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Stimulation of methanogenesis in anaerobic digesters treating leachate from a municipal solid waste incineration plant with carbon cloth

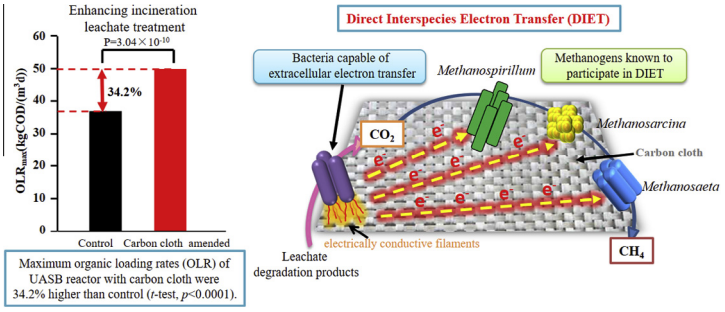
Yuqing Lei [a], Dezhi Sun [a], Yan Dang [a,*], Huimin Chen [a], Zhiqiang Zhao [c], Yaobin Zhang [c], Dawn E. Holmes [b]

[a] Beijing Key Laboratory for Source Control Technology of Water Pollution, Engineering Research Center for Water Pollution Source Control and Eco-remediation, College of Environmental Science & Engineering, Beijing Forestry University, 35 Tsinghua East Road, Beijing 100083, China
[b] Department of Physical and Biological Sciences, Western New England University, 1215 Wilbraham Rd, Springfield, MA 01119, United States
[c] Key Laboratory of Industrial Ecology and Environmental Engineering (Dalian University of Technology), Ministry of Education, School of Environmental Science and Technology, Dalian University of Technology, Dalian 116024, China

HIGHLIGHTS

- UASB reactor with carbon cloth permitted a 34% higher OLR than control.
- VFAs were more efficiently transformed in the carbon cloth-amended reactor.
- Bacteria with extracellular electron transfer capabilities were enriched.
- Methanogens known to participate in DIET were enriched on the carbon cloth.
- These results are applicable to practical leachate treatment projects.

GRAPHICAL ABSTRACT



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ABSTRACT

Bio-methanogenic digestion of incineration leachate is hindered by high OLRs, which can lead to build-up of VFAs, drops in pH and ultimately in reactor souring. It was hypothesized that incorporation of carbon cloth into reactors treating leachate would promote DIET and enhance reactor performance. To examine this possibility, carbon cloth was added to laboratory-scale UASB reactors that were fed incineration leachate. As expected, the carbon-cloth amended reactor could operate stably with a 34.2% higher OLR than the control (49.4 vs 36.8 kg COD/(m^3 d)). Microbial community analysis showed that bacteria capable of extracellular electron transfer and methanogens known to participate in DIET were enriched on the carbon cloth surface, and conductivity of sludge from the carbon cloth amended reactor was almost twofold higher than sludge from the control (9.77 vs 5.47 μS/cm), suggesting that microorganisms in the experimental reactor may have been expressing electrically conductive filaments.

1. Introduction

Leachate generated by MSW incineration plants is a typical organic wastewater with extremely high concentrations of organic matter (Demirel and Scherer, 2008; Ye et al., 2011). Bio-methanogenic reactors are relatively effective at treatment and recovery of energy from fresh leachate. However, accumulation

Abbreviations: OLRs, organic loading rates; DIET, direct interspecies electron transfer; MSW, municipal solid waste; COD, chemical oxygen demand; BOD, biochemical oxygen demand; VFAs, volatile fatty acids; GAC, granular activated carbon; UASB, up-flow anaerobic sludge bed; EGSB, expanded granular sludge bed; VSS, volatile suspended solids; TSS, total suspended solids; HRT, hydraulic retention time; PCR, polymerase chain reaction; STP, standard temperature and atmospheric pressure.

* Corresponding author.
E-mail addresses: yandang@microbio.umass.edu, dangyan.p@gmail.com (Y. Dang).

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of VFAs and drops in pH during the treatment process can inhibit methanogenesis and significantly reduce treatment efficiencies (Liu et al., 2010; Ye et al., 2011). Therefore, it is important to explore methods that can enhance performance of bioreactors treating wastewaters with high OLRs such as MSW incineration leachate.

Methane produced by anaerobic digesters treating MSW leachate results from complex interactions between multiple bacterial and archaeal species. Fermentative microorganisms break down complex organic matter to polymers, which are then hydrolyzed by other organisms to their monomeric subunits (amino acids, monosaccharides, long chain fatty acids) and fermented to alcohols, aldehydes and VFAs (Miyamoto, 1997). These compounds are then degraded by other groups of bacteria to acetate, hydrogen, and carbon dioxide, which can be used as substrates for acetoclastic and hydrogenotrophic methanogenesis (Demirel and Scherer, 2008). In addition to the more traditional methanogens that use fermentative end-products for methanogenesis, other populations of methanogens can generate methane by directly accepting electrons from bacteria that are capable of extracellular electron transfer (Rotaru et al., 2014a,b). This syntrophic partnership between bacteria capable of extracellular electron transfer and electron accepting methanogens is referred to as DIET (Demirel and Scherer, 2008; Stams et al., 2006).

Over the years, much research on anaerobic bioreactors has focused on interspecies hydrogen transfer within these systems (Stams et al., 2006). However, recent studies have shown that a significant proportion of methane generated by anaerobic digesters can be attributed to DIET and that stimulation of DIET within these systems can enhance methane output (Rotaru et al., 2014b). Studies have also shown that DIET can be stimulated in bioreactors by the addition of non-biological conductive materials such as magnetite (Kato et al., 2012; Li et al., 2015), biochar (Chen et al., 2014b), GAC (Liu et al., 2012), and carbon cloth (Chen et al., 2014a; Zhao et al., 2015).

Most studies that have shown that DIET can be facilitated by addition of conductive materials have focused on degradation of simple substrates, such as ethanol (Rotaru et al., 2014b), acetate (Lee et al., 2016), propionate (Cruz Viggi et al., 2014) and butyrate (Li et al., 2015). However, to the best of our knowledge, no studies have examined how conductive materials can stimulate DIET in anaerobic digesters treating incineration leachate which has a much more practical significance. Incineration leachate contains much higher concentrations of complex organic matter and VFAs than the simple substrates fed to the bioreactors mentioned above (Ye et al., 2011). Based on the significantly more complex nature of fresh leachate, it is possible that microbial interactions involved in methane production by anaerobic bioreactors treating incineration leachate differ from those previously studied. Therefore, analytical and molecular approaches were used to investigate whether addition of carbon-cloth to laboratory-scale UASB reactors treating MSW incineration leachate could enhance bio-methanogenic treatment and promote DIET.

2. Materials and methods

2.1. Inoculated sludge and fresh leachate

Laboratory-scale UASB reactors were inoculated with 360 mL anaerobic suspended sludge collected from a full-scale UASB reactor treating food wastewater in Beijing, China. The VSS/TSS ratio of this sludge was 0.66. Fresh leachate collected from an MSW-to-energy incineration plant in Beijing, China was stored at 4 °C. Composition of the leachate is shown in Table 1.

Table 1
Characteristics of leachate from a MSW incineration plant in Bejing, China.

Item	mg/L	Item	mg/L
COD	70,390–75,480	Ca	3275–5827
BOD_5	39,250–46,458	Na	1278–2349
NH_4^+-N	1042–1395	Mg	463–1598
TN	1330–2179	Fe	59.1–679.9
TP	104.6–163.8	Mn	4.56–43.5
Cl^-	3978–7153	Zn	13.9–36.5
SO_4^{2-}	1833–2907	Pb	1.11–7.61
pH	4.58–6.42 (no unit)	Ni	0.91–2.3

2.2. Reactor design

Two identical continuous-flow UASB reactors with working volumes of 1200 mL were operated stably at 33 ± 1 °C. A 10 L gas-sampling bag was attached to the gas outlet from the three-phase separator found at the top of each reactor. Ten pieces of carbon cloth (Kureha enemical; $10 \times 5 \times 0.1$ cm^3, 100 cm^2 for the front and back area, conductivity of 4.35 S/cm) were suspended in sludge at the bottom of experimental reactor. Diluted leachate (raw leachate diluted with tap water as needed) was added to both reactors with a peristaltic pump. Reactors were continuously fed leachate at an HRT of 0.8 day and OLRs were gradually increased from 3.1 kg COD/(m^3 d) to 73.3 kg COD/(m^3 d).

2.3. Analytical methods

The dichromate reflex method was used to monitor the COD of various samples (Moore et al., 1949). The BOD_5 of samples was determined with the dilution method, and solids (TSS and VSS) were measured in a Gooch crucible according to standard methods (APHA, 2005). VFAs (acetate, propionate and butyrate) were measured with high-performance liquid chromatography (Waters e2695, USA) with an eluent of 8 mM H_2SO_4 according to previously described methods (Nevin et al., 2008). The gas volume in the gas-sampling bag was measured with a digital mass flow meter (FMA4000; Omega; USA) every 12 h.

Methane and carbon dioxide were detected with a gas chromatograph (Agilent 7890A, USA) equipped with a flame-ionization detector and Porapak Q column with detector and injection port temperatures of 260 °C and 60 °C, respectively (Lei et al., 2016). The methane production rate, expressed as L(STP)/d was calculated according to methods reported by Borja et al. (2004). pH was recorded with a pH meter (HACH, USA), conductivity of the carbon cloth was measured with a conductivity meter (Leici, DDS-307, China), and conductivities of suspended sludge samples were measured with previously described methods (Zhao et al., 2016c).

2.4. High-throughput sequencing

Sludge samples (1.0 g) and carbon cloth material (1×1 cm^2) were collected at the end of operation when the reactors were still in stable condition (control reactor on day 42 and carbon cloth reactor on day 51). All samples were washed with 2 mL phosphate-buffered saline (PBS; 0.13 M NaCl and 10 mM Na_2HPO_4 at pH 7.2) by centrifugation at 4000 rpm for 2 min. The supernatant was then discarded and pellets were resuspended in 1 mL PBS buffer and 122 μL MT buffer (MP Bio). Lysing Matrix E (MP Bio) (0.5 g) was added to the suspension and samples were homogenized for 1 min at 5.5 m/s in a mini-beadbeater (Biospec, USA). Samples were then centrifuged for 15 min at 13,200 rpm and total DNA was extracted with the FastDNA kit for soil (QBIOgene, CA) according to the manufacturer's instructions. A micro volume spec-