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Stillage reflux in food waste ethanol fermentation and its by-product accumulation

Hongzhi Ma [a,b,*], Jian Yang [a,b], Yan Jia [a,b], Qunhui Wang [a,b], Yukihiro Tashiro [c], Kenji Sonomoto [c]

[a] Department of Environmental Engineering, University of Science and Technology, Beijing 100083, China
[b] Beijing Key Laboratory of Resource-oriented Treatment of Industrial Pollutants, Beijing, China
[c] Department of Bioscience and Biotechnology, Graduate School of Bioresource and Bioenvironmental Sciences, Kyushu University, 6-10-1 Hakozaki, Higashi-ku, Fukuoka 812-8581, Japan

HIGHLIGHTS

- Stillage reflux fermentation used for ethanol production from food waste.
- Lactic acid and sodium chloride were accumulated in food waste ethanol fermentation.
- Lactic acid affected more on ethanol production than sodium chloride.

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ABSTRACT

Raw materials and pollution control are key issues for the ethanol fermentation industry. To address these concerns, food waste was selected as fermentation substrate, and stillage reflux was carried out in this study. Reflux was used seven times during fermentation. Corresponding ethanol and reducing sugar were detected. Accumulation of by-products, such as organic acid, sodium chloride, and glycerol, was investigated. Lactic acid was observed to accumulate up to 120 g/L, and sodium chloride reached 0.14 mol/L. Other by-products did not accumulate. The first five cycles of reflux increased ethanol concentration, which prolonged fermentation time. Further increases in reflux time negatively influenced ethanol fermentation. Single-factor analysis with lactic acid and sodium chloride demonstrated that both factors affected ethanol fermentation, but lactic acid induced more effects.

1. Introduction

The sustainable development of the economy of a society results from the importance given to energy and environment. As a renewable source of clean energy, fuel ethanol is of strategic significance to relieve the shortage of petroleum resources and reduce environmental pollution (Gírio et al., 2010). At present, ethanol production is focused on fermentation, which accounts for more than 90% production in the ethanol market (Zi et al., 2013). Agricultural products, such as corn, wheat, potato, sugar cane, and molasses, are often used as raw materials for ethanol fermentation by yeast (Pandey, 2004). Besides the suitable substrate for ethanol fermentation, the proper control for the pollution caused in ethanol fermentation is another issue should be taken good care of. It is reported that 10 tons of waste water including stillage and other waste water caused by washing or clean would be produced during the production of 1 ton of ethanol, this has become a serious challenge for the industrial development of fuel ethanol (Zhao et al., 2014; Jiménez et al., 2003).

The ethanol industry mainly used the food plant as the substrate which was not encouraged due to the threat for human desire for food. The low cost waste with high carbon source was the research hot all over the world. Among them, food waste has drawn the research attention due to its high volume and nutrient level. Food waste in developing countries represents a much higher fraction in the production of total municipal solid waste (MSW) compared to developed countries. The percentage of food waste in China and Bangladesh could be more than 50%, even as high as 75% (Hossain et al., 2014; Qing et al., 2010), while in USA and France, the percentage would be among 20–30%.

Such a high volume of solid waste also had the characteristic of easy to be degraded and deteriorated, thus finding a proper treatment and resource technology for food waste was of great importance. In Asian countries such as South Korea, Taiwan and Japan, more than 40% of the food waste is processed into compost, swine

* Corresponding author at: Department of Environmental Engineering, University of Science and Technology, Beijing 100083, China.

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feed, and fish feed to partly substitute for chemical fertilizers and conventional feed ingredients (Takata et al., 2012). Also there are researchers who utilized food waste to produce VFA-rich liquids (acidogenic liquid), which was used as denitrification carbon sources (Jiang et al., 2013). Biomethane, biodiesel, lactic acid and ethanol were also chosen as the product from food waste, the energy and resource utilization were mainly concerned by the researchers. But different technologies had their merits and shortcomings (Karmee, 2016; Su et al., 2013; Qing et al., 2010; Ma et al., 2009). Since most fermentation had the similar challenges facing the utilization of food waste. Such as the stress by the ingredient in the food waste (acid and salt), the high volume of fermentation waste, to find a resource technology which could overcome the difficulties is important. Generally saying to adopt the energy saving process is feasible in case of cost. Most fermentation processes are conducted under sterilization conditions (Zhang et al., 2007). Open fermentation without sterilization can not only avoid maillard reaction and furfural inhibition upon sterilization, but this process also provides advantages of reduced energy consumption, minimal equipment requirement, and simple operation. To date, open lactic acid fermentation from food waste has been reported (Wang et al., 2003). Inoculated lactic acid can be used for bacteriostatic action on food waste, and subsequent ethanol fermentation presents no negative effects. Simultaneously, lactic acid can be converted into pyruvate and then generate ethanol (Ma et al., 2014). Furthermore, lactic acid can be used for sterilization and maintenance of ethanol yield. These applications could also be used for ethanol fermentation, since the food waste had undergone cooking process, the traditional liquefying process could be omitted from the thermodynamic analysis (Zhang et al., 2012). Food waste fermentation will produce stillage equally, and treatment technologies including flocculation, biological fermentation, and stillage recycle fermentation are available for stillage (Kiran et al., 2014). Recycle fermentation uses stillage obtained from saccharified food waste fermentation can replace water in the next batch of ethanol fermentation. The most important advantages of stillage recycle fermentation technology is the reduced pollution and the complete utilization of rich proteins, amino acids, and various metal ions in stillage, as well as the improvement of ethanol yield (Kim et al., 1997). Our previous study indicated that fermentation is affected after five stillage recycles, resulting in the reduction of ethanol concentration and the extension of fermentation time by thrice of the original. A study showed that humic acid and protein substances, which accumulated in stillage, are likely to affect ethanol fermentation (Su et al., 2015).

Relative studies have been conducted on the effect of stillage recycling on ethanol fermentation. Zi's research studied stillage total-reflux recycle fermentation from corn and the stress of stillage recycle on self-flocculation yeast (Zi et al., 2010). They also studied the metabolic products such as glycerin trehalose under stress effect and found propionate to be the principal inhibitor of reflux (Zhang et al., 2010). Further research successfully alleviated the effect of propionic acid by non-sterilized fermentation because propionic acids were mainly derived from sterilization. Corresponding research on organic acids and metal ions in multiple reflows from fermentation broth indicated that ORP control can alleviate the influence of stillage to some extent. These studies serve as references for stillage recycling of ethanol from food waste (Liu et al., 2013).

However, few studies have been conducted on by-product accumulation of ethanol fermentation from food waste, considering the unique property of high salts in food waste (Ma et al., 2016). Moreover, there existed research indicated that lactic acid easily accumulates in food waste (Wang et al., 2009). Furthermore, food waste without vaccination can generate substantial amount of lactic acid in the anaerobic process. However, this notion lacks related evidence on whether much lactic acid will affect ethanol fermentation and change the metabolic products.

The present study investigated the industrialized production of fuel ethanol and free yeast continuous fermentation with and without stillage recycle through saccharification of liquid from food waste by enzymatic hydrolysis as substrate in automatic fermentation tank. Moreover, this study explored the influence of stillage recycle on ethanol production from food waste and the accumulation of potential inhibitors (lactic acid and sodium chloride) in technological processes. In addition, this study determined the effects of stillage recycling on ethanol production from food waste to enhance the application of the process. Therefore, revealing the inhibition mechanism of stillage recycle is fundamental to ethanol fermentation.

2. Methods

2.1. Raw materials

Food waste was collected from a dining room in the University of Science and Technology Beijing, China. Corresponding parameters was showed in our previous publications (Zhang et al., 2012). Dry yeast (Anqi Company, China) was used for fermentation and was activated by dissolving it into glucose broth at 2% concentration for 2–3 h in a thermostatic shaker culture at 35 °C

2.2. Ethanol fermentation

After pretreatment, food waste was mixed with water at a ratio of 2:1. For example, 1000 g of food waste was mixed with 500 g of water, with the addition of 100 U/g glucoamylase (Ao Bo Xing Company, China). The solution was saccharified at 60 °C for 6 h, and the liquid was then centrifuged for 10 min with a rotation rate of 4000 r/min. Supernatant was stored at 0–4 °C for the following fermentation. During fermentation, the supernatant was inoculated with yeast at a size of 10%. Broth was sampled every 12 h, and the corresponding fermentation parameters and by-products were determined.

2.3. Method for stillage reflux fermentation

Fermentation broth discharged from the fermenter was collected and distilled to remove ethanol and volatile by-products. The remaining stillage with non-volatile by-products was recycled for saccharification and broth preparation. The broth was then used for ethanol fermentation as mentioned in Section 2.2, and the stillage reflux was utilized for a maximum of seven times.

2.4. Single-factor analysis of lactic acid and sodium chloride

During single-factor experiment, 70 g/L glucose solution was used as the substrate, which was added a variety of lactic acid concentrations (initial lactic acid concentration of 0 g/L, followed by 20 and 40 g/L). To produce ethanol, Angel yeast was added to the substrate and sampled every 8 h. To induce a sodium chloride effect, 70 g/L glucose solution was added with different sodium chloride concentrations (0.02, 0.06, and 0.10 mol/L) in conical flasks. Angel yeast was again added to produce ethanol and sampled every 8 h.

2.5. Analytical method

Fermentation was performed in a fermentation tank (BLBIO-mini-1GC), which was sampled at a certain time, and 5 ml of fermentation broth was obtained at each time. After centrifugation