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In situ observation of the growth of biofouling layer in osmotic membrane bioreactors by multiple fluorescence labeling and confocal laser scanning microscopy

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ARTICLE INFO

Article history:
Received 25 September 2014
Received in revised form
16 February 2015
Accepted 21 February 2015
Available online 5 March 2015

Keywords:
Osmotic membrane bioreactor
Forward osmosis membrane
Biofouling
Fluorescence labeling
Confocal laser scanning microscopy

ABSTRACT

Since the concept of the osmotic membrane bioreactor (OMBR) was introduced in 2008, it has attracted growing interests for its potential applications in wastewater treatment and reclamation; however, the fouling mechanisms of forward osmosis (FO) membrane especially the development of biofouling layer in the OMBR are not yet clear. Here, the fouled FO membranes were obtained from the OMBRs on days 3, 8 and 25 in sequence, and then the structure and growing rule of the biofouling layer formed on the FO membrane samples were in-situ characterized by multiple fluorescence labeling and confocal laser scanning microscopy (CLSM). CLSM images indicated that the variations in abundance and distribution of polysaccharides, proteins and microorganisms in the biofouling layer during the operation of OMBRs were significantly different. Before the 8th day, their biovolume dramatically increased. Subsequently, the biovolumes of β-D-glucopyranose polysaccharides and proteins continued increasing and leveled off after 8 days, respectively, while the biovolumes of α-D-glucopyranose polysaccharides and microorganisms decreased. Extracellular polymeric substances (EPS) played a significant role in the formation and growth of biofouling layer, while the microorganisms were seldom detected on the upper fouling layer after 3 days. Based on the results obtained in this study, the growth of biofouling layer on the FO membrane surface in the OMBR could be divided into three stages. Initially, EPS was firstly deposited on the FO membrane surface, and then microorganisms associated with EPS located in the initial depositing layer to form clusters. After that, the dramatic increase of the clusters of EPS and microorganisms resulted in the quick growth of biofouling layer during the flux decline of the OMBR. However, when the water flux became stable in the OMBR, some microorganisms and EPS would be detached from the FO membrane surface.

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http://dx.doi.org/10.1016/j.watres.2015.02.048
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1. Introduction

Since the concept of osmotic membrane bioreactor (OMBR) combining forward osmosis (FO) with activated sludge process was proposed in 2008, it has attracted growing interests for its potential applications in wastewater treatment and reclamation. In OMBRs, wastewater flows across an FO membrane from the activated sludge to the high-osmotic-pressure draw solution driven by the transmembrane osmotic pressure difference. Compared to conventional membrane bioreactors (MBRs) using hydraulic pressure as the driving force, OMBRs have a lower fouling propensity and less energy consumptions (Cornelissen et al., 2008, 2011; Achilli et al., 2009; Yap et al., 2012). Besides that, higher quality pure water is obtained in the OMBR due to the better retention of FO membrane (Cornelissen et al., 2008, 2011; Achilli et al., 2009; Yap et al., 2012; Qiu and Ting, 2013), although subsequent separation for purified water from the draw solution is needed where wastewater reclamation is targeted.

Generally, FO process is considered to have a lower fouling tendency due to its lack of hydraulic pressure and lower flux conditions (Lutchmiah et al., 2014). Nevertheless, severe FO fouling may still occur in OMBRs as a result of the FO membrane's direct contacts with an activated sludge, a liquid with highly complex nature and consisting of many foulants such as microorganisms, organics and inorganics. Despite that flux decline in OMBRs is typically dominated by the salt accumulation effect (Cornelissen et al., 2008; Xiao et al., 2011; Lay et al., 2012; Wang et al., 2014a), the effect of fouling in OMBRs shall not be overlooked. For example, Zhang et al. (2012b) found that FO membrane fouling contributed to 45% of the total flux decline during the long-term OMBR operation. In fact, the fouling layer formed on the FO membrane surface severely reduced the mass transfer coefficient and thus enhanced the external concentration polarization (ECP) in addition to directly decreasing the water permeability induced by the increase of membrane resistance (Cornelissen et al., 2008; She et al., 2012; Linares et al., 2014; Wang et al., 2014a). Greater attention to OMBR fouling is needed in order to better understand its mechanisms and mitigation methods.

Several studies have focused on the influences of sludge properties on membrane fouling and the foulant compositions of the cake layer in OMBRs. Zhang et al. (2012a) investigated the impacts of different sludge types on the FO membrane fouling in OMBRs and found that the solute and bound polysaccharides had negative relationships with the flux decline rate while the bound and solute proteins had positive impacts. With regard to the characterization of the membrane foulants, Lay et al. (2011) reported that the extracellular polymeric substances (EPS) with small numbers of scattered bacterial cells were found in a thin gel-like secondary layer on the FO membrane surface, but no mature biofilm was formed. Subsequently, Qiu and Ting (2014) further demonstrated that small sludge floc/particles and EPS (in particular, proteins) were enriched in the FO membrane fouling layer according to the analyses of the membrane foulants. Meanwhile, the FO membrane-associated biofilms were characterized by confocal laser scanning microscopy (CLSM) and rRNA gene-tagged pyro-sequencing (Zhang et al., 2014b). Based on these results on the characterization of the membrane foulants, it could be concluded that the biofouling integrated the EPS and bacterial cells is a main contributor to OMBR fouling.

In addition to the characterization of membrane foulants, analyses on the development of biofouling layer on the FO membrane surface are also essential to provide a comprehensive understanding of the biofouling mechanism and thus potential control strategies for OMBRs. Currently, one of the best approaches for nondestructive in situ examination of biofilm is the application of fluorescent probes (Yun et al., 2006; Yu et al., 2011). Many researchers have pointed out that CLSM combined with multiple fluorescence labeling is a powerful tool for investigating the structure, distribution and function of biofilm constituents at the microscale (Yun et al., 2006; Meng et al., 2010; Dominiak et al., 2011; Wang et al., 2012b; Hassan et al., 2014; Linares et al., 2014). Thus, the objective of this study is to in-situ investigate the growing mechanisms of the biofouling layer on the FO membrane surface in the OMBR by multiple fluorescence labeling and CLSM. To the best knowledge of the authors, this is the first study for analyzing the growth of biofouling layer in an OMBR system. In this study, the fouled FO membranes were collected from the OMBRs temporally on days 3, 8 and 25, and the multiple fluorescent staining combined with CLSM was used for analyzing the distributions of α- and β-D-glucopyranose polysaccharides, proteins and total cells and the structure of biofouling layer on the FO membrane surface.

2. Materials and methods

2.1. Experimental set-up and operating conditions

Three identical bench-scale OMBRs named OMBR1, OMBR2 and OMBR3 were operated in parallel in this study. As shown in Fig. 1, an FO membrane cell and a separated bioreactor with an effective volume of 0.9 L were used in each OMBR system. The membrane cell had two same channels (length, width and height of 75, 45 and 40 mm) divided by a unique plate-and-frame FO membrane with an effective membrane area of 34 cm^2. The cellulose triacetate (CTA) FO membranes with an embedded polyester screen mesh (HTI, Albany, OR) were applied in the OMBRs. Considering the fact that membrane orientation plays a significant role in FO applications, the "active layer facing feed" (AL-FS) orientation was employed in order to avoid aggravating fouling especially pore-clogging in the support layer (Lay et al., 2011). The draw solution flew from the upper channel to the draw tank, while the activated sludge was recirculated from the bioreactor to the bottom channel. As for the bioreactor, the municipal wastewater was continuously pumped in, and the aeration of 0.5 $m^3\ h^{-1}$ was provided through an axial perforated tube. In order to maintain a constant water level in the bioreactor, the influent pump was controlled by a water level sensor. In each OMBR system, analytical grade sodium chloride (NaCl) with a concentration of 1.0 M was used as the draw solution, which was recirculated at a flow rate of 0.28 L min^{-1} from a 2 L glass reservoir through the upper channel and back to the reservoir. A conductivity control system was used to control the draw

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