

## Evaluation of fouling prevention methods in a submerged membrane bioreactor treating domestic wastewater

Konstantinos Azis, Spyridon Ntougias, Paraschos Melidis\*

Laboratory of Wastewater Management and Treatment Technologies, Department of Environmental Engineering, Democritus University of Thrace, Vas. Sofias 12, Xanthi 67132, Greece, Tel. +30-25410-79372; emails: pmelidis@env.duth.gr (P. Melidis), kazis@env.duth.gr (K. Azis), sntougia@env.duth (S. Ntougias)

Received 9 April 2019; Accepted 27 August 2019

---

### ABSTRACT

Membrane fouling is considered as the main drawback that currently restricts use of membrane bioreactors (MBR) in domestic wastewater treatment. Microbiological factors such as extracellular polymeric substances (EPS) and soluble microbial products (SMP) have a critical impact on sludge filterability of membrane surfaces. The effects of operating parameters and sludge characteristics on fouling were examined in a pilot-scale MBR treating domestic wastewater under alternating aeration and intermittent feeding conditions. EPS and SMP were measured under various operating conditions and their effects on membrane fouling were assessed. Both glycerol and biosolids addition resulted in carbohydrate EPS and SMP increase, whereas operating mixed liquor temperature above 19°C drastically reduced membrane flux as the consequence of the enhanced microbial activity. The first transmembrane pressure derivative (dTMP/dt) proved to be a suitable indicator of critical flux since an acute raise in dTMP/dt occurred. The implementation of various fouling prevention methods, such as increased backwash flow rate, cross flow aeration, in situ mechanical cleaning, anoxic/aerobic phase duration ratio and offline intensive chemical cleaning, resulted in the improvement of membrane permeability (P). Moreover, the increase in anoxic/aerobic phase ratio accelerated membrane fouling.

*Keywords:* Extracellular polymeric substances (EPS); Soluble microbial products (SMP); Membrane resistance (R); TMP first derivative; Anoxic/aerobic cycle; Wastewater treatment

---

### 1. Introduction

Fouling in membrane treating wastewater increases energy requirement and minimizes membrane lifespan, resulting in higher operating costs [1]. Microbial aggregates deposited on membrane pores increase membrane resistance, creating a more intense and irreversible membrane clogging [2]. The key factors that affect the permeability are the membrane material, the mixed liquor characteristics, the strength and the composition of the influent. Moreover, the main operating parameters, such as the sludge retention time (SRT) and food-to-microorganism ratio (F/M ratio),

highly affect membrane fouling [3–5]. Temperature is a bio-kinetic factor, has a direct impact on biosolids generation [3] and subsequently on membrane fouling [6]. Nonetheless, the temperature is not the only parameter that influences membrane resistance to filtration [6–8]. Sludge characteristics and in particular extracellular polymeric substances (EPS) and soluble microbial products (SMP) concentrations affect membrane resistance [1,9], increasing membrane cleaning frequency. A range of factors, such as composition of wastewater, SRT, mixed liquor suspended solids (MLSS) and bacterial growth rate, have strong impact on EPS and SMP levels [5,10].

---

\* Corresponding author.

Fouling control can be achieved by altering the operating conditions and optimizing bioprocesses as well as by applying physico-chemical cleaning methods. An extensive number of techniques has been implemented to restrict flux decline [5,11]. Emerging fouling prevention methods includes the regulation of the air flow velocity [12], the increase in backwash flow rate [13–15], the implementation of intermittent aeration strategies [16–18] and the performance of the filtration process below the estimated critical flux [19]. Intermittent aeration is a well-established method to control membrane fouling with reduced energy requirement [12,16,19]. Mechanical scouring practices can decrease energy consumption in aerobic membrane bioreactor (MBRs). On the other hand, mechanical fouling mitigation approaches are limited, therefore in this study, pneumatic cleaning of the membrane by air sparging was investigated. Membrane cleaning through utilization of the mechanical properties of a sponge is an alternative way to control fouling. [20,21]. Moreover, in situ and ex situ mechanical cleaning practices can improve cleaning efficacy [22].

Chemical cleaning is implemented in emergency fouling incidents, which are known as irreversible fouling, where strongly absorbing deposits are removed [23]. Organic fouling is due to proteins, polysaccharides, humic acids and other organic molecules [24]. However, the frequency of chemical cleaning can be minimized by applying the abovementioned physical methods. An alternative approach to prevent membrane fouling on long-term basis is bioreactor operation under the critical flux [25], which is commonly determined by the flux-step method [25–27]. In particular, some studies have suggested that the critical flux in MBR systems should be set at the two-third of the maximum flux to sustain effective membrane operation, where both transmembrane pressure (TMP) and flux profile are linearly correlated [28].

The scope of this study was to prevent membrane fouling in MBRs for extended time period by controlling critical flux and consequently the effects of EPS and SMP on membrane permeability under various operating conditions, including temperature and anoxic/aerobic cycle ratio. Moreover, the appropriate fouling prevention methods should be followed are proposed in relationship to membrane resistance.

## 2. Materials and methods

### 2.1. Experimental setup and operating conditions

A pilot-scale MBR system that consists of a 40 L of feeding tank, a 100 L of bioreactor and an 80 L of external membrane tank was installed (Fig. 1). A BE/M-25 CE ROVER brass pump was placed within the two tanks, resulting in the recirculation of the activated sludge from the external MBR tank back to the cylindrical bioreactor by operating 20 s every 3 min at 28 L min<sup>-1</sup>, corresponding to a mean flow rate of 187 L h<sup>-1</sup>. A radial piston stirrer was setup in the main bioreactor and a blower supplied air into a fine bubble diffuser plate at a rate of 1.5 L m<sup>-1</sup>. The start-up of MBR system was achieved through the addition of mixed liquor from the wastewater treatment plant of Xanthi, Greece. The cycling feeding at the beginning of the anoxic phase enhanced the simultaneous removal of the nutrients (C, N and P) [29]. The MBR was operated under an organic loading rate of 0.27 ± 0.02 g biochemical oxygen demand (BOD<sub>5</sub>) g VSS<sup>-1</sup> d<sup>-1</sup>. During the entire experimental

period, sludge was not wasted out and the corresponding sludge age SRT was identical with the exact operating day.

The external tank was equipped with three flat-sheet ultrafiltration membranes (Microdyn Nadir UP-150, Wiesbaden, Germany). More details about the membrane technical specifications can be found in Azis et al. [29]. Fine bubble aeration diffuser was located in the membrane tank to provide air crossflow through an air-blower, thus preventing membrane clogging [30]. The level of dissolved oxygen was maintained within 2 and 3.5 mg L<sup>-1</sup> in both tanks during the aerobic phase. Internal mixing of the activated sludge inside the membrane tank was achieved by a submersible pump operating at 2,500 L h<sup>-1</sup>.

The input flow rate ( $Q_{in}$ ) was 0.18 m<sup>3</sup> d<sup>-1</sup>, corresponding to a hydraulic retention time of 1 d. MBR flow rate ( $Q_{ef}$ ) ranged between 0.048 and 0.18 m<sup>3</sup> d<sup>-1</sup>, depending on the membrane fouling events. The temperature of the mixed liquor was fluctuated from 15°C to 22°C. The TMP was monitored by a transducer and the obtained data were logged per min via the Modscan32 software. The permeate and back-flushing duration were 8 and 1 min, respectively, with intermediate relaxation periods of 30 s (total filtration cycle, 10 min). In order to control the membrane fouling, the pilot-plant was operated below the critical flux. Critical flux was estimated by the flux-step method [27].

Measurement of membrane resistance and permeability was based on Darcy's law (Eqs. 1–3):

$$R = \frac{\text{TMP}}{J \times n} \quad (1)$$

$$P = \frac{J}{\text{TMP}} \quad (2)$$

$$R = P^{-1} \times n^{-1} \quad (3)$$

where  $R$  is the membrane resistance (m<sup>-1</sup>);  $P$  is the permeability (L m<sup>-2</sup> h<sup>-1</sup> bar<sup>-1</sup>); TMP is the transmembrane pressure (mbar or bar, as appropriate);  $J$  is the permeate flux (L m<sup>-2</sup> h<sup>-1</sup>); and  $n$  is the activated sludge viscosity (kg m<sup>-1</sup> s<sup>-2</sup>).

### 2.2. Cleaning methods

Mechanical cleaning was applied either by water pressure or by the aid of a sponge with thin folds, removing membrane surface particles. Taking advantage of the sponge geometry and the sponge material characteristics, successful removal of the build-up cake layer from membrane sheets occurred. The alternating aeration approach was applied under the following anoxic/aerobic phase duration ratios (min/min): 30/60, 60/60 and 90/60. As an additional fouling prevention method, the backwash flow rate was increased from 200 to 400 mL min<sup>-1</sup> and the air flow rate was elevated from 6 to 8 L min<sup>-1</sup>. The membrane module was subjected to ex situ intensive chemical cleaning by applying 500–1,000 mg L<sup>-1</sup> NaOCl solution for a period of 24 h. The membrane tank remained inactive during this cleaning period, where biosolids were transferred to the main bioreactor.

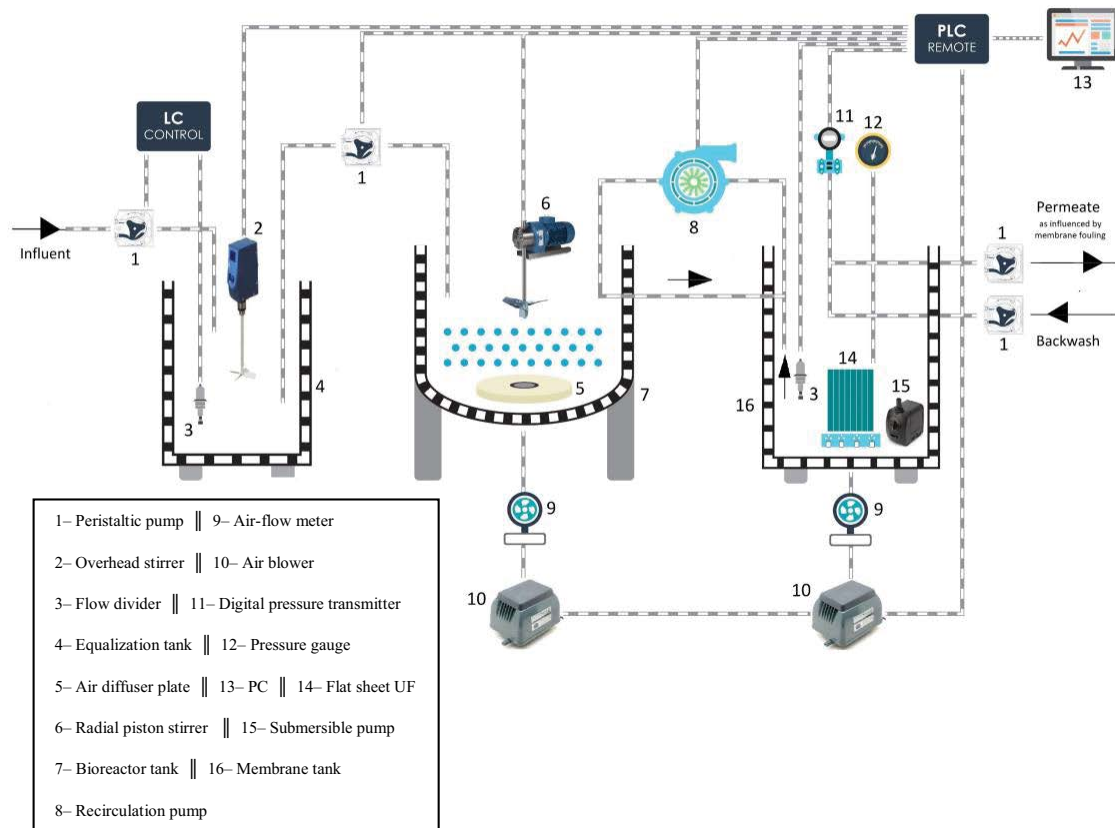


Fig. 1. MBR system configuration.

### 2.3. Control system

Membrane filtration was controlled and supervised on real time basis. A supervisory control and data acquisition system (SCADA) was in communication with a programmable logic controller tasked to monitor TMP. SCADA also managed the suction duration through online data with the aim to achieve an effective filtration performance. In addition, fouling prevention was online monitored and controlled.

### 2.4. Wastewater characteristics

The MBR was fed with domestic wastewater originating from the student Campus of Democritus University of Thrace in Xanthi. The average wastewater characteristics and their standard errors were as follows: pH,  $7.27 \pm 0.03$  mS  $\text{cm}^{-1}$ ; electrical conductivity,  $1.35 \pm 0.05$  mS  $\text{cm}^{-1}$ ;  $\text{BOD}_5$ ,  $170 \pm 10.3$  mg  $\text{L}^{-1}$ ; total chemical oxygen demand (COD),  $315 \pm 18.8$  mg  $\text{L}^{-1}$ ; soluble COD,  $123 \pm 6.48$  mg  $\text{L}^{-1}$ ; total Kjeldahl nitrogen,  $65.4 \pm 3.7$  mg  $\text{L}^{-1}$  and  $\text{NH}_4^+\text{-N}$ ,  $42.4 \pm 3.21$  mg  $\text{L}^{-1}$ .

### 2.5. Analytical methods and experimental setup

$\text{BOD}_5$ , COD, ammonium and nitrate nitrogen, total suspended and volatile suspended solids (VSS), and the other physico-chemical parameters measured were estimated every week to evaluate the performance of the pilot-plant [31].

Determination of EPS and SMP concentrations, which are dealing with the fouling of the membrane, were conducted under various experimental conditions. Samples were collected from the MBR's main bioreactor twice a week and their EPS and SMP proteins and polysaccharide concentrations were determined by using the modified Lowry [32] and the Dubois [33] method, respectively. EPS extraction was carried out by the cation exchange resin method, while the measurement of SMP was performed after centrifugation at 4,000 rpm for 10 min [34]. SMPp and EPSp concentrations are expressed as mg BSA  $\text{g}^{-1}$  VSS, bovine serum albumin (BSA), while SMPc and EPSc as mg glucose  $\text{g}^{-1}$  VSS.

All the experiments were carried out in a temperature-controlled room. A presentation of each experimental setup is given in Table 1. In the respective setups, a sludge volume of 10 L with a biosolids concentration of 9 g  $\text{L}^{-1}$  was added in the bioreactor, whereas COD was increased by 200 mg  $\text{L}^{-1}$ , when glycerol was added.

## 3. Results and discussion

The permeate flux was  $21.0 \pm 0.92$  L  $\text{m}^{-2}$   $\text{h}^{-1}$ , while the MLSS concentration reached up to 8.8 g  $\text{L}^{-1}$ . The permeability was determined to be  $186 \pm 16.8$  L  $\text{m}^{-2}$   $\text{h}^{-1}$   $\text{bar}^{-1}$ , which corresponded to a membrane resistance of  $3.21 \pm 0.22$   $\text{m}^{-1}$ . In the effluent of the MBR system, the  $\text{BOD}_5$  and COD concentrations were  $4.71 \pm 0.24$  and  $17.6 \pm 1.54$  mg  $\text{L}^{-1}$ , respectively.

Ammonium and nitrate nitrogen effluent concentrations were equal to  $1.03 \pm 0.31$  and  $0.89 \pm 0.05$  mg L<sup>-1</sup>, respectively. Notably, the effluent characteristics of the MBR were compliant with the Greek discharge limits for unrestricted irrigation enacted by the Joint Ministerial Decree 145116/11.

### 3.1. Investigation of membrane fouling aspects

#### 3.1.1. EPS and SMP profiles

Protein's EPS and SMP (mg EPSp and SMPp per gram of VSS) remained low, determining values equal to  $0.42 \pm 0.06$  and  $0.20 \pm 0.05$  mg g<sup>-1</sup> VSS, respectively (Fig. 2). The production of EPS and SMP was low, due to the fact that the influent COD was used for energy production and to a lesser extent for cell synthesis [1]. Trussell et al. [35] showed that membrane fouling rate in a submerged MBR treating municipal wastewater was increased by 20-fold, when F/M

ratio elevated from 0.34 to 1.41 g COD g<sup>-1</sup> VSS d<sup>-1</sup>. The same authors also reported that SMP concentration was greatly affected at high F/M ratio, resulting in membrane resistance. These results are in accordance with recent findings [36,37]. Instant addition of glycerol was made at days 78, 90 and 98 in order to improve the denitrification and to study its effect on EPSc and SMPc profiles. The addition of glycerol resulted in EPSc increase up to 19.2 and 10.5 mg g<sup>-1</sup> at day 82 and 102, respectively, whereas SMPc concentration was increased up to 21.5 (day 86), 16.9 (day 95) and 31.1 (day 102) mg g<sup>-1</sup> VSS. The EPSc and SMPc released were decreased to  $2.89 \pm 0.72$  and  $5.50 \pm 0.31$  mg g<sup>-1</sup> VSS, respectively, 3 d after glycerol addition.

Biosolids addition at day 140 resulted in higher EPSc and SMPc concentrations, reaching values of 12.3 (day 144) and 15.2 (day 146) mg g<sup>-1</sup> VSS, respectively. After day 146, SMPc were not further produced and their concentration was gradually declined to the lowest levels. In contrast,

Table 1  
MBR experimental setups

Anoxic/aerobic duration ratio	EPS and SMP profiles		Effect of temperature	Cleaning methods
	Glycerol addition	Biosolids addition		
30/60	–	–	Temperature rise from 15°C to 23°C (days 10 to 50)	–
30/60	days 78, 90 and 98	–	17°C	–
60/60	–	days 140 and 146	17°C	NaOCl (day 139 and 145 <sup>a</sup> )
60/60	–	day 154	17°C	Pressured water (day 154)
60/60	–	–	17°C	Backwash/air flow rate increase (day 175)
60/60	–	–	17°C	NaOCl (day 179 <sup>a</sup> )
90/60	–	–	17°C	Sponge (days 206 and 220)

<sup>a</sup>Estimation of critical flux

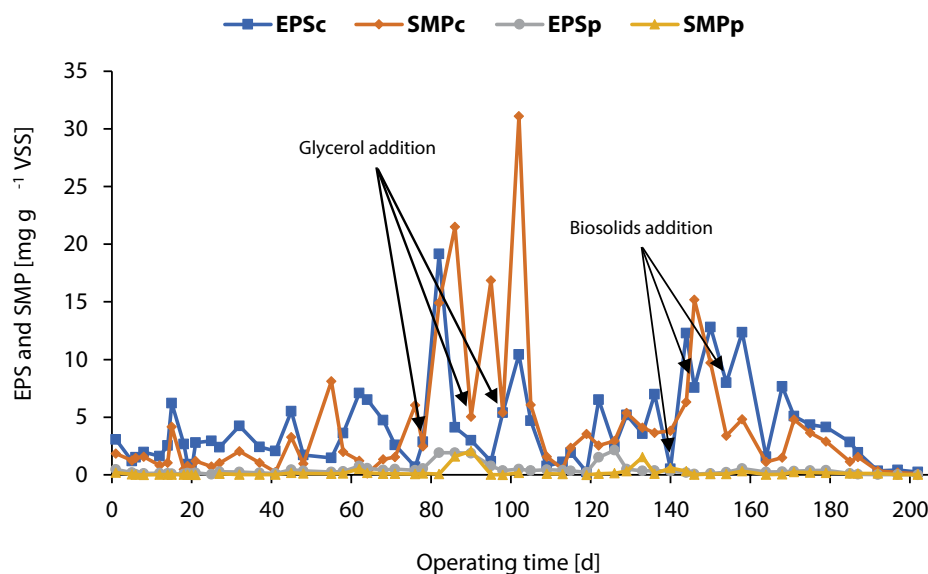


Fig. 2. EPS and SMP concentration profiles. EPSc: carbohydrate EPS; SMPc: carbohydrate SMP; EPSp: proteinous EPS; SMPp: proteinous SMP.

EPSc were more influenced by SMPc, being kept at concentrations as high as 12.3–12.8 mg g<sup>-1</sup> VSS for a period of 2 weeks. Afterwards, their concentrations were reduced to the levels prior to sludge addition.

The proteins' and carbohydrates' content were also measured in the permeate of the MBR, determining values equal to 0.02 ± 0.01 and 0.66 ± 0.29 mg L<sup>-1</sup>, respectively. Proteins were not detected in the permeate, may be due to their possible larger size than membrane pores [38]. Moreover, the SMPp/SMPc ratio was as low as 0.12 ± 0.04, suggesting that carbohydrates constitute a larger fraction than proteins.

SMPc increase can result in the formation of a thin gel layer on membrane surface [5,39], provoking pore blocking and maximizing membrane resistance (Fig. S1). By performing Pearson correlation test, a significant relationship between SMPc and *R* was found (correlation coefficient  $r = 0.681$ ,  $p < 0.01$ ). Fig. 3 illustrates the correlation of membrane resistance with SMPc between day 73 and day 115, where a positive relationship was observed. From day 78 to 86, SMPc were increased from 2.46 to 21.5 mg g<sup>-1</sup> VSS and the membrane resistance from 0.97 to 2.04 m<sup>-1</sup>. From day 91 to 95, SMPc were increased from 5.04 to 16.9 mg g<sup>-1</sup> VSS and the membrane resistance from 1.16 to 2.89 m<sup>-1</sup>. Lastly, SMPc were increased from 5.39 to 31.1 mg g<sup>-1</sup> VSS and the membrane resistance from 1.50 to 3.06 m<sup>-1</sup> from day 99 to 104.

### 3.1.2. Critical flux evaluation

A stable filtration process, which is characterized by a constant flux for an extended time period, can be achieved by operating the MBR below the critical flux [26]. Critical flux was experimentally determined by plotting flux against TMP, whereas dTMP/dt is the slope of the TMP against time (Fig. 4). TMP was gradually elevated for each flux step increase, while dTMP/dt jump from 0.75 to 6.96 mbar min<sup>-1</sup> clearly denoted membrane fouling effects. This dTMP/dt threshold corresponds to the critical flux (32.6 L m<sup>-2</sup> h<sup>-1</sup>), therefore the MBR was operated below this threshold to prevent membrane fouling. However, the critical flux was

reported to be lower than that found in the current study, that is, 13, 16 and 18 L m<sup>-2</sup> h<sup>-1</sup> regarding the data of Psoch and Schiewer [40], Tiranuntakul et al. [28] and Le Clech et al. [26]. However, Nguyen et al. [41] was reported the identical critical flux (33 L m<sup>-2</sup> h<sup>-1</sup>).

### 3.1.3. Effect of temperature on membrane resistance

Fig. 5 presents the influence of the mixed liquor temperature on membrane resistance. By increasing the temperature from 14.8°C to 18.2°C, membrane resistance was steadily increased from 0.83 to 1.98 m<sup>-1</sup>. By exceeding 19°C, the membrane resistance was drastically increased up to 4.63 ± 0.09 m<sup>-1</sup>, thus sharply decreasing flux from 26.4 ± 2.27 to 15.9 ± 1.34 L m<sup>-2</sup> h<sup>-1</sup>. Research works have reported that temperature near 20°C can result in increased level of EPS and SMP, due to the enhanced microbial activity, a fact that can influence membrane fouling [7,42]. Membrane resistance-to-MLSS ratio was 0.30 ± 0.06 m<sup>-1</sup> g<sup>-1</sup> L at temperatures within 14.8°C to 18.2°C, while it was sharply increased up to 1.89 ± 0.39 m<sup>-1</sup> g<sup>-1</sup> L at temperatures over 19°C.

### 3.2. Application of membrane fouling prevention methods

To control reversible and irreversible fouling, aeration scouring and chemical cleaning are proposed as the most widespread and effective methods, respectively [5]. Fig. 6 presents the cleaning methods that can be applied to control reversible and irreversible fouling.

Such strategies are hydraulic cleaning via backwash flow rate increase, cross flow aeration increase, in situ mechanical cleaning, intermittent aeration and offline intensive chemical cleaning, as the ultimate method used for irreversible fouling. At day 141, ex situ chemical cleaning with NaOCl (500 ppm) was implemented to improve the TMP and permeate flux (Fig. 6). In addition, a permeability increase from 40.4 to 183 L m<sup>-2</sup> h<sup>-1</sup> bar<sup>-1</sup> and a TMP decrease from 161 to 125 mbar was observed. Similar to the current findings, Rabuni et al. [24] found that chemical cleaning using NaOCl

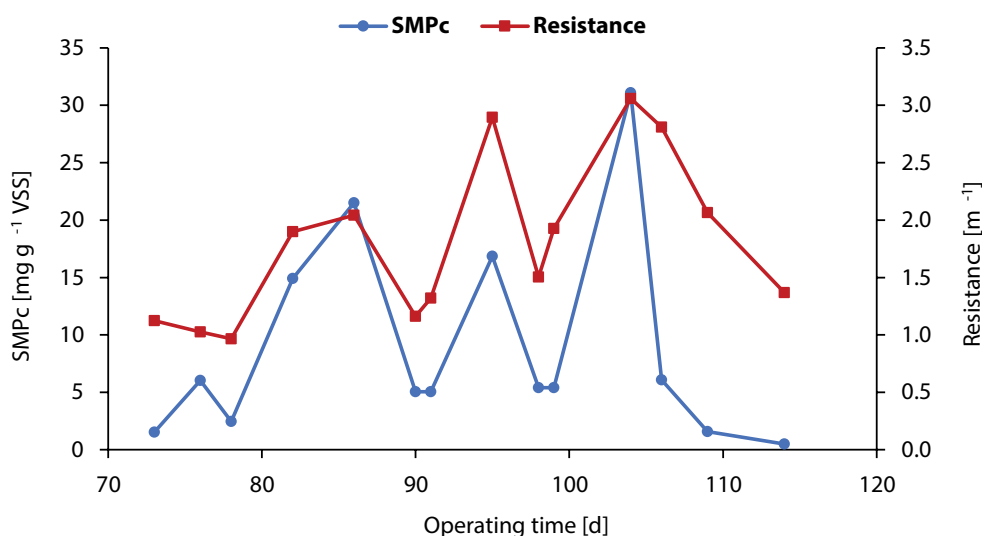


Fig. 3. Correlation between SMPc and membrane resistance. SMPc: carbohydrate SMP.

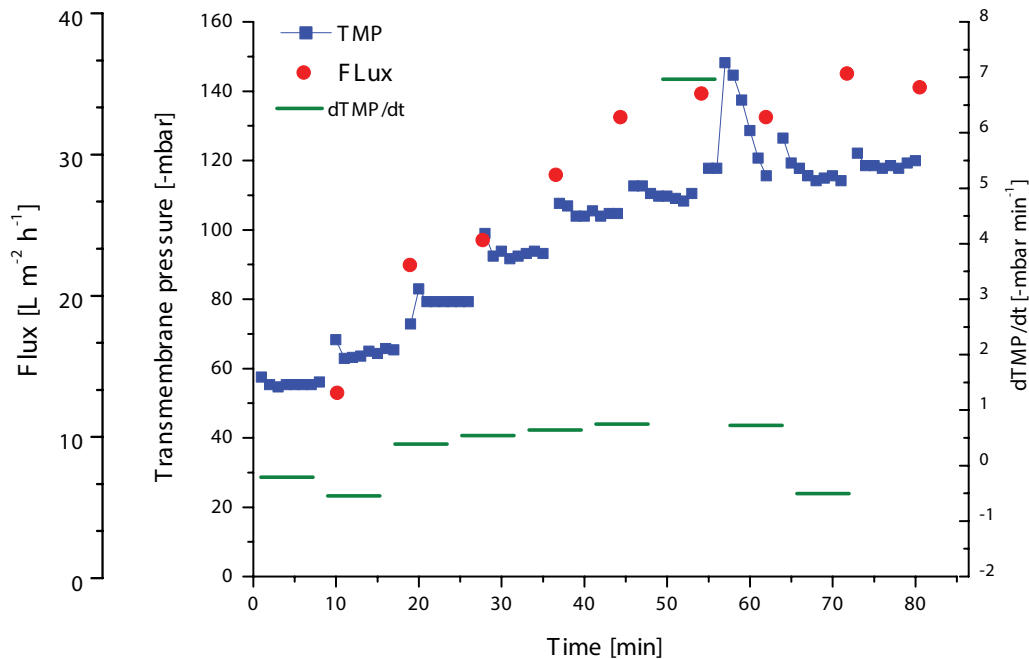


Fig. 4. Critical flux evaluation by  $dTMP/dt$  determination at MLSS concentration of  $4.5 g L^{-1}$ .

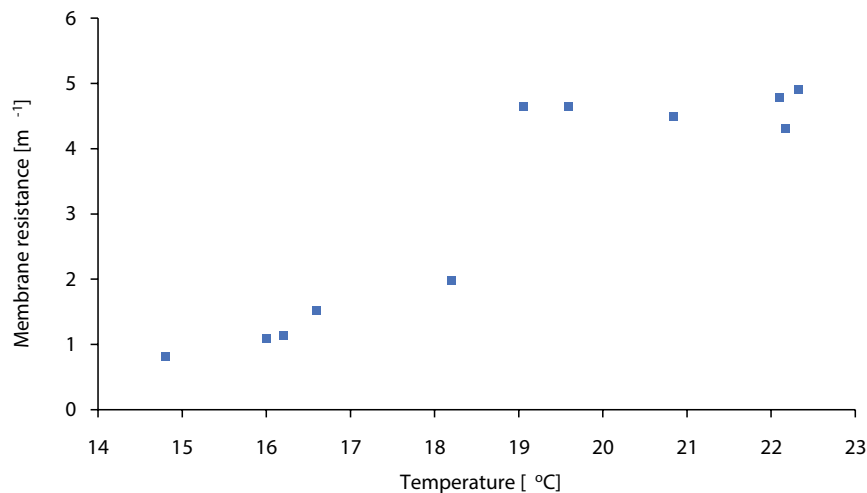


Fig. 5. Correlation between mixed liquor temperature and membrane resistance.

was an effective cleaning method to prevent irreversible fouling. At day 154, membrane was cleaned on site using pressurized water. Moreover, the permeability was increased from  $63.5$  to  $161 L m^{-2} h^{-1} bar^{-1}$  and the TMP was decreased from  $162$  to  $133$  mbar. At day 174, the permeability was increased from  $89.8$  to  $132 L m^{-2} h^{-1} bar$  when both crossflow aeration and backwash flow rate was increased, whereas TMP was slightly decreased from  $150$  to  $141$  mbar. Qaisrani and Samhaber [43] reported that the combination of back-flushing and air-bubbling is the most suitable method for improving membrane performance and shortening cleaning time. To further improve filterability, intense chemical cleaning with  $1,000$  ppm NaOCl was implemented. Thus, the permeability was increased from  $117$  to  $495 L m^{-2} h^{-1} bar^{-1}$

(day 185) and the TMP was decreased from  $150$  to  $62$  mbar. Moreover, in situ mechanical cleaning with sponge was applied, leading to a permeability improvement from  $60.3$  to  $175$  and from  $55.4$  to  $245 L m^{-2} h^{-1} bar^{-1}$  on days 206 and 220, respectively. Meanwhile, the TMP was decreased from  $183$  to  $116$  and from  $169$  to  $103$  mbar, respectively. Mechanical cleaning by sponge has been effectively applied for the membrane cleaning, removing fouling layers in MBRs [22].

### 3.2.1. Effects of anoxic/aerobic cycle on membrane fouling

The application of intermittent aeration is considered as the core of physical cleaning methods in membrane filtration process, reducing the need for chemical cleaning

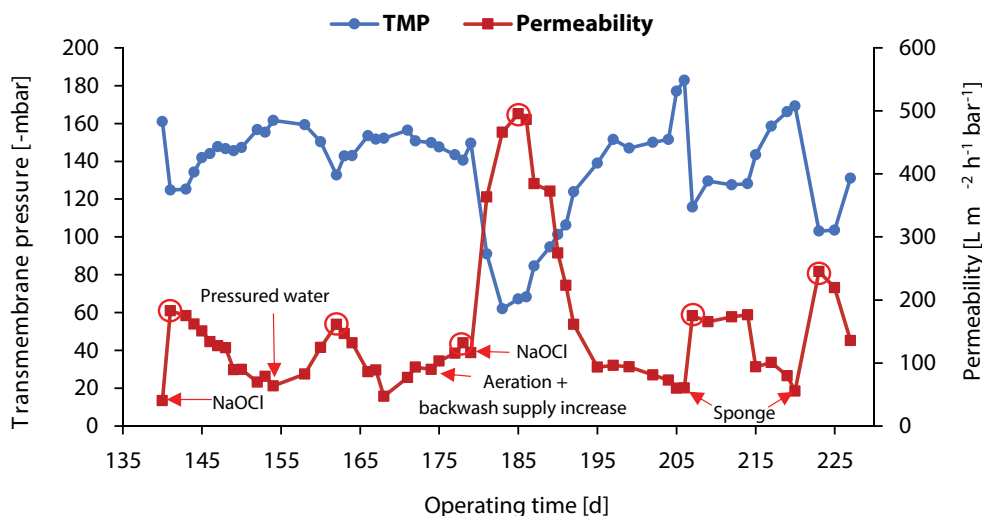


Fig. 6. Effects of cleaning events on membrane fouling prevention.

for the extended period of time [18]. The implementation of anoxic/aerobic phases of 30/60, 60/60 and 90 min/60 min affected membrane performance, as shown in Table 2. By applying anoxic/aerobic phases of 30 min/60 min, TMP, membrane resistance and flux were equal to  $76.9 \pm 2.94$  mbar,  $1.92 \pm 0.25$   $\text{m}^{-1}$  and  $25.5 \pm 2.71$   $\text{L m}^{-2} \text{h}^{-1}$ , respectively. Regarding the implementation of 60 min anoxic/60 min aerobic phase, an increase in TMP and membrane resistance up to  $142 \pm 2.87$  mbar and  $4.32 \pm 0.21$   $\text{m}^{-1}$ , respectively, and a decrease in the permeate flux to  $19.9 \pm 0.71$   $\text{L m}^{-2} \text{h}^{-1}$  was observed. A further increase in the duration of the anoxic phase (90 min anoxic/60 min aerobic) led to an additional decline in membrane efficiency. TMP and membrane resistance were increased up to  $152 \pm 1.20$  mbar and  $5.86 \pm 0.21$   $\text{m}^{-1}$ , respectively, whereas the permeate flux was decreased to  $12.7 \pm 0.38$   $\text{L m}^{-2} \text{h}^{-1}$ . Obviously, the increase in the anoxic phase duration led to membrane fouling acceleration. By changing the anoxic/aerobic phase duration ratio from 0.5 to 1.5, TMP and membrane resistance were increased by two- and three-fold, while permeate flux was decreased by two-fold. Thus, intermittent aeration of 30 min anoxic/60 min aerobic phase optimized membrane filterability and expanded membrane operating duration. According to Campo et al. [44], higher anoxic phase per anoxic/aerobic cycle should be considered as a key factor affecting membrane fouling.

EPS and SMP concentrations were also assessed under anoxic/aerobic duration phase. Due to wastewater feeding at the beginning of the anoxic phase, EPS formation was higher than occurred in the aerobic phase. EPS concentration was increased from  $68.4 \pm 10.2$   $\text{mg g}^{-1}$  VSS at the end of aeration

phase up to  $170 \pm 47.6$   $\text{mg g}^{-1}$  VSS at the end of anoxic phase. Moreover, EPS concentration was increased from  $24.3 \pm 12.4$  to  $68.4 \pm 10.2$   $\text{mg g}^{-1}$  VSS from the beginning to the end of the aerobic phase (Fig. 7a). SMP concentration was also increased from  $77.5 \pm 13.9$  to  $144 \pm 34.4$   $\text{mg g}^{-1}$  VSS from the end of the aerobic phase to the end of the anoxic phase, while SMP concentration was increased from  $17.3 \pm 17.3$  to  $77.5 \pm 13.9$   $\text{mg g}^{-1}$  VSS during the aerobic phase (Fig. 7b).

It is worth noting that the SMP concentration release was higher in the anoxic phase than the aerobic phase. This is due to the hydrolysis of EPS, reaching values of  $144 \pm 34.4$   $\text{mg g}^{-1}$  VSS at the end of anoxic phase, which is in accordance with the data of Capodici et al. [45]. At the aerobic phase, SMP concentration was reduced to  $17.3 \pm 17.3$   $\text{mg g}^{-1}$  VSS (Fig. 7), denoting the advantage of the alternate aeration process. On the other hand, SMP were also released during the aeration phase, but to a lesser extent, in disagreement with the findings of Capodici et al. [45], where SMP were absent in the aerobic phase. In accordance to Capodici et al. [45], longer anoxic phase duration increases cleaning frequency and decreases membrane lifespan.

At anoxic/aerobic phase duration ratio of 0.5, EPS and SMP concentrations remained low, exhibiting values of  $4.48 \pm 1.66$  and  $2.74 \pm 0.16$   $\text{mg g}^{-1}$  VSS, respectively, whereas the permeate flux was the highest detected, that is,  $25.5 \pm 2.71$   $\text{L m}^{-2} \text{h}^{-1}$ . According to Capodici et al. [45], SMP formation is increased by the high metabolic stress applying to the activated sludge during the anoxic phase, which can lead to cell lysis. In particular, SMP release was increased from  $4.48 \pm 1.66$  to  $10.8 \pm 3.62$   $\text{mg g}^{-1}$  VSS during the shift in

Table 2  
Effect of anoxic/aerobic phase duration on membrane fouling

Anoxic/aerobic phase duration ratio	TMP (mbar)	$R$ ( $\text{m}^{-1}$ )	Flux ( $\text{L m}^{-2} \text{h}^{-1}$ )
0.5 (30/60 min)	$76.9 \pm 2.94$	$1.92 \pm 0.25$	$25.5 \pm 2.71$
1.0 (60/60 min)	$142 \pm 2.87$	$4.32 \pm 0.21$	$19.9 \pm 0.71$
1.5 (90/60 min)	$152 \pm 1.20$	$5.86 \pm 0.21$	$12.7 \pm 0.38$

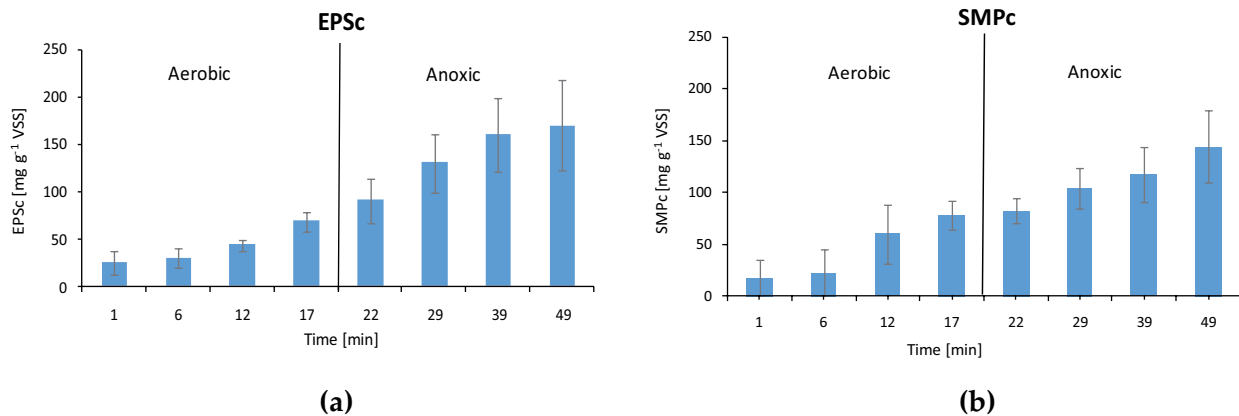


Fig. 7. Determination of (a) EPSc and (b) SMPc at various time points of an anoxic/aerobic cycle. EPSc: carbohydrate EPS; SMPc: carbohydrate SMP.

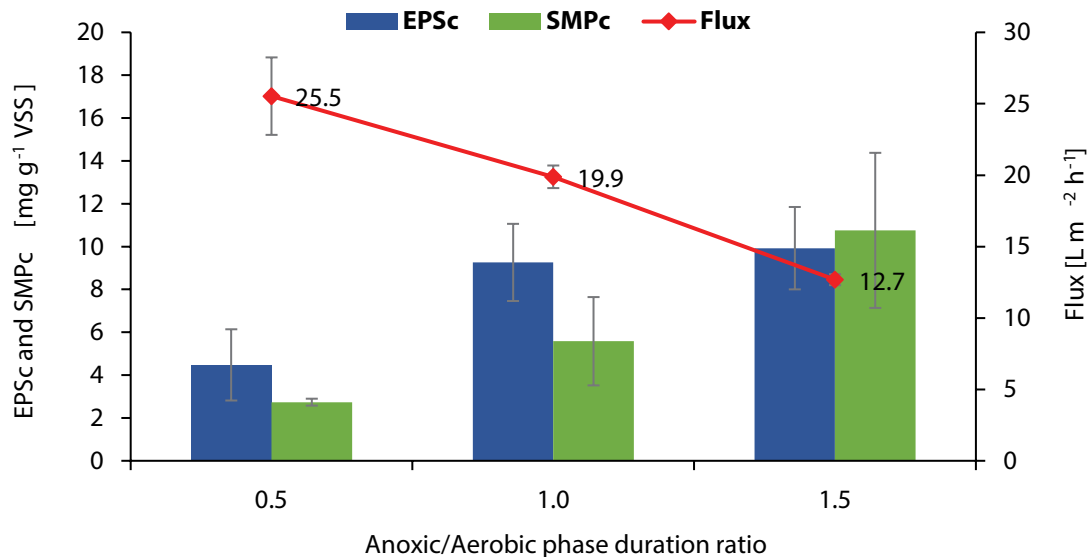


Fig. 8. EPSc, SMPc and permeate flux at various anoxic/aerobic phase duration ratios. EPSc: carbohydrate EPS; SMPc: carbohydrate SMP.

anoxic/aerobic phase from 0.5 to 1.5. At the highest anoxic phase duration examined, SMPc were released as the consequence of EPS hydrolysis [46], owing to the stress induced to microorganisms by the intermittent aeration [44]. EPSc were increased from  $4.48 \pm 1.66$  to  $9.26 \pm 1.80$   $\text{mg g}^{-1}$  VSS when the anoxic/aerobic phase duration ratio was shifted from 0.5 to 1.0. However, further increase of the anoxic/aerobic phase duration ratio to 1.5 did not increase EPSc production (EPSc value of  $9.93 \pm 1.92$   $\text{mg g}^{-1}$  VSS). During anoxic/aerobic phase duration ratio shift, the flux was gradually decreased from  $25.5 \pm 2.71$  to  $12.7 \pm 0.38$   $\text{L m}^{-2} \text{h}^{-1}$  (Fig. 8), indicating a more pronounced effect of SMPc on membrane filterability.

#### 4. Conclusions

The MBR treated effluent was compliant with the Greek discharge limits for unrestricted irrigation. The addition of glycerol resulted in EPSc increase up to  $19.2$   $\text{mg g}^{-1}$

VSS, whereas SMPc concentration was increased up to  $31.1$   $\text{mg g}^{-1}$  VSS. Biosolids addition resulted in higher EPSc and SMPc concentrations, reaching values of  $12.3$  and  $15.2$   $\text{mg g}^{-1}$  VSS, respectively. The  $d\text{TMP}/dt$  was found to be a suitable indicator of critical flux, since a sharp increase in its value was observed at the critical flux point ( $32.6$   $\text{L m}^{-2} \text{h}^{-1}$ ). As the consequence of the increased microbial activity at elevated temperature, the membrane resistance was drastically increased up to  $4.63 \pm 0.09$   $\text{m}^{-1}$  at temperature over  $19^\circ\text{C}$ , thus flux was sharply decreased from  $26.4 \pm 2.27$  to  $15.9 \pm 1.34$   $\text{L m}^{-2} \text{h}^{-1}$ . Implementation of fouling prevention strategies, such as backwash flow rate increase, cross flow aeration increase, in situ mechanical cleaning, anoxic/aerobic phase duration and offline intensive chemical cleaning, resulted in the improvement of membrane permeability. The increase in anoxic/aerobic phase duration ratio increased membrane fouling during MBR operation under intermittent aeration and feeding conditions.



## Acknowledgement

This research is co-financed by Greece and the European Union (European Social Fund-ESF) through the Operational Programme “Human Resources Development, Education and Lifelong Learning” in the context of the project “Strengthening Human Resources Research Potential via Doctorate Research” (MIS-5000432), implemented by the State Scholarships Foundation (IKY). The authors also thank Kouvalakis Kyriakos for his timely help in automation of control system.

## References

- [1] S. Judd, *The MBR Book: Principles and Applications of Membrane Bioreactors for Water and Wastewater Treatment*, 2nd ed., Elsevier, London, 2011, p. 344.
- [2] A. Drews, Membrane fouling in membrane bioreactors—characterisation, contradictions, cause and cures, *J. Membr. Sci.*, 363 (2010) 1–28.
- [3] G. Sabia, M. Ferraris, A. Spagni, Effect of solid retention time on sludge filterability and biomass activity: long-term experiment on a pilot-scale membrane bioreactor treating municipal wastewater, *Chem. Eng. J.*, 221 (2013) 176–184.
- [4] H.J. Lin, M.J. Zhang, F.Y. Wang, F.G. Meng, B.-Q. Liao, H.C. Hong, J.R. Chen, W.J. Gao, A critical review of extracellular polymeric substances (EPSs) in membrane bioreactors: characteristics, roles in membrane fouling and control strategies, *J. Membr. Sci.*, 460 (2014) 110–125.
- [5] F.G. Meng, S.Q. Zhang, Y.T. Oh, Z.B. Zhou, H.-S. Shin, S.-R. Chae, Fouling in membrane bioreactors: an updated review, *Water Res.*, 114 (2017) 151–180.
- [6] P. van den Brink, O.-A. Satpradit, A. van Bentem, A. Zwijnenburg, H. Temmink, M. van Loosdrecht, Effect of temperature shocks on membrane fouling in membrane bioreactors, *Water Res.*, 45 (2011) 4491–4500.
- [7] Z. Ma, X.H. Wen, F. Zhao, Y. Xia, X. Huang, D. Waite, J. Guan, Effect of temperature variation on membrane fouling and microbial community structure in membrane bioreactor, *Bioresour. Technol.*, 133 (2013) 462–468.
- [8] J. Arévalo, L.M. Ruiz, J. Pérez, M.A. Gómez, Effect of temperature on membrane bioreactor performance working with high hydraulic and sludge retention time, *Biochem. Eng. J.*, 88 (2014) 42–49.
- [9] H.-D. Park, I.-S. Chang, K.-J. Lee, *Principles of Membrane Bioreactors for Wastewater Treatment*, 1st ed., CRC Press, Boca Raton, 2015, p. 360.
- [10] Y.H. Shi, J.H. Huang, G.M. Zeng, Y.L. Gu, Y.N. Chen, Y. Hu, B. Tang, J.X. Zhou, Y. Yang, L.X. Shi, Exploiting extracellular polymeric substances (EPS) controlling strategies for performance enhancement of biological wastewater treatments: an overview, *Chemosphere*, 180 (2017) 396–411.
- [11] X.F. Shi, G. Tal, N.P. Hankins, V. Gitis, Fouling and cleaning of ultrafiltration membranes: a review, *J. Water Process Eng.*, 1 (2014) 121–138.
- [12] E. Braak, M. Alliet, S. Schetrite, C. Albasi, Aeration and hydrodynamics in submerged membrane bioreactors, *J. Membr. Sci.*, 379 (2011) 1–18.
- [13] J.L. Wu, P. Le-Clech, R.M. Stuetz, A.G. Fane, V. Chen, Effects of relaxation and backwashing conditions on fouling in membrane bioreactor, *J. Membr. Sci.*, 324 (2008) 26–32.
- [14] K.-J. Hwang, C.-S. Chan, K.-L. Tung, Effect of backwash on the performance of submerged membrane filtration, *J. Membr. Sci.*, 330 (2009) 349–356.
- [15] M. Raffin, E. Germain, S.J. Judd, Influence of backwashing, flux and temperature on microfiltration for wastewater reuse, *Sep. Purif. Technol.*, 96 (2012) 147–153.
- [16] J. Wu, C.D. He, Effect of cyclic aeration on fouling in submerged membrane bioreactor for wastewater treatment, *Water Res.*, 46 (2012) 3507–3515.
- [17] A. Cosenza, G. Di Bella, G. Mannina, M. Torregrossa, The role of EPS in fouling and foaming phenomena for a membrane bioreactor, *Bioresour. Technol.*, 147 (2013) 184–192.
- [18] V.M. Monsalvo, J. Lopez, M.M. Somer, A.F. Mohedano, J.J. Rodriguez, Short-term fouling control by cyclic aeration in membrane bioreactors for cosmetic wastewater treatment, *Desal. Wat. Treat.*, 56 (2015) 3599–3606.
- [19] P. Melidis, S. Ntougias, V. Vasilatou, G. Skouteris, K. Azis, V. Diamantis and A. Alexandridis, Biofouling aspects and critical flux evaluation in an intermittently aerated and fed submerged membrane bioreactor, *Environ. Process.*, 3 (2016) 20–33.
- [20] N.C. Nguyen, S.-S. Chen, H.T. Nguyen, S.S. Ray, H.H. Ngo, W.S. Guo, P.-H. Lin, Innovative sponge-based moving bed–osmotic membrane bioreactor hybrid system using a new class of draw solution for municipal wastewater treatment, *Water Res.*, 91 (2016) 305–313.
- [21] M. Aslam, A. Charfi, G. Lesage, M. Heran, J.W. Kim, Membrane bioreactors for wastewater treatment: a review of mechanical cleaning by scouring agents to control membrane fouling, *Chem. Eng. J.*, 307 (2017) 897–913.
- [22] Z.W. Wang, J.X. Ma, C.Y. Tang, K. Kimura, Q.Y. Wang, X.M. Han, Membrane cleaning in membrane bioreactors: a review, *J. Membr. Sci.*, 468 (2014) 276–307.
- [23] Z.Q. Yin, V.V. Tarabara, I. Xagorarakis, Effect of pressure relaxation and membrane backwash on adenovirus removal in a membrane bioreactor, *Water Res.*, 88 (2016) 750–757.
- [24] M.F. Rabuni, N.M. Nik Sulaiman, M.K. Aroua, C. Yern Chee, N. Awanis Hashim, Impact of in situ physical and chemical cleaning on PVDF membrane properties and performances, *Chem. Eng. Sci.*, 122 (2015) 426–435.
- [25] P. van der Marel, A. Zwijnenburg, A. Kemperman, M. Wessling, H. Temmink, W. van der Meer, An improved flux-step method to determine the critical flux and the critical flux for irreversibility in a membrane bioreactor, *J. Membr. Sci.*, 332 (2009) 24–29.
- [26] P. Le Clech, B. Jefferson, I.S. Chang, S.J. Judd, Critical flux determination by the flux-step method in a submerged membrane bioreactor, *J. Membr. Sci.*, 227 (2003) 81–93.
- [27] S.P. Beier, G. Jonsson, Critical flux determination by flux-stepping, *AIChE J.*, 56 (2010) 1739–1747.
- [28] M. Tiranuntakul, P.A. Schneider, V. Jegatheesan, Assessments of critical flux in a pilot-scale membrane bioreactor, *Bioresour. Technol.*, 102 (2011) 5370–5374.
- [29] K. Azis, C. Vardalachakis, S. Ntougias, P. Melidis, Microbiological and physicochemical evaluation of the effluent quality in a membrane bioreactor system to meet the legislative limits for wastewater reuse, *Water Sci. Technol. Water Supply*, 76 (2017) 1796–1804.
- [30] O.T. Iorhemen, R.A. Hamza, J.H. Tay, Membrane bioreactor (MBR) technology for wastewater treatment and reclamation: membrane fouling, *Membranes*, 6 (2016) 33.
- [31] A.D. Eaton, L.S. Clesceri, M.A.H. Franson, E.W. Rice, A.E. Greenberg, *Standard Methods for the Examination of Water and Wastewater*, 21st ed., American Public Health Association, Washington, D.C., 2005.
- [32] O.H. Lowry, N.J. Rosebrough, A.L. Farr, R.J. Randall, Protein measurement with the Folin phenol reagent, *J. Biol. Chem.*, 193 (1951) 265–275.
- [33] M. Dubois, K.A. Gilles, J.K. Hamilton, P.A. Rebers, F. Smith, Colorimetric method for determination of sugars and related substances, *Anal. Chem.*, 28 (1956) 350–356.
- [34] B. Frølund, T. Griebe, P.H. Nielsen, Enzymatic activity in the activated-sludge floc matrix, *Appl. Microbiol. Biotechnol.*, 43 (1995) 755–761.
- [35] R.S. Trussell, R.P. Merlo, S.W. Hermanowicz, D. Jenkins, The effect of organic loading on process performance and membrane fouling in a submerged membrane bioreactor treating municipal wastewater, *Water Res.*, 40 (2006) 2675–2683.
- [36] L. Hao, S.N. Liss, B.Q. Liao, Influence of COD:N ratio on sludge properties and their role in membrane fouling of a submerged membrane bioreactor, *Water Res.*, 89 (2016) 132–141.
- [37] A.G. Geyik, F. Çeçen, Production of protein- and carbohydrate-EPS in activated sludge reactors operated at different carbon to nitrogen ratios, *J. Chem. Technol. Biotechnol.*, 91 (2016) 522–531.

- [38] J.R.S. Pan, Y.C. Su, C.P. Huang, Characteristics of soluble microbial products in membrane bioreactor and its effect on membrane fouling, *Desalination*, 250 (2010) 778–780.
- [39] G.-P. Sheng, H.-Q. Yu, X.-Y. Li, Extracellular polymeric substances (EPS) of microbial aggregates in biological wastewater treatment systems: a review, *Biotechnol. Adv.*, 28 (2010) 882–894.
- [40] C. Psoch, S. Schiewer, Critical flux aspect of air sparging and backflushing on membrane bioreactors, *Desalination*, 175 (2005) 61–71.
- [41] T.-N.-P. Nguyen, Y.-C. Su, J.R. Pan, C. Huang, Comparison of membrane foulants occurred under different sub-critical flux conditions in a membrane bioreactor (MBR), *Bioresour. Technol.*, 166 (2014) 389–394.
- [42] P. Krzeminski, A. Iglesias-Obelleiro, G. Madebo, J.M. Garrido, H.J.M. van der Graaf and J.B. Jvan Lier, Impact of temperature on raw wastewater composition and activated sludge filterability in full-scale MBR systems for municipal sewage treatment, *J. Membr. Sci.*, 423–424 (2012) 348–361.
- [43] T.M. Qaisrani, W.M. Samhaber, Impact of gas bubbling and backflushing on fouling control and membrane cleaning, *Desalination*, 266 (2011) 154–161.
- [44] R. Campo, M. Capodici, G. Di Bella, M. Torregrossa, The role of EPS in the foaming and fouling for a MBR operated in intermittent aeration conditions, *Biochem. Eng. J.*, 118 (2017) 41–52.
- [45] M. Capodici, G. Di Bella, D. Di Trapani, M. Torregrossa, Pilot scale experiment with MBR operated in intermittent aeration condition: analysis of biological performance, *Bioresour. Technol.*, 177 (2015) 398–405.
- [46] C.S. Lapidou, B.E. Rittmann, A unified theory for extracellular polymeric substances, soluble microbial products, and active and inert biomass, *Water Res.*, 36 (2002) 2711–2720.

### Supplementary information

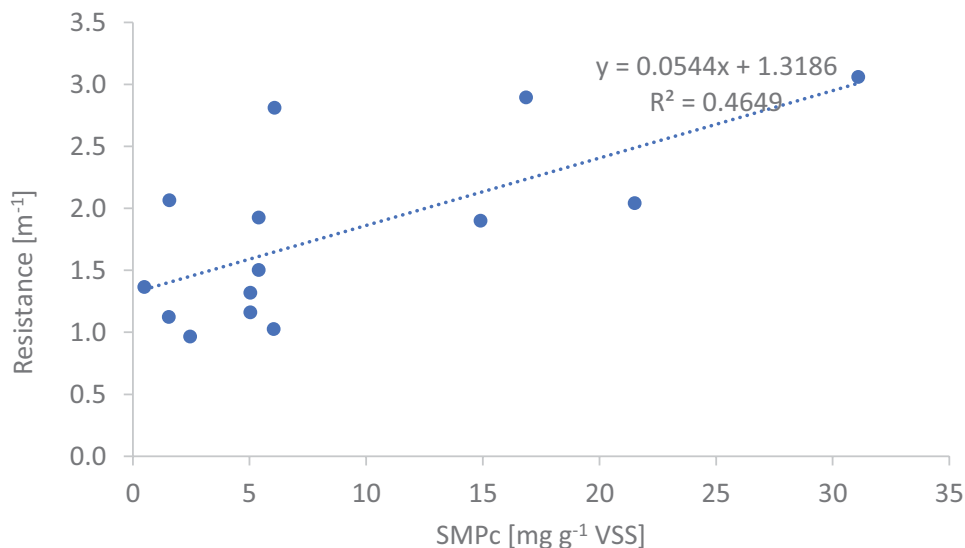


Fig. S1. Relationship between resistance and SMPc. SMPc: carbohydrate SMP.