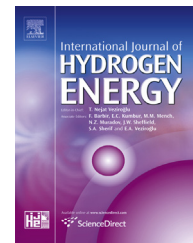


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Biohydrogen generation from anaerobic digestion of food waste

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ABSTRACT

Biohydrogen generated from the anaerobic digestion of a synthetic food waste with constant composition and a real food waste collected in Hong Kong were studied. This study aims at using a monoculture to increase biohydrogen production and determining optimum conditions for maximum biohydrogen production. Among the nine bacteria screened for biohydrogen production, *Escherichia cloacae* and *Enterobacter aerogenes* produced the largest amount of biohydrogen from the anaerobic digestion of synthetic food waste. The optimum anaerobic digestion conditions were determined: initial pH of 7, a water to solids ratio of 5 (w/w), a mesophilic temperature (37 ± 1 °C), and in the presence of 40 mg/L $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$. Anaerobic digestion at the optimum operating conditions using collected food waste with *E. cloacae* as the bacterial source was also performed. By adjusting the pH in the range of 5–6, a specific biohydrogen production of 155.2 mL/g of volatile solids (VS) in food waste was obtained.

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1. Introduction

The disposal of food waste has been receiving a lot of attention for two reasons. First, it takes up the limited landfill space. For example, in Hong Kong, about 3280 ton food waste, which accounts for 36% of the total solid wastes, is dumped to landfills every day and the amount of food waste disposed from restaurants has doubled in the last five years [1]. Second, it is a complete waste to dump a food waste with a high volatile solids (VS) content of 80–97 wt% which is readily biodegradable in an anaerobic process to generate biogas, namely biohydrogen and biomethane [2]. Therefore, converting food waste to energy is a topic pursued by many researchers [3,4].

Most studies used mixed sludge in anaerobic digestion to produce a biogas with 50–60% biomethane but contained little

biohydrogen. The biohydrogen produced by the hydrogen producing bacteria was consumed by hydrogen consuming bacteria such as methanogens to produce biomethane [5]. For example, Chen et al. [6] studied the anaerobic digestion of a mixed food waste collected from a soup processing plant, a cafeteria, a commercial kitchen, a fish farm and a grease trap collection service using anaerobic inoculum from an anaerobic digester. The biogas generated at a food to microorganisms ratio of 0.5 under mesophilic and thermophilic conditions for a residence time of 28 days contained 62% and 64% biomethane, respectively. Similar study had been carried out by Dearman and Bentham [7] using a mixed food waste collected from university kitchens, hospitals and markets, and the biogas generated during the 70 day digestion period contained a composition of biomethane ranging from 55 to 75%.

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Instead of collecting only biomethane from anaerobic digestion, it would be advantageous to collect the biohydrogen generated during anaerobic digestion before being consumed by the hydrogen consuming bacteria. This requires a two-stage reactor where biohydrogen is recovered in the first stage and the residual food waste is further degraded in the second stage of the reactor to produce biomethane.

Pretreated sludge which suppresses the activity of hydrogen consuming bacteria can be used in the first-stage reactor. Various pretreatment methods such as heat-shock [8], acid and base treatment [9] have been used for such purpose. Only biohydrogen and no biomethane was detected in the biogas generated in these studies. Wang and Wan [10] compared these pretreatment methods using a glucose solution and found that heat-shock treatment resulted in the highest biohydrogen production rate and biohydrogen yield.

Treating the mixed sludge by heat or the use of chemicals is costly in industrial applications. An option is to use a single species of bacteria such as *Enterobacter* spp., and *Clostridium* spp. which are capable of generating biohydrogen from food waste through dark fermentation in the first-stage reactor. However, most studies focusing on single species of bacteria used simple sugars such as glucose, xylose, and sucrose, or substrates rich in sugars such as sugarcane wastewater, sugar refinery waste, and orange processing effluent as the nutrient source [11–13]. A real food waste containing proteins and lipids [14] received limited coverage.

This paper studies the biohydrogen generation by a single species of bacteria in anaerobic digestion. As a control, a synthetic food waste with constant composition will be used in preliminary tests to screen out two bacteria, among the nine bacteria tested, that generate the highest amount of biohydrogen. The effect of operating parameters such as the initial pH of food waste, water to solids ratio, temperature, and the addition of inorganic salts on biohydrogen generation of the synthetic food waste will be studied using the two top-performing bacteria. Finally, anaerobic digestion of a real food waste collected locally will be studied with the best performed bacteria under the same optimum conditions.

2. Method and material

2.1. Food waste

The synthetic food waste was composed of rice 6% (w/w), bread 6% (w/w), vegetable 9.6% (w/w), pork 8.4% (w/w), eggs 2.4% (w/w), corn oil 1.2% (w/w), table salt 0.2% (w/w) and water 66.2% (w/w). The synthetic food waste was made up based on the typical carbon to nitrogen ratio and the moisture content of food wastes reported in the literature [2]. Collected food waste was obtained from two Chinese fast food restaurants at the Hong Kong University of Science and Technology. After removing bones and shells, food wastes were ground and mixed thoroughly in a blender (Westinghouse handheld blender) and stored in sealed glass bottles in a refrigerator at 4 °C. Before anaerobic digestion, the food waste was restored to the ambient temperature (22 ± 1 °C).

The characteristics of synthetic and collected food wastes such as total solids (TS), volatile solids (VS), moisture content,

and total phosphorus (TP) concentration were measured according to standard methods [15]. Elemental concentration (C, H, N, S) of the food waste samples were measured by an elemental analyzer (Elementar Analysensysteme GmbH vario EL III CHNS-mode) with sulfanilic acid as the standard. They were dried at 105 ± 5 °C for 24 h and were ground to particles smaller than 0.5 mm before measurement. pH of the food waste was measured by a pH meter (SevenMulti Mettler Toledo).

2.2. Microorganisms

Nine bacteria purchased from the Guangdong Microbial Culture Collection Center in China or the German Collection of Microorganisms and Cell Cultures were used in this study. These strains were selected for the screening study as they are commonly used for biohydrogen generation by anaerobic digestion [11,12]. These strains were grown under anaerobic conditions in various culture media (Table 1). In preparing a culture medium, sodium bicarbonate was added to a culture medium to adjust its pH. The medium was first boiled and was then sparged with N₂ for 30 min to remove any oxygen before autoclave.

2.3. Anaerobic digestion of food waste for biohydrogen production

Anaerobic digestion of synthetic or collected food waste was carried out in culture bottles and the schematic diagram of the experimental setup is shown in Fig. 1. The food waste was first added to a 100 mL culture bottle. Water was then added to obtain the desired water to solids ratio. 1 M NaHCO₃ was used to adjust its initial pH to the target pH. FeSO₄·7H₂O or NiCl₂·6H₂O was used to investigate the effect of inorganic ions on performance. The culture bottle was then purged with pure N₂ for 5 min to ensure anaerobic conditions. Bacteria were then added to the culture bottles such that the ratio of volatile solids in food waste to that in bacteria was kept at 6. The bottles were finally sealed with butyl rubber stoppers and were put in a water bath to keep the culture medium at the desired temperature. Mixing was provided by a stirring magnetic bar in the culture bottle. pH and temperature were measured by a pH and temperature probe. Liquid samples were collected regularly for measuring chemical oxygen demand (COD) and the concentration of volatile fatty acids (VFAs). The volume of biogas was read from the scales on the gas collection tube, and the gas was analyzed for its composition.

For the screening study, 6 g synthetic food waste was used. The screening experiment was carried out at 37 ± 1 °C, an initial pH of 7, a water to solids ratio of 5 and without adding inorganic salts. The experiment was carried out until no biogas was produced for 6 h. Two bacteria that generated the highest amount of biohydrogen were then selected to study the effect of operating parameters on biohydrogen generation. Operating conditions including pH of the initial food waste, water to solids ratio, temperature and the presence of inorganic salts were studied (Table 2). Operating conditions same as the screening study (base case) was first carried out and the operating parameters were then adjusted one at a time. As

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