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Novel process combining anaerobic-aerobic digestion and ion exchange resin for full recycling of cassava stillage in ethanol fermentation

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ABSTRACT

A novel cleaner ethanol production process has been developed. Thin stillage is treated initially by anaerobic digestion followed by aerobic digestion and then further treated by chloride anion exchange resin. This allows the fully-digested and resin-treated stillage to be completely recycled for use as process water in the next ethanol fermentation batch, which eliminates wastewater discharges and minimizes consumption of fresh water. The method was evaluated at the laboratory scale. Process parameters were very similar to those found using tap water. Maximal ethanol production rate in the fully-recycled stillage was 0.9 g/L/h, which was similar to the 0.9 g/L/h found with the tap water control. The consumption of fresh water was reduced from 4.1 L/L (fresh water/ethanol) to zero. Compared with anaerobically-aerobically digested stillage which had not been treated with resin, the fermentation time was reduced by 28% (from 72 h to 52 h) and reached the level achieved with tap water. This novel process can assist in sustainable development of the ethanol industry.

1. Introduction

Recent economic developments in many countries all around the globe have heightened the need for alternative energy resources due to the well-documented drawbacks of fossil fuel (Mood et al., 2013; Shafiei et al., 2014). Fuel ethanol is considered to be a primary fuel candidate for near/long term applications due to its long history, use, and inherent characteristics, such as low toxicity to microbes and environment, low boiling point, high octane number, and comparable energy content (Kumar et al., 2016; Safari et al., 2016). Among the available fuel ethanol feedstock, cassava is very attractive for its natures of low price and high productivity. However, cassava-ethanol plants generate stillage waste at a typical rate of 13–20 L per liter of ethanol produced (Wilkie et al., 2000; Willington and Marten, 1982). Therefore, an important environmental factor in the ethanol production chain is the proper management of stillage, which has a substantial chemical oxygen demand (COD) and acidic and corrosive characteristics (Fuess and Garcia, 2014), due to its high organic matter content. Thus methods for the treatment of stillage, and its economic utilization, need to be implemented in the industry. In conventional cassava-ethanol production processes (Fig. 1a), thin stillage is treated by sequential anaerobic and aerobic digestions, and the anaerobically-aerobically treated stillage (An-Ae-stillage) is directly discharged. An-Ae-stillage is characterized by a dark color, high ash content and high percentages of dissolved organic and inorganic matter (Pant and Adholeya, 2007). Therefore, solving the problem of wastewater management is a matter of great urgency for the ethanol industry (Handelsman et al., 2012; Ryan et al., 2009; Satyawali and Balakrishnan, 2008).

Previous studies (Agler et al., 2008; Alkan-Ozkaynak and Karthikeyan, 2011) found that anaerobic digestion effluent could potentially be used as process water for subsequent ethanol fermentation. In order to reduce energy consumption and operation costs, Zhang and co-works (Zhang et al., 2010b) studied a full recycling process incorporating two-stage anaerobic treatment of distillery wastewater from bioethanol production. Furthermore, Zhang et al. (2010a) studied the effect of utilizing anaerobic digestion of waste effluent on ethanol production in an integrated ethanol-methane fermentation process. In addition, Wang et al. (2013) established and assessed a novel cleaner integrated ethanol-methane fermentation process. In the integrated process, thin stillage was treated by anaerobic digestion and then reused for the following ethanol fermentation. However, ethanol tolerance is clearly influenced by many factors (carbohydrate level, nutrition, temperature, osmotic pressure/water activity, and substrate concentration) (Casey and Ingledew, 1986). Moreover, previous results

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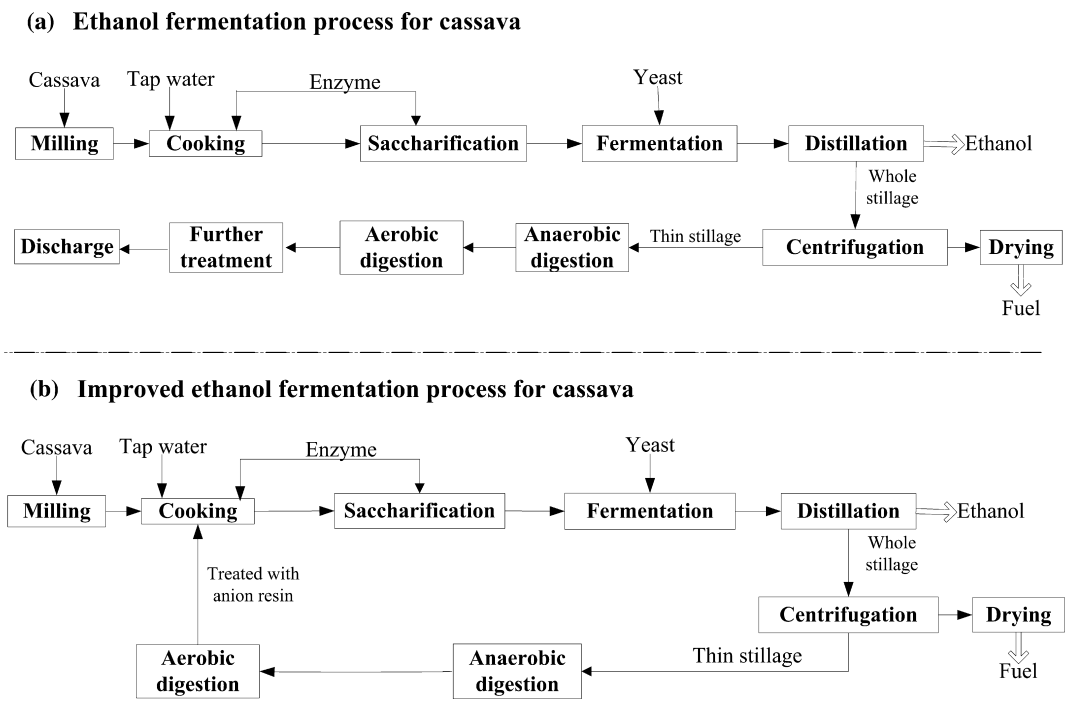

Fig. 1. Process diagram for cassava ethanol production (a) conventional ethanol fermentation process, (b) improved ethanol fermentation process.

showed that the anaerobic digestate had a high buffer capacity with a pH of approximately 8.0, which required considerable sulfuric acid to adjust pH to an optimum value (5.5–6.0) for ethanol fermentation (Yang et al., 2016). An alternative method was therefore required to eliminate acid usage and reduce process costs. Recycling 50–75% of stillage from a previous fermentation was tested successfully, however, at these high levels of stillage recycle, ethanol yield was reduced after three to five runs of closed-loop recycling (Egg et al., 1985; Shojaosadati et al., 1996). In conclusion, methods for reusing wastewater have evolved continuously in response to the need for cleaner production. However, there has been no research on recycling of An-Ae-stillage.

In the conventional ethanol production process, several kinds of wastewater are generated, including thin stillage itself, and anaerobic and aerobic digestion effluents. An-Ae-stillage is directly discharged under the current approach, which not only pollutes the environment but also results in the serious waste of water resources. Moreover, the difficulty of achieving regulatory discharge standards increases operating costs of ethanol plants. To date, little work has been published on the reuse of An-Ae-stillage. In this study, a novel recycling process is described, with the objectives of avoiding environmental pollution and reducing operation costs. In this improved ethanol fermentation process (Fig. 1b), the chloride anion exchange resin is employed to remove the inhibitors contained in the An-Ae-stillage, which may then successfully be reused as process water for subsequent ethanol fermentation batches. This novel process is expected to assist sustainable development of the ethanol industry.

2. Materials and methods

2.1. Microorganism and seed medium

A commercial strain of *S. cerevisiae* for ethanol production (TG1348) was obtained from Henan Tian Guan Co., Ltd (Henan, Nanyang, China). One loopful of *S. cerevisiae* was inoculated into a 500 mL Erlenmeyer flask holding 200 mL of a solution containing (g/L): glucose 20, yeast extract 8.5, $(NH_4)_2SO_4$ 1.3, $MgSO_4{\cdot}7H_2O$ 0.1 and $CaCl_2{\cdot}2H_2O$ 0.06. The flask was incubated on a shaker (200 rpm) at 30 °C for 18 h to produce seed medium.

2.2. Fermentation medium

Fermentation medium was composed of (g/L): glucose 100, $MgSO_4{\cdot}7H_2O$ 0.1, $CaCl_2{\cdot}2H_2O$ 0.06, and urea 0.5, dissolved in the appropriate process water. The pH was adjusted to 4.5 using 30% (w/w) H_2SO_4 or 10% (w/v) NaOH.

2.3. Ethanol fermentation

An-Ae-stillage was provided by Jiangsu Jinmaoyuan Biochemical Engineering Co. Ltd. (Lianyungang, Jiangsu, China). Tap water was used as control. Triplicate fermentations were carried out in 250 mL flasks containing 135 mL of medium. To avoid Maillard reaction, urea was sterilized separately and was added before inoculating. A 10% (v/v) inoculum of seed medium was added to each flask. All fermentations were carried out at 30 °C without shaking.

2.4. Treatment of An-Ae-stillage

At the end of the fermentation, the "beer" was distilled and the whole stillage centrifuged (4000*g*, 20 min). Anaerobic digestion treatment of the thin stillage was done in two-stage anaerobic sequencing batch reactors (ASBRs) by adding thermophilic and mesophilic anaerobic sludge. Nitrogen gas was bubbled through each reactor for 5 min after inoculation to ensure anaerobic conditions. The temperatures for thermophilic and mesophilic ASBR stages were maintained at 55 °C and 35 °C respectively by circulation of heated water through a water jacket. The thin stillage was fed into the thermophilic ASBR using a peristaltic pump in four days. The effluent was centrifuged (4000*g* for 15 min), and the supernatant was pumped into the mesophilic ASBR. The effluent of the mesophilic ASBR was then centrifuged at 4000*g* for 15 min. Both reactors cycles were 1 d total, with 2 h feeding,

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