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# Performance and microbial community of a membrane bioreactor system — Treating wastewater from ethanol fermentation of food waste

## 🔃 Xiaobiao Zhu¹, Mengqi Li², Wei Zheng³, Rui Liu³, Lujun Chen²,³,\*

- 1. Beijing University of Chemical Technology, College of Chemical Engineering, Beijing 100029, China. E–mail: zhuxiaobiao@mail.buct.edu.cn
- 2. School of Environment, Tsinghua University, Beijing 100084, China
- 3. Key Laboratory of Water Science and Technology of Zhejiang Province, Jiaxing 314006, China

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40 Bacterial diversity

Microbial composition

#### ABSTRACT

In this study, a lab-scale biological anaerobic/anaerobic/anoxic/membrane bioreactor (A<sup>3</sup>-MBR) was designed to treat wastewater from the ethanol fermentation of food waste, a promising way for the disposal of food waste and reclamation of resources. The 454 pyrosequencing technique was used to investigate the composition of the microbial community in the treatment system. The system yielded a stable effluent concentration of chemical oxygen demand (202  $\pm$  23 mg/L), total nitrogen (62.1  $\pm$  7.1 mg/L), ammonia (0.3  $\pm$ 0.13 mg/L) and total phosphorus (8.3  $\pm$  0.9 mg/L), and the reactors played different roles in specific pollutant removal. The exploration of the microbial community in the system revealed that: (1) the microbial diversity of anaerobic reactors A<sub>1</sub> and A<sub>2</sub>, in which organic pollutants were massively degraded, was much higher than that in anoxic  $A_3$  and aerobic MBR; (2) although the community composition in each reactor was quite different, bacteria assigned to the classes Clostridia, Bacteroidia, and Synergistia were important and common microorganisms for organic pollutant degradation in the anaerobic units, and bacteria from Alphaproteobacteria and Betaproteobacteria were the dominant microbial population in  $A_3$  and MBR; (3) the taxon identification indicated that Arcobacter in the anaerobic reactors and Thauera in the anoxic reactor were two representative genera in the biological process. Our results proved that the biological A<sup>3</sup>-MBR process is an alternative technique for treating wastewater from food waste.

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#### Introduction

Food waste comes from the kitchens of households, restaurants and public catering rooms, and contains high moisture and high concentrations of organic pollutants. Anaerobic fermentation is an attractive method for food waste treatment, compared to landfills that may cause serious odor and

nuisance, highly polluted leachate wastewater and excessive 54 land occupation (Tawfik et al., 2015; Xie et al., 2014). In 55 anaerobic fermentation, complicated organic substrates are 56 converted into biofuels by bacteria in reactors through 57 hydrolysis, acidogenesis, acetogenesis and methanogenesis 58 (Byun, 2012; Yirong et al., 2015). Recently, anaerobic ethanol 59 fermentation has been considered to be the most effective 60

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<sup>\*</sup> Corresponding author. E-mail: chenlj@mail.tsinghua.edu.cn (Lujun Chen).

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way to obtain renewable energy from food waste (Wu et al., 2015). However, the ethanol fermentation of food waste generates huge amounts of wastewater, which contains high concentrations of inorganic and organic pollutants, such as suspended solids, ammonia, sulfate, phosphorus, saccharides and proteins. The discharge of this wastewater without appropriate treatment would cause serious eutrophication of water bodies. Therefore, it is a critical issue to find an environmentally and economically sound way to treat this wastewater during the application of ethanol fermentation of food waste.

Previous studies on food waste fermentation have mainly focused on the efficiency of biofuel production and the optimization of fermentation processes, but gave little attention to the treatment of fermentation wastewater. The wastewater from food waste fermentation shares many features with distillery wastewater from corn, sorghum or cassava fermentation, but contains more complicated components than the distillery wastewater (Kamalaskar et al., 2010; Wicher et al., 2013). Nowadays, many studies have explored technology for distillery wastewater treatment. However, few have addressed the treatment of wastewater from food waste fermentation. The high loading of multiple organic and inorganic pollutants may cause adverse effects on the treatment efficiency of the fermentation wastewater. Therefore, it is necessary to investigate the properties of the wastewater and design the treatment processes accordingly.

Biological wastewater treatment, which employs microorganisms to degrade pollutants under anaerobic or aerobic conditions, is usually considered a feasible and cost-effective treatment method for both domestic and industrial wastewater. In biological treatment processes, organic and inorganic compounds are transformed into H2O, CO2, N2, etc., achieving the complete mineralization of wastewater pollutants. Researchers have successfully applied biological processes in the treatment of distillery wastewater (Nadia et al., 2015). According to previous research, a combined process involving both anaerobic and aerobic processes is suitable for the treatment of wastewater with high concentrations of organics, such as fermentation wastewater (Shin et al., 2010; Zerrouki et al., 2015). The anaerobic unit can recover bioenergy when it removes the loading of organic pollutants, and the aerobic unit treats the effluent of anaerobic wastewater to reduce the level of organic pollutants further and to remove nitrogen and phosphorus from the wastewater.

The membrane bioreactor (MBR), which relies on the membrane separation of biomass from effluent, is widely used in aerobic biological wastewater treatment. A long sludge retention time and high sludge concentration can be achieved simultaneously in MBRs (Zhao et al., 2009). Therefore, the MBR has a rich microbial community, and the presence of plenty of microorganisms is favorable to the removal of nitrogen and phosphorus and the degradation of organic pollutants. Pretreatment is also necessary, since the fermentation wastewater usually contains high concentrations of suspended solids. Coagulation is the most effective method for the removal of solids, color, and some organic pollutants. The initial pH and dosage of coagulants, which affect the coagulation efficiency, need to be optimized (Ahmad et al., 2006; Fang et al., 2012).

In this study, an anaerobic/anaerobic/anoxic/aerobic- 121 membrane bioreactor (A³-MBR) system was employed to 122 treat food waste fermentation wastewater, and the influence 123 of operation parameters on the efficiency of pollutant 124 removal was investigated. The composition of the microbial 125 community in each reactor of the A³-MBR system was studied 126 by 454-pyrosequencing technology in order to provide useful 127 information relating to the biological treatment process.

#### 1. Materials and methods

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#### 1.1. Wastewater

The wastewater used in the research was obtained from a 132 food waste fermentation process described in previous 133 studies (Thallinger et al., 2009; Ma et al., 2014). In this process,  $Q2\,Q3$  food waste was collected from the dining room of a university 135 in Beijing. The ethanol fermentation of food waste was carried 136 out at pH 4 after the addition of some strains isolated for 137 ethanol production. The ethanol was distilled out after the 138 fermentation. The remaining water discharged from the 139 reactor was called food waste fermentation wastewater. In 140 this study, the water qualities of the raw wastewater were as 141 follows: pH,  $4.0 \pm 0.5$ ; SS (suspended solids),  $1050 \pm 150$  mg/L; 142 COD (chemical oxygen demand),  $15,000 \pm 1000$  mg/L; TOC 143 (total organic carbon),  $5500 \pm 1500$  mg/L;  $NH_3$ -N (ammonia), 144  $225 \pm 30$  mg/L; TN (total nitrogen),  $350 \pm 50$  mg/L; TP (total 145 phosphorus),  $85 \pm 25$  mg/L.

#### 1.2. Coagulation

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Coagulation was employed as the pretreatment process and  $^{148}$  conducted before the biological treatment of the wastewater.  $^{149}$  The pH and coagulant dosage were optimized using 0.5 L of raw  $^{150}$  fermentation wastewater under uniform magnetic stirring at  $^{151}$  room temperature of 23–25 °C. The initial pH was adjusted to  $^{152}$  the desired values using 1 mol/L HCl and 1 mol/L NaOH  $^{153}$  solutions. After 2 min premixing, polyaluminum chloride  $^{154}$  (PAC, purity ( $^{159}$ ) =  $^{186}$ ) was added to the wastewater. Then  $^{155}$  the tested samples were mixed rapidly at 200 r/min for 0.5 min,  $^{156}$  followed by slow stirring at  $^{50}$  r/min for 8 min, and finally  $^{157}$  settled for 30 min. After coagulation, the supernatant ( $^{50}$  mL)  $^{158}$  was collected to determine the residual turbidity and TOC.

#### 1.3. Reactor set-up

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A laboratory-scale anaerobic/anaerobic/anoxic/aerobic MBR 161 ( $A_1/A_2/A_3/MBR$ , or  $A^3$ -MBR) treatment system was applied to 162 treat the coagulation-treated wastewater (Fig. 1). The anaerobic 163 reactors,  $A_1$  (working volume, 0.628 L) and  $A_2$  (working volume, 164 0.628 L), and the anoxic reactor,  $A_3$  (working volume, 0.314 L), 165 were all packed with soft hydrophilic fibers for microorganism 166 adhesion. The aerobic MBR (working volume, 0.628 L) employed 167 a submerged polyvinylidene fluoride hollow fiber membrane 168 module with pore size of 0.02  $\mu$ m. The maximum flux of the 169 membrane module was 9.72 L/m²/hr in this study. Air was 170 supplied by a compressor and dispersed at the bottom of the 171 MBR to provide oxygen for the microorganisms and generate 172 continuous stirring. A part of the MBR effluent was returned to 173

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