Impact of SRT on the performance of MBRs for the treatment of high strength landfill leachate

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ABSTRACT

This study examines the performance and fouling potential of flat sheet (FS) and hollow fiber (HF) membrane bioreactors (MBRs) during the treatment of high strength landfill leachate under varying solid retention times (SRT = 5 to 20 days). Mixed-liquor bacterial communities were examined over time using 16S rRNA gene sequence analysis in an attempt to define linkages between the system performance and the microbial community composition. Similarly, biofilm samples were collected at the end of each SRT to characterize the microbial communities that evolved on the surface of the FS and HF membranes. In general, both systems exhibited comparable removal efficiencies that dropped significantly as SRT was decreased down to 5 days. Noticeably, ammonia and nitrite oxidizing bacteria were not detected at the tested SRTs. This suggests that the nitrifiers were not enriched, possibly due to the high organic and ammonium content of the leachate that led to low TN and NH₃ removal efficiency. The steady-state fouling rate of both membranes increased linearly with the decrease in SRT at an estimated factor of 1.1 and 1.2 for the FS- and HF-MBR, respectively, when the SRT was reduced from 15 to 10 days and from 10 to 5 days. Similar dominant genera were detected in both MBRs, including *Pseudomonas*, *Aequorivita*, *Ulvibacter*, *Taibaiella*, and *Thermus*. *Aequorivita*, *Taibaiella*; *Thermus* were the dominant genera in the biofilms. Hierarchical clustering and non-metric multidimensional scaling revealed that while the mixed liquor communities in the FS-MBR and HF-MBRs were dynamic, they clustered separately. Similarly, biofilm communities on the FS and HF membranes differed in the dynamic bacterial community structure, especially for the FS-MBR; however this was less dynamic than the mixed liquor community.

KEYWORDS

Flat Sheet; Hollow Fiber; Landfill leachate; Membrane bioreactor; 16S rRNA gene sequencing

1. INTRODUCTION

Landfilling remains a common element of a waste management plan and at times the only element due to its relative simplicity and low cost when compared to other waste processing/disposal schemes. However, landfilling is associated with the inevitable formation of heavily polluted leachate generated as a result of biochemical processes and rainwater percolation within landfills and if not properly treated, leachate can threaten the receiving environment (Renou et al. 2008; Kurniawan et al. 2006; El-Fadel et al. 1997). For this context, various biological and physical/chemical technologies have been developed for its treatment. Biological technologies are often applied to treat the bulk of the biodegradable fraction in the leachate, while physical/chemical methods are usually adopted as pre/post treatment or to remove specific recalcitrant pollutants. The promulgation of stringent discharge standards to protect water resources has led to the development of combined biological and physical/chemical separation processes, with the membrane bioreactor (MBR) system increasingly being recognized as the process of choice for the treatment of high-strength wastewater characterized by high content of complex and recalcitrant compounds (Sutherland 2010; Yang et al. 2006). The main advantages of MBRs include their efficient filtration that holds small particles (size $< 0.1 \mu\text{m}$) (Santos et al. 2011), ability to replace the second stage of conventional wastewater treatment (i.e. gravity settling), production of a better quality effluent, and reduction of reactor volume and footprint (Hashisho et al. 2016).

While the literature on the use of various membrane types (e.g. hollow fiber and flat sheet) in MBRs for the treatment of high strength industrial wastewater is relatively rich (Gao et al. 2013; Hai et al. 2005), studies examining the efficacy of various membrane types under varying solid retention time (SRT) for the treatment of high strength stabilized landfill leachate are limited (Hashisho et al. 2016; Remmas et al. 2016; Remmas et al. 2017a, b). In this context, the optimal operational SRT for MBRs remains debatable. Long SRTs are desirable for higher biomass retention, which lead to improved treatment efficiency and dominance of slow growing microorganisms that are able to consume macro-molecules such as polysaccharides, carbohydrates, and proteins (Ahmed et al. 2007; Masse et al. 2006). However, excessively long SRTs can lead to the accumulation of dead and inactive microorganisms, resulting in reduced sludge activity (Han et al. 2005; Huang et al. 2001) and low wasting frequency that can hinder phosphorus removal. In addition, while high SRTs can reduce extracellular polymeric substances (EPSs) and soluble microbial products (SMPs) concentrations in the mixed liquor by promoting starvation conditions in the MBR creating an enabling environment for reduced formation of EPS, excessively high SRTs (50, 70 and 100 d) have been reported to promote microbial lysis that generates EPSs or SMPs that can contribute to fouling (Han et al. 2005; Iorhemen et al. 2016). EPSs are known to interfere with the permeate flow across the membrane through the formation of aggregates of bacteria in biofilms and flocs, the

development of barriers around bacteria, the retention of water, and embedment of bacteria in hydrated gel matrix (Le-Clech et al. 2006). SMPs, on the other hand, can block pores and form a gel structure on the membrane surface that can hinder the filtration process (Rosenberger et al. 2006). Accordingly, whilst high levels of mixed liquor suspended solids (MLSS) and SRTs were adopted in old MBR systems (e.g. up to 30 g/L, 100 days), lower and more controllable levels are being chosen for the newer systems (e.g. up to 16 g/L, 14 days (Jia et al. 2009) in order to decrease fouling propensity and cleaning frequency (Le-Clech et al. 2006).

In this study, common membrane modules, hollow fiber (HF), and flat sheet (FS)¹, were tested in a pre-denitrification MBR system under varying SRTs to assess their effectiveness in treating stabilized high strength landfill leachate and to define potential fouling rates under steady state conditions. During the process, several indicators² were monitored at various compartments (i.e. anoxic and oxic tanks) of the MBR systems. Concurrently, bacterial communities in the mixed liquor and on the membrane surfaces (biofilms) were characterized using 16S rRNA gene sequencing to identify linkages between systems' performance and microbial community composition.

2. MATERIALS AND METHODS

2.1. MBR construction and operation

The experimental setup (Fig. 1) consisted of two 10 L-Plexiglas anoxic tanks (D), stirrer mixers (C) to prevent settlement of solids, and two 50 L-Plexiglas oxic tanks (E, L), with one equipped with FS module (L) and the other with HF module (E). The FS Kubota 203 module consisted of a Chlorinated Polyethylene microfiltration membrane of 0.2 μm nominal pore size and a surface area of 0.1 m^2 , while the HF ZW 10 module consisted of neutral hydrophilic ultrafiltration membrane with 0.03 μm nominal pore size and a surface area of 0.93 m^2 (Table 1). A blower (M), with a rotameter (Omega-FL-3663C) to regulate the airflow from a central air compressor, was attached to each membrane to provide aeration and to help in scrubbing the membrane to minimize potential fouling. Peristaltic pumps (Master Flex 07528-10 and 7550-22) (I, K), with variable speed and reverse operation modes, were used for the permeate suction and recirculation. Both systems were fed with landfill leachate from a single storage tank connected to the denitrification tanks by means of multi-channel peristaltic pumps (A, B). The systems were connected to the drain (H) to allow sludge wastage and hence control the SRT. Two pressure sensors (F) (Omega DPG 1000ADA or

¹ While HF offers the advantage of a high surface-area-to-volume ratio, due to its high packing density; combining compactness, simple and modular reactor design, and leachate treatment by MBRs have generally employed HF membrane modules with a fewer number adopting the FS membrane modules Cui *et al.* (2003); Le-Clech *et al.* (2006).

² Note that data on phosphorus removal achieved by MBR treating stabilized leachate is scarce, with limited to no data on removal achieved by FS MBRs Hashisho *et al.* (2016).

DAR), connected to a digital display, were used to trace variations in the membrane pressure. The latter was displayed on the screen of the pressure sensor and the effluent flux was calculated as the flow per unit area. The permeate flow rate was kept at 9 mL/min, the hydraulic retention time at 100 hours for both membranes and the effluent flux at 5.4 liter/m²/hour (LMH) for the FS and 0.58 (LMH) for the HF modules. Both systems operated simultaneously, while testing different SRT levels at 20, 15, 10, and 5 days (Table 2). The length of operation was selected mainly as a function of the SRT itself, normally 3 SRTs to reach steady state (Brown et al. 2013) – with the exception of SRT = 15 days which appeared to reach steady state earlier (at ~2 SRTs).

Fig. 1 Experimental setup

Table 1 Properties of membrane modules

<i>Indicator</i>	<i>Flat Sheet (ACWA)</i>	<i>Hollow Fiber (Zenon)</i>
Model	Kubota 203	ZW 10
Membrane type	Microfiltration	Ultrafiltration
Materials	Chlorinated Polyethylene	Neutral hydrophilic
Manufacturer	Kubota	GE
Nominal Pore size, μm	0.2	0.03

Table 2 Operating conditions of experimental program

<i>Membrane Type</i>	<i>Surface Area (m²)</i>	<i>Permeate Flow (mL/min)</i>	<i>Effluent Flux (liter/m²/hour, LMH)</i>	<i>Hydraulic Retention Time (hours)</i>	<i>Solid Retention Time (days)¹</i>	<i>Operating Time (days)</i>
Flat Sheet	0.1	9	5.4	100	20	59
					15	26
					10	29
					5	15
Hollow Fiber	0.93	9	0.58	100	20	59
					15	26
					10	29
					5	15

Previous testing was conducted at a constant SRT = 30 days with no fouling or microbial analysis (Hashisho et al. 2016)

Leachate was collected weekly from a locally operated sanitary landfill and transported to the Environmental Engineering Research Center at the American University of Beirut to feed the system. The landfill was established in 1997 as part of an integrated regional solid waste management system and receives over 2000 tons per day of municipal waste composed of a large fraction of organic food waste with high moisture content. The experiment was initiated by filling the reactors with leachate, opening the aeration valves in the oxic tanks, and turning on the mixers in the anoxic tanks at low speed (~150 rpm). The flow rate was increased gradually until reaching an HRT of 100 hours (Alvarez-Vazquez et al. 2004). An anti-foaming agent (Sigma Aldrich) was added (a few drops twice per week during the first month, and once every two weeks afterwards) to control foaming in the membrane tanks. As suggested by the membrane manufacturers, the HF membrane was cleaned using 200 ppm Sodium Hypochlorite (NaOCl) solution as a series of repeated backpulses at room temperature, while the FS membrane was cleaned by gently scraping the solids using a soft cloth also at room temperature.

2.2. Sampling and biochemical analysis

Samples were collected twice a week from the feed and permeate tanks and once a week from all other tanks. They were analyzed according to standard methods (SM) for the examination of water and wastewater (APHA, AWWA, WEF. 2012). The analysis was done for several indicator parameters that included pH (SM 4500-H+ B), BOD₅ (SM 5210 B), total nitrogen (TN, Hach 10072), ammonia (NH₃, Hach 8155), Chemical Oxygen Demand (COD SM 5220 D), total phosphorous (TP, SM 4500 P B (5), E), phosphate (PO₄³⁻, SM 4500-P-E), total suspended solids (TSS, SM 2540 D), and volatile suspended solids (VSS, SM 2540D-E). Pollutant removal data were determined and compared with reported values in the literature. Additionally, the impact of the adopted SRTs on the removal efficiency of the two membranes was assessed using the non-parametric Kruskal-Wallis (KW) statistical test. The Mann-Whitney (MW) non-parametric U test was used to conduct a post hoc multiple comparisons between the different SRTs with the

application of Holm's adjustment. The confidence level for all statistical tests was set to 95% at a significance level of 0.05. Statistical analyses were conducted using the R 3.0 statistical software (Team, R. C. 2013).

2.3. Fouling analysis

Fouling behavior was assessed in terms of the steady-state fouling rate FR_{ss} ($LMH.bar^{-1}.d^{-1}$)³, which is defined as the ratio of effluent flux (F_E ; LMH) to trans-membrane pressure (TMP; bar) per unit time (T; days) (Equation 1). F_E was maintained constant at 5.4 and 0.58 LMH for the FS and HF membranes, respectively. The specific flux (F_S ; $LMH.bar^{-1}$) or permeability was plotted against the days of operation and the corresponding FR_{ss} was determined as the slope of the best-fit linear regression line. The effect of SRT on the fouling performance was assessed by examining FR_{ss} as a function of SRT.

$$FR_{ss} = F_S/T = F_E/(TMP \times T) \quad (1)$$

2.4. DNA extraction, PCR, 16S rRNA gene pyrosequencing and microbial analysis

Bacterial 16S rRNA gene sequencing was conducted to characterize the communities in the FS-MBR and HF-MBR operated at an SRT of 20, 15, 10, and 5 days. For each SRT tested, 1.5 mL of mixed liquor was collected each week from each oxic and anoxic tank. The mixed liquor samples were centrifuged at 10,000 rpm for 10 min. to concentrate the cells, the supernatant was discarded and the pellets were stored at -80 °C for subsequent molecular analysis. Biofilm samples were collected and analyzed at the end of each SRT. The biofilm samples were collected by scrubbing the biofilm attached to the surface of the FS membrane and from one fiber of the HF membrane. The FS membrane was rinsed with a phosphate-buffer saline (PBS) solution prior to scrubbing and the HF was submerged into the PBS solution. The biofilm samples were collected in 1.5 mL sterile tubes and were stored at -80 °C for subsequent molecular analysis. Genomic DNA was extracted using the PowerSoil DNA extraction kit (MO BIO Laboratories, Inc., Carlsbad, CA), according to the manufacturer's instructions. PCR reactions (triplicate) were performed for each extracted DNA sample in a 25 μ L reaction volume using the HotStarTaq Plus Master Mix (QIAGEN, Valencia, CA), 0.25 μ M of each primer, and 20 ng of template DNA. Bacterial 16S rRNA genes were amplified using the bacteria-specific forward primer 341F (5'-Adaptor A-Barcode-CA Linker- CCTACGGGNGGCWGCAG-3') and reverse primer 805R (5'-Adaptor B-TC Linker- GACTACHVGGGTATCTAATCC-3') (Klindworth et al. 2012). PCR was performed

³ Liter/m²/h.bar⁻¹.d⁻¹

using life technologies veritus thermocycler under the following conditions: 94°C for 3 minutes, 28 cycles of 94°C for 30 seconds, 53°C for 40 seconds, and 72°C for 1 minute, and a final elongation step at 72°C for 5 minutes. Following PCR, all amplicon products from different samples were mixed in equal concentrations, purified using Agencourt Ampure beads (Agencourt Bioscience Corporation, MA), and pyrosequenced on the Roche 454 FLX Titanium genome sequencer (Roche, Indianapolis, IN) according to the manufacturer's instructions. The 16S rRNA gene sequences were processed using a proprietary analysis pipeline (MR DNA, Shallowater, TX, www.mrdnalab.com). Raw reads were first demultiplexed to trim the barcodes and primers and then low quality sequence reads defined as outside the bounds of 200 and 1000 bp, sequences containing more than 6 ambiguous base or 6 homopolymers, and sequences with a quality score less than 25 were removed. Sequences were then denoised and chimeras were removed. Sequences were clustered into operational taxonomic units (OTUs) after removal of singletons with a 97% sequence identity threshold. A representative sequence from each OTU was phylogenetically assigned to a taxonomic identity using BLASTn against a curated Green Genes database (DeSantis et al. 2006).

For beta diversity measures, the Bray–Curtis dissimilarity index (Bray and Curtis 1957) was used to determine the percent of dissimilarity in microbial community structure between the various samples at the OTU level. The agglomerative hierarchical clustering (Ward Jr 1963) was used to generate a dendrogram from the Bray–Curtis dissimilarity matrix using the Community Analysis Package 4.0 (PISCES Conservation Ltd, UK). The samples were also compared by Nonmetric Multidimensional Scaling (NMDS) using the statistical software PRIMER 6 (version 6.1.13) and the PERMANOVA+ add on (version 1.0.3). NMDS ordination was generated based on Bray–Curtis dissimilarity index.

3. RESULTS AND DISCUSSION

3.1. Leachate Characterization

The BOD₅/COD ratio of the feed was relatively low (0.08-0.22, Tables 3 and 4), indicating the presence of a considerable portion of slow or non-biodegradable COD, such as humic and fulvic acids (Harmsen 1983; Miller and Clesceri 2003). This is typical of old landfills, where the leachate has attained a certain degree of maturity (Kewu and Wenqi 2008). While the COD, BOD₅, and pH of the leachate fell within reported ranges for old leachate (Table 4), the NH₃ levels of 2200-3200 mg/L were higher than the reported range of 360-2150 mg/L for old leachate (Aziz et al. 2015; Xie et al. 2010). The high N and P content could be attributed to the uncontrolled discharge of high nutrient waste, such as detergents and agricultural waste, typically practiced in developing economies (Chen 1996).

Table 3 Feed BOD₅ and COD under varied SRT

<i>SRT (days)</i>	<i>Operating period (days)</i>	<i>BOD₅ (mg/L)</i>	<i>COD (mg/L)</i>	<i>Feed BOD₅/COD</i>
20	59	724 ±179.83	6088±614.55	0.12±0.02
15	26	750± 202.94	5399± 161.81	0.17± 0.02
10	29	947±78.36	5253±65.19	0.18±0.02
5	15	1003±17.28	5218±62.38	0.19 ±0.00

Table 4 Composition of feed leachate compared with literature reported ranges

<i>Parameter</i>	<i>Unit</i>	<i>Present Study (SRT, days)^a</i>				<i>Leachate</i>	
		<i>20</i>	<i>15</i>	<i>10</i>	<i>5</i>	<i>Old^b</i>	<i>Young^c</i>
pH		8.31-8.49	8.32-8.45	8.2-8.44	8.17-8.25	6.93-9.48	7.26-7.91
BOD ₅	mg/L	406-1020	742-1228	790-1022	988-1024	47-1024	4890-10935
COD	mg/L	5010-6900	5110-5600	5150-5330	5150-5300	290-2860	9520-17760
BOD ₅ /COD		0.08-0.16	0.14-0.22	0.15-0.19	0.19-0.20	0.09-0.4	0.513-0.615
NH ₃	mg N/L	2200-3035	2730-3140	2810-3200	3100-3200	360-2150	1800-4700
TN	mg N/L	2300-3350	2800- 3200	2900-3400	3250-3400	-	2200-5250
TP	mg P/L	14.5-36.5	23-36	19.5-32.5	31.5-34	8-140.8	25-39.5
PO ₄ ³⁻	mg P/L	12-27	20-28	17-28	29-31	-	11-28
TSS	mg/L	510-1390	250-790	470-2830	530-690	156-1374	635-928

^a Sampling and feed characterization: three times / week; ^b Aziz et al. 2015; ^c Rizkallah et al. 2013

3.2. Performance Assessment

Generally, all indicators except for TN exhibited a drop in removal efficiencies as the SRT decreased from 20 to 5 days (Table 5 and Fig. 2), consistent with reported performance degradation under reduced SRT in MBRs treating wastewater (Tan *et al.* 2008). This is normally attributed to the decrease in biomass associated with shorter SRTs. In general, the achieved removal efficiencies were in agreement with reported literature values and at times better, when taking into consideration the strength of the leachate in this study (refer to Table SM-1 in the Supplementary Material for a comparison between the results of this study and those reported in the literature).

Both MBRs achieved comparable COD removal efficiencies across the different SRTs. Efficiencies were found to statistically decrease as a function of SRT (KW chi squared = 27.0 for HF and 29.0 for FS with 3 degrees of freedom; both p-values < 0.05). On average, COD removal decreased from ~50% in FS-MBR and 43% in HF-MBR, at SRT=20 days, to 34% in FS and 19% in HF, at SRT=15 days, and to <20 % in both MBRs at SRT=10 and 5 days (Figure 2a). Across both membranes, the COD removal rates achieved at SRT= 20 days were statistically higher than those observed at the lower SRTs (MW U-test for both MBRs had p-value < 0.05) (Fig. 3-a2 & b2). Literature reported removal efficiencies of COD vary significantly depending on operating conditions and feed concentration. At an influent COD concentration of 2200 mg/L (three folds lower than the current study), Svojitka et al. (2009) reported an equally low COD removal (<30%)

rate at an SRT of 100 days. Yet, higher COD removals from leachate have been reported with combined processes (Alvarez-Vazquez et al. 2004; Jia et al. 2009). Similar to COD removal, BOD₅ removal efficiency tended to statistically increase as a function of SRT for both membranes (KW chi squared =27.4 for HF and 29.8 for FS with 3 degrees of freedom; both with p-values < 0.05) (Fig. 3-a1&b1). Both MBRs achieved their highest BOD₅ removal rates (>90%) at SRT=20 days, which decreased to <65% at SRT=10 and 5 days (Fig. 2b).

The removal of phosphorus compounds was comparable in both MBRs; TP removal efficiencies were found to increase as a function of SRT for both membranes (KW chi squared =12.0 for HF and 21.4 for FS with 3 degrees of freedom; both p-values < 0.05). For the HF-MBR, the Mann-Whitney post-hoc U-test with Holm's correction showed that removal rates achieved at SRT of 20 days were statistically similar to the rates achieved at SRT of 15 days (MW U-test p-value = 0.536). Removal rates at SRT 20 and 15 days were statistically better than those achieved at lower SRTs (MW U-test p-value = 0.036 with SRT 10; and MW U-test p-value = 0.05 with SRT 5) (Fig. 3-a6). As for the FS-MBR, the post-hoc statistical tests showed statistically significant differences in the removal efficiencies at all SRTs, except those at SRT 10 and 15 days (MW U-test p-value p-value 0.31) (Fig. 3-b6). Similarly, the removal efficiency of PO₄³⁻ was found to vary significantly with SRT for both membranes (KW chi squared =14.8 for HF and 16.7 for FS with 3 degrees of freedom; p-values < 0.05), with higher SRTs associated with higher removals (Fig. 3-b5). The observed increase in the removal efficiencies with higher SRT is consistent with reported performance and is commonly attributed to higher biomass concentration. Seo et al. (2000) reported 66% TP removal using a submerged MBR for the treatment of domestic wastewater (COD=216-327 mg/L, TP=3.8-8.1 mg/L or PO₄³⁻=2.4-3.5 mg/L) at SRT of 25 days. Likewise, Ujang et al. (2002) reported 71% PO₄³⁻ removal efficiency at SRT=25 days with a lab-scale MBR fed with synthetic wastewater (COD=625-678 mg/L, PO₄³⁻=10.9-12 mg/L). Lower PO₄³⁻ removal efficiencies (40-50%) were reported at SRT 4 days for the treatment of synthetic wastewater with an influent COD=1496 mg/L and PO₄³⁻=9 mg/L (Isma et al. 2014).

As for TN, both membranes achieved low removal rates ranging between 30 and 35% for the FS-MBR and between 15 and 30% for the HF-MBR across the tested SRTs (Fig. 2c). No statistically significant difference in removal efficiency was found between the different SRTs for the FS-MBR (KW non-parametric test p-value > 0.05) (Fig. 3-b4). Regarding the removal of NH₃, both membranes achieved comparable low removal efficiencies that ranged between 27 and 38%, with no statistical difference observed in the removal rates across the SRTs (KW non-parametric test p-values of 0.5 and 0.3 for the HF and FS, respectively) (Fig. 3-a3 & b3). The low removal rates may be attributed to the inhibiting effect that the high NH₄⁺ concentrations in the feed (>1000 mg/L)

may have had on nitrifiers (Ahmed and Lan 2012; Ince et al. 2013). Microbial data showed the absence of nitrifiers as outlined below. High levels of NH_4^+ in leachate are a result of the lack of a mechanism capable of removing it under the landfills' methanogenic conditions, leading to its accumulation (Kaczorek and Ledakowicz 2006; Kurniawan et al. 2010). Pre-treatment through ammonia-stripping (Hasar et al. 2009), biological fluidized bed with granular activated carbon (Horan et al. 1997), fluidized beds (Imai et al. 1993), or a combination of different technologies such as biological treatment, electro-beam radiation and chemical oxidation (Lin and Chang 2000) have been adopted to achieve removal rates in excess of 80%. As such, establishing a pre-treatment step prior to the MBR seems imperative (Berge et al. 2006) to limit the discharge of high nitrogenous compounds into surface water bodies which releases nitrous oxide into the atmosphere and promotes eutrophication and aquatic toxicity (Philips et al. 2002).

Table 5 Removal efficiencies (%) of various indicators at different SRTs (days)*

SRT	% Removal											
	BOD ₅		COD		NH ₃		TN		TP		PO ₄ ³⁻	
	FS	HF	FS	HF	FS	HF	FS	HF	FS	HF	FS	HF
20	92.4±4.1	90.0±5.5	51.0±10.6	42.9± 8.2	37.7±13.2	36.00±14.13	35.1±13.3	14.6± 6.3	71.8 ± 7.8	65.8±12.4	74.3±12.9	74.3±12.9
15	75.4±9.8	72.3±11.9	33.8± 4.4	18.6± 2.3	37.3±3.7	29.7±7.2	34.7±3.1	30.1± 7.4	59.8±6.9	61.5±13.3	60.7±20.8	68.0±15.7
10	65.0±2.8	61.8±4.1	17.8± 3.6	17.8±2.5	33.6± 3.4	28.6±3.4	30.8±2.2	28.5±1.9	56.9±7.2	53.1±7.8	56.7±5.2	62.7±7.6
5	55.1±3.2	51.2±3.2	14.2± 3.5	15.3±2.1	31.6±2.5	27.3±5.0	29.7± 3.5	26.7±3.7	43.5±4.5	47.0±4.1	46.9±3.3	54.3±4.3
KW test p-value	<0.05	<0.05	<0.05	<0.05	0.3	0.5	0.3	<0.05	<0.05	<0.05	<0.05	<0.05

* Values reported are the mean removal efficiency ± 1 standard deviation, p-values are reported for the non-parametric Kruskal Wallis tests

(e)

(f)

- Feed Concentration (mg/L)
- ▲ HF Removal Efficiency (%)
- △ FS Removal Efficiency (%)
- SRT Solid Retention Time
- COD Chemical Oxygen Demand
- BOD Biochemical Oxygen Demand
- TN Total Nitrogen
- NH_3 Ammonia
- TP Total Phosphorus
- PO_4^{3-} Phosphates

Fig. 2 Temporal variation of feed concentrations and removal efficiencies of various indicators:
(a) COD, (b) BOD, (c) TN, (d) NH_3 , (e) TP, (f) PO_4^{3-} , (g) pH

Fig. 3 Removal efficiencies as a function of SRT and MBR type

3.3. Fouling Assessment

The fouling rates were determined at SRT 15, 10, and 5 days upon reaching steady-state conditions (i.e. over the last SRT period of each run). Fouling rates for SRT 20 were not reported due to faulty readings by the pressure sensor that was noticed towards the end of the experiment. The steady-state membrane fouling rate, defined as the decline in specific flux (permeability) (or slope of the specific flux (permeability) line), was determined under steady-state conditions at 20°C. At SRT=15 days, the steady-state fouling rate of the FS membrane was 1.76 LMH.bar⁻¹.d⁻¹ as compared to 0.18 LMH.bar⁻¹.d⁻¹ for the HF membrane. As the SRT decreased to 10 and 5 days; the steady-state fouling rate increased to 1.99 LMH.bar⁻¹.d⁻¹ and 2.38 LMH.bar⁻¹.d⁻¹ respectively in the FS membrane (Fig. 4) and to 0.22 and 0.27 LMH.bar⁻¹.d⁻¹ respectively in the HF membrane (Fig. 5).

Fig. 4 Flat Sheet Membrane fouling at SRT=15, 10, and 5 days

Fig. 5 Hollow Fiber membrane fouling at SRT=15, 10, and 5 days

The steady-state fouling rate of both membranes increased almost linearly with decreasing SRTs. A similar pattern was observed for both the FS and HF membranes with an increase in the steady-state fouling rate by a factor of 1.1 and 1.2 respectively as the SRT decreased from 15 to 10 days and from 10 to 5 days. Van den Broeck et al. (2012) also reported that MBR fouling varied as a function of SRT, with

higher FS membrane fouling rates (10.8 mbar.d^{-1}) at SRT 10 days as compared to SRT 30 days ($0.311 \text{ mbar.d}^{-1}$) and SRT 50 day ($0.187 \text{ mbar.d}^{-1}$) when the MBR

The steady-state fouling rate of the FS membrane was found to be nearly 10 folds higher than that of the HF membrane across all tested SRTs. This can be attributed mainly to the higher flux in FS-MBR (5.4 LMH) as compared to the HF-MBR (0.58 LMH). This is consistent with previously reported results for the treatment of high strength leachate ($\text{COD} = 4778 \text{ mg/L}$), whereby higher polysaccharide adsorption and soluble EPS retention in membrane pores were reported at higher flux values, thus leading to higher fouling rates (Thanh et al. 2017). Another reason for the difference in fouling rate could be attributed to the pore size whereby pore clogging by large particles normally occurs in large pore size membranes. Accordingly, microfiltration modules (i.e. FS-MBR) are expected to be more prone to fouling than ultrafiltration modules (i.e. HF-MBR) (Choi et al. 2006). In addition, the hydrophobicity of the FS material, compared to the hydrophilic nature of the HF surface, can aggravate fouling (Maximous et al. 2009; Sun et al. 2006; Yu et al. 2005) and results in a faster decline in the filtration rate (Hilal et al. 2005). However, membrane hydrophobicity should be assessed concurrently with other membrane properties such as pore size and morphology. Accordingly, more efforts targeting the direct interaction of the membrane material and foulants are needed (Maximous et al. 2009).

Consistent with reported studies (Trussel et al. 2006; Van den Broeck et al. 2012), the SRT has a direct impact on the membrane fouling rate. Van den Broeck et al. (2012) reported higher FS membrane fouling rates of 10.8 mbar.d^{-1} at SRT 10 days as compared to $0.311 \text{ mbar.d}^{-1}$ at SRT 30 days and $0.187 \text{ mbar.d}^{-1}$ at SRT 50 days at $\sim 16 \text{ LMH}$ membrane flux (Table 6). In addition, higher fouling rates were reported at SRT 30 days in comparison to SRT 50 days, while operating at the same flux (3.37 mbar.d^{-1} vs. $0.644 \text{ mbar.d}^{-1}$ at 22.5 LMH ; $5.242 \text{ mbar.d}^{-1}$ vs. $0.821 \text{ mbar.d}^{-1}$ at 24 LMH ; and 38.7 mbar.d^{-1} vs. $11.15 \text{ mbar.d}^{-1}$ at 27 LMH at SRTs 30 days and 50 days, respectively). Similarly, Trussel et al. (2006) reported that the rate of membrane fouling in HF-MBRs were higher at lower SRTs while operating at a constant flux of 30.6 LMH ($3.65 \text{ LMH.bar}^{-1}.\text{d}^{-1}$ at SRT 2 days, $1.57 \text{ LMH.bar}^{-1}.\text{d}^{-1}$ at SRT 3 days, $0.59 \text{ LMH.bar}^{-1}.\text{d}^{-1}$ at SRT 4 days, $0.39 \text{ LMH.bar}^{-1}.\text{d}^{-1}$ at SRT 5 days, and $0.18 \text{ LMH.bar}^{-1}.\text{d}^{-1}$ at SRT 10 days) (Table 6). Extracellular polymeric substances (EPS) have been recognized as the main fouling components in MBRs and contribute to membrane biofouling (Gao et al. 2013; Kimura et al. 2012). Fouling appears to be more prominent at lower SRTs when the amount of EPS is greater, whereas at high SRTs, the concentration of EPS decreases as the biomass stays longer in the system (Ahmed et al. 2007)).

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Table 6 Fouling rates for the FS- and HF-MBR in comparison with literature reported values for various wastewater streams treated using an MBR system

Wastewater Type	Fouling rates ($m^{-1}d^{-1}$)	Permeate Flux (LMH)	COD influent (mg/L)	HRT (h)	SRT (d)	System	Reference
Municipal WW	1.5×10^{15}	51	-	10	25	MBR	(Van der Marel 2009)
Synthetic WW	$0.5-10.5 \times 10^{12}$	15	700	-	45	HF-MBR	(Bella <i>et al.</i> 2014)
$mbar \cdot d^{-1}$							
Synthetic WW	0.03	0.60-0.75	1000	24	-	HMBR ^a	(Boonyungyuen and Wichitsathian 2014)
	0.23					HF-MBR	
Synthetic WW	9.14	0.60-0.75	1000	24	-	HF-MBR	(Boonyungyuen <i>et al.</i> 2014)
	12.14	1-1.2		12			
	169.92	1.4-1.7		6			
Municipal WW	33.12-472.32	3-22	200	24	30	Tubular MBR	(Brookes <i>et al.</i> 2003)
Raw WW	230.4-3600	52	274.5	4		HF-BF-MBR ^b	(Ivanovic and Leiknes 2008)
Synthetic WW	100.8-316.8	16-121	-	-	-	Tubular-MBR	(Le-Clech <i>et al.</i> 2003)
Municipal WW	0.187	16	457	15-26	50	FS-MBR	(Van den Broeck <i>et al.</i> 2012)
	0.625	20					
	0.644	22.5					
	0.821	24					
	11.15	27					
Municipal WW	0.311	16	457	15-26	30	FS-MBR	(Van den Broeck <i>et al.</i> 2012)
	0.626	20					
	3.37	22.5					
	5.242	24					
	38.7	27					
Municipal WW	10.8	16	457	15-26	10	FS-MBR	(Van den Broeck <i>et al.</i> 2012)
$LMH \cdot bar^{-1} \cdot d^{-1}$							
Municipal WW	0.18	30.6	345	-	10	HF-SMBR ^c	(Trussel <i>et al.</i> 2006)
	0.39				5		
	0.59				4		
	1.57				3		
	3.65				2		
Landfill Leachate	1.76	5.4	5399	100	15	FS-MBR	This study
	1.99		5253		10		
	2.38		5218		5		
Landfill Leachate	0.18	0.58	5399	100	15	HF-MBR	This study
	0.22		5253		10		
	0.27		5218		5		

321 ^aHybrid Membrane Bioreactor; ^b Biofilm Membrane Bioreactor; ^c Submerged Membrane Bioreactor

322 3.4. Microbial Characterization

323 3.4.1. Bacterial community composition in the mixed liquor

324 The results of the 16S rRNA gene sequencing revealed that *Pseudomonas* (7.6%; phylum
325 *Proteobacteria*), *Aequorivita* (7%; phylum *Bacteroidetes*), *Ulvibacter* (3.4%; phylum *Bacteroidetes*),
326 *Taibaiella* (4.5%; phylum *Bacteroidetes*), and *Thermus* (3.7%; phylum *Deinococcus_Thermus*) were the
327 dominant genera detected in the mixed liquor of both MBRs (Fig. 6 and 7). The dominance of
328 *Pseudomonas* has been previously reported in bioreactors and bioelectrochemical reactors treating

leachate (Zhang et al. 2015; Xie et al. 2012). Members of the phylum *Proteobacteria* (42.6%) and *Bacteroidetes* (23.7%) were the dominant phyla in the mixed liquor of both MBRs, with *Gammaproteobacteria* (22.7%) being the dominant proteobacterial class in both the oxic and anoxic tanks (Fig. 6 and 7). The number in parentheses corresponds to the average relative abundance for the whole duration of the experiment and includes both the oxic and anoxic tanks. Bacterial community composition of landfill leachates collected from five landfill sites in China revealed the dominance of *Gammaproteobacteria*, *Bacteroidetes* and *Firmicutes* regardless of the geographic location (Song et al. 2015). The authors also reported the dominance of *Pseudomonas* in most of the samples with relative abundance ranging from 41% to 92%. In a periodically fed MBR treating landfill leachate at different concentrations and operated in cycles of oxic and anoxic phases, the dominant bacterial community detected using Illumina sequencing was candidate phylum Saccharibacteria, representing ~ 50% of the total sequence reads (Remmas et al., 2017a). While very few studies have been reported on the microbial community composition in anoxic-oxic MBRs treating landfill leachate (Remmas et al. 2016; Remmas et al. 2017a, b, d), studies on the treatment of municipal wastewater reported the dominance of *Proteobacteria* and *Bacteroidetes* in aerobic MBR systems operated at different SRTs (Su et al. 2011; Wan et al. 2011). Su et al. (2011) reported the dominance of *Proteobacteria* and *Bacteroidetes* in the suspension of MBR at SRT= 10, and 60 days treating concentrated synthetic municipal wastewater at pH= 7. Likewise, Wan et al. (2011) reported the dominance of *Proteobacteria* and *Bacteroidetes* in the MBR treating municipal wastewater of influent concentrations: COD= 220 mg/L, TN= 26 mg/L, NH₄⁺-N= 19 mg/L, TP=3.9mg/L. The dominance of *Proteobacteria* and *Bacteroidetes* was also reported in a lab-scale bioreactor treating leachate with a COD of 2,200 mg/L and NH₄⁺-N of 1700-2700 mg/L (Xie et al. 2013). It is noteworthy the detection of sequence reads in the current study affiliated to the genus *Halomonas* (0.7%), whose members are known to be halophilic. Recently, bacterial strains phylogenetically affiliated to the genus *Halomonas* were isolated from mature landfill leachate (Remmas et al., 2017b). Also, was interesting the detection of sequences affiliated to the genus *Pusillimonas* (1.2%) in the mixed liquor samples. Members of the genus *Pusillimonas* are reported to be effective degraders and were identified based on Illumina sequences to be one of the major taxa present in aged landfill leachate (Remmas et al., 2017c).

(a) Relative abundance of phylum

(b) Relative abundance of annotated genera

Fig. 6 Relative abundance of bacterial reads retrieved from the nitrifying (oxic) tank of the HF and FS MBR classified at the phylum level (with proteobacterial class). Taxa that represent less than 1% of the total bacterial community composition were classified as “others” (b) Relative abundance of the annotated bacterial genera retrieved from the nitrifying tank. Only bacterial genera with relative abundance >0.2% are presented in this figure.

Fig. 7 Relative abundance of bacterial reads retrieved from denitrifying (anoxic) tank of the HF and FS MBR classified at the phylum level (with proteobacterial class). Taxa that represent less than 1% of the total bacterial community composition were classified as “others” (b) Relative abundance of the annotated bacterial genera retrieved from the denitrifying tank. Only bacterial genera with relative abundance >0.2% are presented in this figure.

The relative abundance of the dominant genera in the mixed liquor, of the anoxic and oxic tanks, varied with respect to time and the type of MBR (i.e. FS vs. HF). In addition, it is noticeable that the change in the evolution of some genera in one MBR was very often accompanied by similar behavior in the other. For instance, in the FS-MBR the proportions of *Pseudomonas* decreased from 7.9 to 2.3% when varying the SRT from 20 to 15 days; similarly, in the HF-MBR the proportions of *Pseudomonas* decreased from 14.4 to 9.9%. Additionally, when the SRT was decreased from 20 to 15 days, the proportions of *Pusillimonas* decreased in the FS-MBR from 5.0 to 0.9% and in the HF-MBR from 1.1 to 0.4%. In contrast, the proportions of *Cellvibrio* increased from 0.3 to 2.5% in the FS-MBR and from 0.3 to 1.4% for the HF-MBR, when the SRT was dropped from 20 to 15 days.

Comparing the proportions of the phylum *Proteobacteria* and *Firmicutes* between the two MBRs, it is apparent that at SRT=15 days *Proteobacteria* dominance (46.3% and 53.2% in the FS-MBR and HF-MBR respectively) was at its highest as compared to all other SRTs. At SRT=10 days, *Firmicutes* dominance was at its highest for both MBRs with 14.2% and 21.3% in the FS-MBR and HF-MBR, respectively. As for *Bacteroidetes*, the proportions of this phyla were at their lowest at SRT=20 days, with 19% and 15.8% in the FS-MBR and HF-MBR, respectively. The absence of a clear trend as a function of SRT indicates that the changing leachate composition is likely to be responsible for the phyla's behavior regardless of SRT. *Alphaproteobacteria* (5-23%), *Betaproteobacteria* (7-21%), *Gammaproteobacteria* (5-38%) were found to be the dominant proteobacterial classes in the oxic tanks of both systems across all tested SRTs (Fig. 6). The anoxic tanks were dominated by *Betaproteobacteria* (3-11%) and *Gammaproteobacteria* (17-39%) (Fig. 7). Note that the change in the evolution of each of these classes in one MBR was often similar to that observed in the other MBR. For instance, in the oxic tanks of the FS-MBR, the proportions of *Gammaproteobacteria* decreased from 13.5% to 10.2% to 8.9% when varying the SRT from SRT=15 days to SRT=10 days to SRT=5 days; similarly, in the HF-MBR compartment, the proportions of *Gammaproteobacteria* decreased from 32.2% to 23.1% to 22.6 at the aforementioned SRTs. The decrease in the *Gammaproteobacteria* proportions was also noticeable in the anoxic tanks with a decrease from 25.2% to 23.5% for the FS-MBR and from 30.4% to 23% in the HF-MBR when SRT was varied from 15 to 10 days. The proportions of *Alphaproteobacteria* in the oxic tank of the FS-MBR increased from 10.3% to 22.7% when the SRT was varied from 20 to 15 days; likewise, the proportions of this class showed an increase from 4.6% to 8.6% at the equivalent SRT. As for *Betaproteobacteria* in the anoxic tanks, the proportion of this class showed a slight increase from 8.5% to 9% in the FS-MBR and from 7.9% to 11.5% in the HF-MBR as SRT was varied from 20 to 15 days. Class *Clostridia* (Phylum *Firmicutes*) was two times more abundant (21%) in the anoxic tank as

compared to the oxic tank (10%). A similar observation was reported in Dao *et al.* (2016), who reported that *Clostridia* dominated in the anaerobic and anoxic ponds treating leachate, with a lower abundance in aerated ponds treating the same influent. Noticeably, ammonia and nitrite oxidizing bacteria were not detected at the tested SRTs suggesting that nitrifiers were not enriched. As discussed previously, this is possibly due to the high organic content of the leachate and the inhibiting effect of high NH_4^+ concentrations in the feed (>1000 mg/L) (Ahmed and Lan 2012; An et al. 2006; Ince *et al.* 2013). Remmas et al. (2016) reported inhibition of ammonia oxidation at high organic loading in an intermittently aerated and fed membrane bioreactor treating landfill leachate. The authors showed that ammonia oxidizing bacteria belonging to the genus *Nitrosomonas* were inhibited, while members belonging to the genus *Nitrosospira* exhibited some tolerance.

3.4.2. Bacterial community composition in the biofilm

Concerning the biofilm samples, the results of the 16S rRNA gene sequencing showed that *Pseudofulvimonas* (3.2%; phylum *Proteobacteria*), *Paracoccus* (1.1%; phylum *Proteobacteria*), *Phyllobacterium* (2.1%; phylum *Proteobacteria*), *Aequorivita* (7.3%; phylum *Bacteroidetes*), *Taibaiella* (7.0%; phylum *Bacteroidetes*), *Thermus* (7.3%; phylum *Deinococcus_Thermus*) and *Candidatus* *Microthrix* (3.0%; phylum *Actinobacteria*) were the dominant genera at the surface of both membranes (Fig. 8b). Yet, the evolution of some of these dominant genera was not accompanied by a similar behavior in the parallel MBR. For instance, the proportions of the genus *Pseudofulvimonas* decreased steadily on the FS membrane with decreasing SRT (from 6.6% at SRT=20 days reaching 3.5% at SRT=5 days), whereas it increased steadily on the HF membrane (from 1.7% at SRT=20 days reaching 4.7% at SRT=5 days). Similarly, the proportions of the genus *Candidatus* *Microthrix* increased steadily on the FS membrane (from 0.4% at SRT=20 days reaching 8.9% at SRT=5 days), whereas it decreased on the HF membrane (from 4.5% at SRT=20 days reaching 0.1% at SRT=5 days).

Proteobacteria and *Bacteroidetes* were the dominant phyla, representing more than 60% of the bacterial community (Fig. 8a). Studies have shown similar dominance of these phyla on the membrane surface of MBRs. Huang et al. (2008) reported the dominance of *Proteobacteria* and *Bacteroidetes* in the biofilm communities of MBR treating municipal wastewater at SRTs of 8 and 30 days. Likewise, Lim et al. (2012) reported the dominance of *Proteobacteria* in the biofilm communities of MBR treating synthetic wastewater with an influent COD of 1200 mg/L at SRT= 30 days. It has been suggested that *Bacteroidetes* are the major contributors to the development of membrane biofilms and thus can have a competitive advantage over other colonizers (Huang et al. 2008). At the class level, *Alphaproteobacteria*

(16 to 25%), *Gammaproteobacteria* (11 to 18%) and *Betaproteobacteria* (2.4 to 12.0%) were among the dominant proteobacterial classes of the biofilm microbial communities of both MBRs at all tested SRTs.

In general, the biofilm communities were more stable to changes in SRT when compared to the mixed liquor communities. Huang et al. (2008) compared the biofouling communities of identical membranes operated under 15 and 30 LMH and at different SRTs (8 and 30 days); they concluded that the imposed membrane flux affected the community structure and composition of biofouling microorganisms. They attributed this difference, between the low and high-flux systems, to the strong convective force that transports bacterial cells towards the membrane surface at higher permeate flux. Their findings are in agreement with our finding that indicates a lack of significant differences in the composition of biofouling communities at different SRTs, given the low fluxes (5.4 LMH for the FS and 0.58 LMH for the HF) used.

3.4.3. *Beta diversity measures*

The bacterial communities were compared using non-phylogenetic measures (Bray-Curtis distance). Beta diversity indicates the partitioning of microbial diversity; therefore, the correlation between the bacterial communities in the mixed liquor and biofilm in the different MBRs (FS vs. HF) were assessed at different SRTs. Despite the fact that both MBRs were operated under the same conditions and fed with the same leachate, hierarchical clustering and NMDS plot revealed that mixed liquor community from the FS-MBR were clustered separately from the mixed liquor community in the HF-MBR (Fig. 9 and 10). Similarly, the biofilm community on the HF membranes was different from the biofilm community on the FS membranes. The bacterial community structure was largely dynamic as can be seen by the NMDS analysis, with a gradual succession away from the initial conditions (i.e. SRT 20) (Fig. 10). This was more pronounced in the HF-MBR when compared to the FS-MBR. Moreover, the mixed liquor community was more dynamic than the biofilm community as can be seen by the wide distribution in the NMDS plot (Fig. 10). Similar to previous MBRs lab-scale studies, the biofilm microbial community was distinct from the mixed liquor community (Lim et al. 2004; Huang et al. 2008; Lim et al. 2012; Zhang et al. 2006) (Fig. 9 and 10).

(a) Relative abundance of phylum

(b) Relative abundance of annotated genera

Fig. 8 Relative abundance of bacterial reads retrieved from the biofouled membranes classified at the phylum (with proteobacterial class). Taxa that represent less than 1% of the total bacterial community composition were classified as “others” (b) Relative abundance of the annotated bacterial genera retrieved from the biofouled membranes. Only bacterial genera with relative abundance >0.2% are presented in this figure. Flat sheet: FS; Hollow fiber: HF.

Fig. 9 A dendrogram generated based on Bray-Curtis distance at 3% cutoff-OTU level.

Fig. 10 Nonmetric multidimensional scaling (NMDS) plot based on Bray-Curtis distance at 3% cutoff-OTU level for samples collected from the nitrifying (Nit) and denitrifying (Den) tanks and membrane surface (Biof) of the HF-MBR and FS-MBR. The numbers on the symbols represent the different SRT values: 20, 15, 10 and 5 days, respectively.

3.5. Economic and Operational implications

The development and adoption of a treatment process is strongly related to its economic viability. As such, MBR systems are less attractive than conventional treatment systems given their high capital and operating costs. However, improvements in membrane production, the development of automated systems (Visvanathan et al. 2000), the decrease in membrane cost (Lin et al. 2012), the implementation of stringent effluent discharge requirements, and the limitation of vacant land in many cities are increasing the economic competitiveness of the MBR technology. This can explain the increase in the number of studies and commercial applications of MBR systems for both industrial and domestic wastewater applications over the past decades (Yang et al. 2006). In addition, the economic viability of the MBR technology has improved, given its reliance on membrane filtration for the separation of treated wastewater. This feature allows it to handle high strength landfill leachate, specifically mature leachate laden with non-biodegradable contaminants.

Several challenges persist when it comes to scaling-up MBR systems, with membrane cleaning, replacement methods, handling of peak flow, and ease of operation being the most serious (Yang et al. 2006). Yet, several success stories have been reported worldwide whereby MBR systems have been operated at a commercial large scale for the treatment of landfill leachate (Cornel & Krause 2006). In this regard, Visvanathan et al. (2000) compared two small-scale wastewater treatment plants adopting conventional activated sludge (CAS) and MBR systems. The latter was reportedly seen more economically competitive in terms of energy consumption, land footprint, and overall sludge generation, and management requirements. Accordingly, quantifying the advantages of the MBR technology improved its economic attractiveness compared to CAS (Visvanathan et al. 2000). In the context of energy savings, anaerobic digesters may have an edge over MBRs in that they do not require aeration. Yet, anaerobic digesters invariably require heating, particularly thermophilic systems, which add to the overall capital and operating costs (Trzcinski & Stuckey 2010). Methane production during the treatment process may offset these costs slightly. Anaerobic MBR systems have indeed been tested for leachate treatment. Although they combine the advantages of both anaerobic and MBR technology, they can be limited by longer startup periods (Kraume & Drews 2010), frequent cleaning, and a higher likelihood of fouling as compared to their aerobic counterparts (Saddoud & Sayadi 2007; Taskan & Hasar 2012). Accordingly aerobic MBR systems have evolved to become a promising technology for the treatment of high-strength wastewater containing complex and recalcitrant compounds (Bilad et al., 2011). However, in the context of nitrogen inhibition, a pre-treatment prior to the MBR is becoming imperative (Berge et al. 2006) to limit the discharge of high nitrogenous compounds into surface water.

4. CONCLUSION

The FS- and HF-MBRs exhibited comparable removal efficiencies for BOD₅ (92% in FS vs. 90% in HF), TP (72% in FS vs. 66% in HF), PO₄³⁻ (74% in FS vs. 75% in HF) and COD (51% in FS vs. 43% in HF) at SRT = 20 days. While better TN removal was achieved with the FS-MBR at SRT= 20 days (35% in FS vs. 15% in HF), ammonia and nitrite oxidizing bacteria were not detected at the tested SRTs, suggesting that nitrifiers were not enriched, possibly due to the high organic and ammonium content of the leachate leading to low TN and NH₃ removal efficiency. The performance of both systems degraded at shorter SRTs. The statistical analysis indicated that the removal efficiencies of TP, PO₄³⁻, COD, and BOD₅ were a function of SRT for both membranes (p-value <0.05); the TN removal also varied as a function of SRT for the HF-MBR (p-value <0.05) but not for the FS-MBR (p-value=0.3).

The fouling assessment showed that the steady-state fouling rate of both membranes increased, almost linearly, with the decrease in SRT. A similar pattern was observed in the FS and HF membranes with an increase in the steady-state fouling rate at a factor of 1.1 and 1.2 when the SRT was reduced from 15 to 10 days and from 10 to 5 days respectively.

The microbial analysis showed that similar dominant phyla were detected in both MBRs with *Proteobacteria*, *Bacteroidetes*, and *Firmicutes* constituting around 80% of the oxic and anoxic mixed liquor community. *Bacteroidetes* and *Proteobacteria* were the dominant groups in the biofilms, forming together more than 60% of the bacterial community at the surface of both membranes. The mixed liquor community in the FS-MBR was clustered separately from the mixed liquor community in the HF-MBR. Similarly, the biofilm community on the HF membranes was different from that on the FS membranes. The bacterial community structure was found to be dynamic, especially for the FS-MBR. Overall, the mixed liquor community appeared to be more dynamic than the biofilm community.

The above findings suggest that membrane bioreactors cannot be conveniently operated at low solid retention times (< 20 days) without drawbacks in terms of removal efficiencies of carbon and nutrients. High nitrogen concentrations in the leachate necessitate pretreatment or post treatment as their removal is challenging with MBRs alone.

ACKNOWLEDGEMENTS

We are indebted to ACWA and GE Power & Water for providing the membranes and for their guidance during the installation of the experimental setup. Special thanks are extended to the Council for Development & Reconstruction for providing access to the Naameh landfill as well as to Lacoco and the landfill management for facilitating and assisting during leachate sample collections.

FUNDING SOURCES

This study was partially funded through the Dar Al-Handasah (Shair & Partners) endowed Chair at the American University of Beirut and the Division of Biological and Environmental Sciences and Engineering at KAUST.

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Supplementary Material

Table SM-1: Performance of MBR in treating landfill leachate

Reference	Location	Scale	Process	Membrane configuration	Influent characteristics			Operational conditions		Removal efficiency		
					COD, mg/L	BOD/COD (age)	NH ₃ , mg/L	HRT days	SRT days	COD %	BOD %	NH ₃ %
This study	Lebanon	Lab	MBR	Sub (FS)	6088	0.12 (O)	2450	4.16	20	51.0	92.4	37.7
				Sub (HF)	6088	0.12 (O)	2450	4.16	20	42.9	90.0	36.00
				Sub (FS)	5399	0.17 (O)	2890	4.16	15	33.8	75.4	37.3
				Sub (HF)	5399	0.17 (O)	2890	4.16	15	18.6	72.3	29.7
				Sub (FS)	5253	0.18 (O)	2994	4.16	10	17.8	65.0	33.6
				Sub (HF)	5253	0.18 (O)	2994	4.16	10	17.8	61.8	28.6
				Sub (FS)	5218	0.19 (O)	3163	4.16	5	14.2	55.1	31.6
				Sub (HF)	5218	0.19 (O)	3163	4.16	5	15.3	51.2	27.3
(Remmas et al. 2017)	Greece	Pilot & Lab	MBR	HF	4190 ± 142	0.17	84.00 ± 3.19	5		45-80		40-50
(Liu et al. 2017)	China	Lab	Two-stage anoxic/oxic	Sub (FS)	4000-20,000		1450-2100	7		80.60		99.04
(Melidis et al. 2016)	Greece	Pilot	Submerged MBR	Sub (HF)	406±78	0.6	58.3±6.2	0.3-1.0	0-100	93	97	91
(Hashisho et al. 2016)	Lebanon	Lab	MBR	Sub (HF)	5978	0.12 (O)	2464	4.16	30	71.4	92.2	47.8
				Sub (FS)	5978	0.12 (O)	2464	4.16	30	68.5	93.2	63.4
(Trzcinski and Stuckey 2016)	United Kingdom		anMBR	Sub				10		90		
Syron et al. 2015	Ireland	Pilot	Membrane Aerated Biofilm Reactor (MABR)	(HF) diluted leachate	1000 to 3000	<0.2	500 to 2500	4.7-7.5		17.5		80-99
(Xue et al. 2015)	China	Lab	MBR	Sub (HF)	783.21	0.32-0.39	41.53			93.61		97.66
(Xue et al. 2015)	China	Lab	MBR	Recirculated (HF)	783.21	0.32-0.39	41.53			93.85		97.86
(Wang et al. 2014)	China	Pilot	Anoxic/Aerobic-MBR	Sub (HF)	3134.88	0.14	434.76			81.5		93.2
(Wang et al. 2014)	China	Pilot	Anoxic/Aerobic-GAC-MBR	Sub (HF)	3134.88	0.14	434.76			89.5		80.5
(Akgul et al. 2013) ^c	Turkey	Lab	UASB+MBR+SHARON+Anammox	(Tub)	28000–37000 1500-2000 ^a	0.7-0.37 (Y), (Y+O)	250-2500		5 d	30-85	75-99	
(Brown et al. 2013)	Canada	Lab	MBR (compost leachate)	Sub (HF)	116000		2720	95	-	99.7		≈100
(Campagna et al. 2013)	Turkey	Full	MBR+NF	Ext (UF)	16360		>400	-	-	84.4		88 ^d
(Ince et al. 2013)	Turkey		Jet loop MBR	Ext (Tub)	13225	0.44 (Y)	2200-2500	1.35 and 2.93	2.16 and 5.33	80-85		3.2-21.3
(Insel et al. 2013) ^c	Turkey	Lab	MBR+ NF+RO	Sub (UF)	18685		1245		30	89		83.5
(Sanguanpak et al. 2013)	Thailand	Pilot	Two-stage MBR (An-Aer)	Sub (HF)	9240	0.629 (Y)	--	1	Inf.	87	99	
(Thanh et al. 2013)	Vietnam	Lab	MBR	Sub	1200–1400 ^a		68 ± 26	3.5-14.6 h	30	Up to 97.5		≤92.0 ± 1.5
(Zhang et al. 2013)	China	Lab	Fenton oxidation+MBR+RO	Sub(HF)	1200-1600 ^a	0.09-0.12	550-725	4	45	83-87.5		72-95
(Boonyaroj et al. 2012a)	Thailand	Pilot	Two-stage MBR (An-Aer)	Sub (HF)	9389	0.746	105-174	1	Inf.	87	97	83-91
(Boonyaroj et al. 2012b)	Thailand	Pilot	Two-stage MBR (An-Aer)	Sub (HF)	9306	0.72 (Y+O)	138	1	Inf.	87	97	90
(Brito et al. 2012)	Brazil	Lab	MBR ^a	Sub(HF)	3942	0.06	1529			74		
(Coban et al. 2012)	Turkey	Full	Ammonia stripping+MBR+NF	Ext (UF)	24000	0.33	2313	-	-	93.75		98
(Hua and Zhang 2012)	China	Full	MBR+NF+RO and MBR+NF+NF	Ext (UF)	30,000	0.5 (Y)	2200 (30000)	-	-	≈97	≈99	98.9-99.6
(Litas et al. 2012)	Greece	Pilot	SMBR (SBR) Mixture of LFL+Synthetic WW 1:1	Sub (FS)	1772	(O)	269	9	-	95		98.2-99.2
(Mahmoudkhani et al. 2012) ^c	Iran	Lab	MBR+RO	Sub (HF)	68250±8000	0.65	1470	15	55	97	99	99.45
(Lv et al. 2012)	China	Pilot	MBR	Sub (HF)	3600-9700	0.31-0.65	200-620 ^a	4		95	100	61.7
(Santinelli et al. 2012)	Italy	Pilot	MBR	Sub (HF)	802		--	-	-			
(Bai et al. 2011)	China	Lab	Anoxic-oxic hybrid MBR Diluted leachate	Ext	500-4500	-	150-1400	-	-	Up to 90		≤60
(Chiemchaisri et al. 2011)	Thailand	Pilot	2-stage MBR (anoxic tank+aerobic MBR)	Sub(HF)	2605–7318	(O+Y) mixed feed	218–1750	0.5 (MBR tank)	-	60-78	99	80-97

Reference	Location	Scale	Process	Membrane configuration	Influent characteristics			Operational conditions		Removal efficiency		
					COD, mg/L	BOD/COD (age)	NH ₃ , mg/L	HRT days	SRT days	COD %	BOD %	NH ₃ %
(Akkaya et al. 2010)	Turkey	Lab	UASB+MBR+MAP	Sub	4250 ^a	(M)	2315.4 ^a	-	-	10-70	-	35
(Trzcinski and Stuckey 2016)	UK	Lab	3-stage (HR-SAMBR-MBR)	Sub (FS)	150-1300	-	-	Variable	300	30-90	-	-
(Li et al. 2010)	China	Pilot	Anaerobic pretreatment/air-lift bioreactor	Ext(UF/Tub)	4670-6700	(Y)	820-960	-	-	87	-	100
(Puszczalo et al. 2010) ^c	Poland	Lab	Mixture of 10% LFL+synthetic WW/SBR	Sub (MF/Cap)	757-1708	0.06(O)	53.5-510	2-3	15	89	>98	>95
(Aloui et al. 2009) ^c	Tunisia	Lab	Stirred tank reactor	Ext (MF/Tub)	7100-8000	0.18(O)	1000-2800	2-3	-	70-77	>90	≈90
(Feki et al. 2009) ^c	Tunisia	Lab	MBR/electrochemical oxidation	Ext(Tub)	6500-8000	0.09 (O)	1500	-	-	61	100	72.8
(Hasar et al. 2009a)	Turkey	Bench	Mixture of LFL+domestic WW	Sub (HF)	8500-14200 +750-2400 ^a	0.4-0.67 (Y)	1100-2150	3.6-6.0 h	5-30	72-99	-	-
(Hasar et al. 2009b) ^c	Turkey	Lab	Ammonia stripping+ coagulation/ flocculation pretreatment + aer/an-MBR+RO	Sub (HF)	8500-19200 ~7300 ^a	0.4-0.7 (Y)	200-1000 ^a	3.6-16.4 h	10-50	60-90	-	87-98
(Jia et al. 2009)	China	Pilot	UASB/MBR	Sub(HF)	40,000-75,000 1440-25600 ^a	0.42-0.52 (Y)	380-1800	0.5	14	98	99	≈100
(Ratanatamskul and Nilthong 2009)	Thailand	Lab	BPAC-MBR	Sub (HF)	5000-6000 1000 ^a	~0.1 (O)	-	1	Inf.	83	-	-
(Svojicka et al. 2009)	Germany	Bench	Compartmentalized activated sludge tank	Ext (UF/Tub)	2200	<0.05	1200	70-170 h	100	≈30	91	90-99
(Jakopović et al. 2008) ^c	Croatia	Pilot	Stirred tank reactor	Sub (HF)	1400-2800	0.46	-	8 h	-	23	-	-
(Sadri et al. 2008)	Canada	Lab	Stirred tank reactor	Sub (HF)	2737-4079	0.11-0.18 (O)	662±176	1-3.5	30, 60	54-78	>97	>99
(Tsilogeorgis et al. 2008)	Greece	Bench	MSBR	Sub (UF/HF)	1391-3977	(O)	200-279	10	infinite	40-60	-	≈100
(Xu et al. 2008)	China	Pilot	Combined anaerobic pre-treatment and MBR	Air-lift Ext (UF/HF)	10,084 9357 ^{a,b}	0.71 (Y)	-	9.5	-	89	>99	-
(Judd et al. 2006)	UK	Lab	MBR	Ext (Tub)	2701	(O)	21.77-588	5	30	-	-	73-99
(Robinson 2007)	UK	Full	3 aerobic biological tanks in series	Ext (UF/Tub)	5000	0.05	2000	-	-	76	>96	≈100
(Sang et al. 2007)	Vietnam	Lab	Stirred tank reactor	Sub (MF)	4000-39,600	>0.68 (Y)	-	50 -infinite	-	84-97	-	-
(Visvanathan et al. 2007)	Thailand	Lab	Thermophilic MBR	Sub	12000±1000	0.39-0.65(M)	1000-1700	1	-	62-79	>97	60-75
(An et al. 2006)	China	Pilot	Anoxic +aerobic zone in tank	Sub (MF)	1500	-	500	8.5	-	75	-	80-99
(Bodzek et al. 2006)	Poland	Lab	Mixture of 10% LFL+synthetic WW	Ext(UF/Tub)	442 ^a	-	390 ^a	-	-	82.4	98.3	62.8
(Canziani et al. 2006)	Italy	Pilot	MBR+MBBR	Sub (Tub)	6,316	0.3 (O)	1000-1500	-	>45	Up to 75	-	≥90
(Chen and Liu 2006)	China	Pilot	Air-lift bioreactor	Air-lift Ext (UF/HF)	4200-15900 ^b	-	-	1.8-12.9	-	70-96	>99	-
(Laitinen et al. 2006)	finnish	Pilot	Dual-tank MBR (SBR +MBR)	Sub (HF)	2200±230	(Y)	210±90	2-5	35-60	>80	>97	>97
(Chaturapruet et al. 2005) ^c	Thailand	Lab	Ammonia stripping pretreatment/stirred tank reactor	Sub (HF)	8000-9000	0.40-0.45(M)	1700-1800	1	-	~70	~ >95	-
(Schwarzenbeck et al. 2004) ^d	Germany	Full	2 reactors in series (denitrification+nitrification)+AC filter	MF	136-1980	~0.2	120	-	-	65	95	97
(Wichitsathian et al. 2004)	Thailand	Lab	Ammonia stripping /stirred tank reactor	Sub (MF/HF)	8000±1000	0.4±0.05 (M)	1700±100	0.66-1	-	60-66 72-76 ^b	94-98	-
(Setiadi and Fairus 2003)	Indonesia	Lab	Stirred tank reactor	Ext(MF/HF)	1800	0.15-0.17	114.8	1	32	31.3	98	66
(Ahn et al. 2002) ^c	S. korea	Full	Aeration basin with anoxic +aerobic parts	Sub(MF/HF)	400-1500	(O)	200-1400	-	-	~38	97	-

Cap: Capillary; Ext: External; FS: Flat Sheet; HF: Hollow Fiber; M: Medium; MF: Microfiltration; O: Old; Sub: Submerged; Tub: Tubular; UF: Ultrafiltration; Y: Young; anMBR: Anaerobic Membrane Bioreactor; NF: Nanofiltration, RO: Reverse Osmosis, UASB: Upflow Anaerobic Sludge Blanket, GAC: Granular activated carbon; MAP: Magnesium Ammonium Phosphate

^a Concentrations after pretreatment or dilution. ^b COD values in terms of the soluble COD. ^c Applied post-treatment to MBR (efficiencies are for MBR only) ^d Combined efficiency for primary clarifier + MBR

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