Pergamon

Wat. Sci. Tech. Vol. 38, No. 2, pp. 177–184, 1998.
IAWQ

0273–1223/98 $19·00 + 0·00

PII: S0273-1223(98)00479-X

DYNAMICS OF METHANOGENIC ACTIVITIES IN A LANDFILL BIOREACTOR TREATING THE ORGANIC FRACTION OF MUNICIPAL SOLID WASTES

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ABSTRACT

A bench-scale investigation was conducted to evaluate the dynamics of methanogenic activities in a landfill bioreactor (LFBR) treating the organic fraction of municipal solid wastes (OFMSW). The specific methanogenic activity (SMA) test was used to measure each individual methanogenic activity on degrading carbohydrate, protein, lipid, butyrate, propionate, and acetate at an interval of one-year. A simple model incorporating three biokinetic parameters, namely lag-phase time, methane production rate and methane production potential, was employed to systematically describe the dynamics of SMA on the anaerobic mineralization of the OFMSW in an LFBR. The results indicate that the model is suitable to describe the dynamics of SMA in the LFBR. According to the dynamics of the SMA, the decomposition of proteins and lipids overruled the stabilization of the LFBR, while the rate of methanogenesis was higher than that of acetogenesis. Comparing the estimation and the observation of methane production rates suggested that the period of the critical incubation time (λ^*) of the protein and the lipid influenced the efficiency of the LFBR.

KEYWORDS

Landfill bioreactor; methanogenesis; organic fraction of municipal solid waste; specific methanogenic activity

INTRODUCTION

Anaerobic digestion of organic solid waste, especially the OFMSW, is of growing importance in the field of solid-waste management. The use of leachate containment, collection and recirculation on a landfill has provided opportunity to transfer a landfill into a controlled bioreactor system. Several types of bioreactors have been developed for the treatment of an OFMSW, including continuous and batch digesters. A batch digester, such as a landfill bioreactor, treating the OFMSW is in essence simpler than a continuous anaerobic digester, and the lower cost per ton of waste treated in a landfill reactor is a consequence of the simple technology applied (ten Brummeler and Koster, 1990; Pohland, 1996). Compared with conventional sanitary landfills, landfill bioreactors provide the potential for more rapid, complete and predictable attenuation of solid waste constituents, enhance gas recovery and utilization, and reduction in health/adverse environmental impacts and attendant liabilities (Pohland, 1997).

Characteristics of methane fermentation of organic refuse samples strongly depend on its chemical nature (Lay et al., 1996a; Lay et al., 1997a). Understanding and recognizing the methane fermentation characteristics of an OFMSW in a landfill bioreactor are, therefore, increasingly important for reducing the pollution potential and enhancing the methane production of OFMSW (Rivard et al., 1989; Mormile et al., 1996). Many attempts have made to enumerate bacterial populations and measure enzyme activity of the bacteria during organic refuse decomposition (Rees, 1980; Jones and Grainger, 1983; Barlaz, 1989). However, to date no attempt has been made to formulate a model describing the dynamics of methanogenic activity by the quantitative measurement of

the substrate utilization on a LFBR treating an OFMSW.

The objective of this study was to examine the dynamics of methanogenic activities during methane fermentation of the OFMSW. For this purpose, the OFMSW stabilization was well progressed in an anaerobic batch digester, namely a landfill bioreactor (LFBR), for measuring the specific methanogenic activities (SMAs) degrading carbohydrate, protein, lipid, butyrate, propionate, and acetate. Furthermore, a model including the parameters of methane production lag-phase time, rate and potential was suggested to systematically describe the dynamics of the SMAs in the LFBR.

MATERIALS AND METHODS

Composition of synthetic OFMSW

A synthetic organic fraction of municipal solid waste (OFMSW) was used as the experimental material for this study. As listed in Table 1, the shredded OFMSW consisted of vegetables (27.0%), fruits (5.4%), meat (4.5%), rice (12.0%), paper (0.01%) and dewatered sludge cake (51.1%). Except for the rice, each component was shredded to less than 2.0 cm in particle size.

Table 1. The composition of synthetic organic fraction of municipal solid waste used in this study.

Components	Composition (%)
Vegetables	27.0
Fruits	5.4
Meat	4.5
Rice	12.0
Paper	0.01
Sludge cake	51.1

Experimental equipment and procedure

The OFMSW was incubated in ten 0.5-liter wide-mouth landfill bioreactors (LFBR, Figure 1). For each LFBR, 300 grams of the shredded OFMSW (Table 1) and 30 grams of seed sludge, obtained from the municipal sewage treatment plant in Sendai City, Japan, were filled to capacity at the beginning of the experiment. Deionized water was added to the OFMSW for adjusting its moisture content to 70% (wt./wt.) to ensure the availability of ample free liquid for leachate recycling. Leachate generation rates ranged from 60 to 120 $mL \cdot d^{-1}$. The LFBRs were operated with a weekly leachate recycle and maintained at a constant temperature of 41 °C, the optimum temperature for mesophilic refuse decomposition (Hartz et al., 1982). The solids in ten LFBRs was sequentially transferred into vials to measure each individual specific methanogenic activity degrading the carbohydrate, protein, lipid, and short-chain volatile fatty acids (VFAs) at an interval of one-year.

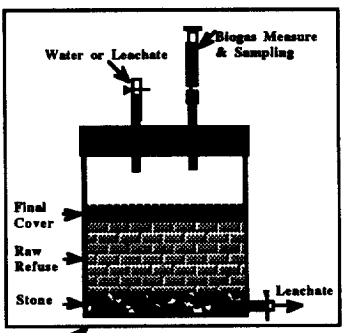

Figure 1. The schematic graph of the landfill bioreactor used in this study.

Specific methanogenic activity

The rates of methanogenesis by sample, usually incubated in a serum vial, can be readily measured and effect of organic matter decomposition on these rates can be assessed. The potential of the methanogens in question to degrade an added methanogenic substrate or their precursors can be determined (Shelton and Tiedje, 1984). Thus, the specific methanogenic activity (SMA) test was performed with a 120 mL vial at 41 °C. Glucose, peptone, linseed oil, acetate, propionate and butyrate were used as the individual substrate to determine the SMA of the sludge inside the LFBR. Among them, glucose, peptone and linseed oil were used as the typical substrates of the carbohydrates, proteins and lipids, respectively (Ogimoto and Imai, 1980). The composition of the incubation media is listed in Table 2 (Li and Noike, 1992) and a mixture gas containing 80% N_2 and 20% CO_2 was used as the headspace gas. The vials were then incubated in a rotary cell culture (Figure 2) rotated at 1.5 rpm for providing better contact among samples, nutrients and microorganisms. The volume of biogas was measured with glass syringes according to the approach reported by Owen et al. (1979). All these operations proceeded under an atmosphere of 80% N_2 and 20% CO_2 using the Hungate technique (Miller and Wolin, 1974)

Table 2. Medium composition of SMA test.

Components	Concentration
Organic substrate	1.0 $g \cdot L^{-1}$
KH_2PO_4	0.4 $g \cdot L^{-1}$
K_2HPO_4	0.4 $g \cdot L^{-1}$
NH_4Cl	1.0 $g \cdot L^{-1}$
Mineral solution [a]	10.0 mL
Vitamin solution [b]	10.0 mL
$NaHCO_3$	4.0 $g \cdot L^{-1}$
$MgCl_2 \cdot 6H_2O$	0.21 $g \cdot L^{-1}$
Cysteine $HCl \cdot H_2O$	0.5 $g \cdot L^{-1}$
$Na_2S \cdot 9H_2O$	0.25 $g \cdot L^{-1}$
Resazurine	0.002 $g \cdot L^{-1}$
pH	7.0-7.2

(a) Contains, in grams per liter of distilled water: nitrotriacetic acid, 4.5; $FeCl_2 \cdot 4H_2O$, 0.4; $CoCl_2 \cdot 6H_2O$, 0.12; $Alk(SO_4)_2$, 0.01; NaCl,1.0; $CaCl_2$, 0.02; Na_2MoO_4, 0.01; $MnCl_2 \cdot 4H_2O$, 0.10; H_3BO_3, 0.01; $CuSO_4 \cdot 5H_2O$, 0.01; $NiCl_2 \cdot 6H_2O$, 0.02.
(b) Contains, in milligrams per liter of distilled water: biotin, 2; folic acid, 2; pyridoxine HCl, 10; thiamine HCl, 5; riboflavin, 5; nicotinic acid, 5; DL-calcium pantothenate, 5; vitamin B12, 0.1; *p*-aminobenzoic acid, 5; lipoic acid, 0.5.

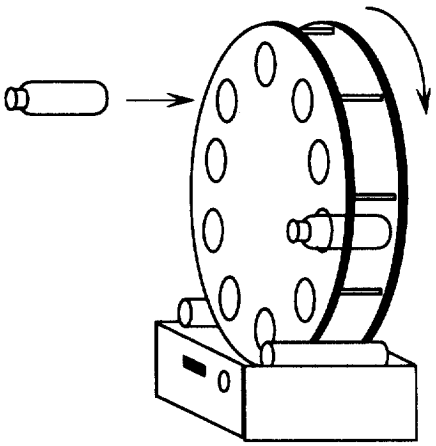

Figure 2. A rotary cell culture device used in this study.

to assure an anaerobic condition.

The specific methane production rate in each vial experiment using each individual substrate was defined as the specific methanogenic activity degrading the corresponding substrate. In order to evaluate the SMA, the modified Gompertz equation shown by Eq. (1) (Lay et al., 1996a; 1997a,b) was used to fit the experimental data in each vial experiment and the SMA was then obtained by dividing the methane production rate by the VS amount of the tested sludge samples.

$$M = P \cdot \exp\left\{-\exp\left[\frac{R_m \cdot e}{P}(\lambda - t) + 1\right]\right\} \quad (1)$$

where M is the cumulative methane production (mL), t is the incubation time (day), λ is the lag-phase time (day), P is the methane production potential (mL), R_m is the methane production rate ($mL \cdot d^{-1}$), and e is exponential 1 in a vial test.

The dynamics of specific methanogenic activity, Ri(t), for degrading each individual substrate in an LFBR was described using Eq. (2), which was the derivative of the cumulative methane production with respect to the incubation time (Lay et al., 1996b).

$$R_i(t) = R_{m,i} \cdot \exp\left\{2 - \exp\left[1 + \frac{R_{m,i} \cdot e}{P}(\lambda_i - t)\right] + \frac{R_{m,i} \cdot e}{P}(\lambda_i - t)\right\} \quad (2)$$

where P_i is the methane production potential of component i (mL), $R_{m,i}$ is the maximum methane production rate

of component i ($mL \cdot gVS^{-1} \cdot d^{-1}$), and λ_i is the lag-phase time of component i (day).

Data analysis

The function of the 'solver' in Microsoft Excel version 5.0, by converging the residual sum of square between the experiment and the estimation to a minimum value, was used to estimate the parameters of these models. All parameters were judged by the diagnosis procedure according to the approach reported by Wen et al. (1994) including the Student-t test, Dubin-Watson test (D), and Kalmogorov-Smirnov test (D_n).

Analytical method

The percentage of methane and carbon dioxide in biogas was analyzed using a gas chromatograph (Shimadzu 8A) equipped with a thermal conductivity detector (TCD) and a 2-m stainless column packed with activated carbon (60/80 mesh). The operational temperatures of the injection port, the oven and the detector were 140, 120 and 140 °C, respectively. Helium was used as the carrier gas at a flow rate of 30 $mL \cdot min^{-1}$. Calibration was made using 99.99% methane standard gas. The detection limit of the TCD was 0.1% for methane. The concentrations of the VFAs were determined by a second gas chromatograph of the same model, equipped with a frame ionization detector (FID) and a 2-m glass column packed with KOCL-FM (60/80 mesh). The operational temperatures for the injection port, the oven and the FID were 160, 140 and 160 °C, respectively. Before the analysis of the VFAs, phosphoric acid was added to control the pH of the samples. The concentrations of total solids (TS) and volatile solids (VS) were determined by a 10-mL sample in the 105 °C and 550 °C ovens, respectively, according to the procedures described in the Standard Methods (APHA et al., 1992). The pH of samples were determined by a pH meter.

RESULTS AND DISCUSSION

Measurement of SMA

To measure the SMAs for degrading different substrates of the LFBR samples at various incubation time, a total of 42-SMA test experiments were carried out on the substrates of lipid, protein, carbohydrate, acetate, propionate and butyrate. Subsequently, equation (1) was used to fit the experimental data of the cumulative methane production curve in each individual vial test. In this study, the specific methane production rate for a certain substrate was defined as the SMA of that substrate. As an example, a nonlinear fitting plot using Eq. (1) is shown in Figure 3. Table 3 summarizes the best values of the SMAs obtained by the data analyses. The

Table 3. Summary of the SMA data degrading each individual substrate.

Incubation time (days)	SMA ($mL\ CH_4 \cdot\ gVS^{-1} \cdot\ d^{-1}$)					
	Acetate	Propionate	Butyrate	Lipid	Protein	Carbohydrate
25	2.4 (0.1)	4.8 (0.2)	7.2 (0.1)	1.2 (0.5)	2.4 (0.1)	24.0 (0.5)
70	9.6 (0.8)	9.6 (0.5)	9.8 (0.4)	2.4 (0.6)	6.0 (0.2)	67.2 (0.4)
120	12.0 (0.5)	10.8 (0.4)	12.5 (0.2)	6.0 (0.7)	7.2 (0.1)	48.0 (0.6)
150	19.2 (0.8)	12.0 (0.3)	15.1 (0.5)	7.2 (0.8)	19.2 (0.5)	38.4 (0.8)
200	33.6 (0.6)	14.4 (0.6)	19.7 (0.6)	24.0 (0.9)	28.8 (0.9)	14.4 (0.3)
250	38.4 (0.5)	21.6 (0.7)	23.5 (0.9)	4.8 (0.5)	16.8 (0.4)	12.0 (0.5)
300	24.0 (0.8)	16.8 (0.2)	28.8 (1.0)	2.4 (0.2)	4.8 (0.2)	7.2 (0.2)

Note: *Data are the mean of two replicate cultures.*
Values in parentheses represent standard deviation.

calculated values of D and D_a for determining the random normal distribution of the residuals ranged, respectively, from 1.1 to 2.9 and 0.05 to 0.20, which were less than the table values of 3.05 and 0.36, indicating that all residuals had a random normal distribution of less than 5% random error. In addition, all of the t-values were larger than that of $t_{0.975}(16) = 2.14$ (table value), where t = b/SE(b), whereas SE(b) and (b) represent, respectively, the standard error of b and all parameters to be estimated. This indicates that the parameters were taken to be statistically significant at a confidence interval of 95% (Box et al., 1978).

Dynamics of specific methanogenic activities in the LFBR

Figure 4 shows the dynamics of SMAs degrading different substrates in the LFBR, illustrating that the SMAs rose to a peak and gradually declined. Equation (2) was used to quantitatively describe the dynamics curves of SMA in the LFBR. The best values of the parameters, P_i, $R_{m,i}$ and λ_i, for fitting the experimental data were determined using the same analysis procedure as used for Eq. (1). Here, the $R_{m,i}$ represented the maximum

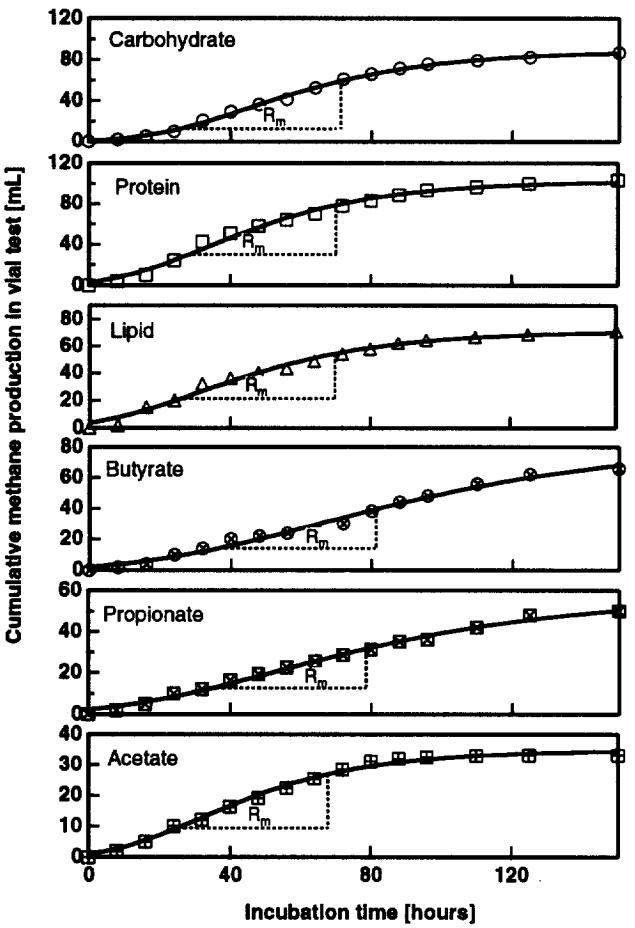

Figure 3. The determination of SMA for degrading various typical substrate. The R_m is the methane production rate in Eq. (1).

specific methanogenic activity, SMA_m. In Figure 4, the marks and the solid lines represented experimental data and the fitted model predictions by Eq. (2), indicating that Eq. (2) was suitable for describing the dynamics of methanogenic activity on a certain substrate, such as protein, lipid, carbohydrate and each individual VFA. The evaluated values of these key parameters were summarized in Table 4. In Table 4, the λ_i of lipid, 160 days, was larger than that of protein, 145 days, while both of them were significantly larger than those of carbohydrate and VFAs. Similar results were obtained for a batch high-solids sludge digestion. The lag-phase time of methane production might reflect a period of hydrolysis/fermentation, which corresponds to the rate of hydrolysis/fermentation of an anaerobic microbial process (Lay et al., 1997b). During this period, the pH value in the leachate of the LFBR was around 6.0 or below. The pH of the system is important for the optimal growth of proteins/lipids-degrading bacteria. For example, in the continuous culture study of entile ruminal contents, the proteins-degrading bacteria are washed out of the system as the pH dropped to 5.5 (Dürre and Andreesen, 1982). This result was due to the fact that the mediation of exo-enzymes was excreted by fermentative bacteria. The protein is degraded to amino acids, the carbohydrate is transformed into soluble sugars and the lipid is converted to long chain fatty acids and glycerine (Gujer and Zehnder, 1983). Moreover, the results were in agreement with those previously obtained from a mesophilic batch digester fed with rice and potato, in which the main component is carbohydrates, at different moisture contents (Lay et al., 1996a), the growth of methanogens in the low moisture content of solid-beds were inhibited by the low pH or high level of VFA. In Figure 4, the SMA approaches to the SMA* at a critical incubation time, λ^*. Each λ^* for the substrate of acetate, propionate, butyrate, carbohydrate, protein and lipid was 95, 100, 94, 92, 190 and 200 days. Apparently, the λ^* of the protein and the lipid were larger than those of acetate, propionate, butyrate, and the carbohydrate. Considering

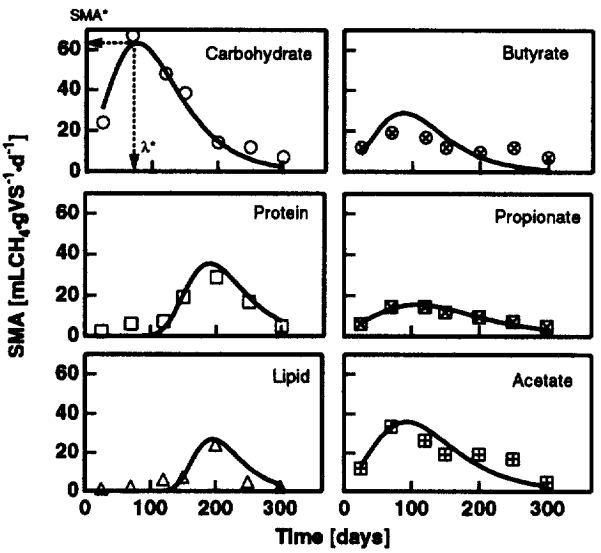

Figure 4. Dynamics of SMA degrading each individual substrate in the LFBR.

Table 4. Summary of Key Parameters for Describing Dynamics Curves using Eq. (2).

		Acetate	Propionate	Butyrate	Carbohydrate	Protein	Lipid
$R_{m,i}$	(mL CH_4• gVS^{-1}• d^{-1})	36.0	15.6	28.8	39.0	28.0	20.0
λ_i	(days)	33.0	30.0	36.0	25.0	145.0	160.0
λ^*	(days)	95.0	100.0	94.0	92.0	190.0	200.0

these results together with the λ, the decomposition of proteins and lipids might overrule the LFBR treating the OFMSW. Additionally, as listed in Table 4, the SMA_m of acetate was higher than those of propionate and butyrate. In general, the products of acetogenesis, such as propionate and butyrate, are converted into the final products for methane production: acetate, hydrogen and carbon dioxide. There is a symbiotic relationship between acetogens and methanogens due to the fact that the activity of acetogens depend on the substrate input and the activity of the methanogens (Shink, 1989). As a result, the examination of the methanogenic activities of acetate, propionate and butyrate suggested that the rate of methanogenesis was higher than that of acetogenesis during the anaerobic mineralization of the OFMSW.

Methane production rate in the LFBR

The LFBR stabilization is a dynamics and microbially-mediated process. Because proteins, lipids and carbohydrates are the main fermentative components of the OFMSW, the overall methane production rate in the LFBR can be defined as the summation of methane production rates on each component including the protein, lipid and carbohydrate. In order to elucidate the relationship between the decomposition of the OFMSW and the methane production rate in the LFBR, the methane production rate estimated using Eq. (2) was compared with that of observation (Figure 5). An examination of Figure 5 reveals that the correlation coefficient, r2, between the methane production rate of the observation and that of estimation was larger than 0.90, illustrating that the former approximately coincided with the latter. By comparing Figures 4 and 5 reveals that the first peak of the methane production rate in Figure 5 was mainly caused by the degradation of the carbohydrate, while the second peak was mostly caused by those of the protein and the lipid. It has been reported that the LFBR stabilization needs to proceed through the methane fermentation phase and into the final maturation phase (Pohland, 1996). Obviously, in this study, the stabilization of the OFMSW in an LFBR was significantly influenced by the conversion of the protein and the lipid into methane. Thus, the period of λ* (refers to Figure 4) of the protein and the lipid of the sludge contained in the LFBR significantly overruled the efficiency of the LFBR treating the OFMSW.

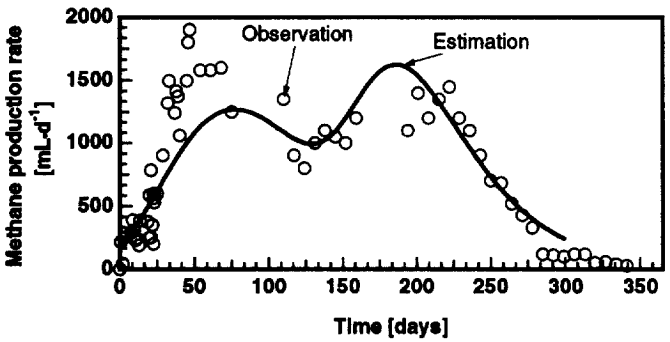

Figure 5. Observed and estimated methane production rates in the LFBR.

CONCLUSIONS

Based on the results and discussion, the principle conclusions derived from this investigation are as follows:

(1) The simple model used in this study was suitable to describe the dynamics of methanogenic activity for the LFBR treating the OFMSW. By using this model, the key parameters, maximum methane production rate and lag-phase time, for the OFMSW decomposition can be exactly estimated.

(2) According to the dynamics of methanogenic activities and each individual substrate degradation conducted in this study, the decomposition of proteins and lipids might overrule the stabilization of the LFBR treating the OFMSW; while the rate of methanogenesis was higher than that of acetogenesis.

(3) Experimental and estimated data, confirmed by the dynamics of methanogenic activity, suggested that the period of λ^* of the protein and the lipid of the sludge contained in the LFBR significantly influenced the efficiency of the LFBR treating the OFMSW.

ACKNOWLEDGMENTS

The authors wish to thank Mr. S. Agatsuma for his excellent technical assistance. During the time much of this work was performed, Y.Y. Li was an associate professor in the Dept. of Civil Engineering, Faculty of Engineering, Tohoku University. We would also like to thank Prof. S. Ghosh for his discussions on the research proposal.

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