Effects of Thermal Pretreatment and Trace Metals on High-rate Thermophilic Anaerobic Digestion of Sewage Sludge

M. Takashima*, E. Nakakihara** and R. Ikemoto**

* Department of Architecture, Civil and Environmental Engineering, Fukui University of Technology, 3-6-1 Gakuen, Fukui 910-8505, Japan (E-mail: *takasima@fukui-ut.ac.jp*)
** Graduate School of Natural Science and Engineering, Kanazawa University, Kakuma, Kanazawa 920-1192, Japan (E-mail: *nakaeri@staff.kanazawa-u.ac.jp*)

Abstract
This study attempted high-rate thermophilic anaerobic digestion of sewage sludge at 10 days HRT, by applying thermal pretreatment of waste activated sludge (170°C and 1hr) and trace metal addition (0.49 mg-Ni/L and 0.54 mg-Co/L). The thermal pretreatment not only increased VS and VSS destruction by 6% and 9%, respectively, but also improved dewaterability of digested sludge drastically. The trace metal addition showed negligible effect on the overall performance, except for the reduced VFA concentrations in the digested sludge. DGT analyses suggested that 70-88% of soluble Ni and only 5-10% of soluble Co be present in a bioavailable form. Microbial analyses based on PCR-DGGE detected the archaea, *Methanosarcina sp.* and *Methanosarcina thermophila*, in the digested sludge.

Keywords
DGT; sewage sludge; thermal pretreatment; thermophilic anaerobic digestion; trace metals

INTRODUCTION

Utilization of waste biomass has been propelled worldwide, particularly from the point of view of global warming. In this regard, anaerobic digestion is superior technology to enable not only the reduction of waste biomass, but also the recovery of energy and resources. For sewage sludge, the technology development has been continued to overcome its poor biodegradability and to accelerate the rate of anaerobic microbial reactions and thus reduce the digester volume. For instance, high-solid digestion, thermophilic digestion and pretreatment of influent sludge are of practical importance. Researches are still undertaken towards the anaerobic digestion handling higher loads.

In this study, we attempted the high-rate thermophilic anaerobic digestion at a short HRT of 10 days. For this purpose, thermal pretreatment of waste activated sludge and addition of trace metals were employed. The former method, suggested in the 80s by Hiraoka et al. (1981), can destroy the waste activated sludge with a refractory nature, and has a merit to reuse the heat for maintaining the digester temperature as well. Among the essential and important trace metals, Ni and Co were selected for supplementation, and others were assumed to be present enough in sewage sludge. The PCR-DGGE method was also applied to analyze Archaeal community in the thermophilic digesters.

METHODS

Sewage sludge and thermal pretreatment

The sewage sludge used was a mixed primary and waste activated sludge. Each sludge was taken from a municipal combined wastewater treatment plant in Fukui City, Japan, further thickened centrifugally to about 4.5% TS in the laboratory, and mixed at the ratio of 3:2 in Run 1. In Run 2, the waste activated sludge was thermally treated at 170°C for 1 hour, using an autoclave (TZA100-15K-LG, Unicontrols), and then mixed with the primary sludge at the same ratio as Run 1.

Experimental apparatus and operation

Two glass-made separable flasks of 3.0 L were used as the anaerobic thermophilic digesters. They were maintained at 55^{o}C and rotated at about 100 rpm. The sewage sludge was fed hourly with a tubing pump (7553-80, Cole-Parmer) to set the HRT at 10 days. The biogas was measured with a wet gas meter (WS-1A, Sinagawa). One of the two reactors was used as a control. The other was added 1.5 mL/d of a metal solution. The metal solution contained $NiCl_2 \cdot 6H_2O$ and $CoCl_2 \cdot 6H_2O$ whose amount was equivalent to the concentration of 0.49 mg-Ni/L and 0.54 mg-Co/L in the feed.

Analytical procedures

Most of the analyses were performed in accordance with Standard Methods (APHA/AWWA/WEF, 1998). VFA and biogas were analyzed with a LC applying a post pH-buffered method (LC-6A, Shimadzu) and a GC with a thermal conductivity detector (GC-9A, Shimadzu), respectively.

Dewaterability. The dewaterability of digested sludge was evaluated with the capillary suction time (CST; Standard Methods 2710G, ultimately expressed as sec-CST per g/L-SS) as well as the water content of centrifuged digested sludge (2,000g and 5 min).

Metals. A polarized Zeeman atomic adsorption spectrometry (Z-5010, Hitachi) was used to determine the concentration of the trace metals examined. The sludge samples for the total metal concentration were prepared by the nitric acid-hydrochloric acid digestion method (Standard Methods 3030F). To estimate the bioavailable concentration of the metals, DGT (Diffusive gradients in thin-films) method was applied with DGT samplers (DGT Research).

Archaeal community. On the last day of each run, DNA of the digested sludge was extracted using UltraClean Soil DNA Kit (Mo Bio Laboratories). The archaeal 16S rRNA partialgenes of the extracted DNA were amplified through PCR (2720Thermal cycler, Applied Biosystems) with the combinations of primers (Arc357f-GC and Arc691r; Watanabe et al., 2004), isolated through DGGE (Dcode; Bio Rad Laboratories), and sequenced using the BigDYE Terminator Kit (ver.3.1, ABI) and ABI PRISM 3130xl.

RESULTS AND DISCUSSION

For each run, the anaerobic digesters were operated for 70 days. With the average and standard deviation of the last six weeks data, the main results are summarized in Table 1.

Effects of thermal pretreatment of waste activated sludge

From Table 1, the destruction percent obtained was 48.3-49.3% for VS and 54.4-54.5% for VSS in Run 1, and, based on the sewage sludge without the thermal pretreatment of waste activated sludge, 54.0-55.2% for VS and 63.5-64.0% for VSS in Run 2. The application of thermal pretreatment improved VS by about 6% and VSS by about 9%. Approximately, the thermal pretreatment itself resulted in the destruction of 3% TS, 4% VS, 44% SS, 51% VSS and 8% COD of waste activated sludge. Gas production was 0.403-0.407 NL/gVS in Run 1 and 0.423-0.434 NL/g-VS in Run 2, resulting in an increase by about 6% with the thermal pretreatment. In the start-up period operated at 20 days HRT, recorded were VS destruction of 58.2%, VSS destruction of 64.8% and gas production of 0.449 NL/g-VS. Combined with the thermal pretreatment, the performance at 10 days HRT became comparable to that at 20 days HRT. These results were subjected to a statistical analysis using *t*-test at the significance level of 0.05. It shows a statistical significance between Run 1 and Run 2 for VS and VSS destruction. Therefore, the positive effect of thermal pretreatment on organic solid destruction was confirmed statistically.

There have been some reports on the thermophilic anaerobic digestion of high-solid sewage sludge (Kiyohara et al., 1998; Moen et al., 2003; Ferrer et al., 2010). And they showed relatively consistent VS destruction of 55.3-57.3% at 10 days HRT, 65.6% at 20 days HRT and 70.6% at 40 days HRT with 4.5-6% sewage sludge. The comparison to these references shows that the destruction percent obtained in this study appears to be lower. On the other hand, the total VFA concentration in the digested sludge varied significantly; around 200 mg/L for Kiyohara et al. (1998), >1,000 mg/L for Moen et al. (2003), and >5,000 mg/L for Ferrer et al. (2010).

Table 1. Influent and digested sludge

	Run 1			Run 2		
	Influent sludge	Digested sludge		Influent sludge[a]	Digested sludge	
		Control	Trace metal		Control	Trace metal
TS (g/L)	43.6±2.9	27.1±0.6	27.6±0.7	47.3±4.1	26.9±2.3	27.7±1.3
SS (g/L)	39.3±2.7	22.3±0.9	22.8±1.3	42.2±4.1	20.9±2.1	21.2±2.2
VS (g/L)	33.1±2.2	17.1±0.6	16.8±0.6	37.7±2.9	16.9±1.6	17.3±1.1
VSS (g/L)	28.8±2.5	13.1±0.5	13.1±0.8	33.4±2.8	12.2±1.2	12.0±1.3
COD (g/L)	55.2±3.5	26.6±1.2	26.6±0.6	62.8±1.5	28.1±3.1	27.6±2.2
Sol. COD (g/L)	3.2±1.0	1.3±0.4	1.1±0.2	4.0±1.5	4.6±0.6	4.1±0.6
Total VFA (mg-COD /L)[b]	—	90±45	29±18	—	668±293	520±270
Biogas production (NL/g-VS)	—	0.403±0.031	0.407±0.026	—	0.423±0.046	0.434±0.049
CH_4 (%)	—	60.6±1.2	60.6±0.8	—	59.9±1.1	59.8±0.9
CO_2 (%)	—	36.4±0.4	36.1±0.6	—	36.7±0.7	36.7±0.7
VS destruction (%)	—	48.3±3.3	49.3±2.6	—	55.2±3.1	54.0±2.9
VSS destruction (%)	—	54.5±3.0	54.4±3.4	—	63.5±3.4	64.0±3.6
COD destruction (%)	—	51.8±2.1	51.7±2.6	—	55.3±4.2	56.0±3.3
COD recovery (%)	—	90.1±6.0	90.7±6.2	—	87.4±5.7	87.7±6.3
CH_4 recovery (COD%)	—	41.8±2.6	42.3±2.4	—	42.6±2.8	43.6±3.3

a) Before waste activate sludge was thermally treated. b) Sum of acetate, propionate, i- and n-butyrate and i- and n-valeric acid.

As shown in Fig. 1, soluble COD, color, CST per SS and water content of centrifuged digested sludge, were changed significantly in Run 2. Soluble COD and color were increased nearly by 4 times. Since total VFA also increased as shown in Table 1, the increase of soluble COD is attributable to VFA and color materials. In Run 2, CST per SS was reduced to half and water content was decreased by about 10%. Therefore, even if thermal pretreatment is conducted to only waste activated sludge, dewaterability can be improved greatly.

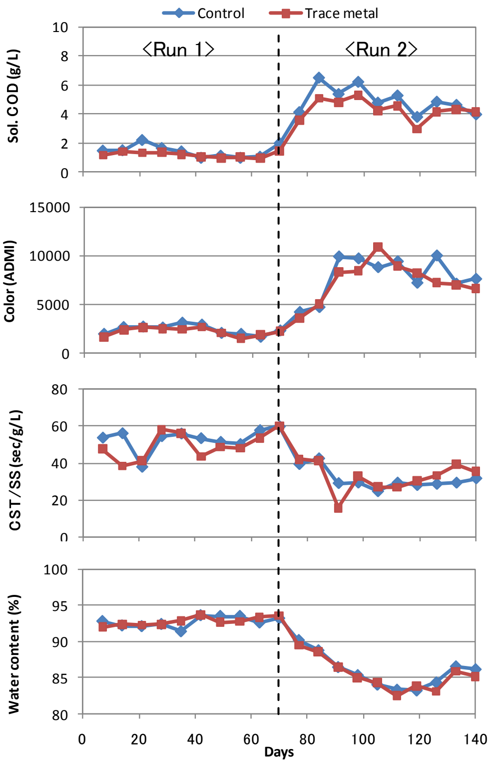


Figure 1. Time course of sol. COD, color, CST/SS and water content after centrifugation of digested sludge.

Effects of trace metals and their bioavailability

Solid reduction and biogas production were almost identical between the control and trace metal addition. The VFA concentration tends to be lower with the trace metal addition, but it was statistically significant only in Run 1. Thus, it is considered that the trace metals were supplied from the sewage sludge almost sufficiently.

Compared with the requirement reported by Takashima et al. (2011), 0.049 mg-Ni/g-COD and 0.054 mg-Co/g-COD, the Co content in this study, 0.021 mg-Co/g-COD, could be insufficient. In addition, as shown in Table 2, 70-88% of soluble Ni was DGT-labile, whereas was 5-10% of soluble Co in the digested sludge. Thus, Co may be suspected to be low bioavailability and even low content for anaerobic thermophilic microorganisms.

Archaeal community

Figure 1(a) shows the DGGE banding profile. There is no difference in banding pattern between the control and trace metal addition, suggesting that a similar archaeal community was existent. Figure 1(b) shows the phylogenetic tree of the arcaeal 16S rRNA sequences detected. The DNA sequences of bands A1-A2 were most closely related to *Methanosarcina sp.* and *Methanosarcina* *thermophile.* According to the review by de Vrieze et al. (2012), *Methanosarcina sp.* can utilize hydrogen, and the metabolic pathway involving acetate oxidation to hydrogen and hydrogen-utilizing methanogenesis may be advantageous over the acetate-utilizing methanogenesis. Further detailed analyses are needed for the microbial community and metabolic pathways.

CONCLUSIONS

In this paper, we focused on the thermal pretreatment of waste activated sludge and trace metal addition in the thermophilic, high-solid anaerobic digestion operated at 10 days HRT.

1) The thermal pretreatment of waste activated sludge (170^{o}C, 1 hour) improved VS destruction by about 6% and VSS destruction by about 9%, which was close to the performance at 20 days HRT. Although soluble COD and color of the digested sludge was deteriorated, dewaterability of the digested sludge was improved drastically.

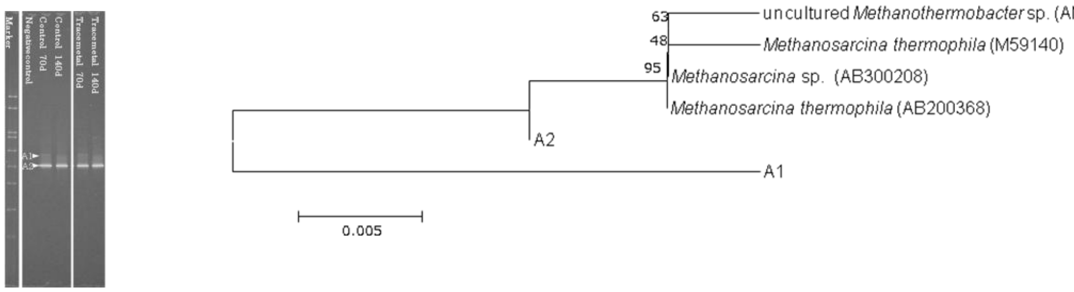
2) The addition of Ni and Co was useful for reducing the VFA concentration of the digested sludge, while showing nil effects on the solid destruction.

3) The DGT analysis showed that 70-88% of soluble Ni and 5-10% of soluble Co were DGT-labile in the digested sludge. Co was suspected to be low bioavailability and even low content.

4) The microbial analysis based on PCR-DGGE showed the archaeal community of less versatile, and detected *Methanosarcina sp.* and *Methanosarcina thermophila* in the digested sludge.

Table 2. Trace metal concentrations in digested sludge.

	Ni				Co			
	Run 1		Run 2		Run 1		Run 2	
	Control	Trace metal	Control	Trace metal	Control	Trace metal	Control	Trace metal
Soluble conc. (μg/L)	123±15	127±16	138±9	154±25	17.2±1.0	22.4±2.4	15.5±1.4	22.2±2.4
DGT-labile conc. (μg/L)	97±17	113±20	96±9	112±22	0.9±0.2	1.4±0.3	0.9±0.8	2.1±2.1
% DGT-labile conc. in soluble conc.	79±5	88±5	70±10	73±14	5±1	6±2	6±6	10±11
DGT-labile conc. for trace metal addition against that for control	—	1.2	—	1.2	—	1.5	—	2.3



(a) DGGE banding profile (b) Phylogenetic tree of Archaea

Figure 2. Results of Archaeal community analysis.

REFERENCES

Ferrer, I., Vazquez, F. and Font, X. 2010 Long term operation of a thermophilic anaerobic reactor: Process stability and efficiency at decreasing sludge retention time. *Bioresource Technology* **101**, 2972-2980.

Hiraoka, M., Takeda, N., Ohsumi, K. and Oku, K. 1981 Study on anaerobic digestion system of sewage sludge (I). *Environmental Technology*. **10**(9), 691-701 (in Japanese).

Kiyohara, Y., Li, Y., Miyahara, T., Mizuno, O. and Noike, T. 1998 Treatment characteristics of high strength sewage sludge by thermophilic anaerobic digestion. *Proceedings JSCE*. No. 601/VII-8, 35-43 (in Japanese).

Moen, G., Stensel, H. D., Lepistö, R. and Ferguson, J. F. 2003 Effect of solid retention time on the performance of thermophilic and mesophilic digestion of combined municipal wastewater sludges. *Wat. Environ. Res.* **75**, 539-548.

Takashima, M., Shimada, K. and Speece, R. E. 2011 Minimum requirements for trace metals (Fe, Ni, Co and Zn) in thermophilic and mesophilic methane fermentation from glucose. *Wat. Environ. Res.* **83**, 339-346.

Watanabe, T., Asakawa, S., Nakamura, A., Nagaoka, K. and Kimura, M. 2004 DGGE method for analyzing 16S rDNA of methanogenic archaeal community in paddy field soil. *FEMS Microbiology Letters* **232**, 153-163.