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Efficient production of short-chain fatty acids from anaerobic fermentation of liquor wastewater and waste activated sludge by breaking the restrictions of low bioavailable substrates and microbial activity



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ABSTRACT

An efficient approach of bioconverting the organic wastes in liquor wastewater (LW) and waste activated sludge (WAS) to valuable short-chain fatty acids (SCFAs) *via* anaerobic fermentation was explored. The maximal SCFAs concentration was 5400 mg COD/L with approximate 80.0% acetic and propionic acids under optimized conditions (LW/WAS ratio 1:1, pH 8 and fermentation 4 d). Mechanisms investigation found that the fermentation of LW/WAS made up the drawbacks of sole WAS fermentation by improving the bioavailable substrates and low C/ N ratio to stimulate the microbial activities. The bioconversion efficiency of substrates for SCFAs generation was therefore enhanced. The humic acids present in LW could also play positive roles in SCFAs promotion. Moreover, the performance of LW/WAS fermentation was highly correlated with appropriate fermentation pH. The fermentative bacteria responsible for SCFAs production were highly enriched and the activities of key hydrolases, acid-forming enzymes and ATP concentration were greatly improved at pH 8.

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Table 1

Characteristics of WAS and LW used in this study.

Index	TSS (mg/L)	VSS (mg/L)	TCOD (mg/L)	SCOD (mg/L)	Total proteins (mg COD/L)	Total carbohydrates (mg COD/L)	Humic acid (mg/L)	рН
WAS	17050 ± 225	8525 ± 150	17015 ± 180	235 ± 25	8028 ± 168	1312 ± 85	65 ± 10	6.7 ± 0.2
LW	-	-	170336 ± 1080	150123 ± 965	20025 ± 728	22617 ± 600	9200 ± 225	3.4 ± 0.2

1. Introduction

Liquor is one of the most featured industries in China which brought great economic benefits. The liquor industry is growing fast in recent years and the production has been estimated to be 13.6 million tonnes in 2016 (Luo et al., 2018). However, as the unavoidable byproduct, large amounts of liquor wastewater (LW) were also generated during the brewing process. It has been reported that over 15 tonnes of LW were produced for 1 tonne liquor production (Luo et al., 2018). LW was characteristic of high concentrated organic wastes, which was mainly composed of sugars (glucose and fructose), followed by ethanol, glycerol, esters and polyphenolic compounds (Mosse et al., 2011). The chemical oxygen demand was commonly up to 10,000-20,000 mg/L (Zhang, et al., 2014). Huge environmental problems, such as pollution of ground and surface water, vegetation damage and odors occurrence, would be caused by LW if without proper disposal (Chapman et al., 2001). In fact, due to the rapid expansion of liquor industry, LW has become the second largest pollution source of light industries to aquatic environment (Xue, 2011).

However, the large mixture of various organic matters present in LW contained high energy which was also the potential sources for resource recovery (Dong et al., 2015). A promising approach to dispose the LW is bioconversion of these wastes into value-added products, by which the minimization of negative environmental impact and recycle of energy could be simultaneously accomplished. Fortunately, several successful attempts of LW reutilization have been made. For example, a pilot-scale energy self-sufficient microbial fuel cell system for wastewater treatment has achieved with the LW as substrates (Dong et al., 2015). Also, high concentration of industrial xanthan has been reported to be obtained from LW with specific microorganisms (Roncevic et al., 2016).

Recently, the recovery of short-chain fatty acids (SCFAs) from organic wastes via anaerobic fermentation has attracted much attention. It could deal with the problem of carbon sources shortage in wastewater treatment plants (WWTPs) to improve the operating performances of biological nutrient removal (Li et al., 2011; Luo et al., 2015). Waste activated sludge (WAS), which was the byproducts of activated sludge process in WWTPs, was once considered as the most attractive substrates for SCFAs production and has have been extensively investigated (Chen et al., 2017; Zhang et al., 2017). It was expected to offer a green and sustainable operational mode in WWTPs by simultaneously reutilizing the carbon sources in WAS and stabilizing the WAS with substantial economic benefits (Wang et al., 2017a; Zhao et al., 2017). However, WAS is a complex mixture of particulate organic and inorganic matters and various microorganisms (Ormeci and Vesilind, 2000). The content of organics in WAS is relatively low and is usually cemented and flocculated together by extracellular polymeric substances (EPS). The effectiveness and efficiency of WAS anaerobic fermentation for SCFAs production were commonly restricted by the inefficient solubilization and hydrolysis of particulate organics to soluble substances, which were considered as rate-limiting steps (Chen et al., 2007). On the contrary, the LW is rich of soluble organic substrates but lacks enough functional microorganisms to bioconvert them. It seems quite attractive to take advantage of the indigenous microorganisms in WAS to recover valuable products from LW. Besides, the efficient production of SCFAs from WAS was also hampered by the drawback of low carbon to nitrogen mass ratio (C/N) in WAS, which was closely related with the metabolism activities of acid-forming microorganisms (Feng

et al., 2009). Whether the organics-enriched LW could make up the drawbacks of sole WAS anaerobic fermentation by providing sufficient substrates and improving the low C/N ratio to stimulate the activities of fermentation bacteria for efficient SCFAs production is unknown. Also, the exploration of key parameters and mechanisms in regulating the performance of anaerobic fermentation of LW/WAS mixture were few. Therefore, the scientific attempt on the investigations on the anaerobic fermentation of LW/WAS mixture for SCFAs production should be very useful in addressing the issues of LW and WAS disposal and carbon sources deficiency in WWTPs.

Therefore, the aim of this paper was to examine the possibility of anaerobic fermentation of LW/WAS mixture for efficient SCFAs production. The influences of LW/WAS ratios and fermentation pH on the SCFAs production were firstly explored. Then the key mechanisms for the efficient SCFAs production by LW/WAS fermentation were illustrated from the perspectives of substrates improvement and microbial activities (activities of key enzymes, ATP concentration and abundances of fermentative bacteria) under different fermentation conditions.

2. Material and methods

2.1. Sources and characteristics of WAS and LW

The WAS used in the tests was withdrawn from the secondary sedimentation tank of a municipal WWTP in Nanjing, China. It was firstly concentrated by settling at 4 °C for 24 h, and then filtrated from stainless steel mesh (2.0 mm). The raw LW was obtained from a white liquor manufacturer in Jiangsu, China. The main characteristics of WAS and raw LW were shown in Table 1. Before use, the raw LW was diluted 10folds. Then the total COD of LW and WAS were almost the same.

2.2. Influences of LW/WAS ratio on the SCFAs production during fermentation

The batch tests were conducted in 7 identical serum bottles with a working volume of 600 mL. Firstly, 350 mL of LW/WAS mixing substrates was inoculated into 5 bottles at the volume ratio of 4:1, 2:1, 1:1, 1:2 and 1:4, respectively. The left two was respectively added with 100% LW and 100% WAS to explore the respective performance of sole fermentation for SCFAs production. The total COD in each reactor was similar which was about 17000 mg/L. The fermentation pH was controlled at pH 7 by the addition of NaOH (4 mol/L) or HCl (4 mol/L). After being flushed with pure nitrogen gas to eliminate oxygen, all fermentation reactors were capped with rubber stoppers, sealed, and placed in an air-bath shaker at stirring speed of 180 revolutions per minutes (rpm) and temperature 35 °C. By determining the SCFAs concentrations and composition in different fermentation systems, the effects of LW/WAS ratio on the SCFAs production were obtained.

2.3. Influences of pH on the SCFAs production during LW/WAS fermentation

The effects of pH on the SCFAs production were explored in 8 identical serum bottles with a working volume of 600 mL. 350 mL of LW/WAS mixing substrates at the ratio of 1:1 was inoculated into each bottles. The fermentation pH was controlled in the range of 4–10 by the addition of NaOH (4 mol/L) or HCl (4 mol/L). The left one without pH controlled was set as the control. After being flushed with pure nitrogen

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