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Full Length Research Paper

Digestion of cattle manure and biodegradable kitchen waste to increase biogas production using rumen fluid as inoculums

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Anaerobic co-digestion strategies are needed to enhance biogas production when treating certain residues such as cattle/pig manure. Co-digestion of food waste with animal manure or other feedstocks with low carbon content can improve process stability and methane production. In this study, anaerobic digestion and co-digestion of cattle manure with organic kitchen waste using rumen fluid as inoculums have been experimentally tested to determine the biogas potential. Co-digestion substantially increased the biogas yields by 24 to 47% over the control (organic kitchen waste and dairy manure only). The highest methane yield of 14,653.5 ml/g-VS was obtained with 75% organic kitchen waste (OKW) and 25% cattle manure (CM) additions. In contrast, addition of 75% cattle manure caused inhibition of the anaerobic digestion process, and its cumulative methane yield was 23% lower than that with 25% cattle manure addition.

Key words: Cattle manure, co-digestion, methane, organic kitchen waste, rumen fluid.

INTRODUCTION

Energy is one of the most important factors for human development and to global prosperity. The dependence on fossil fuels as primary energy source has led to global climate change, environmental degradation, and human health problems. 80% of the world's energy consumption still originates from combusting fossil fuels (Goldemberg and Johansson, 2004). Yet the reserves are limited; means do not match with the fast population growth, and their burning substantially increases the greenhouse gas (GHG) concentrations that contributed for global warming and climate change (Schamphelaire and Verstraete, 2009). So, bio-energy (energy production from biomass) can be seen as one of the key options. Among the many bio-energy related processes being developed, those processes involving microorganisms are especially promising, as they have the potential to produce renewable energy on a large scale, without disrupting strongly the environment or human activities (Rittmann, 2008).

Anaerobic digestion (AD) is a technology widely used for treatment of organic waste for biogas production. Anaerobic digestion that utilizes manure for biogas production is one of the most promising uses of biomass wastes because it provides a source of energy while simultaneously resolving ecological and agrochemical issues. The anaerobic fermentation of manure for biogas production does not reduce its value as a fertilizer supplement, as available nitrogen and other substances

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remain in the treated sludge (Alvarez and Liden, 2007).

Ethiopia has a large population of dairy and beef cattle, generating large amounts of surplus manure that can be used in biogas plants to produce renewable energy. However, the high water content, together with the high content in fibers, are the major reasons for the low methane yields when cattle manure is anaerobically digested, typically ranging between 10 and 20 m^3 CH_4 per tone of manure treated (Angelidaki and Ellegaard, 2003).

Studies demonstrated that using co-substrates in anaerobic digestion system improves the biogas yields due to the positive synergisms established in the digestion medium and the supply of missing nutrients by the co-substrates (Wei, 2000). In a study carried out by Adelekan and Bamgboye (2009) on the different mixing ratios of livestock waste with cassava peels, the average cumulative biogas yield was increased to 21.3, 19.5, 15.8 and 11.2 L/kg TS, respectively for 1:1, 2:1, 3:1 and 4:1 mixing ratios when cassava peel was mixed with cattle waste. In another report, co-digestion of cow dung with pig manure increased biogas yield as compared to pure samples of either pig or cow dung. Comparing to samples of pure cow dung and pig manure, the maximum increase of almost seven and three fold was respectively achieved when mixed in proportions of 1:1 (Muyiiya and Kasisira, 2009). Co-digestion with other wastes, whether industrial (glycerin), agricultural (fruit and vegetable wastes) or domestic (municipal solid waste) is a suitable option for improving biogas production (Amon et al., 2006; Macias-Corral et al., 2008; El-Mashad and Zhang, 2010; Marañón et al., 2012).

Food waste is a desirable material to co-digest with dairy manure because of its high biodegradability (Zhang et al., 2006, 2011; Li et al., 2010; Wan et al., 2011). Study on the biogas production potential of unscreened dairy manure and different mixtures of unscreened dairy manure and food waste using batch digesters at 35°C showed that the methane yield of unscreened manure and two mixtures of unscreened manure and food waste (68/32 and 52/48), after 30 days of digestion, was 241, 282 and 311 L/kg VS, respectively (El-Mashad and Zhang, 2010).

In a study conducted by Zhu et al. (2011), they used different food wastes, including expired creamer; expired beer; slaughterhouse waste (SW); and fat, oil, and grease (FOG), and these food substances were co-digested with dairy manure to determine the methane potential. According to the result, co-digestion substantially increased the methane yields by 2.0 to 4.6 times over the control (dairy manure only).

This study was initiated to investigate the feasibility of biogas production from the different wastes that are generated from Haramaya University and the aims of the present research work were to determine the optimal conditions and mixing ratios for improved production of biogas using co-digestion of cattle manure and solid organic kitchen waste and also identify the key parameters influencing the increase of biogas and methane yield.

MATERIALS AND METHODS

Sample collection and preparation

Fresh cattle manure (CM) from beef and dairy farm, fresh organic kitchen waste (OKW) from staff lounge, and rumen fluid (RF) from the slaughterhouse were collected from Haramaya University compound. 2 kg of fresh cattle manure was collected from eight randomly selected cattle from beef and dairy farms for five consecutive days. In these sites there are special feeds and normal grazing cattle. The special feeds are provided with special type of feeding program that includes silage, concentrate, hay forage, agricultural residues and different grass types, byproducts from Harar Brewery and Hamaressa Food Complex, etc. On the other hand normal grazers are not provided with special type of feeding program rather they graze grasses in the field and get only fodder and agricultural residues. Finally the CM from both types of cattle (special and normal grazers) was sorted and dried separately on a plastic tray using direct sunlight for two days. 3 kg of fresh organic kitchen wastes were also collected from the staff lounge similarly for five consecutive days. The OKW was sorted manually to prevent the inclusion of unwanted and possibly contaminant materials (such as detergents, sand, bones etc.) and then dried with direct sunlight for two days.

Following the methods suggested by Wendland et al. (2006), separately dried cattle manure from special feeds and normal grazers were mixed by weighing equal amount from each source and shredded using shredder (Fritsch- Adam Baumuler model 80a-4S114 type) to an average particle size of 2 mm and kept in a refrigerator at 4°C. The shredded small sized cattle manure and organic kitchen waste were mixed separately with water in 1:5 (solid waste: water) volume ratio, in order to maintain the total solid in the digester between 8 to 15%, which is the desired value for wet anaerobic digestion.

Inoculum preparation

Following the recommendation of Aurora (1983), due to the presence of higher content of anaerobic bacteria in the rumen of the ruminant animals and the abundance of rumen waste disposal from the nearby slaughterhouse, rumen fluid was used as inoculum for anaerobic co-digestion of cattle manure and organic kitchen waste.

Experimental set-up and design

A completely randomized experimental design was used in a 5 × 4 replicated laboratory experiment and it was conducted in a series of five plastic tanks with 2 L capacity which was used as a laboratory scale anaerobic digesters at mesophilic temperature (30 ± 8°C). The working volume of each digester was 1.6 L. In each digester, rumen fluid was used as inoculum. The TS and VS/ TS of the inoculum used were 1.03% (wet basis) and 63.9%, respectively. Each digester was purged for 5 min (300 mL/min) with inert gas (N_2) to create an anaerobic environment. Food waste, cattle manure and their mixtures were separately examined in mono and co-digestion respectively. The characteristics of the different experiments are shown in Table 1. In co-digestion, the amount of organic kitchen waste as well as that of cattle manure in each

Table 1. Properties of organic kitchen waste, cattle manure and rumen fluid (mean ±SD).

Parameter	Organic kitchen waste	Cattle manure	Rumen fluid
pH	5.51±0.129	7.19±0.215	7.45±0.114
MC (%)	82.95±0.169	84.59±0.40	98.98±0.01
TS (Wt %)	17.05±0.169	15.42±0.40	1.03±0.01
VS (Wt %)	15.89±0.52	12.68±0.63	0.66±0.01
VS/TS ratio	93.18±2.54	82.23±2.04	63.9±0.45

Table 2. Properties of cattle manure and organic kitchen waste before digestion (mean value ± SD).

	Parameter (before digestion)				
Mixture	pH	MC (%)	TS (%)	VS (%)	VS/TS (%)
A1	6.95±0.030	86.15±0.128	13.85±0.128	12.85±0.403	92.75±2.398
A2	7.45±0.071	87.46±0.314	12.54±0.314	10.27±0.503	81.9±1.403
A3	7.32±0.065	87.09±0.490	12.83±0.353	10.81±0.470	84.2±1.354
A4	7.19±0.051	86.83±0.358	13.17±0.358	11.34±0.445	86.03±1.159
A5	7.09±0.025	86.42±0.274	13.58±0.274	11.89±0.389	87.5±1.587

digester was varied when it was added. The FW/CM ratios (based on VS) of digestion A3, A4, A5 were designed as 0.3, 1 and 3, respectively, corresponding to the organic kitchen waste and cattle manure amounts of 25:75, 50:50 and 75:25 g-VS/L. In digestion A1, organic kitchen waste was digested alone at the load of 100 g-VS/L, whereas in digestion A2, cattle manure was digested alone at the load of 100 g-VS/L as a control group. Thus, to determine the performance of co-digestion, the co-digestion of A3, A4 and A5 was compared with mono-digestion groups of A1 and A2. In addition, to provide mixing of the digester contents, all digesters were shaken manually for about 1 min once a day prior to measurement of biogas volume.

Measurement of biogas yield

Biogas was collected by water displacement method. In order to prevent the dissolution of biogas in the water, brine solution was prepared. Following the method suggested by Elijah et al. (2009), an acidified brine solution was prepared by adding NaCl to water until a supersaturated solution was formed. Three to five drops of sulphuric acid were added to acidify the brine solution. As biogas production commenced in the fermentation chamber, it was delivered to the second chamber which contained the acidified brine solution. Since the biogas is insoluble in the solution, a pressure build-up and provides the driving force for displacement of the solution. Thus the displaced brine solution was measured to represent the amount of biogas produced. The biogas volume was calculated daily and was transformed into the volume at Standard Temperature and Pressure (STP) condition.

Chemical analysis

The pH, TS and VS of organic kitchen waste and cattle manure samples were measured according to the standard methods (APHA, 1998). The pH values of each digester were monitored in five days interval using digital pH meter (HANNA Model pH-211). Following the method of Radtke et al. (1998) and Yu and Fang (2002), the pH values of the contents of digesters were buffered between 6.8 and 7.4 by the addition of hydrated calcium carbonate. The VS content of the liquor was subsequently measured. The values of VS destructions were calculated based on total mass balances of VS in each digester before and after the digestion test with subtracting the VS contents of the control digesters from that of the testing digester.

RESULTS AND DISCUSSION

Pre-digestion characteristics of substrates

Table 2 summarizes the values obtained in the pre-digestion characteristics of the five feed stocks. As it is shown, there is a considerable amount of variation in the composition of feed mixtures, which is due to the variability in the composition of the samples of the different substrates taken over the experimental period. The content in volatile solids of cattle manure and organic kitchen waste ranged between 9.8-10.8% and 12.4-13.3%, respectively (average values of 10.3 and 12.9%, respectively). On a dry matter (TS) basis, organic kitchen waste contained higher VS than cattle manure. The higher VS content of organic kitchen waste (13 g/kg), compared with that of manure (10 g/kg), means relatively higher energy content, which is desirable from an economic standpoint with regards to biogas energy production. The VS/TS ratios were 82 and 93% for cattle manure and organic kitchen waste, respectively.

Before inoculation the mean pH values of CM and OKW were 7.19 and 5.51, respectively; however, after they are inoculated with rumen fluid, the inoculum mean pH values of the two control groups (A1 and A2) were

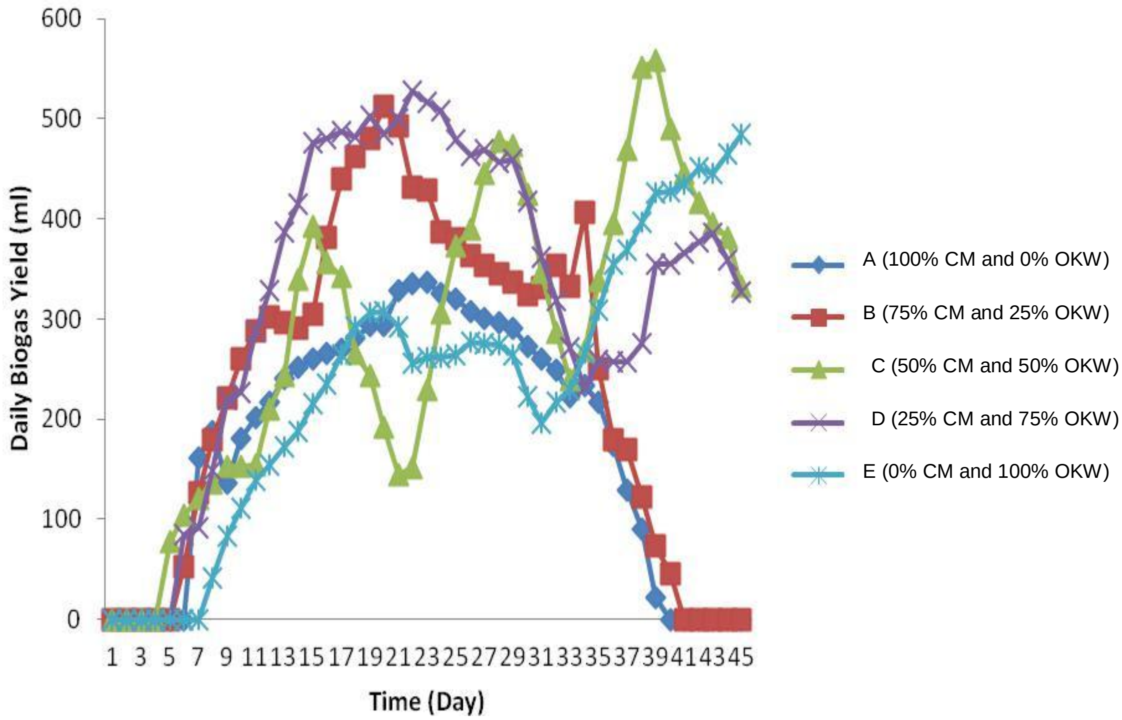

Figure 1. Daily mean biogas yield of digester D in 45 days.

increased. This indicates that the rumen fluid used for this study have had a good buffering capacity as it was also reported earlier (Girma et al., 2004; Forster-Carneiro et al., 2008; Montusiewicz et al., 2008; Uzodinma and Ofoefule, 2008).

Biogas production rate

On average, biogas productions from digesters A2, A3, A4, A5, and A1 were detected on the 7th, 6th, 5th, 6th, and 8th days respectively. The results showed that the co-digestion of samples with the three mix ratios (A4, A5, and A3) produced biogas earlier than the two pure substrates (A1 and A2) that were used as control groups. From the three mix groups, digester A4 produced biogas much faster, followed by digester A5 and A3. This might be due to the attribution of the positive synergetic effect of the co-digestion of CM and OKW in providing more balanced nutrients, increased buffering capacity, and decreased effect of toxic compounds. Digestion of more than one kind of substrate could establish positive synergism in the digester (Mata-Alvarez et al., 2000; Li et al., 2009; Jianzheng et al., 2011). The rapid initial biogas production in digester A4 might be also due to shorter lag phase growth, the availability of readily biodegradable organic matter in the substrate, and the presence of high content of the methanogens.

Biogas production

Biogas production was used mainly as an indication of optimum production and the development of favorable conditions for microbial activity during the digestion process. The daily methane production from the control and digesters are shown in Figure 1. The average daily biogas yield observed from the five digesters (A1, A5, A4, A3, and A2) were 176.77, 237.85, 284.76, 325.63, and 236.18 mL/g-VS, respectively. As compared to digesters A1 and A2, digesters A5, A4, and A3 produced the 1st, 2nd, and 3rd highest volume of biogas on each day during the 45 days of experiment, respectively (Figure 1). The higher biogas production from these mixtures could be due to the balanced (nutrient to microorganism) composition, and stable pH which was attained from the inoculation with rumen fluid and mixing ratios used. On the other hand low average daily biogas production observed from digesters A1 and A2 containing pure 100% OKW and 100% CM, attributed to the unbalanced nutrient to microorganism ratio, and unstable pH value. After the gas production was started and stabilized, digesters A4, A5, and A1 produced the least amount of daily biogas on the 5th, 6th, and 8th, days of the run, respectively. The observed least gas yield from these digesters might be due to the production of volatile fatty acids by the microorganism which hinders the releasing of the biogas. This is in agreement with the report of Budiyono et al. (2010) who also observed low level of

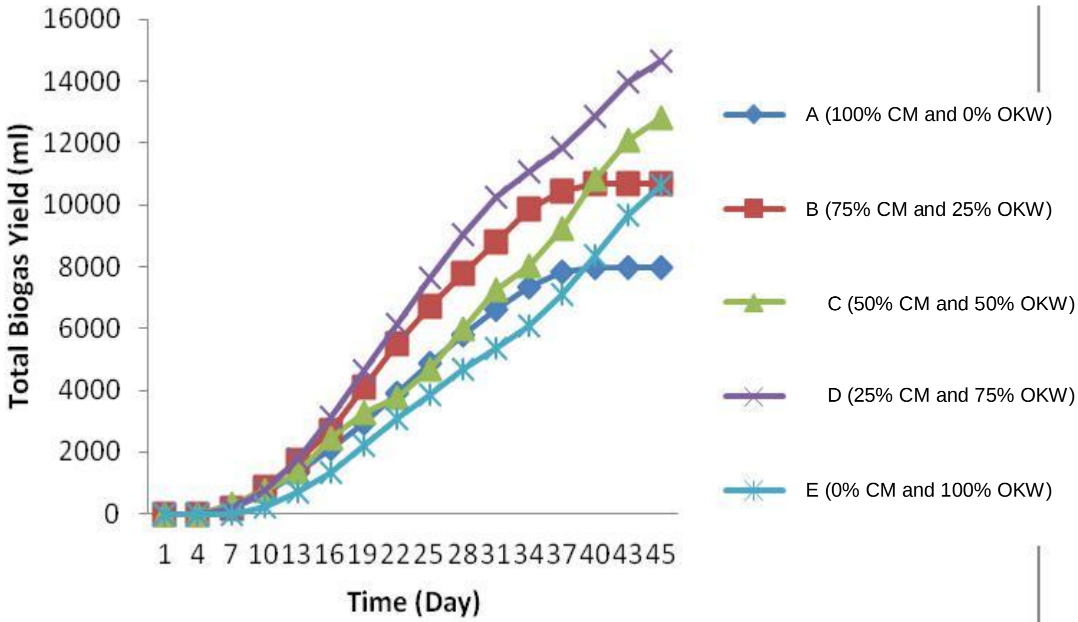

Figure 2. Mean cumulative biogas yield of all samples within 45 days.

biogas production due to the lag phase of microbial growth during these periods of the run.

The cumulative biogas productions of the five samples in all experiments were averaged and the mean cumulative biogas production and total gas production were summarized in Figure 2. As compared to the single anaerobic digestion of the two pure samples, the co-digestion of the three mix ratios produced higher volume of biogas. The total gas produced from the co-digestion of the three mixed samples (A3, A4, and A5) was indicated in Figure 2. From the co-digestion of A3, A4 and A5; 24.12, 37.91 and 47.13% more biogas was produced respectively than the two pure samples used as control. This might be due to mixing of cattle manure with organic kitchen waste provided balanced nutrients, buffering capacity, appropriate C/N ratio and sufficient anaerobic microorganisms. Moreover, the cumulative biogas yield of sample A5 is greater than sample A4 which is greater than sample A3. This might be attributed to the increased content of organic kitchen waste from 25 to 50% and to 75% (Amirhossein et al., 2004; Jianzheng et al., 2011). This result was in accordance with those obtained with co-digestion of 75% brewery waste and 25% sewage sludge (Babel et al., 2009).

Biodegradation during anaerobic digestion

In order to determine which matter in what amount was utilized from the initial feed during the 45 days of retention time and to correlate with the rate and amount of biogas produced, the digestate from each digester were characterized (Table 2). It is important to maintain the pH of an anaerobic digester between 6 and 8; otherwise, methanogen growth would be seriously inhibited (Gerardi, 2003). In this study, the initial pH of all the digesters was in the range of 6.95 to 7.45 even with the addition of acidic food wastes (like injera) indicating the buffering capacity of the cattle manure. But finally the pH showed a significant increase and it was in the range of 7.66 to 8.47. This was predicted because the VFAs produced by acidogens during the start up phase were consumed by methanogens and transferred to the methane. Generally, pH increase accompanies increasing biogas production because methanogens consume VFAs and generate alkalinity. In addition there occurs a decrease in VS and VS/TS ratio and this might be due to the biodegradation and conversion of VS into biogas through the microbial acidogenesis and methanogenesis. At the beginning of the digestion process the average total solids (TS) and volatile solids (VS) content of substrates in all digesters were high (Table 3). But, at the end of the 45 days anaerobic digestion period the contents of both TS and VS were highly reduced and this is attributed to their consumption by fermenting and methanogenic bacteria.

The efficiency of anaerobic co-digestion of cattle manure and organic kitchen waste was evaluated in terms of TS and VS reduction as the amount of dry matter and organic compounds. Table 4 presents the amount of TS, and VS biodegradation and conversion into biogas per mg, TS and VS removed in the anaerobic co-digestion processes of cattle manure with organic kitchen waste at an ambient temperature of 30 ± 8°C.

Table 3. Properties of cattle manure and organic kitchen waste after digestion (mean value ± SD).

	Parameters (after digestion)				
Mixture	**pH**	**MC (%)**	**TS (%)**	**VS (%)**	**VS/TS (%)**
A1	7.66 ± 0.264	94.05 ± 1.067	5.95 ± 1.067	4.71± 0.721	79.16 ± 5.041
A2	8.47 ± 0.173	96.76 ± 0.462	3.24 ± 0.462	0.89 ± 0.307	27.47 ± 6.322
A3	8.27 ± 0.191	96.58 ± 0.486	3.42 ± 0.486	1.28 ± 0.369	37.43 ± 5.824
A4	8.04 ± 0.174	95.96 ± 0.539	4.05 ± 0.539	2.05 ± 0.394	50.62 ± 3.495
A5	7.86 ± 0.236	95.28 ± 0.788	4.72 ± 0.788	2.83 ± 0.568	59.96 ± 4.403

Table 4. Organic matter degradation and biogas yield from each digester.

Treatments	Organic matter composition and its removal						Biogas yield		
	Total solids (mg/vol)			Volatile solids (mg/vol)			Total (ml)	ml/mg TS removed	ml/mg VS removed
	Initial TS(mg)	Removed			Removed				
		Mg/Vol.	%/Vol.		Mg/Vol.	%/Vol.			
A1	22,160	12,640	57.04	20,560	13,024	63.35	10,628.3	0.84	0.82
A2	20,064	14,880	74.16	16,432	15,008	91.33	7,954.8	0.54	0.53
A3	20,528	15,056	73.34	17,296	15,248	88.16	10,703.3	0.71	0.70
A4	21,072	14,592	69.25	18,144	14,864	81.92	12,814.3	0.88	0.86
A5	21,728	14,176	65.24	19,024	14,496	76.20	14,653.5	1.03	1.01

Biodegradation of TS and VS was high in samples containing high proportion of CM and decreases as the proportion of OKW in the mix ratio increases. With gas production rate of 1.03 ml/mg TS or 1.01 ml/mg VS removed from the biodegradation of 14,176 mg (65.24%) of the initial TS, or 14,496 mg (76.20%) of the initial VS, digester A5 gave the 1st highest cumulative biogas yield of 14,653.50 ml/g-VS. The result showed that in digester A4 and A5 there was a direct relationship between total biogas yield and gas production rate per each milligram of total solids and volatile solids removed. This might be because, the digestion process in these two digesters had more balanced acidogenesis and methanogenesis and the VS removed were utilized for biogas produce more efficiently than the other levels. Similar results were reported by Joung et al. (2008) from the anaerobic co-digestion of swine manure and food waste.

Digester A2 was observed with the highest percentage of TS and VS removal; however, it produced the least cumulative biogas yield of 7,954.75 ml/g-VS. This might be because of the presence of only cattle manure that is inoculated with rumen fluid. Since both cattle manure and rumen fluid are partially digested in the guts of the ruminants less biogas production from cattle manure within short retention period can be attributed to its relatively lower organic content than organic kitchen waste. Generally, it was observed that the TS and VS removal rates were affected by the different mixing ratios of cattle manure with organic kitchen waste and the hydraulic retention time. This suggests that high concentration of anaerobic bacteria content in rumen fluid and cattle manure works effectively to degrade organic matter composed in organic kitchen waste. So the results of this study imply that the biodegradability of organic matter and cumulative biogas yield was improved by co-digesting cattle manure with organic kitchen waste using rumen fluid as inoculum.

Co-digestion performance and synergistic effect

The co-digestion of three mix ratios (75:25, 50:50 and 25:75) of rumen fluid inoculated CM with OKW was performed and biogas productions from the biodegradation of organic matter were compared with pure cattle manure and organic kitchen waste as the controls. As the result indicated, the co-digestions of the three mixes showed improved biogas production rates and achieved higher cumulative biogas production than the two pure samples. This higher biogas production from digesters A3, A4, and A5 with mixed substrates of rumen fluid inoculated cattle manure and organic kitchen waste was due to the increased carbon content of OKW and high concentration of anaerobic bacteria content of cattle

Table 5. Synergistic effect of co-digestion of cattle manure and organic kitchen waste.

Treatments	Percentage of CM /OKW	Cumulative biogas yield				
		Cattle manure (ml)	Co-digestion (ml)	Organic kitchen waste (ml)	Increase (ml)	Increase (%)
A1	0:100	0.00		10,628.25		
A2	100:0	7,954.75		0.00		
A3	75:25	5,966.06	10,703.25	2,657.06	2,080.13	24.12
A4	50:50	3977.38	12,814.25	5314.13	3522.74	37.91
A5	25:75	1988.69	14,653.5	7971.19	4693.62	47.13

manure and rumen fluid. In other words this might be due to synergistic effect of CM to OKW (Table 5). The synergistic effect is mainly attributed to more balanced nutrients, increased buffering capacity, and decreased effect of toxic compounds (Li et al., 2009; Danqi, 2010; Jianzheng et al., 2011). More balanced nutrients in co-digestion would support microbial growth for efficient digestion, while increased buffering capacity would help maintain the stability of the anaerobic digestion system.

As it is shown on Table 5, from the co-digestion of cattle manure and organic kitchen waste with 75:25, 50:50, and 25:75 mix ratios 24.12, 37.91 and 47.13% additional biogas production was obtained, respectively when it is compared with that of the mono-digestions. It is evident from this result that digestion of more than one kind of substrate could establish positive synergism in the digester and provides more balanced nutrients as well as buffering capacity thus enhance the anaerobic digestion process and bio-energy production.

Identification of mix ratio for highest biogas production

As the proportion of OKW in the mix ratio increases from 0 to 25% to 50% and to 75% biogas yield was increased by 24.12, 37.91 and 47.13%, respectively. Thus, digester A5 with mix ratio of 25% CM and 75% OKW produced the highest volume of biogas (Figure 2). This might be due to the high organic content of OKW coupled with the supply of suitable microorganisms and missing nutrients by the rumen fluid and CM make the carbon to nitrogen ratio within the desired range.

Conclusions

Organic kitchen wastes co-digested with cattle manure improved the biogas potential compared to cattle manure alone. The co-digestion of rumen fluid inoculated CM and OKW with mix ratio of 50:50, gives biogas yield earlier and highest average daily and cumulative biogas yield were obtained from the co-digestion of rumen fluid inoculated CM and OKW with 25:75 ratio. The 25:75, 50:50 and 75:25 mix ratios of CM and OKW gave from 24.12 to 47.13% additional biogas yield and cumulative gas production was enhanced by 1.01-1.84 times. Thus, as compared to the mono-digestions of pure CM and pure OKW anaerobic co-digestion of rumen fluid inoculated CM and OKW in 25:75, 50:50, and 75:25 mix ratios enhances both the rate and amount of biogas yield.

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