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Short Communication

Continuous fermentation of food waste leachate for the production of volatile fatty acids and potential as a denitrification carbon source

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HIGHLIGHTS

- Effects of HRT and pH on fermentative production of VFAs from FWL were examined.
- The VFA yield was revealed to be much more significantly affected by pH than HRT.
- The HRT and pH for maximum VFA yield were estimated and experimentally validated.
- The fermentation filtrate outperformed conventional C sources for denitrification.

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ABSTRACT

This study investigated the simultaneous effects of hydraulic retention time (HRT) and pH on the continuous production of VFAs from food waste leachate using response surface analysis. The response surface approximations ($R^2 = 0.895$, $p < 0.05$) revealed that pH has a dominant effect on the specific VFA production (P_{TVFA}) within the explored space (1–4-day HRT, pH 4.5–6.5). The estimated maximum P_{TVFA} was 0.26 g total VFAs/g COD_f at 2.14-day HRT and pH 6.44, and the approximation was experimentally validated by running triplicate reactors under the estimated optimum conditions. The mixture of the filtrates recovered from these reactors was tested as a denitrification carbon source and demonstrated superior performance in terms of reaction rate and lag length relative to other chemicals, including acetate and methanol. The overall results provide helpful information for better design and control of continuous fermentation for producing waste-derived VFAs, an alternative carbon source for denitrification.

1. Introduction

Volatile fatty acids (VFAs) are short-chain fatty acids composed of six or fewer carbon atoms and are volatile at atmospheric pressure (Lee et al., 2014). These carboxylic acids have many applications in the production of bioplastics, biochemicals, and biofuels and can also be used as energy and carbon sources for biological removal of nutrients (Chang et al., 2010). Commercial production of VFAs has depended largely on chemical routes using petrochemicals as raw materials. Although biological production of VFAs via fermentation is attracting increasing attention with growing concerns about environmental pollution and energy crisis, the use of sugars or starch from edible crops as the main substrate raises the issues of economic viability and food security. Using more economical feedstocks is therefore a key to resolving this limitation, and acidogenic fermentation of organic waste, such as food waste and sewage sludge, has been explored as a promising approach to more environment-friendly and economical production of VFAs. Such an approach can also contribute to the minimization of waste disposal.

One of the most practical uses of waste-derived VFAs is providing carbon source for denitrification. Biological nitrogen removal is generally accomplished by aerobic nitrification (i.e., oxidation of NH_4^+ to NO_3^-) followed by anoxic denitrification (i.e., reduction of NO_3^- to N_2). Denitrification is a heterotrophic bioprocess, while nitrification is an autotrophic bioprocess, and therefore, the availability of usable carbon sources is a crucial factor for effective nitrogen removal. However, available organic carbon is often not sufficient in municipal wastewater, and carbon sources are externally added to increase the C/N ratio for denitrification. Adding external carbon sources (most often methanol) is an expensive operation that substantially increases the operating costs of wastewater treatment plants (Jiang et al., 2013). As the demand

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for nitrogen removal increases with the introduction of more stringent effluent standards worldwide, there is a critical need to develop sustainable and economical carbon sources for denitrification such as waste-derived VFAs (Frison et al., 2013).

Production of VFAs by mixed-culture fermentation has been extensively explored using various organic wastes as feedstock (Chang et al., 2010; Jiang et al., 2013; Lee et al., 2014). In anaerobic mixed-culture processes, VFAs are readily utilized by microbes and mineralized to CH_4 and CO_2 via methanogenesis. Therefore, continuous acidogenic fermentation can be achieved by controlling the operating parameters to facilitate hydrolysis and acidogenesis while preventing methanogenesis. Such process control can be of particular importance when using readily utilizable organic wastes, which often contain diverse microbes in abundance, as feedstock. Hydraulic retention time (HRT) and pH, regarded as among the most influential operating conditions on the functional attributes of a bioprocess, have often been employed as control factors to facilitate the desired fermentation process while preventing methanogenesis (Min et al., 2005; Lim et al., 2008; Jiang et al., 2013; Lee et al., 2014; Kumar et al., 2015).

This study aims to examine the simultaneous effects of HRT and pH on the fermentative production of VFAs from food waste leachate (FWL), a sustainable fermentation feedstock, and locate the optimum operating conditions for maximum VFA yield. Given the high organic and nutrient contents of FWL and the increasing production of food waste worldwide (Thi et al., 2015), FWL is an attractive feedstock for bioconversion. Correlations between HRT or pH and the VFA productivity were analyzed by response surface analysis (RSA), an effective statistical tool to investigate simultaneous effects of multiple variables and to construct response models. Additionally, the fermentation filtrate recovered from the culture under the estimated optimum conditions was examined for its potential as an external carbon source for denitrification.

2. Methods

2.1. RSA design and FWL fermentation

A total of 11 continuous fermentation trials were conducted under nine different HRT and pH conditions according to the face-centered response surface design (Fig. S1 and Table 1). The design consisted of nine experimental runs with the center point being triplicated. The explored ranges of HRT and pH were set to 2.5 ± 1.5 days and 5.5 ± 1.0, respectively, with reference to the literature (Han and Shin, 2002; Min et al., 2005; Jiang et al., 2013). RSA was carried out as previously described (Jung et al., 2016) through a sequential procedure of fitting the experimental data to increasingly complex polynomials and selecting the best-fit model using Design Expert 7 software (Stat-Ease, Minneapolis, USA). In this study, specific VFA production (P_{TVFA}), defined as the amount of total VFAs (C_2–C_6) produced per unit mass of substrate chemical oxygen demand (COD) fed (g total VFAs/g COD_f), was chosen as the dependent variable of interest for RSA approximation. The detailed design information for RSA is provided as both actual and coded values for ease of comprehension in Table 1.

Table 1
Experimental design for response surface analysis and observed data.

Run	HRT (days)		pH		P_{TVFA} (g total VFAs/g COD_f)	
	Actual	Coded	Actual	Coded	Observed	Predicted[a]
1	1	−1	4.5	−1	0.06	0.04
2	4	1	4.5	−1	0.08	0.06
3	1	−1	6.5	1	0.24	0.22
4	4	1	6.5	1	0.22	0.20
5	1	−1	5.5	0	0.09	0.11
6	4	1	5.5	0	0.08	0.11
7	2.5	0	4.5	−1	0.06	0.10
8	2.5	0	6.5	1	0.24	0.26
9[b]	2.5	0	5.5	0	0.18 (0.01)	0.16

[a] Predicted model response.
[b] Center point was triplicated. Observed value is presented as the average with standard deviation in parentheses.

For each fermentation trial, a continuous reactor with a working volume of 1 L was operated anaerobically at a mesophilic temperature of 35 ± 2 °C. FWL, which was used as a substrate for fermentation, was collected from the bottom drainage of a food waste storage hopper at a local biogas plant digesting 100 tons of food waste and 50 tons of cattle manure daily. Each reactor was initially filled with FWL and started without inoculation. Steady-state data were collected for each trial, and when the reactor showed a stable performance after at least three turnovers of the HRT, the different operating conditions were compared. The observed data were used to model the response of P_{TVFA} to variations in HRT and pH within the design boundary. To experimentally validate the model prediction, triplicate reactors were run under the estimated optimum operating conditions for maximum P_{TVFA}.

2.2. Batch denitrification tests

Denitrifying sludge collected from an anoxic tank of a full-scale biological nitrogen removal process was used as inoculum. Fermentation filtrate (pore size, 0.2 μm) was recovered from the FWL fermenters run at the estimated optimum HRT and pH. The mixture of the filtrates was examined for its potential as a denitrification carbon source in comparison with acetate, methanol, and a commercial product (OC, ETEC Korea, Korea) used in the full-scale facility from which the inoculum biomass was obtained. Batch denitrification tests were conducted in serum bottles for each carbon source in triplicate. Each bottle was prepared with synthetic nitrate wastewater (final concentration, 50 mg NO_3^-–N/L as KNO_3), a carbon source to test (final concentration, 350 mg COD/L), and denitrifying sludge (inoculation ratio, 2% v/v) in a total working volume of 100 mL. The concentration of inoculated denitrifying biomass was 172 mg volatile suspended solids (VSS)/L. The initial COD/N ratio was set at 7 in the test bottles with reference to the literature (Xie et al., 2012). In addition, a negative control without an added carbon source was prepared in triplicate to compensate for the background denitrification. All bottles were flushed with nitrogen to remove headspace oxygen and sealed with rubber stoppers and aluminum crimps. The initial pH of the mixture ranged between 7.3 and 7.7 in the test bottles, and no pH adjustment was made. All bottles were incubated at room temperature with intermittent manual shaking at 3-h intervals. Samples for monitoring the denitrification process were taken from each bottle using a syringe with a needle. The observed NO_3^-–N and NO_X–N removal profiles were further described by fitting the observed denitrification data to a modified Gompertz equation (Eq. (1)) for each carbon source tested

$$S_0 - S = S_0 \cdot \exp\left[-\exp\left\{\frac{R_m \cdot e}{S_0}(\lambda - t) + 1\right\}\right] \quad (1)$$

where S_0 is the initial nitrogen concentration (mg/L), S is the residual nitrogen concentration at time t (mg/L), R_m is the maximum nitrogen removal rate (mg/L d), λ is the lag phase length (h), and t is the incubation time (h).

2.3. Analytical methods

The COD concentration was measured using an HS-COD-MR kit (Humas, Daejon, Korea). Total nitrogen and total phosphorus con-