\ NL_{biogas}/g_{SVin}$ and are below the literature range for similar organic wastes (e.g. 0.65–0.86 m^3/kgSV, Mähnert *et al.* 2005). The limited bioconversion could be reasonably due to the limited degradation time, within which only the rapidly hydrolysable fraction of the whole biodegradable organic matter would have been effectively converted. This statement is supported by the fact that, at the end of the test, the GPR was still relevant and constant, indicating the presence of a residual biodegradable organic matter and it is also in agreement with the generally accepted idea that the degradation rate of crops and grass residues is limited by the first disintegration and hydrolysis phases. The complex shape of the cumulative gas production reflects the

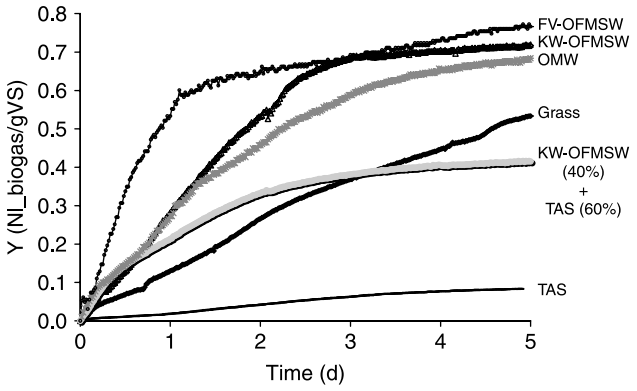

Figure 5 | Example of specific biogas production registered during batch tests on various organic substrates.

Table 3 | Experimental results of lab-scale tests on the different organic substrates in Figure 5

Parameter	Unit	TAS	FV	KW	Grass	OMW
TS	[%]	5.3	12	29.3	34	4.7
VS/TS	[%]	67	88	96	89	90
F/M	[gCOD/gCOD]	0.55	0.07	0.08	0.09	0.03
St-biogas yield	[NLbiogas/gVS_{fed}]	0.08	0.79	0.72	0.55	0.69
CH_4/biogas	[%]	65	60	62	58	64
StBMP	[$NLCH_4/gVS_{fed}$]	0.05	0.47	0.45	0.32	0.44

complexity of the substrate, possibly including fractions characterised by different rate of hydrolysis. By plotting the average GPR versus the initial substrate concentration for the three tests (Figure 6, on the right), a linear trend can be observed, suggesting that a first order degradation kinetic is taking place, with a kinetic constant of 0.044 NLbiogas/gSVin/d. The average methane partial pressure in the biogas, as measured at the end of the test, was 63 ± 3%.

The stBMP of the centrate of the organic fraction of the municipal solid wastes was found to be 0.31 $NL_{CH4}/gCOD_{in}$, corresponding to an overall biomethanization, within the 1-week test, of 88%. The methane partial pressure in the evolved biogas was 64%. The trend in the biogas production (Figure 7, the left) showed a clear bending point at 1.3 d, when, most likely, the soluble and already acidified fraction of the whole COD was fully converted. The slower biogas production measured later on is possibly limited by the

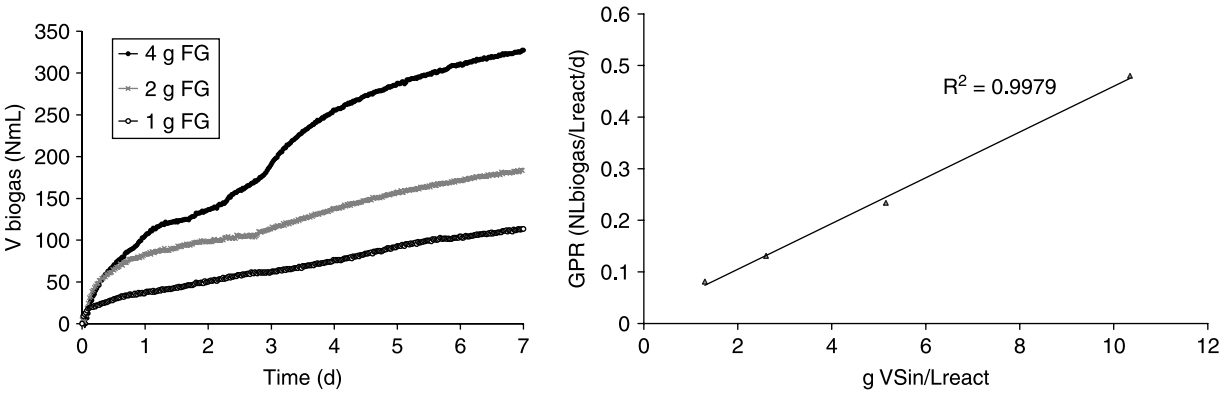

Figure 6 | Net biogas production in test with increasing doses of grass (left), Average gas production rate (GPR) in test with increasing doses of grass (right).

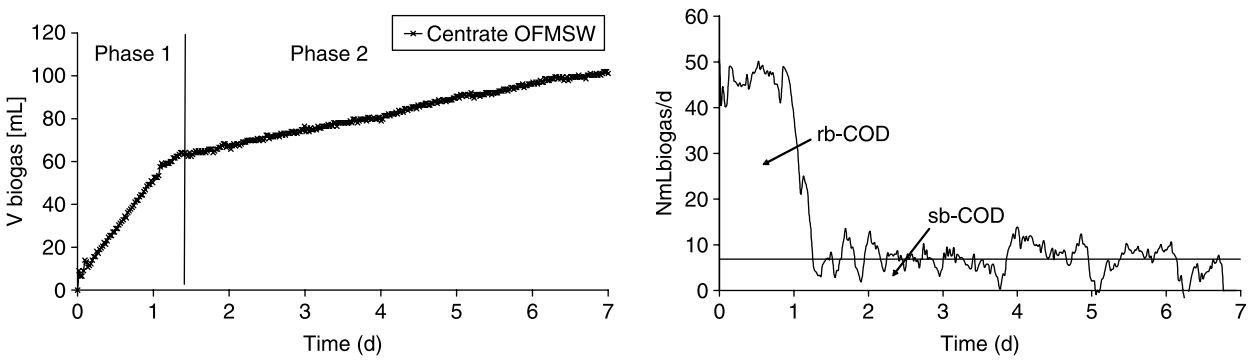

Figure 7 | left Net biogas production in test with 2 ml of C-KW (left), net biogas production rate in test with C-KW (right).

acidogenic phase of complex soluble components (sugars, proteins and lipids). The fraction of rapidly degradable COD in the centrate can be computed by subtracting the constant biogas production estimated after 36 h to the one measured during the initial high-rate degradation (Figure 7, on the right), thus spotting the biogas assigned to the rapidly degradable fraction. By assuming a constant biogas composition equal to the final one, a 48% of the centrate COD resulted to be biomethanised within 1.3 d.

CONCLUSIONS

The present study was aimed at setting up and testing an experimental procedure to evaluate the anaerobic degradability of complex substrates by short term (4–7 days) batch tests using the OxiTop Control manometric system.

Preliminary tests were carried using acetate and glucose as reference substrate to check the reliability of the system. The average anaerobic degradability was 99% for acetate and of 83% for glucose, well in accordance with what expected from stoichiometric reactions. Results of many triplicate tests confirm the high repeatability of the method.

Results of short-term batch tests (performed with low F/M value to shorten the digestion time) on several organic substrates (thickened activated sludge, olive mills wastewaters, kitchen wastes, fruit and vegetable wastes, fresh grass) were in good agreement with literature data.

We can conclude that the proposed lab-scale procedure, allows operators to obtain, in 4–7 days, a rough indication of substrate anaerobic biodegradability achievable under real operating conditions.

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