A low energy gravity-driven membrane bioreactor system for grey water treatment: permeability and removal performance of organics

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Abstract

Synthetic grey water was treated with a low-pressure gravity-driven membrane bioreactor (GDMBR) system. The system was operated without any direct shear at the membrane surface and without any cleaning or flushing. In order to reduce energy consumption, one reactor was operated without aeration and the results were compared with an aerated reactor. Although the dissolved oxygen content was low (0.4-0.6 mg/L) in the non-aerated system, a stable permeability was observed at a level of around 20 L/m²hbar (flux of 1 L/m²h). The fouling resistance was dominated by the bio-fouling layer, which could be removed hydraulically. In comparison to the aerated system, the bio-fouling layer grown

without aeration exhibited a lower biological activity, was thicker and had a lower surface roughness. Furthermore, the stable permeability of the aerated system was higher (40 L/m²hbar), which could be explained by a lower content and load of extracellular polymeric substances (including extracellular polysaccharides and proteins). In comparison to filtration through a non-fouled membrane, the rejection of biopolymers and humic substances was strongly enhanced, which shows that the bio-fouling layer acts as a secondary membrane. The energy consumption was 0.02-0.04 kWh/m³, which is substantially less than for normal MBRs, and even less than for the traditional activated sludge process. These results show that grey water can be treated in a membrane reactor without any cleaning and flushing and even without any aeration, which makes the process suitable for decentralized wastewater treatment and considerably reduces the energy consumption.

Graphical abstract

Keywords: low-pressure membrane system; grey water treatment; aeration; flux stabilization; bio-fouling layer; energy consumption

1. Introduction

Due to water shortage, the reuse of wastewater gains more and more attention in many parts of the world, including industrial and developing countries. Grey water is defined as the wastewater generated in households, including effluents of showers, baths, wash basins, laundry- and kitchen sinks, but not the toilet wastewater (black water) [1, 2]. It represents the major volume of the domestic wastewater (60-75%) with a relatively low content of nutrients and pathogens [2]. The reclaimed grey water should meet four criteria for reuse: hygienic safety, aesthetics, environmental tolerance and economic feasibility. In order to reach these goals, different approaches have been applied including physical, chemical and biological processes. Among these methods, membrane processes appear to be a very attractive solution due to the high effluent quality [3] and small footprint.

However, membrane processes also have several disadvantages, such as membrane fouling and required cleaning and maintenance during long term operation. Consequently, regular backwashing, flushing and chemical cleaning are applied to recover the required flux. Furthermore, pumps, control processes, pre-treatment and auxiliary equipment are indispensable, which gives rise to management, operation and maintenance and therefore limits the application for decentralized wastewater treatment. To address these issues, a low pressure gravity-driven membrane process (GDM) has been developed recently, which is operated without any cleaning or flushing at pressures in the order of 0.04 bar [4, 5]. This process is suitable for decentralized water treatment with simple operation and maintenance, low energy consumption and little auxiliary equipment. Previous studies on GDM show that the permeate flux stabilizes at 3-15 L/m²h depending on the water quality after several months of operation due to the formation of a highly permeable and heterogeneous bio-fouling layer, in which biological activity by a broad range of different organisms leads to a relatively

permeable fouling layer with stable flux during extended operation times [4, 6, 7]. The GDM technology was also combined with aeration for the treatment of grey water, whereby submerged membranes were applied to provide an activated sludge reactor [8]. Wash water recycling was tested using this technology in an autonomous toilet [9]. Furthermore, the impact of shear stress was investigated with synthetic grey water. Results show that the shear stress changed the properties of the bio-fouling layer, and resulted in a thin but dense fouling layer with high EPS content, and thus the flux was not stable, while flux stabilization did occur in the low-shear system [8]. In addition, the GDM technology was also successfully applied for the pretreatment of seawater before reverse osmosis [10, 11].

As stated above, the fouling layer adhered on the membrane surface plays a very important role in the flux stabilization of the GDM system. The properties of fouling layer do not only determine the flux, but also influence the quality of the permeate. Chomiak et al. [12] found that the “biofilm + membrane” composite retained a larger amount of biodegradable foulants than the membrane alone, due to the activity of the biofilm. Derlon et al. [13] found that a young biofilm on membranes increased the permeate quality due to the degradation of AOC (> 80%). However, the hydrolysis of the organics accumulated on membrane surface increased the AOC content of the permeate in older biofilms. Similar findings were reported in membrane bioreactor systems (MBR). The soluble COD within the MBR can be further reduced by the cake layer deposited on the membrane surfaces during the filtration process, and the rejection efficiency is probably due to the degree of sieving and/or adsorption onto the fouling layer [14-16]. However, not many studies are available on the fate of specific organic compounds or organic fractions in the mixed sludge and in the permeate, on which fractions are retained by the fouling layer or biofilm, and which fractions would pass through. Understanding the fate of the fractions can further guide water reclaim processes, for example in respect of the control of disinfection by-products.

It is known that oxygen is necessary for the metabolism of microorganisms, and that

aeration is responsible for around 70-80% of the energy consumption of wastewater treatment [17]. Therefore, the treatment of grey water without any active aeration would result in a considerable reduction in energy consumption. So far, several studies have focused on the impact of DO concentration on the membrane fouling in the MBR process [18-23]. However, none of these investigations reported how the DO concentration influenced the fate of organics in MBRs. It was shown before that DO determined the structure and properties of bio-fouling layer [18] and also that the properties of sludge were influenced by the DO [24, 25]. It can be expected that the properties of the fouling layer (or biofilm) might influence the removal of organics. However, no studies are known on the treatment of grey water in membrane reactors without any aeration.

The aims of this study were therefore: (1) to evaluate if it is possible to operate a grey-water membrane reactor in a stable manner without any aeration; (2) to understand how the oxygen content influences the properties of bio-fouling layer (thickness, surface roughness, bioactivity and EPS contents and distribution etc.); (3) to evaluate the role of bio-fouling layer on the permeability of low-pressure membrane bioreactor system; (4) to identify the fate of organic carbon fractions in the mixed liquor as well as in the permeate, and further to elucidate how the fouling layer enhances the removal efficiency of organics during long term filtration. To this end, a non-aerated low-pressure gravity-driven membrane bioreactors (GDMBR) was operated with synthetic grey water during several months, and the results were compared with an aerated GDMBR as described before (Ding et al. 2016).

2. Materials and methods

2.1 GDMBR setup

Two gravity-driven membrane bioreactors were operated in parallel for 120 days of operation. The working volume of the reactors was 9 L (Fig. 1). Reactor 1 (GDMBR1) was operated without aeration (low DO), while Reactor 2 (GDMBR2) was operated with aeration

(high DO) using an indirect aeration to avoid aeration shear stress at the membrane surface as described in Ding et al. [8]. The aeration in GDMBR2 was adjusted at a flow rate of 60 L/h, resulting in a DO concentration of 6.0-6.5 mg/L, whereas the DO concentration in GDMBR1 was in the range of 0.4-0.6 mg/L. In both reactors, a magnetic stirrer was used to avoid sludge settling and provide mixing of the reactor contents. The trans-membrane pressure (TMP) was controlled at 50 mbar by keeping the liquid level constant using an overflow tank. The overflow was recycled back to the feed pump in order to avoid loss of suspended sludge (Fig. 1).

Flat sheet membranes were UP150 from Microdyn Nadir (Wiesbaden, Germany), made of polyethersulfone with a nominal cut-off of 150 kDa. Three membrane plates were used in each reactor. The total membrane surface per reactor was 0.06 m². Virgin membranes were cleaned for 24 h in nanopure water to remove chemical reagents. Both systems were located in a room with a constant temperature of 20 °C. There was no sludge discharge during the whole operation period (except for the sludge samples for testing), and therefore sludge retention time was almost infinitely in both systems. The feed water was synthetic grey water, as described in Ding et al. [8], representing mixed shower and kitchen grey water after preliminary settling and screening. The feed solution contained in total COD 300 mg/L, total organic carbon (TOC) 100mg/L, NH₄⁺-N 30 mg/L and TP 5 mg/L.

2.2 Analytical methods

2.2.1 Organic carbons

The TOC and Dissolved Organic Carbon (DOC) concentrations of mixed liquor and permeate were measured by an automatic total organic carbon analyzer (TOC-V, Shimadzu, Japan). The various natural organic matter (NOM) fractions were analyzed by using Liquid Chromatography coupled to an Organic Carbon Detector (LC-OCD) [26, 27]. This method

allows for the quantification of biopolymers (MW >10 kDa), humic substances (MW≈1000-3500 Da), building blocks, low-MW organics and neutrals.

2.2.2 Dissolved oxygen (DO) measurement

A dissolved oxygen meter (Oxi 340i-WTW, Germany) was used to measure the DO concentration near the membrane surface in each reactor. The biofilm adhered on the reactor walls was removed mechanically and resuspended before each measurement.

2.2.3 Biomass quantification and EPS measurement

The bio-fouling layer adhered on the membrane surface was characterized in terms of the mass of volatile and total solids at the end of the experiment. The detail approach can be found in the literature [12]. Extraction of extracellular polymeric substances (EPS) from suspended sludge and the bio-fouling layer was conducted by the heating method as described by Adav and Lee [28]. The concentration of proteins was tested by the bicinchoninic acid (BCA) method according to Smith et al. [29]. The concentration of polysaccharides was measured by the anthrone-sulfuric method [30].

2.2.4 ATP measurement

ATP content was measured in the suspended sludge in the GDMBR and in the bio-fouling layer attached on the membrane surface using the BacTiter-Glo (Promega Corporation) kit and protocol. The steps were based on the literature [31-33]: 100 µL of suspended sludge sample was mixed with 100 µL of reagent. For the analysis of the bio-cake layer, this layer was scraped carefully from the membrane surface by glass stick at the end of the operation time and resuspended with nanopure water, and then homogenized. The reaction was carried out at 38 °C with 10 s incubation time on a vortex mixer. All samples were measured in duplicate. The ATP concentration was normalized by the VSS concentration or total COD of

the sludge that was used for the ATP-assay.

2.4 Morphology of fouling layer

2.4.1 Optical Coherence Tomography (OCT)

Optical Coherence Tomography (OCT) (model 930 nm Spectral Domain, Thorlabs GmbH, Dachau, Germany) was employed to characterize the physical structure of the bio-fouling layer. At the end of the filtration experiment, membrane modules were taken out from the reactor, and carefully placed on the OCT stage for measurement. Matlab® (MathWorks, Natick, US) was used to analyze the images. More details can be found in a former publication [7].

2.4.2 Confocal laser scanning microscopy (CLSM)

Confocal laser scanning microscopy (CLSM, LeicaSP5, Wetzlar, Germany) was used to investigate the development of the bio-fouling layer structure and the EPS distribution. Membrane modules were also taken out for CLSM analyses at the end of GDMBR operation. The fouled membranes were first fixed with 2.5% formaldehyde solution (Sigma Aldrich, P252549) in water during 0.5 hours in the dark, washed twice with nanopure water and cut in several pieces of 1 cm². Then fouling layer samples were stained with different dyes, incubated for half an hour at 20°C in the dark and washed again with pure water. Total DNA content (intracellular and extracellular) of the fouling layer was stained with 2µg/ml 4'-6-diamidino-2-phenylindole (DAPI), representing the total bacterial biomass (Figure S1: blue). Concanavalin A – Alexa488 conjugate (C11252 Thermo Fisher Scientific) was used to stain the α -d-mannose and α -d-glucose groups of biopolymers to represent the EPS (extracellular polysaccharides) (Figure S1.green). Sypro Orange (170-3120, Bio-Rad) was used to target all the proteins. Each piece was imaged on three randomly chosen positions. Z-stacks were rebuilt to 3D images using Imaris (Bitplane, Zürich, Switzerland).

2.5 Filtration resistance distribution

Darcy's law was applied to evaluate the filtration resistance as described previously (Ding et al. 2016). The total resistance (R_t) consists of membrane resistance (R_m), resistance of hydraulically reversible (R_r) and hydraulically irreversible fouling (R_{ir}), and can also be composed of membrane resistance (R_m), resistance of the pore blocking (R_p) and cake layer (R_c). R_m was determined by measuring the flux of the virgin membrane with nanopure water. R_{ir} and R_r were calculated by testing the flux before and after flushing the membrane using nanopure water. R_{cp} and R_c were determined by measuring the flux before and after cleaning the membrane gently by a sponge.

3. Results and discussion

3.1 Permeability and structure / composition of fouling layer

3.1.1 Permeability (Flux) development in a non-aerated and an aerated reactor

Fig. 2 shows the flux trends of the non-aerated reactor (GDMBR1), in comparison to the aerated reactor (GDMBR2). It can be roughly divided into two stages. In the first stage, the permeability decreased sharply from 1080 L/m²hbar at t=0 in both reactors to around 20 L/m²hbar in GDMBR1 (low DO) and to around 40 L/m²hbar in GDMBR2 (high DO) until day 50. During the second stage, the permeability remained stable at around 40 L/m²hbar (corresponding to a flux of 2 L/m²h) in GDMBR2 and around 20 L/m²hbar in GDMBR1 (corresponding to a flux of 1 L/m²h), respectively, from day 50 until the end of the operation. The phenomenon of flux stabilization was first observed by Peter-Varbanets et al. in a dead-end gravity-driven membrane filtration system with a value of 4-10 L/m²h for the filtration of river, lake or diluted wastewater [4]. The stabilization of flux is related to the development of heterogeneous structures in the bio-fouling layer, due to biological activity including predation in the fouling layer [4, 6, 7]. Besides, they also found that DO-limiting conditions

resulted in a lower flux in GDM systems during a filtration period of 30 days [5]. Similarly, Jabornig and Podmirseg [34] also reported flux stabilization during grey water treatment with values of 1-2 L/m²h in an aerated gravity-driven hollow fiber membrane systems. In the latter study, the energy demand was reported to be < 1.4 kWh/m³, which is significantly lower than for normal MBRs.

As for the grey water treatment using MBR systems, Bani-Melhem et al. [35] also performed a study on a constant pressure MBR for grey water treatment. Due to the short period of each stage (7 days) and membrane cleaning, no flux stabilization was observed. The permeability of their system on Day 7 of each stage was in the range of 15-38 L/m²hbar, which was lower than the stable value of our long-term aerated GDMBR system. Jabornig [36] used an aerated moving bed biofilm membrane reactor (microfiltration membrane with a pore size of 0.2 µm) for grey water treatment, and after 80 days the flux fluctuated around 5 L/m²h under a constant pressure of 200 mbar (corresponding to a permeability of 25 L/m²hbar, which was nearly the same as our non-aerated system).

Most studies on submerged, conventional MBRs treating municipal and domestic wastewater report membrane flux values between 5-40 L/m²h, with a TMP increase during time [37]. Sun et al. [38] observed the characteristics of a full-scale MBR for municipal wastewater treatment with backwashing and chemical cleaning once a week, and the flux was fixed at 10 L/m²h. TMP hovered at 250 mbar and 600 mbar during the summer and winter season respectively, corresponding to permeabilities of 40 L/m²hbar and 17 L/m²hbar. During the summer the permeability was nearly the same as our aerated GDMBR, however, it was even lower than the non-aerated GDMBR in winter season. Sabia et al. [39] reported that the permeability was equivalent to 64 L/m²hbar in a pilot-scale MBR. The reason for this relatively high value is presumably the high membrane pore size of 0.2 µm, and the fact that backwashing and chemical cleaning were also applied in their system. In a study by M. Zahid et al. [40], the flux was 16.3 L/m²h, and the TMP ranged from 200-800 bar with back flushing

and physical cleaning. Hence, when the TMP reached values larger than 300 mbar and 900 mbar, the permeability would be lower than our aerated MBR and non-aerated MBR, respectively. On basis of the present investigations, a flux stabilization was observed, implying that no progressive fouling took place during 120 days. However, pilot tests will be required to investigate operation over longer periods with real greywaters and changeable influent qualities.

The low permeate flux in our system was because of the low constant TMP. The applied TMP (50 mbar) was also much lower compared with those observed MBR operated at constant flux. Increasing the constant gravity-driven pressure is expected to increase the permeate flux, but not the permeability. Therefore, although the flux values in our study were only 1 and 2 L/m²h in the non-aerated and aerated GDMBR, the permeabilities were still higher than the reported MBR for grey water treatment (even higher than other normal MBR systems) accompanied with low energy consumption.

3.1.2 Filtration resistance distribution

In order to investigate how hydraulic resistances were distributed, the membrane modules of the non-aerated reactor (GDMBR1) were taken out for resistance distribution analyses at the end of the experiment (day 120). As shown in Fig. 3, the total resistance (R_t) was $20.0 \times 10^{12} \text{ m}^{-1}$, which is considerably higher than in the aerated system as published before ($9.7 \times 10^{12} \text{ m}^{-1}$, Ding et al. 2016). The reversible fouling (R_r) represented a large percentage of R_t , with a value of $19.2 \times 10^{12} \text{ m}^{-1}$ and $9.2 \times 10^{12} \text{ m}^{-1}$ in the non-aerated and aerated reactor, respectively. The irreversible fouling (R_{ir}) accounted for a small percentage of R_t with a value less than $1 \times 10^{12} \text{ m}^{-1}$ in both systems. Therefore, low DO conditions significantly increased the reversible resistance and led to a high total filtration resistance. While this reversible fouling is very loosely attached, simple physical cleaning (such as flushing or draining) after long operation is sufficient to restore the flux in this type of low-

pressure membrane system regardless of the DO concentration.

In addition to hydraulic reversible and irreversible fouling, the values for cake layer resistance (R_c) and pore blocking resistance (R_p) were determined by mechanical cleaning of the membrane using a sponge. It can be seen from Fig. 3 that the total resistance is dominated by R_c with a value of $19.4 \times 10^{12} \text{ m}^{-1}$ in low-DO GDMBR, and $9.2 \times 10^{12} \text{ m}^{-1}$ in high-DO GDMBR, while R_p was significantly smaller with $0.3 \times 10^{12} \text{ m}^{-1}$ and $0.1 \times 10^{12} \text{ m}^{-1}$ in GDMBR1 and GDMBR2, respectively (previous data from Ding et al. (2016)). This means that the cake layer was the dominant resistance. Although the GDMBR and conventional MBR are quite different regarding operation and fouling, the cake layer is also dominating the fouling resistance in MBRs, as reported by Bani-Melhem et al. for a MBR system treating real grey water [35]. Similarly, it was found that the cake sludge dominated the fouling resistance but prevented pore blocking in a full-scale MBR [41]. Additionally, low DO concentrations resulted in a higher cake layer resistance, which is in line with the research of Jin et al., showing that the specific cake resistance under low DO conditions was 21-36 times higher than that under high DO conditions, and that this difference may be attributed to the difference in the fraction of small particles present in the biofilms between low and high DO conditions [18].

3.2 Membrane fouling: role of the bio-fouling layer

To better understand the phenomena of the permeate flux decline and under different DO concentration, the properties of bio-fouling layer adhered on the membrane surface were analyzed.

3.2.1 Morphology of the fouling layer

Membrane modules were taken out from each reactor for observation using optical coherence tomography (OCT) on day 35 and day 120, respectively, and systematic image

analysis was performed as described in Section 2.4.1. It can be seen from Fig. 4 (a, b, e and f) and Table 1 that the thickness of the bio-fouling layer in both reactors increased with operation time. In GDMBR1 (low DO), the average thickness increased from 413 μm (day 35) to 790 μm (day 120), while it increased from 275 μm (day 35) to 344 μm (day 120) in GDMBR2 (high DO). Additionally, the surface roughness of the fouling layer increased from 3.4 μm to 72 μm in GDMBR1, and from 22.4 μm to 121 μm in GDMBR2. From these results, it can be concluded that the DO concentration influenced the thickness of the bio-fouling layer; lower DO led to a thicker but less rough bio-fouling layer on the membrane surface.

OCT observation was also used to analyze the residual fouling layer after hydraulic cleaning and chemical cleaning. Fig. 4 (c, d, g and h) shows that most of the fouling layer can be removed by gentle flushing of the membrane surface, whereby the thickness of the fouling layer was diminished from 790 μm to 8.9 μm in GDMBR1 (low DO) and from 344 μm to 7.4 μm in GDMBR2 (high DO). In both reactors, hardly any further change in the fouling layer structure was observed after chemical cleaning, because the layer was already practically completely removed by hydraulic cleaning. Therefore, it can be concluded that the bio-fouling layer was relatively loosely adhered on the membrane surface in the low-pressure GDMBR systems, so that hydraulic flushing is sufficient for release of the major part of the fouling layer, regardless of the DO content. These results also correspond with the resistance analysis reported in Section 3.1.2, that the cake layer contributed to the main filtration resistance, and that this cake resistance was hydraulic reversible.

3.2.2 Biomass, EPS content and activity of the fouling layer

Quantification of biomass adhered on the membrane surface was conducted at the end of the operation. The suspended sludge and the bio-fouling layer from each reactor were taken out for EPS extraction, and the concentrations of protein and polysaccharide were measured. As seen in Table 2, the amount of biomass of the fouling layer (gVSS/m^2) was more than 2

times higher at low DO (GDMBR1), which indicates that more organic compounds and cells accumulated on the membrane surface compared with high DO (GDMBR2). This corresponds with the fact reported above that the fouling layer was thicker at low DO. Furthermore, the EPS content of the bio-fouling layer in GDMBR1 (284.6 mg/gVSS) was more than two times higher than in GDMBR2 (122.9 mg/gVSS). This implies that the surface load of EPS was more than five times higher at low DO (3074 and 578 mgEPS/m² in GDMBR1 and GDMBR2, respectively). Both proteins and polysaccharides in GDMBR1 were higher than those in GDMBR2. Additionally, the EPS contents (including proteins and polysaccharides) of suspended sludge in GDMBR1 was higher than GDMBR2. The higher amount of biomass with higher EPS contents attached on the membrane is likely one of the reasons for the low stable flux under low DO condition, because the biomass (including colloidal EPS) is considered to be the main foulant in MBR-like systems [42].

To better understand how the DO concentration influenced the distribution of EPS in the bio-fouling layer, CLSM analyses were conducted (Fig. S1), with red, green and blue signals representing proteins, polysaccharides and bacterial cells (DNA), respectively. As shown in Fig. S1 (a and b), more non-colored areas were observed in GDMBR2 than in GDMBR1. Assuming that the non-colored areas correspond to voids, this suggests that the fouling layer under low DO condition was more compact than in the high DO system, and it indicates a higher EPS content (proteins and polysaccharides) of the biofouling layer at low DO. This is also confirmed by the EPS quantification results (Table 2). Fig. S1 (c, d, e, f, g, h, i and j) shows the optical cross-sections of the bio-fouling layers. Under high DO conditions, the bacterial cells, proteins and polysaccharides were distributed homogeneously along the thickness of the layer. However, the proteins and polysaccharides were located mainly near the membrane surface at low DO conditions, which indicates that the EPS adhere strongly to the membrane; the bacterial cells were more inclined to the top of the layer.

The ATP content of the bio-fouling layer in GDMBR1 and GDMBR2 was determined as

0.84 and 1.1 nMATP/mgL⁻¹VSS, respectively. Similar tendencies were observed in the mixed liquor sludge, as shown in Fig. S2. The ATP content increased with time in both reactors, and remained steady after day 50. The values were always higher in GDMBR2 (high DO) than in GDMBR1 (low DO), which confirms that low DO resulted in a lower level of biological activity as compared to high DO conditions.

3.3 Effect of DO on organics removal

3.3.1 Sludge concentration and activity

Fig. S3 shows the suspended sludge concentrations versus time. While no sludge was inoculated at the start, the concentrations in both reactors increased gradually from 0 to 600 mg/L in GDMBR1 (low DO) and 0 to 700 mg/L in GDMBR2 (high DO). The reason for the higher sludge concentrations in GDMBR2 was the shorter hydraulic retention time (HRT), caused by the higher flux in GDMBR2. The activity of the suspended sludge was determined by ATP measurement (for TOC concentrations see Fig. 5 in Section 3.2.2). The ATP contents in both reactors increased sharply during the first 30 days, and the values were always higher in GDMBR2 than GDMBR1. The ATP contents remained stable afterwards at around 1.5 and 2.0 nM/mgL⁻¹ COD in GDMBR1 and GDMBR2, respectively. This implies that the biological activity was well developed in the GDMBR system, and that a higher DO resulted in a higher biological activity. Williamson and Nelson [43] also found that a significant increase in viability (ATP) occurred in bench-scale activated sludge reactors with increasing DO concentrations up to 15 mg/l.

3.3.2 TOC removal

Fig. 5 presents the removal of organics under high DO and low DO conditions. The TOC concentration in the feed of both reactors was around 100 mg/L during the whole operation. The TOC concentrations in the permeates were around 10 mg/L on day 20 in both reactors,

and afterwards the concentrations in GDMBR1 were always higher than in GDMBR2. After day 20, the concentrations in both reactors continued to decrease gradually, and reached values around 5 mg/L on day 50. It should be noticed that on day 50 the values in GDMBR1 were still slightly higher than in GDMBR2.

As for the mixed liquor, the DOC concentration decreased sharply from 80 mg/L to 20 mg/L after 22 days in both reactors. The DOC continued to decrease to values below 10 mg/L in both reactors, although the decrease was initially more rapid in GDMBR2. A possible reason for this is that high DO led to a higher metabolic activity of the microorganisms in the sludge. Similar results were reported by Dong [44], showing that the COD removal efficiencies in a membrane aeration/filtration combined bioreactor were 94.5% and 96% with DO concentrations of 0.5 mg/L and 4 mg/L, respectively.

3.3.3 Fate of organic carbon fractions

To better understand how DO is influencing the removal of organics, LC-OCD was used for detailed characterization of the organics in the feeds as well as in the mixed liquor and permeate effluent. Fig. 6a shows the LC-OCD profiles of feed water (day 45). The chromatograms can be divided into five fractions: (i) biopolymers (BP), (ii) humic substances (HS), (iii) building blocks (BB), considered to be natural breakdown products of humics, (iv) low molecular weight humics and acids (Low-MW organics) and (v) neutrals [27]. These fractions are quantified in Table S1. As shown in this table, the feed was mainly composed of neutrals (102 mg/L, 97.8% of the total DOC) while the humic substances content was 1.0 mg/L (1.0%), which was in accordance with the feed water recipe described in Section 2.2. The BP peak could not be determined in the feed water because the cellulose was present as colloidal suspended organics, and the prefilter before the SEC column removes colloidal material.

Fig. 6b and c show the LC-OCD profiles of mixed liquors and effluents on day 45 and on

day 114, respectively, and the integration results are presented in Table S1 and Table S2 in the Supplementary Material. The mixed liquor (supernatant of the sludge) also contained five main peaks. In comparison with the profile of feed water, DOC contents in GDMBR1 and GDMBR2 were 12.1 mg/L and 7.1 mg/L, respectively, which was significantly lower than the feed (104.9 mg/L) on day 45. The neutrals peak almost disappeared in both reactors, whereas the BP content was 6.0 mg/L and 3.2 mg/L in low DO and high DO reactors, respectively, while practically no BP was present in the feed water. This clearly indicates that the sludge degraded the low molecular weight organics (such as glucose, amino acids and fatty acids) and released extracellular biopolymers (EPS).

The DOC contents in GDMBR1 and GDMBR2 on day 114 were 6.9 mg/L and 5.9 mg/L, respectively, which were lower than on day 45. The neutrals peaks were still hardly observed in both reactors; the value of BB peaks decreased from 6.0 mg/L to 1.1 mg/L in GDMBR1 and from 3.2 mg/L to 0.9 mg/L in GDMBR2. The BP fraction, which is considered to be composed mainly of polysaccharides and proteins, in the sludge supernatant might originate from bound EPS, which can be rejected by membranes [45]. Furthermore, the peaks of HS and BB in both reactors increased slightly on day 114 as compared with day 45. Humics and building blocks are the metabolic products of microorganisms, which are non-biodegradable organics, and therefore, accumulated slightly in the reactors with time. From the TOC results presented in Section 3.3.2, it can be concluded that the oxygen content influenced the concentration of organics in the mixed liquor, and the LC-OCD results clearly show that a higher DO leads to a lower concentration of biopolymers, humics and building blocks in the mixed liquor (Table S1 and S2).

Comparing the permeate with the feed solution, the DOC removal rates were 94.6% and 95.6% in GDMBR1 on day 45 and day 114, and 96.2% and 96.7% in GDMBR2 on day 45 and day 114 (Table S1 and S2). The main fraction removed was the neutrals fraction. As stated above, the molecular weight of the neutrals is 0.35-0.5 kDa, so membrane rejection

cannot be the reason for this high removal. Therefore, it can be concluded that the neutrals of the feed were removed by biodegradation or by the presence of the bio-fouling layer (see section 3.3.4), and the efficiency of this removal process increased with time in both reactors. Additionally, the biopolymer fractions in the effluents (E1 and E2) decreased from 1.2 mg/L and 0.56 mg/L (day 45) down to 0.05 mg/L and 0.06 mg/L (day 117), suggesting that the removal efficiencies for large molecular weight organics improved during long term operation in GDMBR.

These results are well in line with the analyses of ATP and EPS (Section 3.2.2), which indicate that high DO conditions resulted in a high biological activity, while the activated sludge consumed more organics and released less EPS in the bulk at high DO. At low DO, there is more BP in the bulk than at high DO while the BP contents in the effluents are similar, which indicates that more non-biodegradable BP (=EPS) was accumulated in the fouling layer. Correspondingly, the EPS content in the fouling layer is higher at low DO.

3.3.4 Role of the bio-fouling layer: impact on organics removal

As discussed in Section 3.3.3, the removal of biopolymers increased with increasing operation time. To investigate the underlying mechanisms, the mixed liquor was taken out from the reactors and filtrated by the virgin UF membrane under 50 mbar on day 114. The organic composition of these samples, as measured by LC-OCD, is shown in Fig. 7. UF1 and UF2 were the permeate of mixed liquid from GDMBR1 and GDMBR2 directly filtrated by new membrane; E1 and E2 were the effluent of GDMBR1 and GDMBR2. We can clearly see the peaks of biopolymers, humics, BB and LMW acids in the permeate of UF; however, only humics, BB and LMW acids peaks were observed in the effluent of the GDMBRs. The same membrane (nominal cut-off 150 kD) used in the GDMBRs was used for ultrafiltration of the mixed liquor. The UF membrane rejected the biopolymer fraction by 44.0 – 47.7%, while the rejections in the GDMBRs at low and high DO were 93.9% and 95.9%, respectively (Table 3).

Also for humic substances, an increased rejection was observed in the GDMBR as compared to the UF alone (48.1-51.6% vs. 31.2-38.1%). A plausible explanation for this increased rejection can be the fact that the bio-fouling layer adhered on the membrane surface acted as a secondary barrier, enhancing the rejection of biopolymers and humic substances. Increased retention by cake layers, a phenomenon also designated as “secondary membrane” or “dynamic membrane”, has been reported by Ersahin et al. [46]. This principle was also applied to reduce membrane fouling [47], by a secondary membrane acting as a layer in which foulants are removed by adsorption or screening. Our results, showing increased removal of fouling compounds (biopolymers) by the bio-fouling layer, correspond to this general behavior of secondary membranes. Similarly, Kang et al. [48] demonstrated that membrane biofilms acted as a secondary filtration barrier for both low (<1 kDa) and high (>100 kDa) molecular weight organic matter during long-term MBR operation using real domestic wastewater. While humic substances are considered as non- or slowly biodegradable, biopolymers removal in principle also can take place by biodegradation. For example, Chomiak et al. [12] showed that hydrolysis and biodegradation of dextrans (MW 150 – 2000 kDa) took place in membrane biofilms.

No increased rejection, or even a decreased rejection in presence of the bio-fouling layer, was found for building blocks, LMW-humics and neutrals. These fractions can originate from the influent, from metabolic products or from degradation of biomass (e.g., the concentration of the humics fraction in the reactor was higher than in the feed). A decreased rejection or even a negative rejection can be explained by degradation and leaching processes within the bio-fouling layer. Similar results were published by Derlon et al. [13]: Biofilms initially removed assimilable organic matter (AOC), but the biofilm started to leach AOC after extended operation resulting in an elevated AOC content in the permeate.

In summary, the low-pressure GDMBR exhibited a good removal efficiency of organics for grey water treatment, and the removal efficiency of the Biopolymers and Humics

increased with operation time. Additionally, the high DO content system always achieved better removal efficiencies of organics, although the difference became less significant during long term operation.

3.4 Fouling mechanisms

As discussed in section 3.2.2, more biomass was accumulated on the membrane surface which resulted in a thicker bio-fouling layer containing more biopolymers and EPS under low DO conditions. Furthermore, we found that the mixed liquor of the low-DO system contained more biopolymers (section 3.3.4 and Fig. 7). Soluble microbial products (such as biopolymers) play an important role in membrane fouling under both aerobic and anaerobic conditions [49-52]. Thus, we can assume that deposition of biopolymers from the mixed liquor contributed to the higher cake layer resistance under low DO conditions. Additionally, the bio-activity of the fouling layer was lower than at high DO conditions. This can potentially be due to the action of higher organisms (metazoans) at high DO, having less activity at low DO. In previous investigations it has been described that predation in the bio-fouling layer leads to higher porosity, higher surface roughness and lower hydraulic resistance [6, 7, 53, 54]. This corresponds well with results of surface roughness, while the roughness were smaller in low DO condition (Section 3.2.1). Therefore, it is plausible that, in addition to the fouling layer composition, a lower level of predation under low DO conditions contributed to the lower permeate flux in comparison to the high DO case.

Furthermore, the EPS content within the fouling layer (proteins and polysaccharides) was higher under low DO conditions, which is in line with the fact that a higher amount of biopolymers was removed by the membrane under low DO conditions (Table 3: R1-E1 = 1076 $\mu\text{g/L}$; R2-E2 = 858 $\mu\text{g/L}$). Similarly, Yoon Kim et al. [20] found that a higher amount of EPS and lower porosity for the biofilm in a low-DO MBR was the key factor leading to increased fouling. SMP or EPS were related to the higher metabolic stress that anoxic

conditions exerted on the biomass [55]. Actually, it has been widely reported that EPS (including proteins and polysaccharides) is a main foulant for the membranes [49, 56, 57]. Interestingly, our results also show that the DO content influenced the spatial distribution of EPS in the bio-fouling layer (Fig. S1), which has not been reported before. When DO was sufficient in the bulk mixed liquor, the bacterial cells, proteins and polysaccharides were distributed homogeneously along the layer. However, the EPS (proteins and polysaccharides) were mainly located inside the layer and near the membrane surface, when DO was limited. The probable reason for this is that bacterial cells were more inclined to the top of the layer to obtain more oxygen from the bulk sludge to meet the requirement of metabolism. This is in agreement with the fact that the oxygen level does not only influence the metabolisms but also the bacterial community in the reactor [58].

3.5 Energy consumption analyses

Based on the characteristics of GDMBR, there is no energy cost for backwashing and physical cleaning. Therefore, as seen in equation (1), the total energy consumption (E_{total} , kJ) for treating per m^3 water only consists of the energy for lifting the water to the inlet by pumping (E_1 , kJ), the energy for recycling the overflow mixed liquor (E_2 , kJ) and the energy of the aeration for the whole system by the air compressor (E_3 , kJ)

$$E_{total} = E_1 + E_2 + E_3 \quad (1)$$

$$E_1 = \frac{W_1}{\eta_1 \eta_2} = \frac{m_1 g h}{1000 \cdot \eta_1 \eta_2} = \frac{\rho V g h}{1000 \cdot \eta_1 \eta_2} \quad (2)$$

$$E_2 = n E_1 \quad (3)$$

$$E_3 = W_2 = \frac{m_2}{AE} \quad (4)$$

As seen in equation (2), (3) and (4), E_1 is the energy consumption for pumping the wastewater, kJ; W_1 is effective work for pumping the wastewater (kJ); η_1 is the coefficient of

electrical energy transforming into mechanical energy; η_2 is the efficiency of the pump; m_l is the mass of the wastewater pumped (kg); V is the volume of wastewater pumped (L); g is gravitational acceleration (m/s^2); ρ is the density of the wastewater (kg/m^3); h is the height for pumping the feed (m). E_2 is energy consumption for recycling the overflow back to the system (kJ); n is the reflux ratio. E_3 is the energy consumption of the air compressor (kJ); m_2 is the mass of the oxygen (kg); AE is aeration efficiency (kgO_2/kWh). In this study, ρ was assumed as 1000 kg/m^3 , n was 4; g was 9.81 m/s^2 , η_1 and η_2 were both set as 0.60; the lifting height was set as 0.5 m (corresponding a TMP of 50 mbar); AE was set at $0.3 \text{ kgO}_2/\text{kWh}$, which is based on Grady Jr et al. [59]. Thus, the energy consumptions of GDMBR1 and GDMBR2 can be calculated as 0.019 kWh/m^3 and 0.040 kWh/m^3 , respectively.

The energy consumption of traditional wastewater treatment is reported to be 0.3-0.4 kWh/m^3 [60]. Table 4 shows a list of operation parameters and energy consumptions of MBRs for wastewater and grey water treatment. These MBRs are usually operated under constant flux mode, and the flux ranges from 8-25 $\text{L/m}^2\text{h}$ in pilot and full scale processes [61-63]. The energy consumption of these MBR fluctuates from 0.64-6.1 kWh/m^3 for wastewater treatment. Santasmasas et al. reported that the energy consumption was 2.0 kWh/m^3 for a larger MBR grey water treatment unit with nominal capacity of 3696 L/day [64]. Jabornig developed a fixed fiber biofilm membrane process (also without backwashing and cleaning) with an energy consumption of 1.4 kWh/m^3 for household grey water treatment [34, 36]. As for the grey water treatment, the energy cost would be lower than 1.6 kWh/m^3 . In our GDMBR systems, the energy consumptions were even lower than the traditional activated sludge process ($0.02\text{-}0.04$ vs. $0.3\text{-}0.4 \text{ kWh/m}^3$). As discussed in Section 3.1.1, although the flux of GDMBR was quite low, the permeability was more comparable with the normal constant-flux mode MBR. In addition, the operation of GDMBR is less complicated since no backwashing, physical cleaning and chemical cleaning is required.

GDMBR represents a relevant alternative for the decentralized wastewater treatment and

reuse in rural area of developing and transition countries. The required capacity for domestic grey water is approximately 200 L/d per family. Assuming the flux stabilized at 2 L/m²h (high DO condition), the required membrane area will be 4.2 m²; If the flux is stable at 1 L/m²h (low DO case), the required membrane area is 8.4 m² per household.

Evidently, the lower energy consumption goes to the expense of membrane area: the flux in the aerated GDMBR is around 4-10 times lower than in conventional MBR, while the flux in the non-aerated GDMBR is again 2 times lower than in the aerated GDMBR. Thus, while energy consumption decreases, the required membrane area is increasing. However, the price of membranes is expected to decrease further in future due to developments in material science and increase in production scale. Furthermore, the costs of auxiliary pipes, valves, equipment and process control is expected to be lower for the GDMBR system, especially for the non-aerated system, in view of the absence of a suction pump, cleaning cycles and aeration. The choice of which type of system is preferable depends on scale and local conditions. In general, GDMBR seems to be more preferential for decentralized applications, where energy availability might be critical, and where the membrane area is less decisive. However, the decision on the choice of the most optimal setting will have to be taken for each specific case in consideration of the local conditions.

4. Conclusions

We investigated the possibility to operate a membrane reactor for grey water treatment in a passive manner, i.e. without aeration, without cleaning or backwashing, and in a gravity driven mode at a low transmembrane pressure (50 mbar). It was shown that a stable permeability and flux could be obtained during extended operation, although this permeability is lower than in a similar reactor with aeration (20 vs. 40 L/m²hbar). At both DO conditions, the main resistance was caused by a hydraulically reversible cake layer.

The bio-fouling layer adhered on the membrane surface plays a crucial role for the flux

development and removal of organics. At low DO, more biopolymers were rejected from the mixed liquor and a thicker bio-fouling layer developed on the membrane surface with a higher hydraulic reversible resistance and higher cake layer resistance. The surface load of EPS in the fouling layer (mg EPS/m^2) was more than five times higher at low DO. A lower biological activity, a thicker fouling layer and smaller surface roughness were observed at low DO than at high DO. Furthermore, comparison with a normal ultrafiltration showed that the presence of the fouling layer resulted in an enhanced rejection of high-MW compounds (biopolymers) and of humic substances, indicating that the bio-fouling layer acted as a secondary membrane. No significant rejection of low-MW compounds (building blocks and acids) was observed in presence of the bio-fouling layer. In some cases, even a net production of this fraction was observed, which is probably related to hydrolysis and leaching processes taking place within the bio-fouling layer.

GDMBR is a simple process which can be operated with low maintenance, without any backwashing, physical flushing, chemical cleaning or sludge discharge. For the grey water composition studied here, the process can even be operated without any aeration during extended periods of time, which makes this process suitable for decentralized wastewater treatment and water reuse in developing countries. Although the flux is significantly lower compared to normal MBRs, the permeability is still comparable. In the low-DO process the energy consumption was only 0.019 kWh/m^3 , which means the process requires substantially less energy than conventional MBRs or activated sludge processes. Further research is needed to investigate the potential fields of application, as well as methods to increase the stable flux value.

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Fig. 1: Schematic diagram of the two membrane systems investigated: (a) GDMBR1: without

aeration (low DO) and (b) GDMBR2: with aeration (high DO)

Fig. 2: Permeability as a function of time at low DO (GDMBR1) and at high DO (GDMBR2).

Fig. 3: Distribution of filtration resistances of GDMBR1 on day 120: (a) Reversible fouling resistance (R_r); Irreversible fouling resistance (R_{ir}); Cake layer (R_c); Pore blocking resistance (R_p); and membrane resistance (R_m).

Fig. 4: Typical OCT images of bio-fouling layer developed on the membrane surface on day 35, day 120, after hydraulic cleaning and after chemical cleaning. Red arrows indicate the fouling layer-membrane interface

Fig. 5: TOC concentrations in mixed liquor and effluent of GDMBR1, operated without aeration, and GDMBR2, operated with aeration (the latter based on previously published data (Ding et al. 2016)).

Fig. 6: Effect of DO on removal of organics. LC-OCD chromatograms of: (a) influent (synthetic feed water); (b) mixed liquors and permeates on Day 45; (c) mixed liquors and permeates on Day 114. R1 and R2: mixed liquid samples from GDMBR1 and GDMBR2; E1 and E2: effluents of GDMBR1 and GDMBR2

Fig. 7: Effect of fouling layer on the effluent composition. UF1 and UF2 were the permeates of mixed liquid from GDMBR1 and GDMBR2 directly filtrated by a new membrane; E1 and E2 were the effluents of GDMBR1 and GDMBR2.

Table 1: Fouling layer thickness and roughness as calculated from OCT image analyses in GDMBR1 (low DO, no aeration) and GDMBR2 (high DO)

Samples	Thickness (μm)		Roughness (μm)	
	GDMBR1	GDMBR2	GDMBR1	GDMBR2
Day 35	413 $\pm$ 2.0	275 $\pm$ 15*	3.4 $\pm$ 2.0	22.4 $\pm$ 5.6*
Day120	790 $\pm$ 50	344 $\pm$ 70*	72.0 $\pm$ 4.1	121 $\pm$ 16*
After Hydraulic cleaning	8.9 $\pm$ 1.8	7.4 $\pm$ 1.4*	--	--
After Chemical cleaning	5.4 $\pm$ 1.6	7.4 $\pm$ 1.4*	--	--

*The numbers shown in italics represent previously published data (Ding et al. 2016)

Table 2: EPS, ATP and biomass in mixed liquor and in biofouling layer, analyzed on day 120

(n=2)

Sludge sample	mgEPS/gVSS	mgPr/gVSS	mgPs/gVSS	ATP content nM/ mg/LVSS	Biomass on the membrane (gVss/m ²)
GDMBR1 sludge	191.3 $\pm$ 5.3	159.7 $\pm$ 1.7	20.6 $\pm$ 0.4	--	--
<i>GDMBR2 sludge</i>	<i>106.3$\pm$4.6 *</i>	<i>78.0$\pm$3.5*</i>	<i>12.7$\pm$1.5*</i>	--	--
GDMBR1 fouling layer	284.6 $\pm$ 13.2	184.3 $\pm$ 11.3	42.6 $\pm$ 8.3	0.84 $\pm$ 0.03	10.8 $\pm$ 0.6
<i>GDMBR2 fouling layer</i>	<i>122.9$\pm$8.8*</i>	<i>103.7$\pm$0.7*</i>	<i>24.7$\pm$2.1*</i>	1.1 $\pm$ 0.05	4.7 $\pm$ 0.9

*The numbers shown in italics represent previously published data (Ding et al. 2016)

Table 3: Effect of cake layer on the removal of organics (LC-OCD analyses of organics on Day 114). R1 and R2: mixed liquor samples; UF1 and UF2: permeate samples from the mixed liquor using a virgin UF membrane; E1 and E2: effluent samples of GDMBR1 and GDMBR2, respectively (concentrations in $\mu\text{g/L}$)

Sample	DOC	BP	HS	BB	LMWha	Neutrals
R1	6996	1122	2496	1599	148	1631
	Percentage %	(16.0)	(35.7)	(22.9)	(2.1)	(23.3)
R2	5934	914	2108	1439	113	1360
	Percentage %	(15.4)	(35.5)	(24.3)	(1.9)	(22.9)
UF1	4845	587	1716	914	238	990
	Percentage %	(13.2)	(38.6)	(20.6)	(5.4)	(22.3)
UF2	3808	512	1304	968	132	893
	Percentage %	(13.4)	(34.2)	(25.4)	(3.5)	(23.5)
E1	4625	46	1208	1083	278	2010
	Percentage %	(1.0)	(26.1)	(23.4)	(6.0)	(43.5)
E2	3416	56	1093	1056	126	1084
	Percentage %	(1.6)	(32.0)	(30.9)	(3.7)	(31.7)
Removal (UF1/R1)%	30.7	47.7	31.2	42.8	--	39.3
Removal (UF2/R2)%	35.8	44.0	38.1	32.7	--	34.3
Removal (E1/R1)%	33.9	95.9	51.6	32.3	--	--
Removal (E2/R2)%	42.4	93.9	48.1	26.6	--	--

Table 4: Literature values for the energy consumption of MBRs for sewage treatment as well as energy consumption for the GDMBR process with and without aeration. EC represents the energy consumption per m³ wastewater

	References	Capacity per day (m ³ /d)	Flux (L/m ² h)	Permeability (L/m ² hbar)	EC (kWh/m ³)
	Gil et al [58]	10	19-25	63-83	4.9-6.1
	Fenu [59]	5500	22	37-110	0.64
MBR for wastewater treatment	Komesli [60]	170	8.3-13	27-43	1.1-2.53
	Jabornig [34]	0.2	5	25	<1.3
	Jabornig [32]	0.4-1.6 × 10 ⁻³	1-2	7	<1.4
	GDMBR1	2.9 × 10 ⁻³	2.0	40	0.019
	GDMBR2	1.4 × 10 ⁻³	1.0	20	0.040

Highlights

- Greywater was treated in GDM without any backwash, flushing and cleaning
- The permeability stabilized after approximately 50 days
- The lower flux at low DO was caused by higher content of EPS in the fouling layer
- The biofouling layer caused an increased rejection of biopolymers and humics
- The energy consumption of the reactors was 0.02 - 0.04 kWh/m³