Organic micro-pollutants’ removal via anaerobic membrane bioreactor with ultrafiltration and nanofiltration

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ABSTRACT

The removal of 15 organic micro-pollutants (OMPs) in synthetic municipal wastewater was investigated in a laboratory-scale mesophilic anaerobic membrane bioreactor (AnMBR) using ultrafiltration and AnMBR followed by nanofiltration (NF), where powdered activated carbon (PAC) was added to enhance OMPs removal. No significant effects of OMPs spiking and NF connection on bulk organics removal and biogas production were observed. Amitriptyline, diphenhydramine, fluoxetine, sulfamethoxazole, TDCPP and trimethoprim showed readily biodegradable characteristics with consistent biological removal over 80%. Atrazine, carbamazepine, DEET, Dilantin, primidone and TCEP showed refractory characteristics with biological removal below 40%. Acetaminophen, atenolol and caffeine showed a prolonged adaption time of around 45 d, with initial biological removal below 40% and up to 50–80% after this period. Most readily biodegradable OMPs contained a strong electron donating group. Most refractory OMPs contained a strong electron withdrawing group or a halogen substitute. NF showed consistent high rejection of 80–92% with an average of 87% for all OMPs, which resulted in higher OMPs removal in AnMBR-NF than in AnMBR alone, especially for refractory OMPs. Limited sorption performance of PAC for OMPs removal was mainly due to low and batch dosage (100 mg/L) as well as the competitive sorption caused by bulk organics.

Key words | anaerobic membrane bioreactor, electron donating/withdrawing group, municipal wastewater, nanofiltration, organic micro-pollutants

INTRODUCTION

Organic micro-pollutants (OMPs) have received increasing attention in recent years due to their potential harmful effects on public health and aquatic ecosystems. Eliminating OMPs in wastewater treatment systems is an important solution to control OMPs discharge that may impact ecosystems and potentially follow the water cycle back to drinking water sources. The aerobic membrane bioreactor (MBR) with conventional microfiltration (MF) or ultrafiltration (UF) has proved to be a more promising way to eliminate OMPs than the conventional activated sludge process. The MF/UF membrane filtration can completely reject microbial flocs in the bioreactor, which can result in high biomass concentration, long sludge retention time (SRT) and promote a diverse microbial community, thus enhancing OMPs removal (especially those with moderate biodegradation characteristics) (Weiss & Reemtsma 2008; Sipma et al. 2010). However, there are only few studies on OMPs removal in an anaerobic MBR (AnMBR), where the different microbial community and redox conditions compared with the aerobic MBR may promote OMPs removal. Wang et al. (2014) reported the effective removal of five polycyclic musks in an AnMBR and biotransformation as the dominant removal mechanism. Monsalvo et al. (2014) investigated the biological removal of 38 OMPs in an AnMBR and focused on advanced characterization of the relative amount of OMPs accumulated within the fouling layers.
formed on the membranes. Wijekoon et al. (2015) demonstrated a relationship between hydrophobicity and specific molecular features (e.g., electron withdrawing group (EWG), electron donating groups (EDGs)) of OMPs and their removal efficiency in an AnMBR from a pool of 27 OMPs. Especially, they found those OMPs containing nitrogen and sulphur were better removed in an AnMBR than in an aerobic MBR, possibly due to nitrogen or sulphur reducing bacteria in the AnMBR.

As a potential development, a novel AnMBR with nanofiltration (NF) may be proposed to enhance OMPs removal for wastewater reuse. NF can directly reject most OMPs effectively (Yangali-Quintanilla et al. 2010; Chon et al. 2012) and thus result in a long retention time of OMPs in a bioreactor to facilitate their biodegradation. A laboratory-scale AnMBR using UF was first set up in this study to investigate the removal of 15 OMPs (including pharmaceutically active compounds, personal care products, household chemicals and pesticides) from synthetic municipal wastewater. Then, an NF system was connected to form a hybrid AnMBR-NF process, where OMPs removal was further investigated. Finally, a batch dose of powdered activated carbon (PAC) was added to the anaerobic bioreactor to assess its effect on OMPs removal.

**MATERIALS AND METHODS**

**AnMBR set-up**

A laboratory-scale AnMBR set-up (Figure 1) consisted of a completely mixing bioreactor (Applikon Biotechnology, The Netherlands) with an effective volume of 2 L controlled by level sensor (temperature 35 ± 1 °C, pH 7 ± 0.1, stirring speed 200 ± 2 rpm, hydraulic retention time 12 h) and a side-stream crossflow hollow fibre UF membrane module (polyvinylidene fluoride, nominal pore size 30 nm, filtration area 310 cm², on/off 9/1 min, flux 6 L/m²/h, crossflow velocity 0.2 m/s). The AnMBR was operated alone in Phase 1 (1–30 d), where synthetic municipal wastewater with a chemical oxygen demand (COD) of 400 ± 10 mg/L consisting of food ingredients, chemical compounds and trace metals (the detailed recipe presented in Wei et al. (2014)) was treated at a flow of 4 L/d. Fifteen OMPs including pharmaceutically active compounds, personal care products, household chemicals and pesticides were spiked into the synthetic municipal wastewater at a concentration of 10–20 μg/L for individual OMPs (Table 1).

In Phase 2 (31–70 d), a Sepa CF II membrane cell system (Sterlitech Corporation, USA) holding a flat-sheet NF90 membrane (Dow/Filmtec, USA) was used in daily batch crossflow (0.1 m/s) constant-pressure (10 bar) filtration mode to filtrate the AnMBR permeate (4 L), producing 2 L permeate for final discharge and 2 L concentrate as affluent back to the AnMBR to form a semi-continuous AnMBR-NF process (Figure 1). In order to maintain the same organic loading rate in terms of both bulk organics (0.8 gCOD/L/d) and OMPs (20–40 μg/L/d for individual OMPs) in the AnMBR alone in Phase 1, concentrated synthetic municipal wastewater with double COD (i.e., 800 mg/L) and OMPs (i.e., 20–40 μg/L for individual OMPs) concentration was treated at a flow of 2 L/d in the AnMBR-NF in Phase 2. Here, the daily batch NF concentrate of 2 L was added into the concentrated synthetic municipal wastewater to maintain the total flow of the AnMBR at 4 L/d.

In the last 10 d of AnMBR-NF in Phase 2, a batch dose of PAC was added to the anaerobic bioreactor at 100 mg/L to assess its effect on OMPs removal. No excess sludge was wasted during the whole operation except in sampling for biomass measurements (weekly 5 mL) and batch OMPs sorption/biodegradation tests (60/75 mL at 30/61 d, respectively), resulting in an SRT in the bioreactor of around 2,800 d for the experimental period. Before the experiments, the AnMBR had been operated for more than 4 months to investigate the sustainable organic loading rate for this synthetic municipal wastewater (Wei et al. 2014).

**Figure 1** | Schematic diagram of the laboratory-scale AnMBR (Phase 1: 1–30 d) and AnMBR-NF (Phase 2: 31–70 d) set-up.

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**Table 1** | Concentration of OMPs used in synthetic municipal wastewater.

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Analytical methods

The OMPs removal as well as bulk organics removal, biogas production and biomass evolution were monitored during this study. OMP samples spiked with the corresponding isotopes were pre-concentrated via solid phase extraction (Dione Autotrace 280 solid-phase extraction instrument and Oasis cartridges) first, and then measured via liquid chromatography (Agilent Technology 1260 Infinity Liquid Chromatography unit, USA) followed by mass spectrometry (AB SCIEX QTRAP 5500 mass spectrometer, Applied Biosystems, USA) (Alidina et al. 2014). Commercial COD kits (TNT series, Hach Company) were used to measure the COD of influent and permeate based on the method of rapid digestion (150 °C, 2 h) followed by colorimetric measurement. Biogas composition and volume (reported under a temperature of 25 °C and pressure of 1 atm) were measured according to the gas bag method (Ambler & Logan 2011) based on gas chromatography. Biomass concentration in terms of mixed liquor suspended solids (MLSS) and mixed liquor volatile suspended solids (MLVSS) was measured according to the standard method of glass fibre filtration followed by sequential drying at 105 and 550 °C (APHA, AWWA & WEF 2005).

Removal definitions

The OMPs used in this study have a molecular weight less than 500 Dalton (i.e., an equivalent diameter of around 1–2 nm), while the nominal pore diameter of the UF membrane used in this study is 30 nm. Thus, we assume the direct OMPs rejection by UF membrane would be negligible. For both OMPs and bulk organics, the biological removal (including biodegradation and sorption) was defined as the ratio of feed mass minus UF permeate mass versus feed mass for both the AnMBR alone in Phase 1 and the AnMBR-NF in Phase 2. In the AnMBR alone in Phase 1, it can be calculated according to Equation (1), where $R_b$ is biological removal, $C_f$ is feed concentration, $C_{pU}$ is UF permeate concentration, $V_f$ and $V_{pU}$ are feed and permeate volume per day, respectively, and $V_f = V_{pU} = 4$ L.

$$R_b = \frac{C_f \times V_f - C_{pU} \times V_{pU}}{C_f \times V_f} = \frac{C_f - C_{pU}}{C_f}$$ (1)

### Table 1 | OMPs characteristics used in this study

<table>
<thead>
<tr>
<th>Compound</th>
<th>Application</th>
<th>CAS number</th>
<th>Formula</th>
<th>MW*</th>
<th>Charge**</th>
<th>logD**</th>
<th>EDG*</th>
<th>EWG*</th>
<th>C_f*</th>
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<tr>
<td>Carbamazepine</td>
<td>Anticonvulsant</td>
<td>298-46-4</td>
<td>C_{15}H_{12}N_2O</td>
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<td>Dilantin</td>
<td>Anticonvulsant</td>
<td>57-41-0</td>
<td>C_{15}H_{12}N_2O_2</td>
<td>252</td>
<td>N</td>
<td>1.41</td>
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<td></td>
<td>11</td>
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<tr>
<td>Primidone</td>
<td>Anticonvulsant</td>
<td>125-33-7</td>
<td>C_{12}H_{14}N_2O_2</td>
<td>218</td>
<td>N</td>
<td>0.83</td>
<td>-CONH_2</td>
<td></td>
<td>14</td>
</tr>
<tr>
<td>Sulfamethoxazole</td>
<td>Antibiotic</td>
<td>723-46-6</td>
<td>C_{10}H_{11}N_3O_3S</td>
<td>253</td>
<td>-</td>
<td>-0.22</td>
<td>-NH_2</td>
<td>-SO_3H</td>
<td>10</td>
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<tr>
<td>Trimethoprim</td>
<td>Antibiotic</td>
<td>738-70-5</td>
<td>C_{16}H_{19}N_4O_3</td>
<td>290</td>
<td>N, +</td>
<td>0.27</td>
<td>-NH_2, -OR</td>
<td></td>
<td>10</td>
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<td>Amitriptyline</td>
<td>Antidepressant</td>
<td>50-48-6</td>
<td>C_{20}H_{23}N</td>
<td>277</td>
<td>+</td>
<td>2.28</td>
<td>-NR_2</td>
<td></td>
<td>10</td>
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<tr>
<td>Fluoxetine</td>
<td>Antidepressant</td>
<td>54910-89-3</td>
<td>C_{17}H_{18}F_3N_2O</td>
<td>309</td>
<td>+</td>
<td>1.15</td>
<td>-NHR, -OR</td>
<td>-CF_3</td>
<td>10</td>
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<tr>
<td>Acetaminophen</td>
<td>Analgesic</td>
<td>103-90-2</td>
<td>C_4H_3NO</td>
<td>151</td>
<td>N</td>
<td>0.47</td>
<td>-OH, -NHCOR</td>
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<td>16</td>
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<td>Diphenhydramine</td>
<td>Antihistamine</td>
<td>58-73-1</td>
<td>C_{17}H_{21}NO</td>
<td>255</td>
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<td>1.25</td>
<td>-NR_2, -OR</td>
<td></td>
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<td>Atenolol</td>
<td>Beta-blocker</td>
<td>29122-68-7</td>
<td>C_{14}H_{22}N_3O_3</td>
<td>266</td>
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<td>-2.09</td>
<td>-OR, -OH</td>
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<td>Caffeine</td>
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<td>58-08-2</td>
<td>C_6H_10N_2O_3</td>
<td>194</td>
<td>N</td>
<td>-0.63</td>
<td>-R</td>
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<td>DEET</td>
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<td>134-62-3</td>
<td>C_{12}H_{17}NO</td>
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<td>-CONR_2</td>
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<td>TCEP</td>
<td>Flame retardant</td>
<td>115-96-8</td>
<td>C_4H_{12}Cl_3O_4P</td>
<td>285</td>
<td>N</td>
<td>1.47</td>
<td>-OR</td>
<td>-Cl</td>
<td>10</td>
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<tr>
<td>TDCCP</td>
<td>Flame retardant</td>
<td>13674-87-8</td>
<td>C_4H_{12}Cl_3O_4P</td>
<td>431</td>
<td>N</td>
<td>3.27</td>
<td>-OR</td>
<td>-Cl</td>
<td>10</td>
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<td>Atrazine</td>
<td>Herbicide</td>
<td>1912-24-9</td>
<td>C_6H_{14}ClN_5</td>
<td>216</td>
<td>N</td>
<td>2.64</td>
<td>-NHR</td>
<td>-Cl</td>
<td>10</td>
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</table>

*MW, molecular weight, Dalton; D, pH-dependent n-octanol–water distribution ratio; EDG, electron donating group; EWG, electron withdrawing group; C_f, feed concentration in the AnMBR alone in Phase 1, μg/L.

In the AnMBR-NF in Phase 2, with recycling NF concentrate to the AnMBR, biological removal can be calculated according to Equation (2), where \( R_{b,N} \) is biological removal for the N day, \( C_{f,N} \) is feed concentration for the N day, \( C_{pU,N-1} \) is UF permeate concentration for the N-1 day, \( C_{pN,N-1} \) is NF permeate concentration for the N-1 day, \( C_{pU,N} \) is UF permeate concentration for the N day, \( V_{f,N} \) is feed volume for the N day, \( V_{pU,N-1} \) is UF permeate volume for the N-1 day, \( V_{pN,N-1} \) is NF permeate volume for the N-1 day, \( V_{pU,N} \) is UF permeate volume for the N day, and \( V_{f,N} = 2 \text{ L} \), \( V_{pU,N-1} = V_{pU,N} = 4 \text{ L} \), \( V_{pN,N-1} = 2 \text{ L} \), \( C_{f,N} = 2C_f \).

\[
R_{b,N} = \frac{(C_{f,N} \times V_{f,N} + C_{pU,N-1} \times V_{pU,N-1} - C_{pN,N-1} \times V_{pN,N-1})}{C_{f,N} \times V_{f,N} + C_{pU,N-1} \times V_{pU,N-1} - C_{pN,N-1} \times V_{pN,N-1}}
\]

(2)

The total removal was defined as the ratio of feed mass minus UF permeate mass versus feed mass for the AnMBR alone in Phase 1 and feed mass minus NF permeate mass versus feed mass for the AnMBR-NF in Phase 2. According to this definition, the total removal was the same as biological removal for the AnMBR alone in Phase 1 (i.e., Equation (1)) while in the AnMBR-NF in Phase 2 it included biological removal and direct rejection by the NF according to Equation (3), where \( R_{t,N} \) is total removal for the N day, \( C_{p,N} \) is NF permeate concentration for the N day, \( V_{p,N} \) is NF permeate volume for the N day, \( V_{p,N} = 2 \text{ L} \), and others are the same as Equation (2).

\[
R_{t,N} = \frac{C_{f,N} \times V_{f,N} - C_{p,N} \times V_{p,N}}{C_{f,N} \times V_{f,N}} = \frac{C_{f,N} - C_{p,N}}{C_{f,N}}
\]

(3)

NF rejection was defined as the ratio of UF permeate mass minus NF permeate mass versus UF permeate mass according to Equation (4), where \( R_N \) is NF rejection for the N day and the others are the same as in Equations (2) and (3).

\[
R_N = \frac{C_{pU,N} \times V_{pU,N} - C_{p,N} \times V_{p,N}}{C_{pU,N} \times V_{pU,N}}
\]

(4)

### RESULTS

#### Biomass evolution

From Figure 2, the biomass concentration showed a little decrease initially followed by a slow increase during Phase 1, and a gradual decrease to a somewhat stable value during Phase 2. MLVSS was in the range of 3.9–5.4 g/L during the whole operation. MLVSS/MLSS was around 0.8, indicating a stable sludge composition.

#### Bulk organics removal

A high and stable COD removal around 97% and permeate COD below 20 mg/L were achieved during the AnMBR alone in Phase 1 (Figure 3). Adding NF filtration in Phase 2 resulted in a slight COD accumulation in the UF permeate due to the high rejection (around 60–80%) of refractory organics (e.g., humic-like substances from soluble microbial products). Thus, a slight decrease in biological COD removal from the AnMBR was observed (97 to 92%), but total removal from the AnMBR-NF system was maintained at a very high level (around 99%). COD in the UF permeate showed an initial significant reduction after PAC addition followed by a rapid increase to the level before PAC addition, indicating the limited sorption performance for bulk organics removal, which was mainly due to a low and batch PAC dosage (100 mg/L).
Biogas production

Figure 4 shows there was a stable biogas composition (methane 70–80%, carbon dioxide less than 5% and nitrogen 20–25%) and methane production (150–250 ml/gCOD) during the whole operation. These results were in agreement with previous results from the same AnMBR treating the same synthetic municipal wastewater (Wei et al. 2014), indicating no significant effects of OMPs spiking and NF connection on anaerobic performance.

OMP removal

Figures 5 and 6 show the OMPs concentration in UF permeate and OMPs biological removal in the whole operation. From Figures 5(a) and 6(a), amitriptyline, diphenhydramine, fluoxetine, sulfamethoxazole, TDCPP and trimethoprim showed no significant accumulation in the UF permeate after NF connection, and had readily biodegradable characteristics with over 80% biological removal (total removal in AnMBR-NF over...
98%). From Figures 5(b) and 6(b), atrazine, carbamazepine, DEET, Dilantin, primidone and TCEP showed a significant accumulation in the UF permeate after NF connection due to their high rejection by NF and refractory characteristics, with below 40% biological removal (total removal in AnMBR-NF 30–80%). From Figures 5(c) and 6(c), acetaminophen, atenolol and caffeine showed a prolonged adaptation time of around 45 d, initial biological removal of below 40% and up to 50–80% after this period (corresponding total removal in AnMBR-NF 80–95%). The effects of compound characteristics, NF rejection and PAC sorption on OMPs removal are further analysed in the next section.

**DISCUSSION**

**Compound characteristics**

First, no intrinsic correlation between the biodegradability and application of OMPs could be observed. For example, amitriptyline and fluoxetine, which are antidepressants, showed readily biodegradable characteristics while the flame retardants TCEP and TDCPP showed different biodegradability, which might be related to their molecular structure characteristics.

Compound characteristics (e.g., charge, hydrophobicity, halogen substitute, electron donating/withdrawing group, ring number, etc.) were regarded as the key factors affecting its biodegradability and removal in the aerobic MBR (Tadkaew et al. 2011). Those authors found that strong hydrophobic compounds (logD > 3.2) showed good removal mainly via sorption in the aerobic MBR. In this study, TDCPP with logD > 3.2 as well as four compounds with positive charge (amitriptyline, diphenhydramine, fluoxetine, trimethoprim) also showed consistent high biological removal (over 80%) during the whole operation. However, positive charged atenolol showed low biological removal (below 20%) in the AnMBR alone in Phase 1. The atenolol concentration in the UF permeate (Figure 5(c)) was low at the beginning of the AnMBR alone in Phase 1 (7.9 μg/L at day 1).
but increased rapidly in the initial few days (16.1 μg/L at day 3, 18.8 μg/L at day 7, close to the feed concentration of 20 μg/L), which demonstrated the occurrence of initial sorption onto sludge flocs followed by the rapid saturation of the sludge flocs due to their limited sorption capacity. Therefore, their biological removal could be mainly attributed to biodegradation rather than sorption and/or partition onto sludge flocs, especially considering the long-term operation (70 d) without wasting sludge in this study. Although a positive charge and/or high logD resulted in a longer contact time between compounds and microorganisms (thus benefiting their adaption and biodegradation), their main contribution was the initial removal from the liquid phase by sorption and/or partition onto sludge flocs.

Further analysis indicated the potential dependence of OMPs’ biodegradability on an electron donating/withdrawal group and the halogen substitute in molecular structure. All readily biodegradable compounds in this study contained a strong EDG (–NH₂, –NHR, –NR₂, –OR). All refractory compounds in this study contained a strong EWG (–CONH₂, –CONHR, –CONR₂) or halogen substitute (–Cl). These findings were in agreement with OMP studies in aerobic MBRs (Hai et al. 2011; Tadkaew et al. 2011) and AnMBRs (Monsalvo et al. 2014; Wijekoon et al. 2015). For some compounds (sulfamethoxazole, fluoxetine, TDCPP, TCEP, atrazine) containing both a strong EDG and a strong EWG or halogen substitute, biodegradability might be dependent on the relative strength of their electron donating and withdrawing capability.

**NF contribution**

From Figure 7, the tight NF membrane used in this study with a molecular weight cut-off around 200 Dalton showed a consistently high rejection of 80–92% with an average of 87% for all compounds tested. After NF connection, the total removal for all compounds in the AnMBR-NF was better than in the AnMBR alone, especially for refractory compounds, indicating the contribution of direct OMP rejection by NF. For the compounds with prolonged adaption time (acetaminophen, atenolol, caffeine) in this study, NF rejection resulted in not only a longer retention time in the bioreactor, thus enhancing their biological removal even without considering potential adaption effects, but also a higher compound concentration (more than twice that before NF connection) in the bioreactor, which might reach the threshold to activate their biological degradation and thus shorten their adaption time.

**PAC sorption**

Similarly to bulk COD in UF permeate, all refractory compounds’ concentration in UF permeate showed significant reduction initially after PAC addition, followed by a gradual

![Figure 7](http://iwaponline.com/jwrd/article-pdf/6/3/362/376365/jwrd0060362.pdf)
increase to the level before PAC addition (Figure 5(b)). This indicated a limited sorption performance for OMPs removal, which was mainly attributed to low and batch dosage (100 mg/L) as well as competitive sorption by bulk organics. Thus, the PAC application method (e.g., continuous addition, high dosage, sludge discharge) should be investigated to enhance the removal of refractory compounds. In order to avoid competitive sorption from the PAC by bulk organics and take full advantage of NF rejection (for both bulk organics and OMPs), granular activated carbon sorption might be used as a cost-effective post-treatment for NF permeate (Nguyen et al. 2013). Alternatively, advanced oxidation processes (e.g., ozone) could be a promising greener post-treatment for OMPs elimination, because sorption can only transfer the OMPs from water to activated carbon rather than mineralize them completely (Altmann et al. 2014).

CONCLUSIONS

Among 15 OMPs spiked in synthetic municipal wastewater treated by a mesophilic AnMBR process in this study, six compounds (amitriptyline, diphenhydramine, fluoxetine, sulfamethoxazole, TDCCP and trimethoprim) showed readily biodegradable characteristics, with biological removal of over 80%, six compounds (atrazine, carbamazepine, DEET, Dilantin, primidone and TCEP) showed refractory characteristics with biological removal below 40%, and three compounds (acetaminophen, atenolol and caffeine) showed a prolonged adaption time of around 45 d. Due to the complex molecular characteristics of OMPs and the limited number of OMPs used in this study, it was not possible to draw a distinct conclusion on the correlation between molecular structure and biodegradability. Generally speaking, the most readily biodegradable compounds contained a strong EDG while the most refractory compounds contained a strong EWG or halogen substituent. NF filtration enhanced the retention time of OMPs in the AnMBR, and thus their overall removal via biodegradation as well as through direct rejection, especially that of refractory compounds, was significantly improved. Limited PAC sorption performance for OMPs removal was mainly due to low and batch dosage (100 mg/L) as well as the competitive sorption caused by bulk organics. Although better performance of AnMBR-NF compared to AnMBR alone was observed, it would still not be an absolute barrier for OMPs discharge. Coupling post-treatment like sorption (e.g., activated carbon) and/or oxidation (e.g., ozone) would be necessary to remove the refractory OMPs.

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