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Thermophilic anaerobic co-digestion of separated solids from acidified dairy cow manure

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ABSTRACT

This study examined the potential for partly substituting dairy cow manure (DCM) with solids from solid to liquid separation of acidified dairy cow manure (SFDCM) during thermophilic anaerobic digestion. Three different substituting levels with a maximum of 30% substitution were tested. All digesters substituting DCM with SFDCM showed a stable biogas production with low volatile fatty acid concentrations after a short transition period. An increased methane yield in terms of digester volume compared to DCM alone was obtained with increasing amount of SFDCM and about 50% more methane was achieved when 30% of DCM was substituted with SFDCM. The digestates were subsequently digested in a post digestion, during which the methane yield increased proportionally with increasing amounts of SFDCM. It can be concluded that SFDCM is a suitable biomass for co-digestion and can be used to increase methane yield in terms of digester volume at ratios up to at least 30%.

1. Introduction

Treating animal manure by anaerobic digestion (AD) is of increasing interest since it has the simultaneous advantages of preventing greenhouse gas emission and producing renewable energy (Sommer et al., 2004). However, in many commercial digestion plants the biogas yield from animal manure is often too low to be economically feasible. Animal manure is often having too high water content and is also low in easily degradable carbon to produce methane (Hamelin et al., 2011). A method to increase methane yield in term of volume from animal slurry is by increasing the volatile solid (VS) concentration by substituting the liquid manure with the solid fraction of manure from solid to liquid separation (Møller et al., 2007a; Hamelin et al., 2011). Other than increasing biogas production per unit mass, this strategy can provide other advantages such as reducing transport cost of slurry from farms to centralized biogas plants, reducing the thermal demand to maintain digester temperature (Asam et al., 2011), increasing the value of digested material as fertilizer (Kaparaju and Rintala, 2008), and avoiding the use of crops as co-substrate leading to competition of land use between energy crops and food production (Searchinger et al., 2008).

Currently, a new manure acidification technology to reduce ammonia emission from slurry has been developed in Denmark. The acidification process takes place in a treatment tank by controlled application of concentrated sulphuric acid (96% H_2SO_4) to reach pH 5.5 (Eriksen et al., 2008). Because of the addition of concentrated sulphuric acid, acidified manure has a very high sulphate content and low pH value, therefore utilization of acidified manure as substrate for AD might cause process failure. During an AD process, sulphate is reduced to sulphide by the sulphate reducing bacteria (SRB) (Gerardi, 2003). The sulphate inhibition takes place in two phases (Chen et al., 2008); the first being the competition of SRB with other microorganisms such as methanogens, acetogens or fermentative microorganisms for acetate, H_2, propionate and butyrate (Colleran et al., 1995). Secondly, the inhibition comes from the toxicity of sulphide to some microorganisms (Colleran et al., 1998). In addition, dissolved sulphide in the liquid phase of AD is mainly in the form of un-ionized H_2S and as HS^- (Hullshoff Pol et al., 1998), where H_2S can easily permeate the cell membrane of microorganisms causing denaturation of proteins within the cytoplasm producing sulphide and disulphide cross-linking between polypeptide chains (Siles et al., 2010).

Studies to investigate the substitution of slurry with solid fractions from solid to liquid separation of manure in AD have been carried out previously (Møller et al., 2007a; Asam et al., 2011). However experiments to evaluate co-digestion of solid fractions derived from acidified dairy cow manure (SFDCM) have to our knowledge not yet been investigated. Therefore, this work investigated the utilization SFDCM as a co-substrate in thermophilic (50 °C) AD. The aims of this study are to: (1) determine the ultimate methane yield of dairy cow manure (DCM), acidified DCM,

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SFDCM and the liquid fraction of acidified DCM, (2) investigate the performance of thermophilic digesters treating substrates with different concentrations of SFDCM, and (3) evaluate the residual methane yield of digestates from a digesters treating different concentrations of SFDCM.

2. Methods

2.1. Experimental set up

In the study, 3 digester set-ups were tested, namely: batch, continuous and post digestion. The batch experiments were implemented to assess the ultimate methane yield of DCM, acidified DCM, SFDCM and the liquid fraction of acidified DCM. The continuous experiment was implemented to assess the methane yield and evaluate the digester performance with different concentrations of SFDCM .The post digestion experiments were implemented to determine the residual methane potential in the digestate manure from the continuous experiment. The batch and post digestion experiments were conducted in batch assay using 0.5 L infusion bottles.

2.1.1. Batch and post digestion experiment

The batch digestion experiments were conducted in batch assay using 0.5 L infusion bottles with the method described by Møller et al. (2004). The inoculum to substrate ratio was 1.2 ± 0.2 in terms of VS. Batch digesters containing solely inoculum served as a control to measure the gas production from the inoculum, which was subtracted from the gas production of the experimental batch digesters. Each digester was sealed using butyl rubber stoppers and aluminium caps and the headspace was flushed with 99.9% nitrogen for 2 min. Batch assays were done in triplicates and ran for a period of 70 days at 50 °C. The post digestion experiments were conducted with a similar method as the batch experiment except that inoculum was not used.

2.1.2. Continuous experiment

The continuous experiment was conducted using four identical intermittently stirred digesters (R1, R2, R3, R4) with 6.2 L total capacity and a 4.2 L working capacity, 14 days hydraulic retention time (HRT) and maintained at 50 °C. The mixing system operated at 60 rpm for 15 min every hour. The experiment was started by filling all digesters with 3.9 L of inoculum and 0.3 L of DCM and daily feeding with 0.3 L of DCM (after first removing 0.3 L g of digestate from a port at the base of the digesters) continued for the next 21 days giving 14 days average HRT. The digesters were fed through a tube, the outlet of which was submerged under the substrate level to avoid air ingress during the feeding process. Treatments started after the 21 days start up period, and consisted of substituting DCM with 10% (R2), 20% (R3) and 30% (R4) of SFDCM. One of the digesters (R1) continued to be fed solely with DCM. The three concentrations of SFDCM corresponding to 34.4%; 54.1% and 66.9% of the VS fed to R2, R3, and R4, respectively. The experiment continued for a period of four HRT corresponding to 56 days in total.

2.2. Inoculum and substrate

Inoculum for the batch experiment was sourced from the active commercial biogas digester at Research Centre Foulum, Denmark, which operates at a thermophilic temperature (52 °C). The commercial digester treats pig manure, cattle manure, maize silage and industrial by-product. Prior to use as inoculum, digested slurry from the commercial biogas digester was kept at 50 °C for 3 weeks to ensure that the major part of the remaining organic material was converted to biogas. The digested slurry was separated using a sieve (500 μm serial number 5564470 D-42759 Haan, Germany) and only the liquid fraction was subsequently used to inoculate the batch tests in order to get a uniform inoculum and to further minimize the inoculum biogas production. Inoculum for the continuous experiment was also from the active commercial biogas digester at Research Centre Foulum, Denmark as described previously. However, in this case the digested slurry was transferred directly from the commercial biogas digester to the laboratory scale digesters. The inoculum properties can be seen in Table 1.

The substrates used for the batch experiment were DCM, acidified DCM, SFDCM and liquid fraction of DCM. SFDCM and DCM were used as substrates for the continuous experiment. The separation technique used was a screw press (Fibre Master, Germany) with 0.5 mm screen size. DCM was taken from cows in the lactation period and was collected from a slurry storage pit at Research Centre Foulum, Denmark. Acidified DCM, SFDCM and liquid fraction of DCM were taken from dairy cows in the lactation period and were obtained from a Danish farm using acidification technology developed by Infarm A/S Aalborg, Denmark. Acidification of manure was performed by addition of 96% H_2SO_4 at a ratio of about 5 kg sulphuric acid t^{-1} slurry to reach pH 5.5 (Eriksen et al., 2008). Manure from the sludge pit was transferred to the 20 m^3 treatment tank in which acid was added at the bottom of the treatment tank. In order to minimize foam formation, manure was aerated during acid addition. A portion of the acidified manure in the treatment tank was pumped back to the slurry storage pit. These flushing processes took place 6–10 times per day. After this process, about a 15 cm depth of slurry was left in the slurry storage pit. The rest of the acidified manure from the treatment tank was transferred to the slurry storage tank (Kai et al., 2008).

The substrates used for batch and continuous experiments were collected on a single occasion and stored at −20 °C. DCM was kept in a 20 L plastic sealed barrel and SFDCM was kept in 5 L sealed plastic container at −20 °C until use.

In the post digestion experiment, the substrates were sourced from the continuously running digesters after running for a period of 45 days corresponding to 3 times of HRT. The substrate properties can be seen in Table 1.

2.3. Analytical methods

Biogas production from the continuous experiment was measured on a daily basis by collecting the gas in aluminium coated gas packs. In all experiments, biogas was measured using an acidified water displacement method. Gas samples were analyzed for

Table 1
Inoculum and substrate properties.

Inoculum/substrate	Sulphate (mg L^{-1})	Total VFA (mg L^{-1})	TAN (mg L^{-1})	Total N (g L^{-1})	VS (%)	TS (%)	pH
Inoculum (batch and post digestion)	n.a.	65	500	n.a.	2.2	3.6	8.30
Inoculum (continuously experiment)	n.a.	296	1500	n.a.	2.4	3.2	7.73
DCM	1099	4835	1300	2.7	6.3 ± 0.15	7.6 ± 0.15	6.89
Acidified DCM	8877	5102	1600	3.4	6.7 ± 0.03	8.5 ± 0.03	5.98
SFDCM	4259	2983	800	4.3	29.8 ± 0.08	32.1 ± 0.13	6.44
Liquid fraction acidified DCM manure	9842	5457	1700	3.5	3.8 ± 0.02	5.6 ± 0.02	6.04

n.a: Not available.

CO_2 and CH_4 content twice a week in the continuously running experiment and every time the biogas production was measured in the batch and the post digestion experiments. The gas composition was analyzed using a Perkin Elmer Clarus 500 gas chromatograph equipped with a Thermal Conductivity Detector and a Turbomatrix 16 Headspace auto sampler. Methane and CO_2 were isolated using a 12′ × 1/8″ Hayesep Q 80/100 Column, He was used as the carrier gas at 30 mL min^{-1}, and the injection port, oven, and detector temperature were 110, 40, and 150 °C, respectively.

Volatile fatty acids (VFA) (C_2–C_5) were determined twice a week by means of gas chromatograph (Hewlett Packard 6850A) with flame ionization detector (FID). The column was an HP-INNOWax, 30 m × 0.25 mm × 0.25 μm. The carrier gas was He. The temperature of the column was gradually increased from 110 °C to 220 °C at the rate of 10 °C min^{-1}.

Hydrogen sulphide (H_2S) in the gas phase was analyzed twice a week using precision gas detector tubes (Kogyo K.K., Kitagawa, Japan). Sulphide and sulphate concentration in the liquid phase was determined colormetrically at 690 nm with a Merck® spectrophotometer (NOVA 60, sulphide test 1.14770.0001, sulphate test 1.00617.0001).

Total solids (TS) were determined once a week by drying at 105 °C for 24 h. Ash was determined by combusting the dried samples at 550 °C for 5 h and VS was calculated by subtracting the ash weight from the TS. Total nitrogen was analyzed using the Kjeldahl standard method (APHA, 1995) and a Kjell-Foss 16200 auto analyzer (Foss Electric, Hillerod, Denmark). Total ammonia nitrogen (TAN) was measured colorimetrically at 690 nm with a Merck® spectrophotometer (NOVA 60, NH_4+ test 1.00683.0001). pH was measured using a pH meter (Knick Type 911, Germany).

Data were statistically analyzed using SAS® software, Cary, NC. Duncan multiple range tests was used in post ANOVA analysis, when differences were found to be significant at the level $P = 0.05$ level.

3. Results and discussion

3.1. Batch experiment

The average ultimate methane yield (*Bo*) from DCM, acidified DCM, SFDCM and liquid fraction of DCM were 281, 203, 264, and 138 L kg VS^{-1}, respectively (Fig. 1). *Bo* of DCM in this experiment is comparable with values used by IPCC (1996) that estimates *Bo* of DCM in developed countries to be 240 L kg^{-1}. In general, lower *Bo* values of the SFDCM compared to whole manure and liquid fraction would be expected (Møller et al., 2007b) and since this is not the case sulphide inhibition might be the reason since the sulphate concentration was lower in SFDCM (Table 1). In Fig. 1, it seems that sulphide inhibition in the digester treating the liquid fraction of acidified DCM was higher than in the digester treating acidified DCM. This can be explained by the fact that the batch tests operate on a fixed VS ratio of inoculum to substrate, and therefore a larger amount of the liquid fraction was added to the same amount of inoculum resulting in a higher concentration of sulphur in the batch digester. Fig. 1 shows that there was a higher biogas production rate in the later incubation period in the digesters treating acidified DCM than in other digesters indicating that there was adaptation of microorganisms to sulphide inhibition in these digesters. It might be caused by the lowering of the sulphide concentration in the liquid phase due to H_2S transfer to the gas phase and the subsequent periodical removal of the biogas production. The same phenomenon was not observed in the digester treating the liquid fraction of acidified dairy cow manure indicating that the microorganisms in these digesters were got higher inhibition than that in the digesters treating acidified DCM which was probably caused by the higher sulphur concentration in these digesters.

3.2. Continuous experiment

3.2.1. Methane production

The methane yield per kg VS added is given in Fig. 2A. During the last 2 days of the starting up period, the mean methane gas yield was 209 ± 5 L kg VS^{-1}. Based on this methane yield, about 74% of the ultimate methane yield (*Bo*) of DCM was obtained in the continuous experiment. This value is comparable with Nielsen and Angelidaki (2008), who reported methane yield from 226–263 L kg VS^{-1} in thermophilic digesters (55 °C) treating cow manure with 15 days HRT. The methane yield per kg VS (Fig. 2A) declined significantly ($p < 0.05$) (Table 2) as the concentration of SFDCM increased. This phenomenon could be explained as a consequence of higher concentrations of SFDCM that has a lower degradability and therefore a lower concentration of easily degradable material per kg VS in the substrate but also sulphur inhibition might play an important role. Similarly, Møller et al. (2007b) found that methane yield per kg of VS was higher in a digester treating pig manure solely compared to a digester treating high amount of solid fraction from pig manure. In addition, Schievano et al. (2008) reported that the potential methane yield relies not only on the VS content of substrate but also on the degree of degradability of organic materials in AD.

This paper found that the methane yield in term of digester volume (L L^{-1}) increased significantly ($p < 0.05$) as the proportion of SFDCM in the substrate increased (Table 2). The methane yield per digester volume during the experimental period is illustrated in Fig. 2B. The methane yield in the digester with

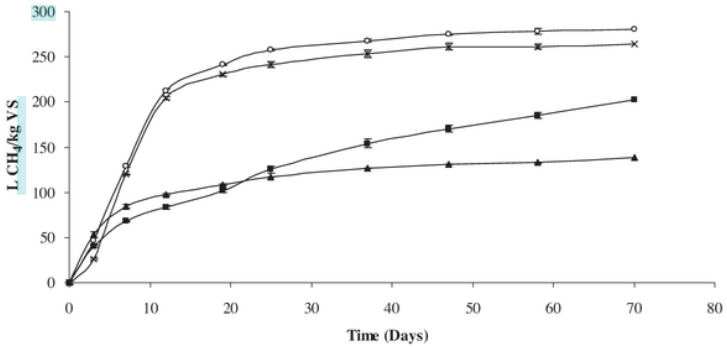

Fig. 1. The ultimate methane yields of: ○: DCM, ■: acidified DCM, ▲: liquid fraction of acidified DCM, and ×: solid fraction of acidified DCM.

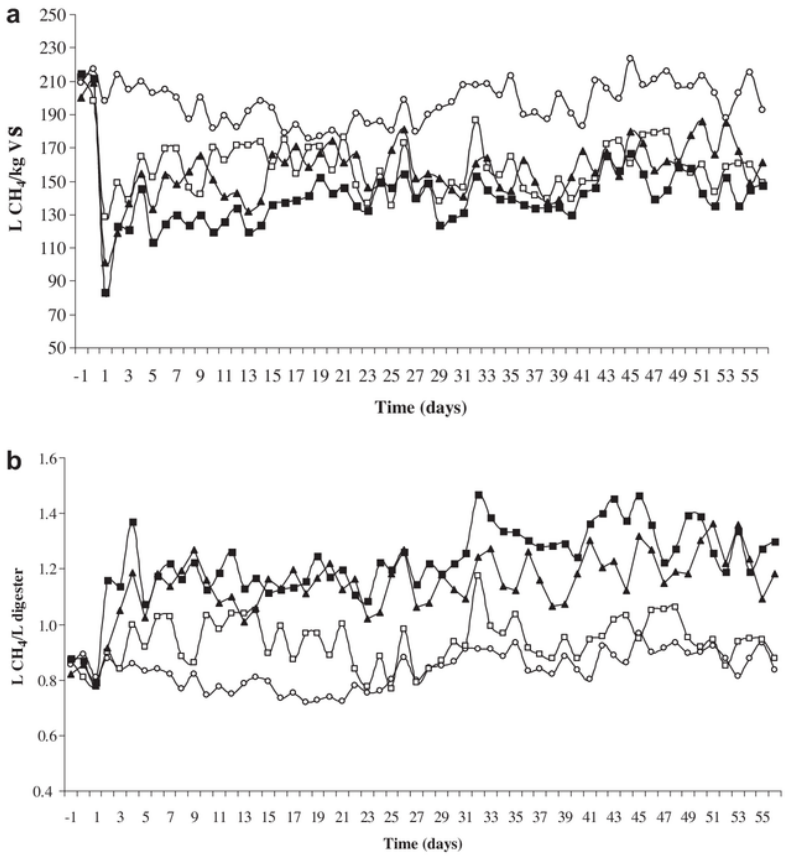

Fig. 2. A. Methane yield per kg VS added with substrate (○: control, □: 10% SFDCM, ▲: 20% SFDCM, and ■: 30% SFDCM). B. Methane yield per digester volume per day (○: control, □: 10% SFDCM, ▲: 20% SFDCM, and ■: 30% SFDCM).

Table 2
Process parameters. The values in each column followed by the same letter are not significantly different ($p > 0.05$).

Treatment	Methane yield				Sulphide (mg L^{-11})	TAN (mg L^{-1})	VS reduction (%)	Methane concentration (%)	H_2S (ppm)	pH
	($LL^{-1}d^{-1}$)	(L kg VS^{-1})	(L kg^{-1} substrate)	(L kg^{-1} digestate)						
Control	0.84 ± 0.1^{a}	196.7 ± 12.0^{a}	11.7	5.5 ± 0.02	58.9 ± 4.5^{a}	1800	35.4 ± 7.5^{a}	62.6 ± 0.9^{a}	401 ± 349^{a}	8.05 ± 0.04^{a}
10% SFDCM	0.94 ± 0.1^{b}	157.4 ± 13.7^{b}	13.2	8.6 ± 0.09	64.1 ± 5.1^{b}	1600	31.1 ± 3.0^{a}	60.1 ± 1.1^{b}	2364 ± 610^{b}	7.97 ± 0.06^{b}
20% SFDCM	1.16 ± 0.1^{c}	155.6 ± 15.8^{b}	16.2	13.3 ± 0.08	67.2 ± 4.7^{b}	1500	30.0 ± 6.3^{a}	58.2 ± 1.4^{c}	3240 ± 850^{c}	7.91 ± 0.05^{c}
30% SFDCM	1.24 ± 0.1^{d}	138.2 ± 14.1^{c}	17.3	15.7 ± 0.03	71.2 ± 7.2^{c}	1500	27.3 ± 8.9^{a}	56.8 ± 2.0^{d}	4100 ± 1005^{d}	7.84 ± 0.06^{d}

30% SFDCM substitution was approximately 50% higher than in the reference without SFDCM addition. This result is comparable with Møller et al. (2007b) who reported that the methane yield in the digester with 60% substitution of pig manure with the solid fraction of pig manure was almost doubled compared to pig manure alone. The enhancement of methane yield per substrate volume caused by substitution of DCM with SFDCM may also be partly attributed to the role of sulphuric acid as a pre-treatment method prior to AD. Dilute or strong acid can be used for pre-treatment, causing the solubilisation of hemicellulose that can in turn increase accessibility of enzymes to the cellulose and promote the formation of volatile organic compound that can be converted to methane (Hendrik and Zeeman, 2009). In addition, Kahar et al. (2010) found that sugar yield from corncob in 0.5% H_2SO_4, a similar concentration of acid as that used in the manure acidification process, and then autoclaved at 122 °C for 20 min was 1.6 times higher than that without acid addition.

3.2.2. Parameters in liquid phase

Fig. 3 show the total VFA concentration in digester R1, R2, R3 and R4. The total VFA concentration increased sharply after SFDCM addition commenced and reaches the peak concentration 17 days after the initial treatment where after the concentration decreased and stabilized at a low concentration after a period of two times the HRT. This can be caused by a sudden increase in the VS loading or by the increasing level of sulphur following the addition of SFDCM. The VS concentration in the substrate during treatment

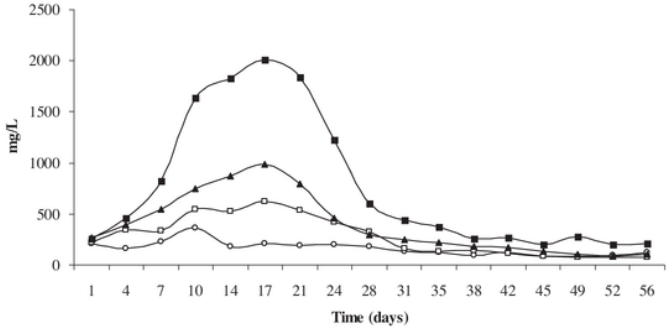

Fig. 3. Total VFA concentration in the digesters (○: control, □: 10% SFDCM, ▲: 20% SFDCM, and ■: 30% SFDCM).

was 6 ± 0.2%, 8 ± 0.4%, 10 ± 0.4%, and 12 ± 0.8% in R1, R2, R3 and R4, respectively. After two HRT following initial treatment, total VFA concentration was stable with a low concentration indicating that digester treating SFDCM as co-substrate up to 30% (67% on VS basis) can operate with a stable process. The high ratio of SFDCM was not expected to cause ammonia inhibition of the process since TAN concentrations during the two last weeks of the continuous experiment (Table 2) were considerably lower than TAN inhibition threshold of about 2.5 g L^{-1} reported by Hashimoto (1986).

The sulphide concentration was significantly ($p < 0.05$) higher as the proportion of SFDCM in the substrate increased (Table 2), but the sulphide concentration in the liquid phase of the digesters was under the sulphide inhibition thresholds of 100–800 mg L^{-1} as dissolved sulphide or approximately 50–430 mg L^{-1} as un dissociated H_2S, as reported by Parkin et al. (1990). The digester pH values were significantly lower ($p < 0.05$) when the proportion of VS in the substrate was increased (Table 2). This was attributed to a lower pH value in the SFDCM than in DCM (Table 1). There was no significant effect from the treatments on the VS reduction ($p > 0.05$) but on average the VS reduction was lower in the digester treating a higher SFDCM concentration (Table 2). This is probably caused by the lower degradability of SFDCM.

3.2.3. Parameters in gas phase

This study found that the methane concentration in the biogas was significantly lower ($p < 0.05$) as the concentration of SFDCM in the substrate increased (Table 2). This phenomenon might be caused by differences in pH value of digestates manure since under high pH condition a higher proportion of CO_2 is dissolved in the digestates slurry causing a higher CH_4 concentration in the gas phase (Møller et al., 2007b). The observed reduction in the methane concentration in the gas is in accordance with this theory since the pH value in the digester was significantly correlated ($p < 0.05$) with increasing concentrations of SFDCM (Table 2).

The average gas phase H_2S concentrations are given in Table 2. A significant increase ($p < 0.05$) of H_2S concentration in the gas phase was observed as the proportion of SFDCM in the substrate increased. The H_2S concentration in the gas phase when treating 30% SFDCM was approximately 10 times higher than in the digester treating only DCM. The concentration of free H_2S in the gas phase of a digester is largely affected by the rate of biogas production. A high rate of biogas production will lead to an increased transfer of H_2S to the gas phase (Elferink et al., 1994) which is in agreement with the work presented here. A high H_2S concentration in the biogas can cause problems such as odour, and corrosion of pumps and pipes (Hullshoff Pol et al., 1998). Therefore more attention should be given to maintenance of scrubber devices for removal of H_2S from biogas produced by digesters treating high concentration of SFDCM since the level of H_2S prior to use in combined heat and power plant should be low. Rasi et al. (2011) reported that H_2S concentration in biogas for traditional boilers and internal combustion is recommended not more than 1000 ppm.

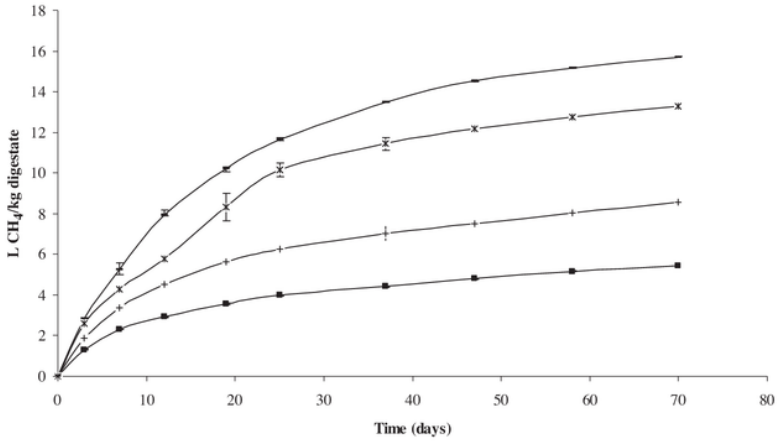

Fig. 4. Residual methane potential of digestates (■: control, +: 10% SFDCM, *: 20% SFDCM, and –: 30% SFDCM).

3.3. Post digestion experiment

When the digestates slurry was subjected to post digestion, the residual methane yield increased with increasing ratios of SFDCM (Fig. 4 and Table 2). Table 2 shows that the residual methane yield in the digestates slurry from the digester with 30% SFDCM was approximately three times higher than the yield found in the digestates manure from digester treating only DCM. Fig. 4 also shows that after 19 days of post digestion, about 65% of the total residual methane yield can be achieved. This result indicates that when using high ratios of SFDCM special emphasis on post digestion is needed to prevent high methane emissions and to gain the full methane potential. An efficient post digestion can thus compensate for some of the reduced digestion efficiency in terms of VS degradation in digesters with a high ratio of SFDCM. The methane concentration in the biogas from digestates slurry shows the same pattern as seen in the continuous digesters with lower concentration with increasing levels of SFDCM. The methane concentrations were 58.3%, 56.5%, 54.7% and 53.6% in R1, R2, R3 and R4, respectively.

4. Conclusions

It has been demonstrated that digesters substituting up to 30% of DCM with SFDCM can run with stable biogas production and low VFA levels. Sulphide and TAN concentrations are below the inhibition thresholds. Methane concentration is significantly lower as the concentration of SFDCM increases. Methane yield in term of digester volume can be increased by approximately 50% in the digesters substituting 30% of DCM with SFDCM. Post digestion of digestates slurry from digesters treating DCM with SFDCM is higher compared to the control indicating that this step will be of increasing importance as the amount of SFDCM is increased.

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