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Relation between sludge properties and filterability in MBR: Under infinite SRT

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Abstract. A laboratory-scale submerged membrane bioreactor (MBR) was continuously operated for 100 d at an infinite sludge retention time (SRT) with the aim of identifying possible relation between the filterability of mixed liquor and sludge properties, such as extracellular polymeric substances (EPS), soluble microbial products (SMP), viscosity of mixed liquor, zeta potential of flocs and particle size distributions (PSD). Research results confirmed that MBR can operate with a complete sludge retention ensuring good treatment performances for COD and NH_3-N. However, the long term operation (about 40 d) of MBR with no sludge discharge had a negative influence on sludge filterability, and an increase in membrane fouling rates with the time was observed. There as a strong correlation between the sludge filterability and the fouling rate. Among the different sludge properties parameters, the concentration SMP and EPS had a more closely correlation with the sludge filterability. The concentrations of SMP, especially SMP with MW above 10 kDa, had a strong direct correlation to the filterability of mixed sludge. The protein fractions in EPS were biodegradable and available for microorganism metabolism after about 60 days, and the carbohydrates in EPS had a significantly negative effect on sludge filterability in MBR at an infinite SRT.

Keywords: membrane bioreactor (MBR); sludge retention time (SRT); activated sludge; soluble microbial products (SMP); extracellular polymeric substances (EPS)

1. Introduction

Membrane bioreactors (MBRs) have been increasingly popular for the treatment of municipal and industrial wastewater in recent years (Tian *et al*. 2015, Zhang *et al*. 2014a). MBR is considered as a good integration of conventional activated sludge system (CAS) and advanced membrane separation, providing many advantages over CAS, such as smaller footprint and better effluent quality, enabling direct potential reuse (Le-Clech *et al*. 2006, Zhang *et al*. 2014a, Tian *et al*. 2015). However, membrane fouling is a major obstacle for wide-spread applications of MBR,

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which significantly reduces membrane life, overall membrane performance and increases operating costs (Meng *et al*. 2006, Tian *et al*. 2015).

Membrane fouling is the physicochemical interaction between the biofluid and membrane to form a cake layer and the adsorption of the dissolved particles into membrane pores and leads to flux decline. As for given membrane materials, sludge properties, especially its filterability, is eventually crucial for MBR permeability and flux maintenance (Sun *et al*. 2014). Therefore, a good filterability of the activated sludge (AS) is a ”must” for a sustainable, reliable and low-cost energe performance in MBRs (Krzeminski *et al*. 2012). Many factors have been reported that might influence the AS filterability in MBR including floc size, mixed liquor suspended solids (MLSS) concentration, viscosity of mixed liquor, and soluble microbial products (SMP) in the supernatant and extracellular polymeric substances (EPS) in the sludge suspension (Judd 2011, Kraume and Drews 2010). In order to maintain a good filterability for practice in MBRs, several operational parameters should be adjusted, the solids retention time (SRT) (and consequently the F/M ratio), has been indicated as a main variable influencing membrane fouling since it influences sludge characteristics and filterability (Duan *et al*. 2014, Krzeminski *et al*. 2012).

Owing to the complete solids retention operated by the membrane separation, MBR can operate at more defined and usually greater SRT than CAS. The MBR tends to be operated with a long SRT to maintain high biomass concentrations, which is beneficial to the reduction of solids production and reactor volume (Pollice *et al*. 2008). Recently, attention has mainly been given to the research field of zero discharge of activated sludge (Villain and Marrot 2013, Huang *et al*. 2001, Masse *et al*. 2006, Pollice *et al*. 2006). Many authors stated the biological applicability of complete sludge retention, reporting high and stable degradation rates and very little sludge production or zero net growth in previous studies (Huang *et al*. 2008, Pollice *et al*. 2004). In fact, the operation at an infinite SRT may lead bacteria to be in low F/M and microorganisms respond to this by acceleration of endogenous respiration, accompanied with the release of organic cellular constituents (such as SMP) by secretion and cells autolysis, which might affect the mixed liquor characteristics and induce changes in the mixed liquid filterability (Zhang *et al*. 2014b). Therefore, the aim of the present research was to elucidate the variation of mixed liquor filterability in MBR with complete sludge retention. For this purpose, a lab-scale submerged MBR without sludge discharge was operated for about 100 days. The AS filterability and sludge properties such as MLSS, EPS, SMP, zeta potential and particle size distribution (PSD) were investigated, and the statistical analysis were used to investigated the impact of sludge properties on the AS filterability. Finally, the mixed liquid characteristics were analyzed by measuring SMP and SMP with molecular weight (MW) fractions, proteins and carbohydrates in the EPS closely related with the AS filterability.

2. Material and methods

2.1 MBR systems

A lab-scale submerged MBR with working volume of 8 L was operated. The schematic of the experimental setup is in Fig. 1. A bundle of U-shaped hollow fiber membranes made of polyvinylidene fluoride (PVDF) with a nominal pore size of 0.2 μm and a filtration area of 0.1 m^2 was submerged in the reactor. Permeate through the submerged membrane module was continuously withdrawn using a peristaltic pump (Model BT-300, Baoding Longer Precision Pump

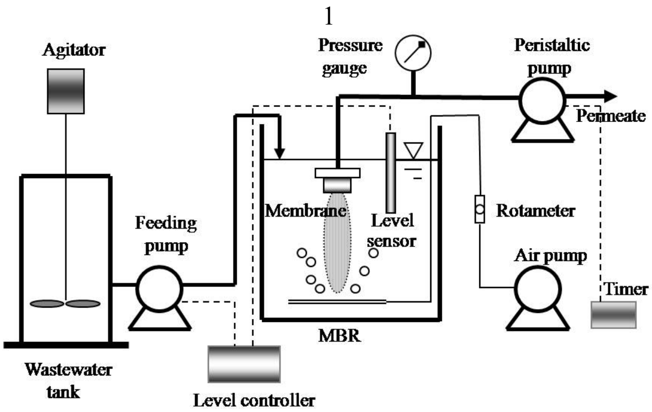


Fig. 1 Schematic diagram of the experimental set-up

Co., Ltd., China) at a constant flux of 10 L/m^2·h, operated with an intermittent mode of 13 min on and 2 min off. Chemical cleaning procedure would be carried out if the trans-membrane pressure (TMP) reached about 30 kPa during the operation. The membrane module was taken out and soaked in 0.5% (v/v) NaClO solution for about 2 h to recover its permeability.

The seed sludge was taken from another lab-scale MBR that had been operated for more than one year. Hydraulic retention time (HRT) was maintained at 6 h. No excessive sludge was discharged except for sampling. Synthetic wastewater was used in order to avoid difficulties in quantification of metabolic products due to variation in the composition and concentration of organic pollutants in the real wastewater. The synthetic wastewater was prepared using glucose, NH_4Cl, and KH_2PO_4 as a source of carbon, nitrogen, and phosphorous, respectively. The recipe could be obtained from our previous study (Zhang *et al*. 2008).

2.2 Filterability

The mixed liquid filterability was investigated in batch tests using Amicon 8400 dead-end cells (Millipore, USA). The cells had a volume of 350 mL and an effective membrane filtration area of 41.8 cm^2. All experiments were at room temperature and a constant 30 kPa pressure. In order to avoid the effect of suspended solid concentration on the MFI analysis, all sludge samples were diluted or concentrated to guarantee the same MLSS (about 5000 mg/L) prior to each bath test. The permeation flux was determined by weighing permeates on an electronic top loading balance connected to a personal computer running an auto-reading program. A new membrane sheet (PVDF, 0.2 μm) was used for each trial.

Under unstirred conditions, MFI was obtained from the plot of t/V versus V (Zhang *et al*. 2014a), which is defined as the gradient of the linear region found in the plot of the well-known cake filtration equation at constant pressure (Delgado *et al*. 2010).

$$\frac{t}{V} = \frac{\mu R_{\mathrm{m}}}{A\Delta P} + \frac{\mu \alpha C}{2A^2 \Delta P} V \tag{1}$$

$$\mathrm{MFI} = \frac{\mu \alpha C}{2A^2 \Delta P} \tag{2}$$

Where t is the filtration time (s), V the permeate volume (L), μ the dynamic viscosity of permeate (Pa s), R_m the intrinsic membrane resistance (m^{-1}), A the area of membrane (m^2), ΔP the applied transmembrane pressure (Pa), α the specific gel resistance (m/kg) and C is macromolecules concentration in bulk solution (kg/m^3).

2.3 Analytical items and methods

Standard analytical methods were used to measure for COD, NH_3-N, MLSS and MLVSS (APHA 2005). The SMP were obtained by centrifuging the mixed liquor at 4 000 r/min for 10 min, followed by filtration through a membrane of 0.45 μm. The SMP were used for fractionation with stirred cell ultrafiltration (UF) membranes (Millipore). Three UF membranes with pore size of 10, 3 and 1 kDa were used and the dissolved organic carbon (DOC) concentration was measured in series. DOC was determined by using a total organic carbon (TOC) analyzer (TOC-VCPH, Shimadzu, Japan). The extraction of EPS from the suspended microbial flocs was carried out using a heating method (Morgan 1990). Carbohydrates were determined according to the phenol-sulfuric acid method (Dubois *et al*. 1956) with glucose as standard. Proteins were determined by the modified Lowry method (Lowry *et al*. 1951) with bovine serum albumin (BSA) as standard. Particle size distributions of flocs were measured with a Malvern Mastersizer (Malvern Instruments, UK). The flocs were first shaken in order to break them into small particles, and the supernatant was sampled for zeta potential measurement using a Malvern zeta analyzer (Zetasizer 2000, UK).

2.4 Statistical analysis

Statistical analyses were carried out using Microsoft Excel built-in statistical functions. Pearson's product momentum correlation coefficient, r_p was used for linear estimations of the strength and direction of linear correlations between two parameters.

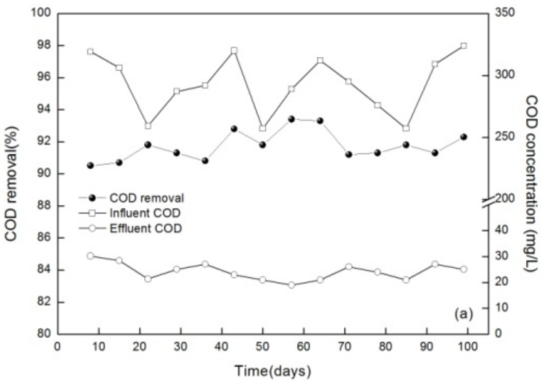
$$r_p = \frac{\sum\left(x - x_{avg}\right)\left(y - y_{avg}\right)}{\sqrt{\sum\left(x - x_{avg}\right)^2\left(y - y_{avg}\right)^2}} \tag{3}$$

where, r_p is the Pearson's product momentum correlation coefficient, (x, y) is a sample of paired data, and x_{avg} and y_{avg} are mean values. Generally, the value of r_p oscillates between-1 and +1, as r_p = -1 or r_p = +1 represents a perfect correlation, and 0 shows no correlation. If $-0.4 < r_p < +0.4$, the correlation can be assumed weak and ignored. The positive r_p shows a direct proportionality, while the negative r_p shows an inverse proportionality.

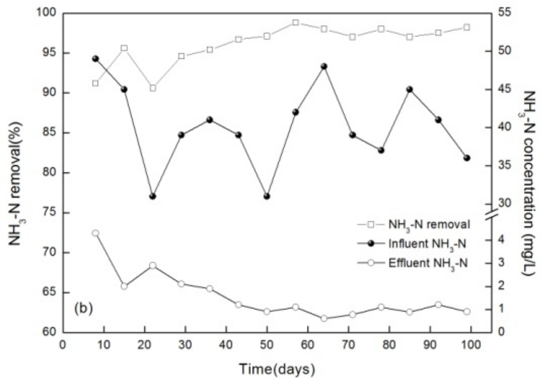
3. Results and discussion

3.1 Performance of the MBR

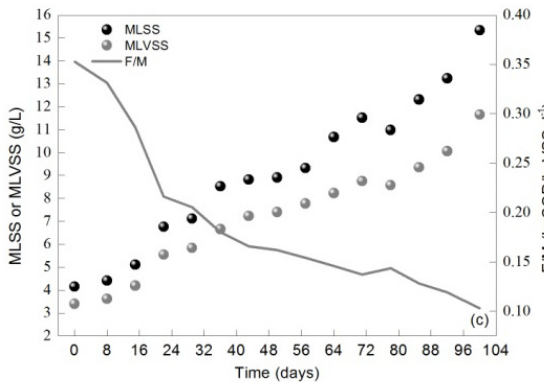
The characteristics of influent and effluent in the MBR were shown in Figs. 2(a) and (b). It could be seen that the influent COD fluctuated from 257 to 324 mg/L, which might be expected that some organic fractions were degraded in the wastewater tank due to the high temperature in the summer, similar phenomena was found in our previously study (Zhang *et al*. 2014b). The



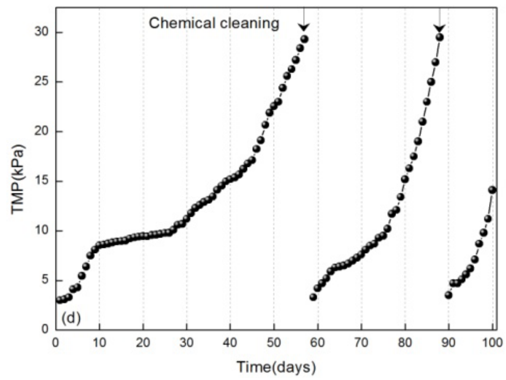
(a) COD concentration and removal efficiency



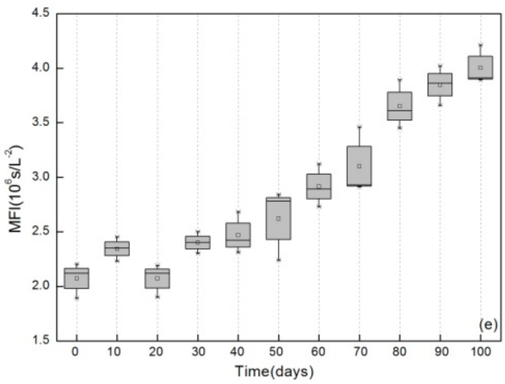
(b) NH_3-N concentration and removal efficiency



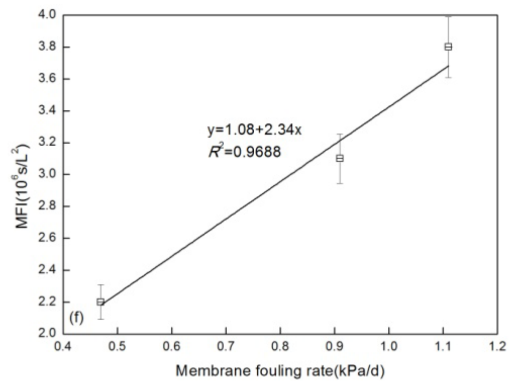
(c) Variation of MLSS, MLVSS, F/M



(d) Change of TMP with time



(e) Variations of MFI



(f) Relationship between MFI and fouling rate

Fig. 2 Performance of the MBR under no sludge discharge

effluent COD varied in range of 19-30.2 mg/L, with the average COD removal rate of > 90% during the entire operation. With respect to NH_3-N, the influent concentration ranged 31 to 49 mg/L and the effluent varied in range of 0.78-3.1 mg/L. It was observed that the NH_3-N removal efficient continuously increased with the operation time, which was explained that the autotrophic nitrifies could proliferate without any loss due to the membrane rejection and completely sludge retention. Moreover, nitrifying bacteria would meet less competition from other heterotrophic microorganisms at a long SRT (Chen *et al*. 2012). Overall, a good effluent quality was observed throughout the complete experiment, indicating that the completely sludge retention did not adversely affect the COD removal efficiency, conversely, which had a benefit for the NH_3-N removal due to enhance growth of nitrifying bacteria.

The variations of MLSS, MLVSS and F/M with the time under a complete sludge retention condition were demonstrated in Fig. 2(c). It was clear that the MLSS concentration increased with the operation time, reaching as high as 15.8 g/L at 100 days. During the whole operation time, the average MLVSS/MLSS ratio of 80% was observed, indicating that the sludge had higher activity and no obvious accumulation of inorganic matter under this condition. Corresponding to the change of the MLVSS concentration, the F/M ratio gradually decreased and finally approached to 0.1 kg COD/(kg VSS d)$^{-1}$ at the day 100. Under such a low loading operation, the majority of microorganisms in MBR suffered limited supply of substrate and were constrained to enter the endogenous respiration state, and thus will influence the AS characteristics (Chen *et al*. 2012). A lower F/M ratio enhances organic removal efficiency and sludge flocculation, thereby increasing the settleability of biomass in the reactor (Li *et al*. 2011). However, if the F/M ratio becomes too low, cell growth is limited and sludge deflocculation may also occur (Ghangrekar *et al*. 2005).

Typical TMP profiles depicting fouling trends for the MBR was shown in Fig. 2d. The membrane fouling rates were computed based on TMP development each day (dTMP/day) through the experimental time. The increase rate of TMP is an important indicator for evaluating the development of membrane fouling in submerged MBRs (Le-Clech *et al*. 2006). Based on the TMP rise, the total operation time could be divided into three stages, the membrane fouling rates were 0.48, 0.93 and 1.11 kPa/d, respectively. The MFI parameter gives an idea of the fouling potential characteristics of the mixed liquor. A lower MFI of the sludge was beneficial to the membrane filterability improvement in the MBR. It is based on the cake filtration mechanism, as recently reported by Jang and co-workers (Jang *et al*. 2006). Fig. 2(e) showed the change of MFI of mixed liquor with operation time. The MFI values ranged from 1.8 to 2.1 × 10^6 s/L^2 before 40 days, suggesting that the changes in mixed liquor filterability were not obvious in the initial stage experiment. It was after 40 days that MFI values started a sharp linearly increased as the time and the maximum MFI had reached to 4.2 × 10^6 s/L^2 at 100 days, which was more than 2-time as higher as that of the initial stage. These results indicated that the long term operation for the MBR with no sludge discharge had a negative influence on the filterability of mixed sludge. The relationship between the fouling rate and the MFI was shown in Fig. 2(f). The linear curve showed a strong correlation between the MFI and the fouling rate with an r-squared value of 0.9688. Therefore, the MFI could be considered as a reliable mixed sludge filterability indicator to predict the extent of membrane fouling rate in MBR filtration process. A previous study showed a similar result (Zhang *et al*. 2014a).

3.2 Correlation between MFI and sludge properties

Lots of experimental studies and practical operations have indicated that the sludge flocs and microbial metabolites were a major cause of membrane fouling (Sponza 2003). The interaction of sludge and metabolites with the membrane surface and pore spaces could cause the increasing filtration resistance, which led to decrease membrane filterability. Table 1 showed the results obtained from statistical analyses of activated sludge parameters in the MBR. The MFI was found highly related to the amount of SMP and EPS, which indicated that the microbial metabolites

Table 1 Correlations between MFI and sludge properties in MBR

Item	MFI (s/L^2)	EPS (mg/L)	SMP (mg/L)	MLSS (mg/L)	Zeta (mV)	Viscosity (mPas)	PSD (μm)
MFI	1						
EPS	0.7765	1					
SMP	0.8235	0.7904	1				
MLSS	0.4521	0.7321	0.2517	1			
Zeta	0.3472	-0.7234	-0.6892	0.0512	1		
Viscosity	-0.5298	0.3786	0.8921	0.6213	-0.2636	1	
PSD	-0.6328	0.7021	-0.6231	0.1574	-0.5879	0.2415	1

greatly influence the AS filterability in MBR without sludge discharge. Compared with EPS (r_p = 0.7765), the concentration of SMP (r_p = 0.8235) had a more significant impact on the filterability, suggesting that the relatively high fouling role played by SMP compared to those of EPS. It was reported that SMP had a more directly influence on membrane fouling than EPS (Zhang *et al*. 2008), a higher SMP content corresponded to a higher filtration resistance (Lee *et al*. 2003, Meng *et al*. 2006).

Apparently, EPS and SMP had also a strong correlation with other sludge properties. For example, SMP had a strong correlation with the viscosity (r_p = 0.7765), while the EPS was closely relevant to the MLSS (r_p = 0.7321), zeta potential (r_p = -0.7234) and PSD (r_p = -0.6328), implying that the microbial metabolites, especially with regard to EPS, had a significant influence on the sludge properties. It was reported that EPS play a significant role in sludge by altering the physicochemical characteristics such as charge, hydrophobicity, and the polymeric properties (Le-Clech *et al*. 2006). Aimed to clarify their role in AS filterability, the effects of the different fractions of EPS or SMP on the filterability should be investigated in details in the following text to understand the fouling mechanisms in MBR without sludge discharge.

3.3 SMP in the supernatant

With the glucose-based substrate was used in this study, the organic residue in the supernatant mainly consisted of SMP. The change of concentration and MW distribution of SMP with the operation time were shown in Fig. 3. The concentration of SMP obviously increased before 20 days, which was probably caused by the sudden changes due to no sludge discharge in the reactor environment. Despite a certain degree of fluctuation from 20 to 60 days, SMP concentration showed a trend of increase to more than 90 mg/L at the day 100 in the supernatant, suggesting the slowly biodegradable substancees tended to accumulate in MBR without sludge discharge. SMP increase could be mainly attibuted to the decrease in the F/M ration, which was ralate to the microbial viability (Chen *et al*. 2012). As the F/M decreased, microbial competition for nutrients resulted in a certain number of microorganisms lost their viability, and the breakdown of dead cells caused a large dissolution of intracellular substances, causing SMP concetration accumualted gradually in the supernatant.

The SMP had a broad spectrum of molecular weight (see in Fig. 3), the fraction of MW < 1 kDa (small molecules) was dominant before 40 days, accounting for 47.4-52.9%. However, the majority of the dissolved species lied in the MW range more than 10 kDa (macromolecules) after

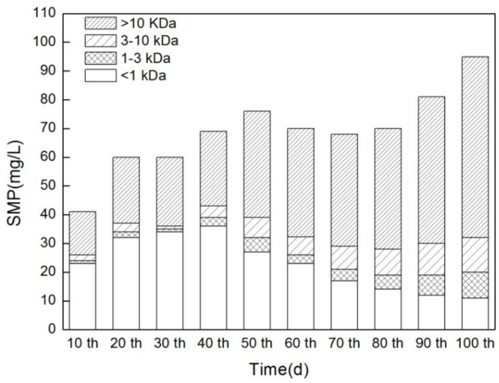


Fig. 3 Change of MW distributions of SMP during the operation time

40 d, for example, the MW > 10 kDa accounted for 66.3% in total SMP at the 100-day. These results indicated that the MW had a changing trend from the small molecules to the macromolecules in the supernantant under no sludge discharge condition. A similar phenomenon of macromolecules accumulation in a MBR suspension had been reported previously (Zhang *et al*. 2008). Moreover, the tendency of macromolecules changing in the supernatant was exactly cioncide with that of the MFI values (see in Fig. 2(e)), suggesting the high MW soluble materials would potentially deteriorate sludge filterability.

To confirm the effect of different factions of SMP on the mixed liquor filterability, MFI values were correlated with concentrations of different MW fractions (Fig. 4). The MFI values were most correlated with the concentrations of SMP with MW > 10 kDa (R^2 = 0.9012), suggesting that membrane fouling was directly related to the concentration of SMP with high MW fraction. During filtration of SMP with high MW fraction, pore adsorption would rapidly block membrane pores, causing more organics to be captured and accumulate on the membrane. On the other hand, the fractions with high MW would also fill the void space of cake layer on the membrane surface, which would increase the cake resisitance.

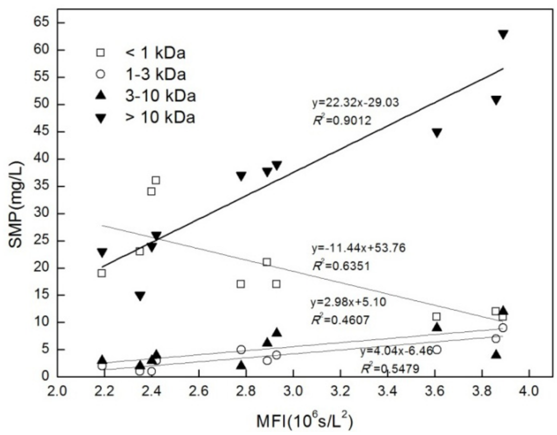


Fig. 4 Correlation between MFI and different fractions of SMP

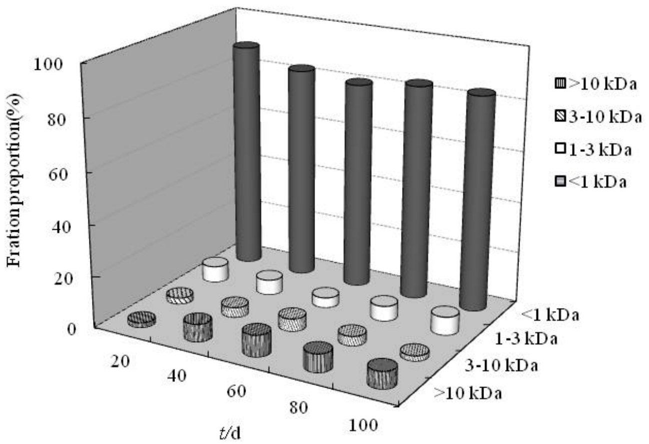


Fig. 5 MW distributions of SMP in the MBR effluent

The membrane fouling might be in close association with solute rejection in supernatant because the membrane fouling could be originated from adsorption of organic species and adhesion of microbial cells at the membrane surfaces (Liang *et al*. 2007). The MW distributions of SMP in the effluent were presented in Fig. 5. The fraction of MW < 1 kDa (small molecules) was dominant over the time, which constituted above 80% of the effluent. It was evident that a greater proportion of SMP with small molecules was not effectively retained by the MF membrane, which also exhibited a little effect on the MFI because of their sizes (see Fig. 4).The fraction of MW > 10 kDa (macromolecules) composed the second dominant fraction, accounting for about 10% after 40 days. The high macromolecules rejection efficiency attributed enhanced membrane rejection to the formation of a fouling layer on the membrane surface that affected molecular sieving (Liang *et al*. 2007), which was also proved that the macromolecules in supernatant had a high potential to foul membrane (see Fig. 4).

3.4 EPS in the sludge flocs

Generally, the amount and composition of EPS is influenced by the microbial species, growth stage, limitation of substrate, and other conditions (Wang *et al*. 2009). Complete sludge retention would induce changes the physiological state and the endogenous respiration state (Villain and Marrot 2013), and thus might affect the composition of EPS. It is generally accepted that carbohydrates and proteins are the major components of EPS (Zhang *et al*. 2014a). During the study period, EPS protein (EPSp) presented higher variable values than EPS carbohydrate (EPSc) became the predominant EPS component (Fig. 6). The average concetration of EPSp was 38.2 mg/L at day 10, which increase gently 93.7 mg/L at day 60. A higher EPSp might be due to the presence of a large quantity of exoenzymes, the biodegradable organic substances (such as glucose in this study) would give rise to a high level of exoenzymes in the EPS matric (Spona *et al*. 2003). After 60 days, the EPSp amount switched its ascending trend into a descending pattern and leveled off at 50.2 mg/L at day 100. The decrease of EPSc content indicated that the protein could be degraded as a carbon source (Sheng *et al*. 2010), as the opertion time increase, the biodegradation process becomed more effective and consequently the concentrations of EPSp were significantly reduced.

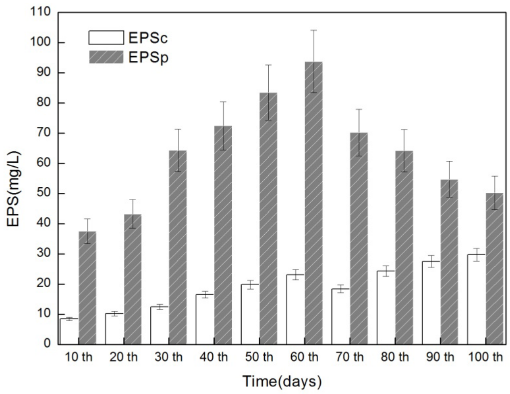


Fig. 6 Change of the concentrations of EPSc and EPSp with operation time

The variation trend of EPSc concentration was similar with EPSp before 60 days, the EPSc amount increased from 8.4 mg/L to 23.1 mg/L with the operation time (from 10 to 60 days). The observed EPSc reduction from 60 to 70 days was lower compared to the respective reduction of EPSp, and then the amount of EPSc continuously increased until the end of experiment. The increase of EPSc could be attributed to the fact that endogenous respiration played a dominant role and the number of deaths increased in this state. Increase in microbial death and intracellular polymers arose from cell lysis and secretion was represented as the increase of EPSc (Wang *et al*. 2009).

The correlation between MFI and the concentration of EPSp and EPSc were shown in Fig. 7. It can be seen that MFI showed a positive correlation to the concentration of EPSc (R^2 = 0.8682), while EPSp showed a weaker correlation through the high amounts (R^2 = 0.1245). Obviously, the EPSc was the major factor in EPS affecting the AS filterability, which was explained that EPSc had a more wide molecular weight distributions as compared to EPSp (Zhang *et al*. 2014a), resluting in a significant role in AS filterability. Moreover, it was aslo reported that EPSc can increase the hydrophobicity of sludge flocs and reduce its floc size and dewaterability (Le-Clech *et al*. 2006), suggesting that maintain the low level of EPSc was necessary.

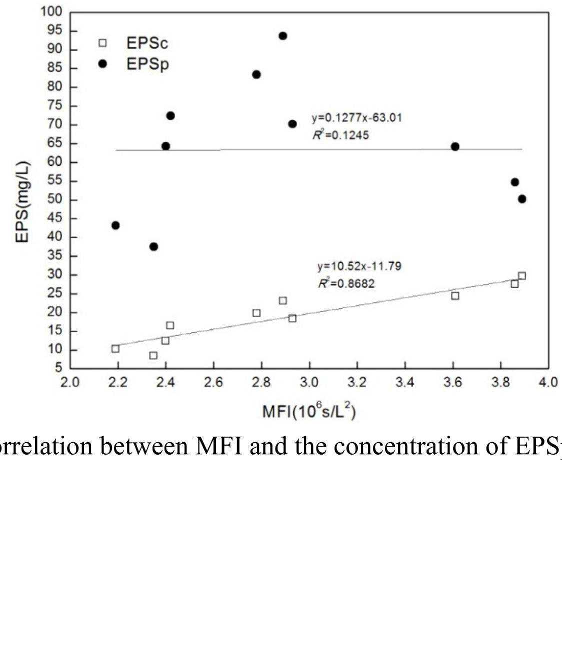


Fig. 7 Correlation between MFI and the concentration of EPSp or EPSc

4. Conclusions

The results confirmed that an infinite SRT had no obviously effect on COD removal, but it benefited for the NH_3-N removal. As the MLSS increased in this operation mode, a low level of F/M had a negative influence on the filterability of mixed sludge, resulting in an increase in membrane fouling rates with the time. Statistical analysis indicated that the AS filterability had strong correlation to the amount of SMP and EPS. For the SMP in the supernatant, the MW distributions had a changing trend from the small molecules to the macromolecules under no sludge discharge condition. The SMP amount, especially the macromolecules with MW > 10 kDa, were correlated positively to sludge filterability. The fraction of MW < 1 kDa (small molecules) was dominant in the effluent. The protein fractions in the EPS were biodegradable and available for microorganism metabolism after about 60 days, and the carbohydrates in the EPS had a significantly negative effect on the sludge filterability in MBR at an infinite SRT.

Acknowledgments

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References

APHA (2005), Standard methods for the examination of water and wastewater, (12th Ed.), American Public Health Association, American Water Works Association, Water Environmental Federation; Washington, USA.

Chen, W., Liu, J. and Xie, F. (2012), "Identification of the moderate SRT for reliable operation in MBR", *Desalination*, **286**, 263-267.

Delgado, L., Marquis, V., Schetrite, S., Leszkowicz, A., Paranthoen, S. and Albasi, C. (2010), "Effect of cytostatic drugs on the sludge and on the mixed liquor characteristics of a cross-flow membrane bioreactor: Consequence on the process", *J. Membr. Sci.*, **347**(1-2), 165-173.

Duan, L., Song, Y., Yu, H. and Xia, S. (2014), "The effect of solids retention times on the characterization of extracellular polymeric substances and soluble microbial products in a submerged membrane bioreactor", *Bioresour. Technol.*, **163**, 395-398.

Dubois, M., Gilles, K.A., Hamilton, J.K., Rebers, P.A. and Smith, F. (1956), "Colorimetric method for determination of sugars and related substances", *Anal. Chem.*, **28**(3), 350-356.

Ghangrekar, M.M., Asolekar, S.R. and Joshi, S.G. (2005), "Characteristics of sludge developed under different loading conditions during UASB reactor start-up and granulation", *Water Res.*, **39**(6), 1123-1133.

Huang, X., Gui, P. and Qian, Y. (2001), "Effect of sludge retention time on microbial behavior in a submerged membrane bioreactor", *Process Biochem.*, **36**(10), 1001-1006.

Huang, L.N., Wever, H.D. and Diels, L. (2008), "Diverse and distinct bacterial communities induced biofilm fouling in membrane bioreactors operated under different conditions", *Environ. Sci. Technol.*, **42**(22), 8360- 8366.

Jang, N., Ren, X. and Cho, J. (2006), "Steady-state modeling of bio-fouling potentials with respect to the biological kinetics in the submerged membrane bioreactor (SMBR)", *J. Membr. Sci.*, **284**(1-2), 352-360.

Judd, S. (2011), "The MBR book: Principles and applications of membrane bioreactors for water and wastewater treatment", Butterworth-Heinemann.

Kraume, M. and Drews, A. (2010), "Membrane bioreactors in waste water treatment status and trends", *Chem. Eng. Technol.*, **33**(8), 1251-1259.

Krzeminski, P., Iglesias-Obelleiro, A., Madebo, G., Garrido, J.M., van der Graaf, J.H.J.M. and van Lier, J.B. (2012), "Impact of temperature on raw wastewater composition and activated sludge filterability in full-scale MBR systems for municipal sewage treatment", *J. Membr. Sci.*, **423**, 348-361.

Le-Clech, P., Chen V. and Fane, A.G. (2006), "Fouling in membrane bioreactors used in wastewater treatment", *J. Membr. Sci.*, **284**(1-2), 17-53.

Lee, W., Kang, S. and Shin, H. (2003), "Sludge characteristics and their contribution to microfiltration in submerged membrane bioreactors", *J. Membr. Sci.*, **216**(1-2), 217-227.

Li, A.J., Li, X.Y. and Yu, H.Q. (2011), "Effect of the food-to-microorganism (F/M) ratio on theformation and size of aerobic sludge granules", *Process Biochem.*, **46**(12), 2269-2276.

Liang, S., Liu, C. and Song, L. (2007), "Soluble microbial products in membrane bioreactor operation: Behaviors, characteristics, and fouling potential", *Water Res.*, **41**(1), 95-101.

Lowry, O.H., Rosebrough, N.J., Farr, A.L. and Randall, R.J. (1951), "Protein measurement with the Folin phenol reagent", *J. Biol. Chem.*, **193**(1), 265-275.

Masse, A.. Sperandio, M. and Cabassud, C. (2006), "Comparison of sludge characteristics and performance of a submerged membrane bioreactor and an activated sludge process at high solids retention time", *Water Res.*, **40**(12), 2405-2415.

Meng, F., Zhang, H. and Yang, F. (2006), "Effect of filamentous bacteria on membrane fouling in submerged membrane bioreactor", *J. Membr. Sci.*, **272**(1-2), 161-168.

Morgan, J.W., Forster, C.F. and Evison, L.A. (1990), "Comparative study of the nature of biopolymers extracted from anaerobic and activated sludge", *Water Res.*, **24**(6), 743-750.

Pollice, A., Laer, G. and Blond, M. (2004), "Biomass growth and activity in a membrane bioreactor with complete sludge retention", *Water Res.*, **38**(7), 1799-1808.

Pollice, A., Saturno, D. and Giordano, C. (2006), "Long term effects of complete sludge retention in a membrane bioreactor for municipal wastewater treatment", *Desalination*, **199**(1-3), 325-327.

Pollice, A., Laera, G., Saturno, D. and Giordano, C. (2008), "Effects of sludge retention time on the performance of a membrane bioreactor treating municipal sewage", *J. Membr. Sci.*, **317**(1-2), 65-70.

Sheng, G.P., Yu, H.Q. and Li, X.Y. (2010), "Extracellular polymeric substances (EPS) of microbial aggregates in biological wastewater treatment systems: A review", *Biotechnol. Adv.*, **28**(6), 882-894.

Sponza, D.T. (2003), "Investigation of extracellular polymer substances (EPS) and physicochemical properties of different activated sludge flocs under steady-state conditions", *Enzyme Microb. Technol.*, **32**(3-4), 375-385.

Sun, F., Lv, X., Li, J., Peng, Z., Li, P. and Shao, M. (2014), "Activated sludge filterability improvement by nitrifying bacteria abundance regulation in an adsorption membrane bioreactor (Ad-MBR)", *Bioresour. Technol.*, **170**, 230-238.

Tian, Y., Li, H., Li, L., Su, X., Lu, Y., Zuo, W. and Zhang, J. (2015), "In-situ integration of microbial fuel cell with hollow-fiber membrane bioreactor for wastewater treatment and membrane fouling mitigation", *Biosens. Bioelectron.*, **64**, 189-195.

Villain, M. and Marrot, B. (2013), "Influence of sludge retention time at constant food to microorganisms ratio on membrane bioreactor performances under stable and unstable state conditions", *Bioresour. Technol.*, **128**, 134-144.

Wang, Z., Chu, J., Song, Y., Cui, Y., Zhang, H., Zhao, X., Li, Z. and Yao, J. (2009), "Influence of operating conditions on the efficiency of domestic wastewater treatment in membrane bioreactors", *Desalination*, **245**(1-3), 73-81.

Zhang, H.F., Sun, B.S., Zhao, X.H. and Gao, Z.H. (2008), "Effect of ferric chloride on fouling in membrane bioreactor", *Sep. Purif. Technol.*, **63**(2), 341-347.

Zhang, H.F., Wang, Z., Zhang, L. and Song, L. (2014a), "Impact of sludge cation distribution pattern on its filterability in membrane bioreactor", *Bioresour. Technol.*, **171**, 16-21.

Zhang, H.F., Gao, Z., Zhang, L. and Song, L. (2014b), "Performance enhancement and fouling mitigation by organic flocculant addition in membrane bioreactor at high salt shock", *Bioresour. Technol.*, **164**, 34-40.