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Fate of pharmaceuticals and their transformation products in integrated membrane systems for wastewater reclamation

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Abstract

The removal of pharmaceuticals (PhACs) present in urban wastewater by membrane bioreactors (MBRs) followed by reverse osmosis (RO) or nanofiltration (NF) membranes has been frequently addressed in the literature. However, data regarding the removal of their main human metabolites and transformation products (TPs) is still scarce. In this study, the presence of 13 PhACs and 20 of their metabolites and TPs was monitored during 2 consecutive years in the different treatment steps of urban raw wastewater (sewer, primary treatment, MBR and RO/NF). Rejection of the selected contaminants when using low pressure NF membranes (NF-90) or RO membranes (ESPA 2) after MBR step was also investigated. MBR achieved a high removal efficiency for the analgesic acetaminophen, which was found at the highest concentrations in the sewer and influent samples (18-74 μg L^{-1}) over the two experimental periods. Those PhACs that were partially removed after the MBR, were almost completely removed (>99%) by the RO membrane working under different process conditions. At a similar average permeate fluxes (18 L m^{-2} h^{-1}), the NF membrane showed high removal efficiencies (>90%) for all of the PhACs and their metabolites, though lower than those achieved by the RO membrane. When the flux of the NF90 membrane was increased to 30 L m^{-2} h^{-1} (while still operating at a feed pressure lower than the RO membrane at 18 L m^{-2} h^{-1}) the performance of the membrane increased, achieving 98% rejection of PhACs.

Keywords

Metabolites, degradation, Membrane Bioreactor, Reverse Osmosis, Nanofiltration, antibiotics

1 Introduction

As human health is increasingly depending on pharmaceutical products, the scientific community together with environmental and public health authorities have made a significant effort in understanding the fate of pharmaceutically active compounds (PhACs) both through engineered urban wastewater treatment processes as well as in the natural environment. This has been reflected in the high number of scientific publications devoted to this subject in the last two decades. On the contrary, modification of legal regulations up to now is basically inexistent. However, water scarcity and water reuse have become essential issues in water resource management worldwide, always considering the conservation of aquatic ecosystems as a final goal and main driver of the involvement of the scientific community.

So far, most of the research carried out regarding the fate of PhACs through MBR processes has mainly focused on the study of parent compounds, neglecting their metabolites. Only recently, the environmental presence of human metabolites and different transformation products (TPs) of these PhACs has been included within the scope of removal efficiency studies of different wastewater treatments technologies [1–3] . It should be considered that, once released onto the environment, these metabolites and TPs can be both innocuous [4], but also nocuous and even more toxic than the original substance against different aquatic organisms. Furthermore, the coexistence of the original drugs with these metabolites and TPs could lead to additive, antagonistic and/or synergetic effects which are hard to predict and that should be investigated. For instance, the assessment of the ecotoxicity of the photoproducts of the anti-inflammatory diclofenac (DCF) or naproxen has provided the evidence that acute and chronic toxicity can be greater for these photoproducts than for the parent compounds[1]. Effective concentration values (EC_{50}) obtained after 15 min assays for the antibiotic sulfapyridine (SPY)

and its acetylated metabolite, N^4-acetylsulfapyridine (acSPY), demonstrated that the marine bacteria *Vibrio fischerii* was more sensitive to the presence of the metabolite than to the original drug, and according the EU legislation (Directive 93/67/EEC), the former could be categorized as toxic[2].

On the other hand, it has been demonstrated that conventional activated sludge (CAS) treatment processes in waste water treatment plants (WWTPs) do not adequately remove many of the most commonly used PhACs such as DCF, the anticonvulsant carbamazepine (CBZ) or the antibiotic sulfamethoxazole (SMX) [5,6]. Different studies have demonstrated that membrane bioreactors (MBRs) are capable of removing moderately biodegradable and hydrophobic trace organic pollutants more efficiently than CAS treatments processes [7]. During the last years, MBR technology has become an accepted alternative to CAS processes for municipal wastewater treatment, not only regarding PhACs removal but also the overall water quality [8]. Furthermore, the combination of MBR technology followed by the reverse osmosis (RO) process or nanofiltration (NF) is typically used when high quality product water is required for planned potable reuse or when a reduction in salinity is required for irrigation reuse applications [9]. Different works have demonstrated that both membrane processes alone yield high rejection rates for PhACs usually >80% [10,11]. Various studies have shown that the combination of the MBR process with RO or NF results in higher removal rates for a wide range of PhACs, present in the wastewater (feed stream) to be treated; these are concentrated into a stream of water known as the concentrate stream (also known as the reject or brine stream) [12], leaving a permeate stream (also known as the product stream) with very low concentrations of the contaminants of interest. A recent work by Dolar et al. [13], which was carried in the same WWTP as this work, showed that removal rates for 22 PhACs by a Ropur TR70-4021-HF-RO membrane were above 99%.

A number of factors affect the removal of PhACs by the MBR-RO/NF process [14] which are not always properly considered when reporting removal efficiencies of the membrane processes involved. These include the membrane type (affecting molecular weight cut-off, surface morphology, hydrophobicity and charge), membrane fouling, membrane process parameters (membrane recovery and average permeate

flux) and feed water quality including temperature, pH and concentration, particularly due to urban activity and weather related events (such as rain periods). Furthermore, a number of studies have also shown that NF and ultralow pressure RO membranes are not perfect barriers for various micropollutants including endocrine disruptors and some PhACs such as SMX or CBZ [15,16]. Both membranes had a negative surface charge in the pH range used in the study [17,18], whereas the hydrophobic nature of a membrane is typically characterised by its contact angle. Alturki et al. [7] measured the contact angle of both the NF90 and ESPA2 membranes for virgin membranes (1 hour use) and after 25 hours of use. Although both virgin membranes could be classified as hydrophilic (contact angle of 42.5º for NF90 and 60.63º for ESPA2), the measurements consistently showed that following filtration of MBR effluent, there was an increase in the membrane surface hydrophobicity after 25 hours of use (contact angle of 77.6º for NF90 and 85.9º for ESPA2). In our case, since the membranes were in operation for a significantly longer period of time, the membranes are expected to be moderately hydrophobic.

It should also be considered that significant savings could be made in terms of energy reduction and chemicals and concentrate disposal costs when using NF in the place of RO membranes [19]. NF membranes are found in similar spiral wound configurations to RO membranes, making the switch during routine membrane replacement a feasible option to make.

Taking all this information into consideration, the objective of the present study is to evaluate and compare MBR-RO and MBR-NF in terms of pollutant removal efficiencies. To this aim, the fate of 13 PhACs together with their main human metabolites and TPs (20) was investigated in both configurations.

2 Methodology

2.1. MBR-RO/NF pilot plant

The MBR-RO/NF pilot plant was installed within the facilities of the full-scale WWTP of Castell-Platja d'Aro (NE Spain). This WWTP treats urban wastewater with a capacity of 35000 $m^3 day^{-1}$ and 175

000 populations equivalent (PO). The whole plant is shown in Figure 1a and consists of pre-screening followed by a bioreactor of 2.26 m^3, divided into four compartments according to the University of Cape Town (UCT) configuration for nutrient removal, as described by Monclús et al. [20] The MBR was installed at the outlet, containing a submerged flat sheet membrane from Kubota with a total surface area of 8 m^2 and a pore size of 0.4µm. The RO system consisted of variable frequency drive (VFD) controlled high-pressure pump and a single 4" x 40" membrane element. The two membranes tested for comparison during this study were a Filmtec NF90 membrane and a Hydranautics ESPA2 (RO) membrane. Both had a negative surface charge and were hydrophobic. The main difference between the two is the molecular weight cut-off (MWCO) (ESPA2 ≈ 100 g mol^{-1}, NF90 ≈ 200 g mol^{-1}) which in practical terms affects the salt retention and the membrane permeability.

Internal concentrate recirculation to the feed of the membrane was employed to increase the system recovery while remaining within membrane supplier's specifications (see Figure 1b). The VFD and the actuated concentrate valve allowed the system to be operated at a constant average permeate flux based on a fixed permeate flow set point of 135 L h^{-1}. Both membranes were operated at a system recovery rate (Q_p/Q_f) of 75% and a flux (Q_p/Membrane surface area) of 18 L m^{-2} h^{-1}; this last value remains within the average system flux of different operating installations, which usually ranges between 16.8 and 21 L m^{-2} h^{-1} [9]. The recovery of the membrane element (Q_p/Q_s), which includes the concentrate recirculation, was set at 12.5% within the manufacturer's guidelines and similar to those used in the operating plants surveyed by Raffin et al. [9], which ranged between 8.6 and 12.7 %. The operating conditions and membrane properties of the RO and NF membrane systems are given in Table S1 in the Supplementary Information (S.I.).

Due to the concentrate recirculation aforementioned, the composition of the feed water to the membrane was a blend of fresh feed water from the MBR and recirculated concentrate (Figure 1b). This meant that the feed concentrations of the solutes were higher than those normally found in ordinary feed water. The

removal efficiencies of the RO and NF membranes were thus calculated using a mass balance through the membrane, as shown in Equation 1:

$$\mathbf{C_m} = \frac{\mathbf{Q_p \times C_p + Q_c \times C_c}}{\mathbf{Q_m}} = \frac{\mathbf{Q_f \times C_f + Q_r \times C_c}}{\mathbf{Q_m}} \quad \textbf{[1]}$$

Where C_m (ng L^{-1}) is PhACs concentration in the membrane system feed water, Q_p (L s^{-1}) is the permeate flow, C_p (ng L^{-1}) is the PhACs concentration in the permeate, Q_c (L s^{-1}) is the concentrate flow, C_c (ng L^{-1}) is the PhACs concentration in the concentrate, Q_m is membrane system feed flow, Q_f (L s^{-1}) is the system feed flow without recirculation, C_f (ng L^{-1}) is the concentration in the feed water without recirculation and Q_r (L s^{-1}) is the recirculated flow.

Equation 2 was used to ensure that the results obtained for the system feed, permeate and concentrate make sense:

$$\mathbf{Q_f \times C_f - Q_p \times C_p = Q_d \times C_c} \quad \textbf{[2]}$$

where Q_d (L s^{-1}) is the drain flow from the membrane system. Finally, equation 3 calculated the membrane rejection as follows:

$$\textbf{Membrane Rejection } (\%) = \left(\mathbf{1 - \frac{C_p}{C_m}}\right) \times \mathbf{100} \quad \textbf{[3]}$$

The higher concentration of the membrane feed can simulate the last element within the multi stage plug-flow membrane configuration typically employed in a full-scale system. On the other hand, the higher fluxes tested for the NF membrane give an indication of the high fluxes expected in the first membranes of a pressure vessel. No anti-scalants or biocides where used in the pre-treatment of the MBR effluent before being fed to the RO or NF membranes, which led to membrane fouling over the experimental period (7000 L m^{-2} of permeate produced). The data acquisition system allowed for the continuous monitoring and control of online flow rates, conductivities, temperature, pH, redox potential (ORP), trans-membrane pressure, air scouring frequency and dissolved oxygen.

2.2. Sampling and sample preservation

Samples were taken during two consecutive years (2013 and 2014) in the WWTP of Santa Cristina d'Aro (Spain). The actual influent to the pilot plant was derived from the main sewer influent to the WWTP from the town of Santa Cristina d'Aro (4600 inhabitants). Sampling was carried out during four weeks in each campaign during the dry weather flow to avoid the dilution of the influent due to a combined sewer system in the town (between April-May 2013 and in May 2014, respectively). Grab samples of MBR influent, MBR effluent/RO feed, RO permeate and RO concentrate were taken twice per week during both experimental periods (four weeks each), always considering a temporal shift between sampling the influent and effluent of the MBR equal to the hydraulic retention time (HRT) of the reactor. Additionally, during the second year, integrated samples were taken from the larger WWTP influent sewer (after a coarse screen) every two hours to obtain the range and variation of concentrations of the target compounds during a 24-hour period.

Water samples were collected in 1 L amber PET bottles (pre-rinsed with Milli-Q water), and filtered on-site through a 1.2 μm microfiber filter followed by a 0.45μm nylon membrane filter. They were stored at 4ºC during transportation and frozen upon arrival to the laboratory.

2.3. Chemicals and reagents

HPLC-grade solvents (water, methanol (MeOH), acetone and acetonitrile (ACN)) and formic acid (HCOOH) (98–100%) were supplied by Merck (Darmstadt, Germany) and Thermo Fisher Scientific (Thermo Scientific, Franklin, MA, US). High purity standards (>99%) of the PhACs acetaminophen (ACM), sulfamethoxazole (SMX), sulfapyridine (SPY), sulfamethazine (SMZ), sulfadiazine (SDZ), venlafaxine (VFX), diazepam (DZP), carbamazepine (CBZ), diclofenac (sodium salt) (DCF), fluoxetine (FXT), metoprolol (MTP), ranitidine (RTD) and the metabolites norverapamil (desVPM), norfluoxetine (norFXT) and 2-OH-carbamazepine (2-OH-CBZ) were purchased from Sigma-Aldrich (St. Louis, MO, USA). High purity standards for the metabolites 4-nitro-sulfamethoxazole (n-SMX), 4'-OH-diclofenac (4-

OH-DCF), diclofenac amide (adDCF), acridone (ACRO), D,L-N-desmethylvenlafaxine (N-desVFX), D,L-O-desmethylvenlafaxine (O-desVFX), N^4-acetylsulfapyridine (acSPY), N^4-acetylsulfamethazine (acSMZ), N^4-acetylsulfamethoxazole (acSMX), nordiazepam (norDZP), 3-OH-acetaminophen (3-OH-ACM), 10,11-epoxy carbamazepine (epo-CBZ), metoprolol acid (MTPA), α-OH-metoprolol (α-OH-HMTP) and O-desmethylmetoprolol (O-DMTP) were purchased from Toronto Research Chemicals (TRC Inc., Ontario, Canada). Verapamil (VPM) was obtained from the European Pharmacopoeia (EP). Isotopically labelled compounds, used as internal standards were purchased from Sigma-Aldrich (fluoxetine-d_5), TRC (verapamil-d_6, diclofenac-d_4, 4'-OH-diclofenac-d_4, sulfamethoxazole-d_4, N^4-acetylsulfapyridine-d_4, N,L-O-desmethylvenlafaxine-d_4 and acetaminophen-d_4), Cerilliant (Texas, U.S.A.) (diazepam-d_5) and from CDN isotopes (Quebec, Canada) (carbamazepine-d_{10} and venlafaxine-d_6). Stock standard solutions for each of the analytes were prepared in MeOH at 1 mg mL^{-1} and stored in the dark at −2 ◦C. Standard solutions of the mixtures of all compounds were made at appropriate concentrations and used to prepare the aqueous calibration curve and also to perform the recovery studies. Stock standard solutions for the internal standards were prepared similarly. Aqueous standard solutions always contained <0.1% of MeOH.

2.4. UHPLC-MS^2 analysis

Analysis of the PhACs was performed adapting the methodology by García-Galán et al. [3]. Briefly, fully automated on-line pre-concentration of samples, aqueous standards and operational blanks was performed using a Thermo Scientific EQuan™ system (Thermo Scientific, Franklin, MA, US), by means of two LC columns, the first for pre-concentration of the sample and the second for chromatographic separation. The flow rate for the chromatographic separation was set to 500 µL min^{-1}, being eluent A ACN, and B UPLC grade water slightly acidified with 0.01% HCOOH. MS/MS analysis were carried out on a TSQ Vantage triple quadrupole mass spectrometer (Thermo Scientific, Franklin, MA, US), equipped with an electrospray (ESI) turbo spray ionization source. The target compounds were analysed in both positive and negative ionization mode simultaneously (polarity switch mode). For increased sensitivity, two

selected reaction monitoring (SRM) transitions per compound were monitored, one for quantitation and the other for positive confirmation. To improve the performance of the triple quadrupole, time-specific SRM windows were adjusted to the retention times of each target compound. A complete list of the target PhACs and their main human metabolites and TPs are given in Table 1. Their physico-chemical properties are given in Table S2 of S.I.

2.5. Other analyses

All samples were analysed according to standard methods [21] for conventional physical and chemical parameters: total suspended solids and volatile suspended solids (TSS & VSS; APHA standard method 2540D), total and soluble chemical oxygen demand (COD; APHA standard method 5220B), dissolved organic carbon (TOC; Shimadzu TOC-V_{CSH} analyzer), ammonium (BÚCHI B-324 distiller, Titrino 719S Methrohm) and total Kjeldahl nitrogen (BÚCHI B-324 distiller, Titrino 719S Methrohm). Nitrites (NO_2^--N), nitrates (NO_3^--N) and phosphates (PO_3^{4-}-P), together with other major ions (Na^+, K^+, Mg^{2+}, Ca^{2+}, Cl^-) were analysed using ion chromatography (Metrohm 761-Compact; APHA standard method 4110B).

3 Results

3.1 MBR performance

The pilot plant was operating in a similar configuration over both experimental periods, maintaining an SRT of 30 days and a flux of 19 L m^{-2} h^{-1} (see Table S1 in S.I.). Dalmau et al. [22] had previously investigated the effect of aeration on biological and filtration performance of the same MBR pilot plant. These authors focused in the modification of the dissolved oxygen (DO) set-point in the aerobic compartment followed by modifications of the air-scour flow rate in the membrane tank. They found that the optimal value was a DO set-point of 0.5 mg O_2 L^{-1} in the aerobic tank and that the activity of nitrifiers was significantly reduced when the DO set-point decreased to lower values. For this reason,

the DO set-point in the aerobic tank was maintained at 0.5 mg O_2 L^{-1}, whereas the air scour flow rate was kept at 10 m^3 h^{-1} to ensure that fouling was not an issue (transmembrane pressure < 30 mbar). The results given in Table 2 show that during both years, the pilot plant was working under nitrifying conditions, consistently reducing NH_4^+ to values < 1 mg L^{-1}. On the contrary, denitrification was very low, particularly during the 2014 campaign. The anaerobic conditions in the first tank of UCT configuration enhanced the development of phosphate accumulating organisms (PAOs), responsible of enhanced biological phosphorous removal (EBPR). The increment of phosphorous removal efficiencies from 42% to 70% is attributed to the increment of the influent COD, one of the limiting factors in EBPR [20] and that was three times higher in 2014 than 2013.

3.2 Occurrence of PhACs in raw urban wastewater and influent

3.2.1 Sewer wastewater and primary treatment

Sewer samples were taken only during the campaign of 2014 (during 24 h, at 2h intervals). During this campaign, 2 of the studied compounds were detected at average concentrations > 1 µg L^{-1} in these samples: one metabolite, MTPA (1.4 µg L^{-1}) and the anti-inflammatory ACM (30.3 µg L^{-1}). Similar concentrations were found in influent samples in the same campaign (see Figure 2). The yearly consumption of ACM is estimated to be 700-1400 tons per year in Spain [23]. ACM is usually amongst the PhACs detected at highest levels, together with other anti-inflammatories such as ibuprofen[5,24]. Variations in the levels of the PhACs studied before and after primary sedimentation (comparison between sewer samples and influent samples, see Figure 1a) ranged from barely no change for the metabolites norDZP (0.19%) and O-desVFX (2.5%), to a reduction of more than 50% of the initial concentration for acSMX (50.36%) (the reduction for the parent compound, SMX, was 47%), DZP (51.2%) and FXT (56.2%). On the contrary, norFXT increased slightly (10.5%) from the sewer to the influent of the WWTP. However, a mass balance with FXT would not be accurate as its concentration was much lower than the metabolite's. DCF concentration remained quite the same during this primary

treatment (increase of 6.6%), whereas its hydroxylated metabolite doubled its concentration during this primary treatment, going from an average value of 452 ng L^{-1} to 904 ng L^{-1}. It is known that human metabolism of DCF yields a 50% of the total dose as 4-OH-DCF and its glucuronide conjugate [25], which could have deconjugated during this first phase of the treatment and reverted to 4-OH-DCF, contributing to the observed increase. This phenomenon was observed for SPY and its acetylated metabolite, acSPY. In this case, acSPY concentration decreased a 12.5% during primary treatment whereas that of SPY increased approximately in the same proportion (10%). A deconjugation of acSPY during this primary treatment could justify these results, as demonstrated in a previous publication by Garcia-Galan et al., who observed the back-transformation of at least a 50% of the initial concentration of the acetylated metabolite in the parent compound in a simulated fixed-bed bioreactor during several days[26].

Figure 3 shows a 24-hour profile of DCF, CBZ, VFX and MTP together with their human metabolites and TPs in the sewer. For all of them, a similar evolution through time is observed for parents and products. Table S3 in SI shows the concentrations measured in the sewer samples during 2014.

3.2.2 Influent wastewater

In 2013, the highest concentrations detected in the influent samples corresponded to the same compounds as in 2014 but at lower concentrations. This could be due, amongst other reasons, to strong rain events registered during April 2013 on the same week that sampling began, and also to a higher precipitation average during the sampling period in that year. As mentioned in the previous section, ACM was detected at the highest concentrations in both campaigns (see Table S4 in S.I.).

Regarding MTP and its metabolites, concentrations found for MTPA (0.6-1.1 μg L^{-1}) were higher than those published by Rubirola et al. [27] who detected levels ranging between 119 ng L^{-1} and 298 ng L^{-1} in influent wastewaters of two WWTPs in Catalonia (Spain). Another publication made an estimation of

the presence of this metabolite in WWTP influents at concentrations around 2 μg L^{-1} [28]. The MTPA/MTP ratios in the influent ranged between 60-80, and are in accordance with the metabolic excretion rates of these compounds, as MTP is excreted in a 60-65% as MTPA, and only a 3-10% is excreted in its original form [28]. Table 3 shows the concentration ratios between the parent drugs and their metabolites along the different treatment steps. As observed, the ratios are generally maintained between the sewer samples and the influent samples for all the target compounds.

3.3 Removal of PhACs by MBR

Out of the 13 parent compounds and 20 metabolites and TPs analysed, 10 parent compounds and 8 of their metabolites were observed at concentrations ten-times greater than their corresponding limit of detection (LOD) in the MBR effluent samples (LODs are given in Table S5 in S.I.). Due to the high number of studied compounds, concentrations below this threshold are not discussed in detail in this section.

Despite being the compound detected at the highest level in the influent samples, ACM was completely eliminated during the MBR treatment in both campaigns (see Figure 4). As they are hydrophilic compounds, adsorption to biomass is usually negligible and biodegradation should be considered the main removal mechanism [29]. On the contrary, concentrations of MTPA in the MBR effluents were higher than those detected in the influent in most cases, with average negative elimination rates of -52% and -42% in 2013 and 2014, respectively. These results could indicate the formation of this compound as biodegradation product of the parent compound MTP; however, as shown in Table 3, MTPA/MTP ratios are maintained in influent and effluent samples (being only slightly higher in the effluent for the samples of 2013), indicating that MTP is also not eliminated during MBR treatment and, similarly to MTPA, concentrations were usually higher in the effluent samples, with average negative elimination rates of -38% and -45% (2013 and 2014 respectively, see Figure 4). Rubirola et al. reported a similar behaviour for MTPA and observed a concentration for MTPA 10 times higher in effluent than in

the influent wastewater in a conventional WWTP. The authors attributed these high concentrations to the generation of MTPA from atenolol, the major β-blocker present in influent wastewaters (up to 2 orders of magnitude higher than MTP) [27]. Indeed, Radjenovic et al. [30] demonstrated in a previous study that MTPA was also a primary degradation product for atenolol in MBR-sludge batch experiments, in which MTPA was detected simultaneously to the immediate degradation of atenolol and reached a 40% of the initial spiked concentration of atenolol after only 1 day. Despite it was out of the scope of this work, atenolol is frequently detected in MBR influent wastewaters[5,24]. Therefore, the higher concentrations of MTPA in the MBR effluent could be attributed to atenolol degradation during treatment.

VFX and their two metabolites also showed negative removals in both campaigns, with higher concentrations after MBR treatment. These results are in accordance with previous studies in which negative elimination rates were obtained after CAS treatment [31,32]. The limited sorption to the biomass of these compounds (low solid-water distribution coefficient (K_d)) and their resilience to biodegradation could explain partly this poor removal. This could also explain the results obtained for DZP and norDZP, with very low or negative elimination rates for both analytes in both campaigns. The same applies for the very low removals observed for CBZ or DCF; in this case, as mentioned in section 3.2.1., it is usually attributed not only to low biodegradability and low K_d values, but also to deconjugation of human phase II metabolites (that may not have been included in the monitoring), parent compound formation by enzymatic reactions from phase I metabolites or desorption from particulate matter [33]. For instance, removal efficiencies of DCF varied from negative values to an elimination of 76%. As mentioned in section 3.2, the SRT during both campaigns was 30 days. According to Clara et al. [34], SRTs > 10 days enhance the biodegradation of PhACs such as DCF, whereas for CBZ there would not be any significant improvement. Indeed, for CBZ only negative removal values were obtained in both campaigns. In the present study, metabolites of CBZ also presented negative elimination rates (data from 2013) and therefore no correspondence could be established. Low removals of CBZ have been frequently

reported by many researchers, and it is usually considered as a non-degradable, recalcitrant compound [35].

The acetylated metabolite of SMX was fully eliminated after MBR treatment in both campaigns. On the contrary, the concentration of SMX was only partially reduced, with removal rates ranging from 57-70% in 2013 and 60-84% in 2014. Xia et al. [36] published similar removal values for SMX in an MBR but demonstrated that longer SRTs resulted in a better efficiency. Trinh et al. [37] estimated a removal of 62% for SMX during MBR treatment (SRT of 15 d) and attributed a 59% of it to biotransformation, neglecting the possibility of adsorption to biomass. Other publications also highlighted biotransformation as the main removal mechanism for SMX [36,38]. Considering these previous results, despite the biodegradation of SMX during MBR, acSMX does not seems to be a potential biodegradation by-product of SMX, as it is not detected in the MBR effluent. A rapid hydrolysis and degradation of the metabolite itself could account for the incomplete removal of SMX in case it reverted back into the parent compound, as demonstrated for SPY[26]. The high removal efficiencies obtained in this work can be attributed to the high SRT and the mixed liquor suspended solids (MLSS) concentrations in MBRs.

3.4 Removal of PhACs by RO and NF

Water from the MBR system provided feed water with a Silt Density Index (SDI) above 5 (15 minutes) consistently, making the water suitable for treatment by spiral wound RO and NF membrane processes in terms of particulate and colloidal matter. During the first 7 days of operation, in which PhACs monitoring took place, the RO membrane required a feed pressure of 6.5 bar, whereas the NF membrane required nearly half the operating feed pressure at 3.3 bar to produce a similar average permeate flux of 18 L m^{-2} h^{-1} at 75% recovery. Table 2 shows the performance of the RO and NF membranes in terms of conventional water quality parameters showing that on average, while the RO membrane had a rejection of 97% of the total dissolved solids (measured as electrical conductivity), whereas the NF membrane yielded a rejection of 92%.

As mentioned in section 3.3, the main removal pathways of the target compounds during MBR treatment are biodegradation and their adsorption into the sludge, whereas their rejection through the submerged membranes can be considered negligible [39]. The fate of those compounds that were not fully removed by the MBR process was then investigated as they passed through RO and NF membranes separately. The rejection results for the RO and NF membranes are given in Figure 5. The RO process resulted in a removal > 99%, for all the compounds evaluated. The few compounds which were still detected in the RO permeate at concentrations ten-times greater than their LOD (see Table S5 in S.I) were O-desVFX (1.59±0.96 ng L^{-1}) which was detected in five out of the seven samples, α-OH-MTP (3.10 ± 3.12 ng L^{-1}) which was detected in four out of the seven samples, MTPA detected in two samples (5.39 ± 8.85 ng L^{-1}), epo-CBZ (0.93 ng L^{-1}), acSPY (1.5 ng L^{-1}) and SMX (5.47 ng L^{-1}) which were each detected in one sample. ACRI (1.35 ng L^{-1}) and RTD (0.02 ng L^{-1}) were also detected but below ten-times their LOD.

The NF membrane showed high removal efficiencies (>90%) for most of the studied compounds, although as expected, these removals were lower than those obtained with the RO membrane. For instance, at a similar flux, O-desVFX was found at 36 ng L^{-1} in the permeate of the NF membrane, a concentration significantly higher than that of the RO permeate. A number of compounds and their metabolites were also detected in the permeate of the NF membrane while being completely removed by the RO membrane (see Table S6 in S.I.).On the contrary, α-OH-MTP, which was also detected at low concentrations in the RO permeate was consistently well below LOD in the NF permeate samples.

As mentioned earlier in the paper, the MWCO for the ESPA2 RO is less than 100 Daltons, whereas that of the NF membrane is approximately 200 Daltons [7]. The compounds with the lowest MW amongst those compounds included in this study were ACM and its metabolite 3-OH-ACM (151 g mol^{-1} and 167 g mol^{-1} respectively) as well as ACRI (179.1 g mol^{-1}) and ACRO (195.2 g mol^{-1}). However, the benefit of integrating MBR with a RO process couldn´t be demonstrated in our study for these particular compounds, as all had been completely removed during MBR treatment. Radjenovic et al. [11]

investigated the rejection of ACM in a full-scale drinking water treatment plant comparing RO (BW30LE-440) and the same NF membrane used in this study (NF90). The results showed that the RO membrane had a rejection of 85.6% for ACM, whereas the NF90 membrane had a lower rejection for ACM (44.6%).

Various studies have shown that there are a number of processes which affect the removal of solutes by membranes including membrane properties, the chemistry of the solution and the physicochemical properties of the contaminants [12,40]. Although separation of trace contaminants by membrane processes is primarily based on size exclusion, net sorption at the membrane-solute interface plays an important role. Various studies have shown that size exclusion is responsible for rejection of uncharged and hydrophilic substances while electrostatic interactions between charged solutes and negatively charged membranes have an important role for charged solutes [7,12].

It is quite clear that the negatively charged RO and NF membranes used in this work had a significant effect on the lower rejection of positively charged compounds, since the few compounds detected above the LOQ in the permeate stream for both membranes were positively charged, including VFX, FXT, MTP and RTD.

When comparing the results to those of Radjenovic et al. [11], we observe that they obtained similar results for SMX and DCF (rejection >99%) working with the NF90 membrane, but also a higher rejection for CBZ (99.7%, at a flux of 22.9 L $m^{-2}h^{-1}$), compared to the results obtained in this study, of only 78% at a flux of 18 L $m^{-2}h^{-1}$ and 89% at a flux of 26 L $m^{-2}h^{-1}$.

A comparison of the capital and operating costs of RO and NF processes was carried out in previous studies. Yangali-Quintanilla et al. [19]showed that using a NF90 membrane a cost saving of 0.07$ m^{-3} (12%) saving could be made when compared to using an RO membrane. As demonstrated in the present study, when the feed pressure of the NF membrane was roughly half of that required to produce the same amount of water with the RO membrane, energy savings made up for a significant part of the savings. Besides, the reduced chemical pre-treatment required to produce the same amount of water

by these membranes also results in a relevant reduction of the costs. In terms of capital, the costs of the installation of a NF system when compared to that of a RO system are lower, specifically in the high pressure loop due to this reduced pressure requirement. Bellona et al. [41]showed similar cost savings in terms of electrical consumption during production and chemicals used for pH adjustment, as well as reduced costs in terms of membrane cleaning (half the cleanings per year, implying less system downtime).

When comparing RO/NF based treatment schemes to non-RO/NF based treatment schemes (which typically involves ozone and biologically activated carbon (BAC) processes), the capital and operational costs of non-RO/NF schemes are significantly lower. In terms of PhAC removal, although RO/NF treatment schemes are higher, treatment processes with ozone and BAC have proved to be more efficient [42] and at a lower cost. Plumlee et al. [43] showed that the total capital cost for a 1 MGD (Million Gallon Per Day) ozone-BAC facility was less than half the price of MF-RO facility, this difference increasing with the capacity of the plant. The operational cost for a 1 MGD ozone-BAC treatment facility was roughly 15% that of a MF-RO facility and, although these prices are specific for the feed water being treated and the application of the effluent following treatment, they give a good indication of the difference in costs between RO/NF and non-RO/NF based treatment schemes. In practice, RO/NF based treatment schemes are used only when the water reuse treatment requires a degree of demineralisation.

3.5 Effect of average permeate flux on PhAC removal

As the flux of the NF90 membrane was increased from a relatively conservative flux of 18 L $m^{-2}h^{-1}$ to an average permeate flux of 26 and 30 L $m^{-2}h^{-1}$ (feed pressure increasing from 3.3 bar to 4.5 and 5.7 bar respectively)the rejection of the studied PhACs, metabolites and TPs also increased. Higher rejection at higher fluxes occurs due to the increased dilution of the constituents of interest within the permeate channel, as more water passed through the membrane. This increase in the rejection as the flux increases is typically observed in RO systems when measuring the concentrations of ionic constituents

(Cl^-, NO_3^-, PO_4^{3-}, SO_4^{2-}, Na^+, K^+, Mg^{2+}, Ca^{2+}) and electrical conductivity. Figure 6 shows the effect on the rejection of the studied compounds when the average permeate flux for the NF membrane was increased from 18 L $m^{-2}h^{-1}$ to 26 L $m^{-2}h^{-1}$ and 30 L $m^{-2}h^{-1}$. The compounds which had rejections lower than 90% at an average permeate flux of 18 L $m^{-2}h^{-1}$, such as CBZ and MTP, increased these rates from 78% to 89% (at 26 L $m^{-2}h^{-1}$) and 97% (at 30 L $m^{-2}h^{-1}$) in the case of CBZ, and from 57% to 63% and 73 % for MTP. Compounds which had moderate rejection rates above 90%, such as VFX, N-desVFX or O-desVFX, increased to high rejection rates (>97%); as expected, those compounds which had high rejection rates (>97%) at the lower flux of 18 L $m^{-2}h^{-1}$ increased to near complete rejection; these include SPY (99% to 100%), SMX (99% to 100%), FXT (96 to 100%), 4-OH-DCF (96% to 99 and 100%), α-OH-MTP (97% to 100%) and VPM (98% to 100%).

Operating the system at fluxes above the typical 17-21 LMH range may not be sustainable over the long term depending on the feed water quality. Even in this narrow range the difference in rejection would be noticeable. Therefore, the flux is a very important parameter when reporting rejection of micropollutants. Operationally, a flux of 18 LMH is conservative and would require minimal chemical addition in the pre-treatment of the RO/NF system and minimal cleaning. As the flux increases to 26 LMH, still within a realistic operational range, pH adjustment of the feed water with H_2SO_4 is required and it would increase the operational costs significantly, due both to the dose and the high price of the acid. Bellona et al. [41] showed that this cost increase due to H_2SO_4 addition, when moving from 17 LMH to 25 LMH, went from 0.03 \$ m^{-3} produced to 0.11 \$ m^{-3}. In operational terms, increasing the flux beyond 26 LMH would increase membrane cleaning and replacement as well as pre-treatment expenses significantly to the point that it may not be feasible.

3.6 Concentration of PhACs in the concentrate stream

The concentrations of the PhACs, metabolites and TPs in the concentrate stream are interesting from two different points of view. The first is the recognition that, although the concentrate stream may

meet discharge limits for conventional regulated parameters such as nutrients (if a biological nutrient removal process is included and it is functioning correctly), the concentrate stream has high concentrations of the target compounds compared to those of the final effluents and even influent of the MBR process. As also noted in the work by Rodríguez-Mozaz et al. [44], the concentrated effluent poses a serious threat to aquatic ecosystems if discharged to water bodies. A number of options are discussed in the review by Pérez-González et al. [45] to reduce the organic pollutant load in this concentrated effluent prior to the release into the environment, by advanced oxidation processes (AOPs) such as ozonation, fenton processes, photocatalysis and photo-oxidation, sonolysis and electrochemical oxidation. The second aspect is that, since this is a concentrated stream, we can identify compounds which had been removed by the MBR treatment to levels below the method LODs for the MBR effluent stream, but which had not actually been fully removed.

The compounds with concentrations > 500 ng L^{-1} in the concentrate stream during the RO and NF sampling campaigns included VFX and its metabolite O-desVFX, norFXT, DCF and 4-OH-DCF and MTPA (see Figure S1 and Table S6 in S.I.).

As expected, for those compounds which had similar concentrations in the RO/NF feed, the concentration of the RO concentrate was higher than that measured in the NF concentrate, due to the higher rejections by the RO membranes. This could be demonstrated by the concentration factor (CF), which is the ratio of the concentration in the membrane concentrate to that in the feed water. In theory, assuming perfect removal of solutes by a membrane, a system recovery of 75% should yield a CF of 4 (1/ (1-0.75)). In our case, the CF depended on the rejection of the particular compound by each of the membranes. For instance, the CF for the RO membrane (the membrane having a rejection closer to 100%) was usually higher than the theoretical value of 4 because concentrate recirculation was employed to comply with the membrane manufacturer operating specifications, as explained in section 2.1. The CFs for the compounds found to have the highest concentrations in the RO & NF concentrate were: VFX: 3.7 to 2.5, O-desVFX: 4.3 to 2.6, DCF: 4.5 to 4.4, 4-OH-DCF: 1 to 2.3, MTPA: 1.6 to 0.5. The CF for norFXT for

the RO membrane could not be calculated since the concentration of the MBR effluent was < LOD, but a concentration of 3046 ng L^{-1} was measured in the concentrate.

4 Conclusion

This work allowed us to compare the removal of selected PhACs, metabolites and their TPs through MBR-RO and MBR-NF processes with real wastewater to understand the extent of the difference in the rejection of such compounds when using low pressure NF membranes in the place of RO membranes. An automated analytical methodology based on LC-LC-MS/MS was used to quantify the concentration of 14 parent compounds and 22 of their metabolites from the sewer to primary treatment, through to the MBR and finally through the RO or NF membranes. The results showed that operating withboth membrane processes consecutively became a highly efficient process to remove all the studied compounds, generating a highly concentrate stream which would require further treatment before being discharged into the natural environment. When comparing the removal efficiencies of the RO and NF membranes, the RO membrane showed near complete removals (>99%) of all the compounds over various process conditions, whereas the NF membrane resulted also in high removal efficiencies but to a lesser extent (> 90%). The choice between using a RO versus a tight NF membrane very much depends on the reuse application and the discharge limits set. The membrane type (molecular weight cut-off and surface morphology), RO/NF process parameters (membrane recovery and average permeate flux) and membrane condition will have a significant effect on the removal of PhACs. Therefore, and despite that the results in this paper are indicative of practical removals expected by the two different types of membranes, the parameters mentioned above will in reality all affect the removal efficiency of a given process. In terms of PhACs and their metabolites and TPs, for instance, if concentrations below 20 ng L^{-1} are acceptable in the permeate water, then using tight NF membranes similar to those utilized in this study would reduce the pumping power required to obtain the same amount of water by nearly half. If lower concentrations are required (i.e. below 5 ng L^{-1}), then a RO membrane will be required. Increasing the flux of the NF

membrane will reduce the concentrations close to those obtained by the RO membrane, but would then require a similar pumping power as the RO membrane and probably suffer in terms of increased membrane fouling and would therefore make sense to keep using an RO membrane at a conservative flux.

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Figure captions:

Figure 1: (a) Scheme of the MBR-RO/NF pilot plant showing the sampling points: (1) sewer, (2) influent tank, (3) MBR permeate tank, (4) RO/NF feed, (5) RO/NF permeate and (6) RO/NF concentrate and (b) detailed scheme of the membrane system showing the inlet flow (Q) and concentration (C) of the system feed (f), membrane feed (m), permeate (p), concentrate (c), recirculation (r) and drain (d).

Figure 2: Measured concentrations (ng L^{-1}) of the studied compounds in WWTP inlet (sewer) and following primary treatment (influent) of the sampling campaign in 2014. Gaps in the figure show that the analyte was not detected.

Figure 3: Concentration profile of diclofenac, carbamazepine, venlafaxine and metroprolol and their corresponding metabolites in the sewer samples over a 24-hour period, and evolution of the chemical oxygen demand (COD) and total Kjeldahl Nitrogen (TKNT) during the same period.

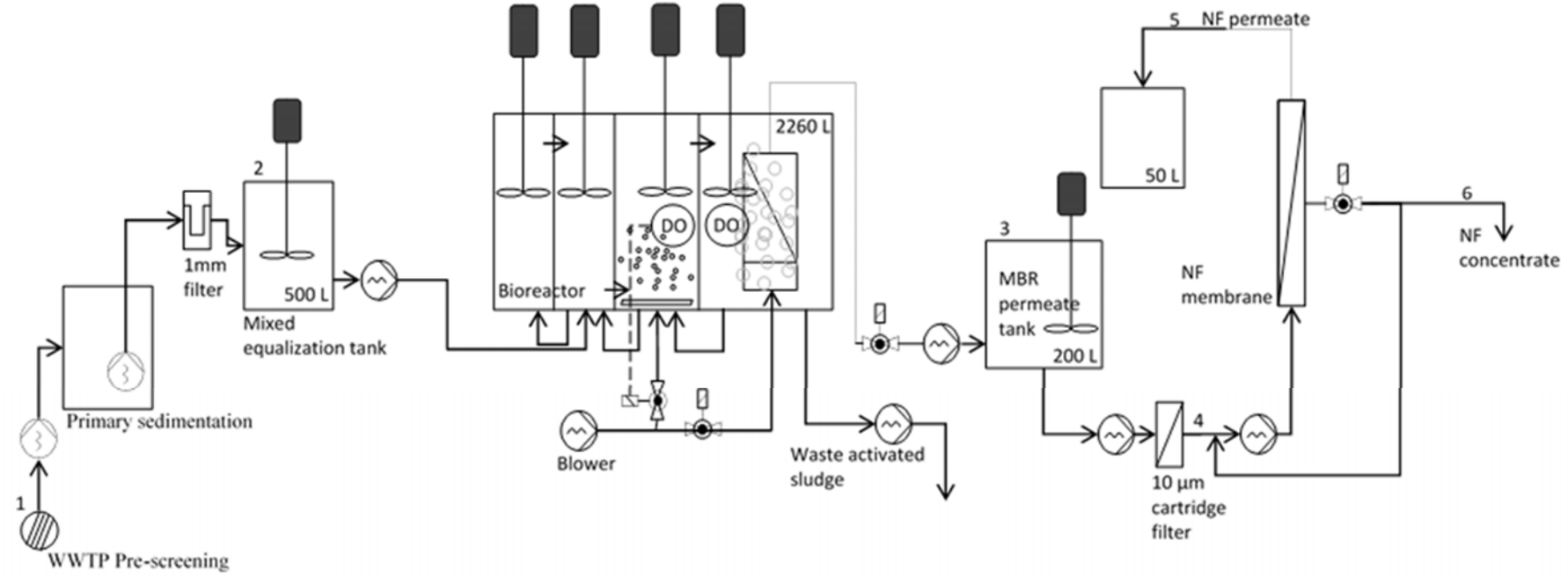
Figure 4: Removal (%) of the studied PhACs, metabolites and TPs during treatment in the MBR.

Figure 5: Removal (%) of the studied PhACs, metabolites and TPs by RO and NF membranes. Gaps in the figure show that the analyte was not detected.

Figure 6: Removal (%) of the studied PhACs, metabolites and TPs by a NF membrane at different average permeate fluxes. Gaps in the figure show that the analyte was not detected.

Figure 1

a.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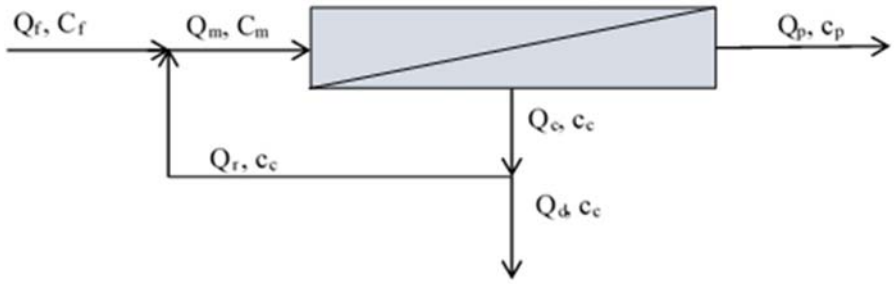
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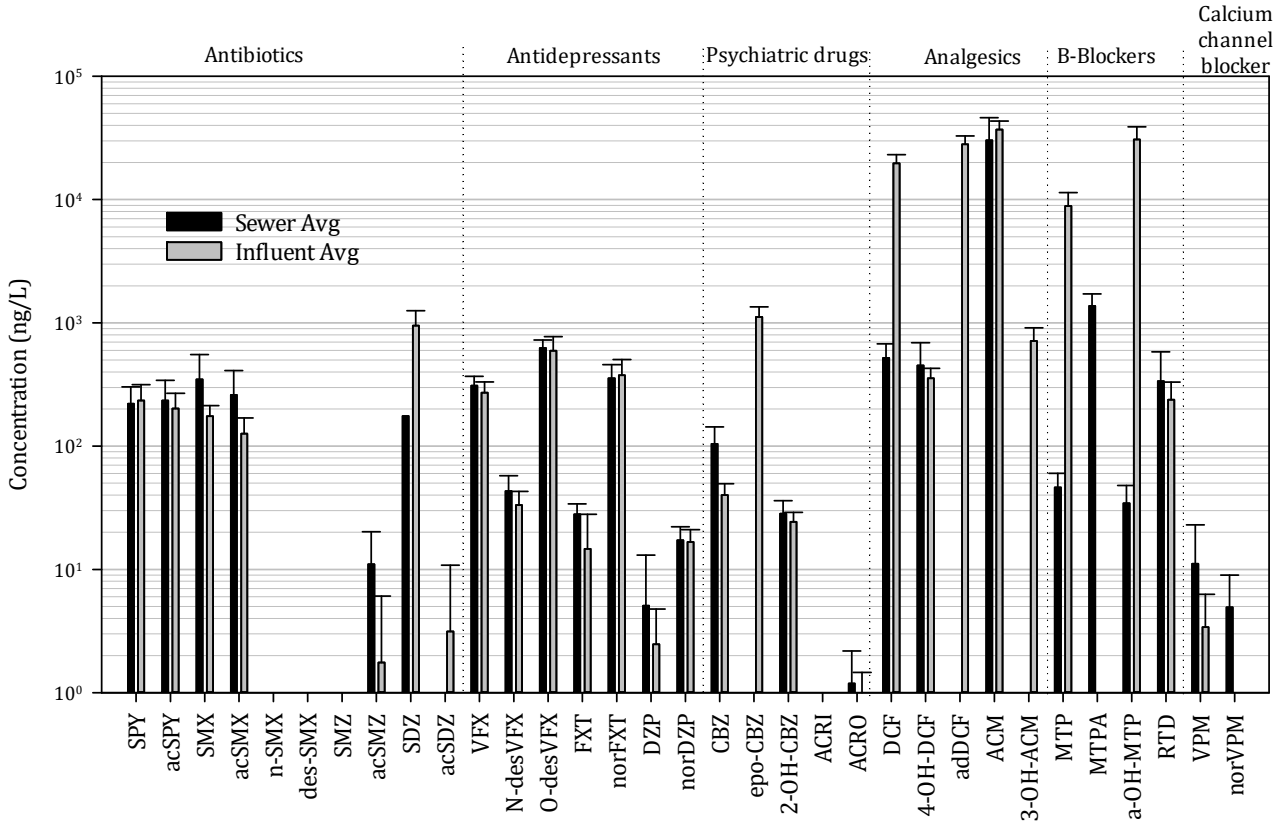
Figure 2:


Antibiotics
Antidepressants
Psychiatric drugs
Analgesics
B-Blockers
Calcium channel blocker
Sewer Avg
Influent Avg
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SMX
acSMX
n-SMX
des-SMX
SMZ
acSMZ
SDZ
acSDZ
VFX
N-desVFX
O-desVFX
FXT
norFXT
DZP
norDZP
CBZ
epo-CBZ
2-OH-CBZ
ACRI
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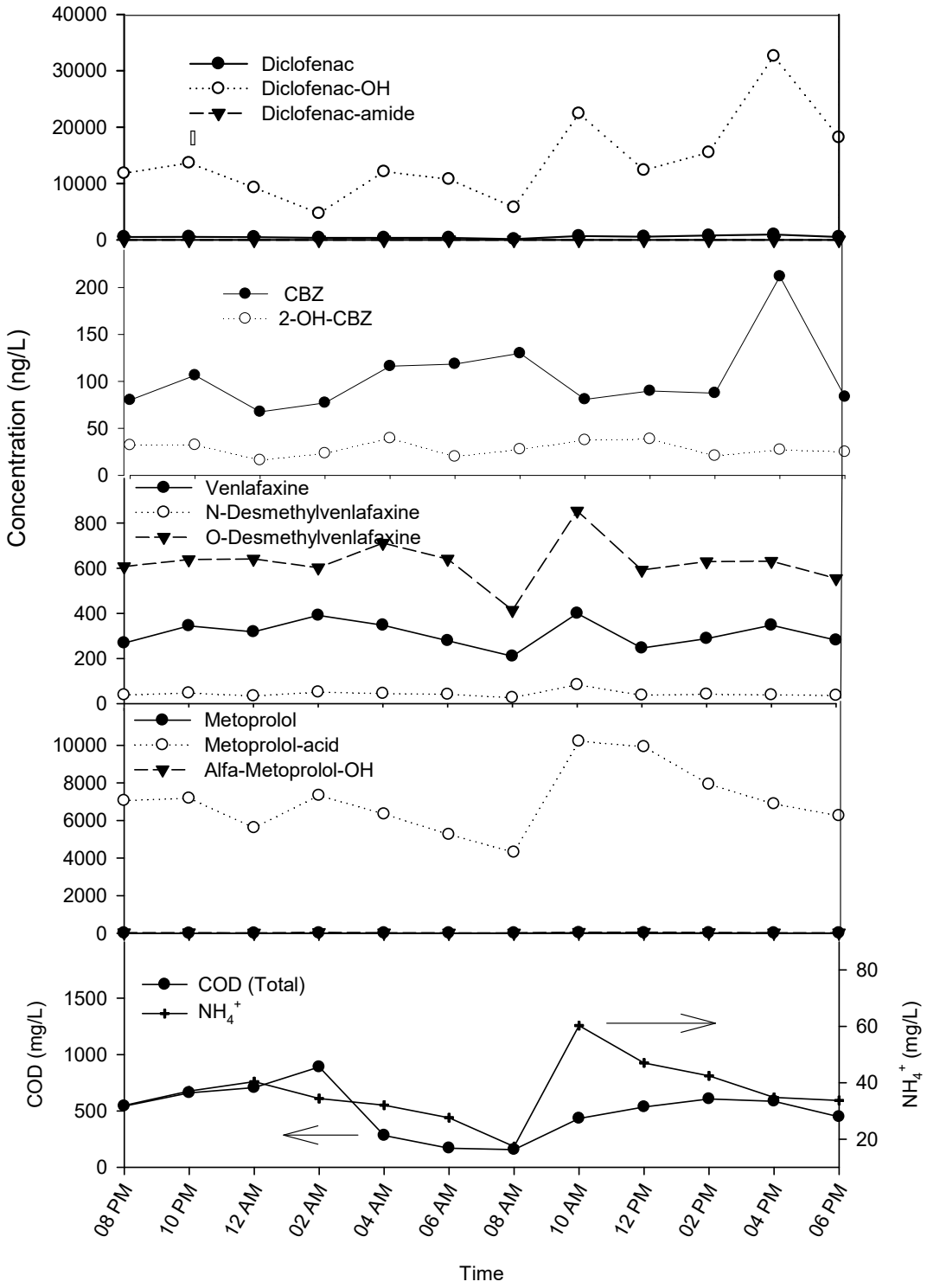
DCF
4-OH-DCF
adDCF
ACM
3-OH-ACM
MTP
MTPA
a-OH-MTP
RTD
VPM
norVPM

Figure 3: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Diclofenac-OH
Diclofenac-amide
CBZ
2-OH-CBZ
Venlafaxine
N-Desmethylvenlafaxine
O-Desmethylvenlafaxine
Metoprolol
Metoprolol-acid
Alfa-Metoprolol-OH
COD (Total)
NH4+
Concentration (ng/L)
COD (mg/L)
NH4+ (mg/L)
Time
08 PM
10 PM
12 AM
02 AM
04 AM
06 AM
08 AM
10 AM
12 PM
02 PM
04 PM
06 PM

Figure 4

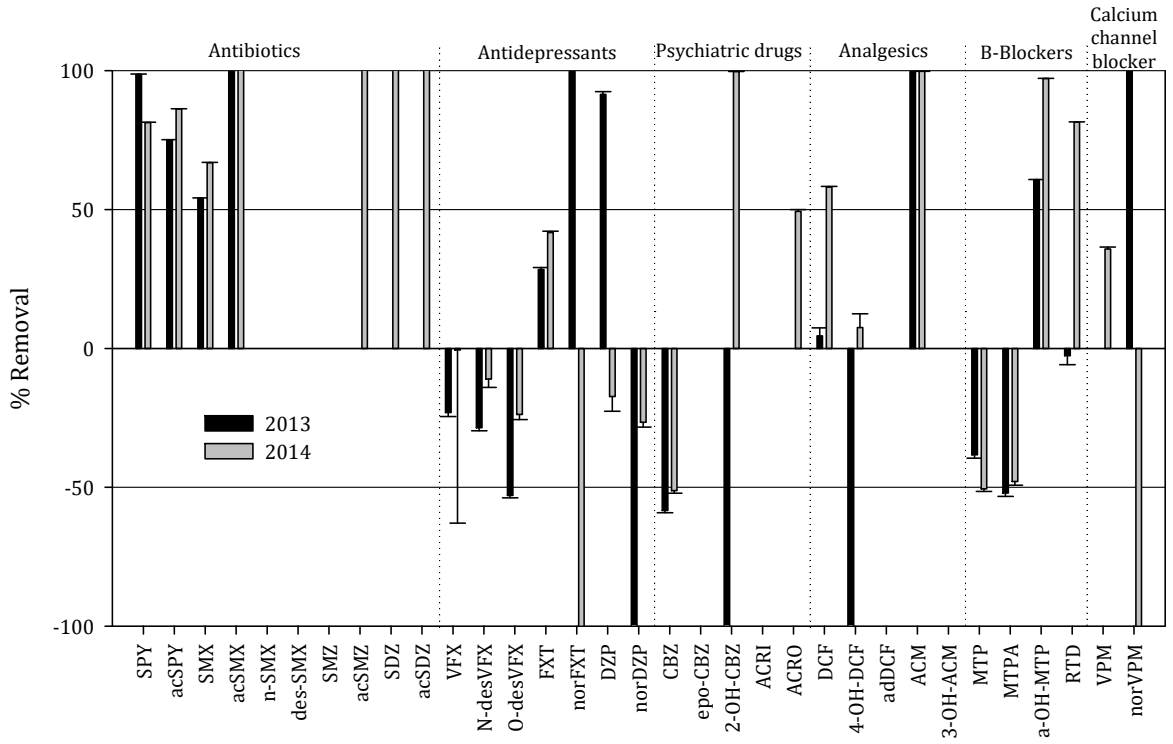
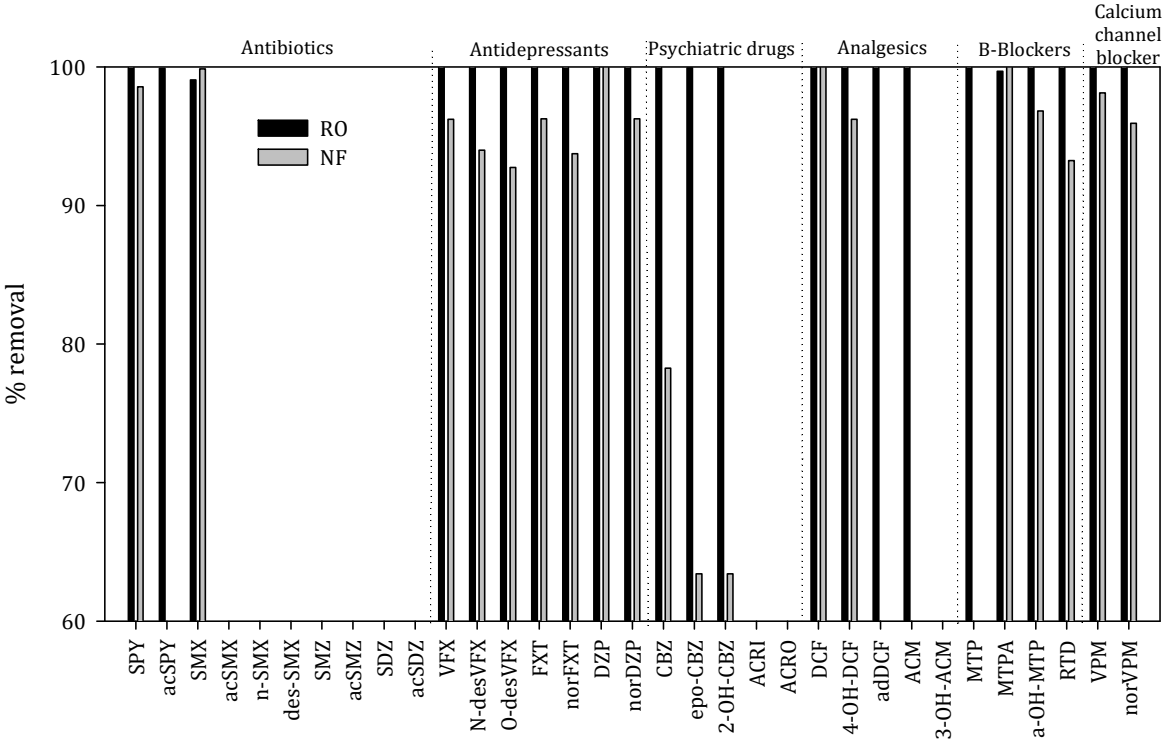

Antibiotics
Antidepressants
Psychiatric drugs
Analgesics
B-Blockers
Calcium channel blocker
% Removal
100
50
0
-50
-100
2013
2014
SPY
acSPY
SMX
acSMX
n-SMX
des-SMX
SMZ
acSMZ
SDZ
acSDZ
VFX
N-desVFX
O-desVFX
FXT
norFXT
DZP
norDZP
CBZ
epo-CBZ
2-OH-CBZ
ACRI
ACRO
DCF
4-OH-DCF
adDCF
ACM
3-OH-ACM
MTP
MTPA
a-OH-MTP
RTD
VPM
norVPM

Figure 5


Antibiotics
Antidepressants
Psychiatric drugs
Analgesics
B-Blockers
Calcium channel blocker
RO
NF
% removal
100
90
80
70
60
SPY
acSPY
SMX
acSMX
n-SMX
des-SMX
SMZ
acSMZ
SDZ
acSDZ
VFX
N-desVFX
O-desVFX
FXT
norFXT
DZP
norDZP
CBZ
epo-CBZ
2-OH-CBZ
ACRI
ACRO
DCF
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adDCF
ACM
3-OH-ACM
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MTPA
a-OH-MTP
RTD
VPM
norVPM

Figure 6

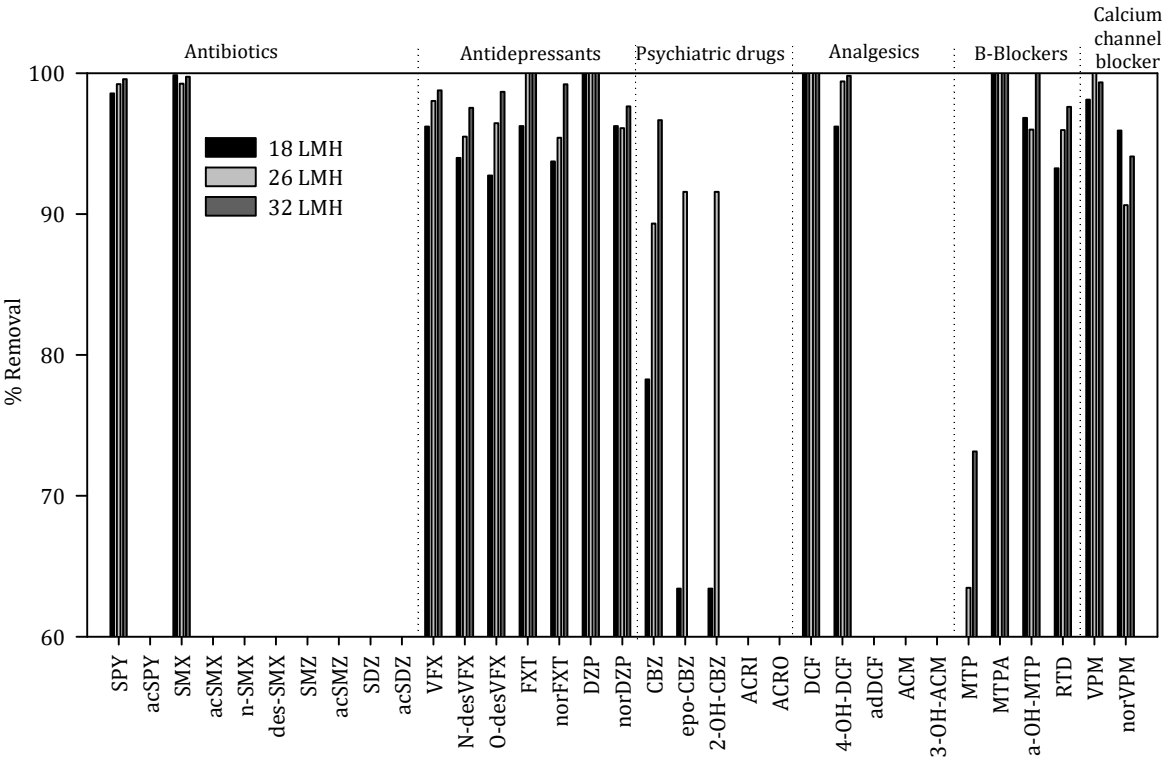
Antibiotics
Antidepressants
Psychiatric drugs
Analgesics
B-Blockers
Calcium channel blocker
18 LMH
26 LMH
32 LMH
% Removal
100
90
80
70
60
SPY
acSPY
SMX
acSMX
n-SMX
des-SMX
SMZ
acSMZ
SDZ
acSDZ
VFX
N-desVFX
O-desVFX
FXT
norFXT
DZP
norDZP
CBZ
epo-CBZ
2-OH-CBZ
ACRI
ACRO
DCF
4-OH-DCF
adDCF
ACM
3-OH-ACM
MTP
MTPA
a-OH-MTP
RTD
VPM
norVPM

Table captions:

Table 1: Pharmaceuticals (PhACs), metabolites and transformation products (TPs) investigated in this study (metabolites and TPs in italics)

Table 2: Average conventional water quality parameters showing MBR, RO and NF performance over the experimental periods

Table 3: Concentration ratios between the investigated pharmaceuticals and their main metabolites and transformation products along the wastewater treatment

Table 1

Therapeutic class	Compound		CAS number	Formula	MW (g/mol)
Antibiotics	Sulfapyridine	SPY	144-83-2	$C_{11}H_{11}N_3O_2S$	249.3
	N^4−acetylsulfapyridine	acSPY	19077-98-6	$C_{13}H_{13}N_3O_3S$	291.3
	Sulfamethoxazole	SMX	723-46-6	$C_{10}H_{11}N_3O_3S$	253.3
	N^4−acetylsulfamethoxazole	acSMX	21312-10-7	$C_{12}H_{13}N_3O_4S$	295.3
	Nitrosulfamethoxazole	n-SMX	29699-89-6	$C_{10}H_9N_3O_5S$	283.3
	Desaminosulfamethoxazole	des-SMX			238.1
	Sulfamethazine	SMZ	57-68-1	$C_{12}H_{14}N_4O_2S$	278.3
	N^4-Acetylsulfamethazine	acSMZ	100-90-3	$C_{14}H_{16}N_4O_3S$	320.4
	Sulfadiazine	SDZ	68-35-9	$C_{10}H_{10}N_4O_2S$	250.3
	N^4-Acetylsulfadiazine	acSDZ	127-74-2	$C_{12}H_{12}N_4O_3S$	292.3
Anti-depressants	Venlafaxine	VFX	93413-69-5	$C_{17}H_{27}NO_2$	277.4
	N−Desmethylvenlafaxine	N-desVFX	149289-30-5	$C_{16}H_{25}NO_2$	263.4
	O−Desmethylvenlafaxine	O-desVFX	93413-62-8	$C_{16}H_{25}NO_2$	263.4
	Fluoxetine	FXT	54910-89-3	$C_{17}H_{18}F_3NO$	309.3
	Norfluoxetine	norFXT	83891-03-6	$C_{16}H_{16}F_3NO$	295.3
	Diazepam	DZP	439-14-5	$C_{16}H_{13}ClN_2O$	284.7
	Desmethyldiazepam	norDZP	1088-11-5	$C_{15}H_{11}ClN_2O$	270.7
Psychiatric drugs	Carbamazepine	CBZ	298-46-4	$C_{15}H_{12}N_2O$	236.3
	10,11-Epoxy−Carbamazepine	epo-CBZ	36507-30-9	$C_{15}H_{12}N_2O_2$	252.3
	2−Hydroxycarbamazepine	2-OH-CBZ	68011-66-5	$C_{15}H_{12}N_2O_2$	252.3
	Acridine	ACRI	260-94-6	$C_{13}H_9N$	179.1
	Acridone	ACRO	578-95-0	$C_{13}H_9NO$	195.2
Analgesics/ anti-inflammatories	Diclofenac	DCF	15307-86-5	$C_{14}H_{11}Cl_2NO_2$	296.2
	4-OH-diclofenac	4-OH-DCF	64118-84-9	$C_{14}H_{11}Cl_2NO_3$	312.2
	Diclofenac−amide	adDCF	15362-40-0	$C_{14}H_9Cl_2NO$	278.1
	Acetaminophen	ACM	103-90-2	$C_8H_9NO_2$	151.2
	3-OH-acetaminophen	3-OH-ACM	37519-14-5	$C_8H_9NO_3$	167.2
B-Blockers	Metoprolol	MTP	37350-58-6	$C_{15}H_{25}NO_3$	267.4
	Metoprolol−acid	MTPA	56392-14-4	$C_{14}H_{21}NO_4$	267.3
	Alfa−Metoprolol−OH	α-OH-MTP	56392-16-6	$C_{15}H_{25}NO_4$	283.4
	Ranitidine	RTD	66357-35-5	$C_{13}H_{22}N_4O_3S$	314.4
Calcium channel blocker	Verapamil	VPM	52-53-9	$C_{27}H_{38}N_2O_4$	454.6
	Norverapamil	norVPM	67018-85-3	$C_{26}H_{36}N_2O_4$	440.6

Table 2

		2013			2014		
MBR Water Quality							
Parameter	Units	Influent	Effluent	Removal	Influent	Effluent	Removal
COD total	(mg/L)	188	16.6	91%	412	45	89%
COD soluble	(mg/L)	65	-		163	-	
TKN total	(mg/L)	29	1.8	94%	55	1.5	97%
NH_4^+	(mg N/L)	18	0.4	98%	34	1.0	97%
NO_3^-	(mg N/L)	0.9	4.7		0	12	
NO_2^-	(mg N/L)	0.4	0.1		0	0	
Total-N	(mg N/L)	30.3	6.6	78%	55	13.5	75%
PO_4^{3-}	(mg/L)	1.4	0.8	42%	3.6	1.1	70%
pH		7.6	-		7.5	-	
T	°C		20			22	
Turbidity	NTU		< 0.3			< 0.3	

RO and NF Water Quality							
Parameter	Units	RO feed	RO permeate	Removal efficiency	NF Feed	NF Permeate	Removal efficiency
Electrical Conductivity	(µS/cm)	909	27	97%	1016	81	92%
Temperature	°C	20.2	-		24	-	
pH		7.1	6.4		7.4	6.5	
Cl^-	(mg/L)	129	2.8	98%	156	14.1	91%
NO_3^-	(mg/L)	3.6	0.3	91%	13.4	4.4	67%
PO_4^{3-}	(mg/L)	0.2	< LOD		0.3	< LOD	
SO_4^{2-}	(mg/L)	31.8	0.06	99.8%	28.1	0.2	99.2%
Na^+	(mg/L)	112	5.1	95%	113	17.1	85%
K^+	(mg/L)	15.9	0.6	96%	20.5	2.7	87%
Mg^{2+}	(mg/L)	13.7	0.03	99.7%	17.6	0.1	99.4%
Ca^{2+}	(mg/L)	113.3	0.9	99.2%	83.8	1.5	98%

Table 3

		Sewer	MBR influent	MBR effluent	RO/NF FEED	RO/NF PERMEATE	RO/NF CONCENTRATE
acSPY/SPY	*2013*	*no data*	1.5	1.2	1.3	0	1.5
	2014	1.06	0.9	1.2	0	0	0
acSMX/SMX	*2013*	*no data*	0.5	0.3	0	0	0
	2014	0.8	0.7	0	0	0	0
norVFX/VFX	*2013*	*no data*	0.2	0.2	0.2	-	0.3
	2014	0.14	0.1	0.1	0.1	0.3	0.1
O-desVFX/VFX	*2013*	*no data*	2.9	3	3.1	1.1	3.6
	2014	2.04	2.2	2.6	2.5	4.2	2.6
N-desVFX/VFX	*2013*	*no data*	0.2	0.2	0.2	-	0.2
	2014	*0.14*	0.1	0.1	0.1	0.3	0.1
norFXT/FXT	*2013*	*no data*	72.4	-	-	-	46.4
	2014	*4.7*	106.4	51.9	49.7	12.5	12.5
norDZP/DZP	*2013*	*no data*	1.7	3.5	56.5	-	3.6
	2014	*7.82*	9.8	12.5	14.2	-	5
4-OH-DCF/DCF	*2013*	*no data*	0.7	1.2	1.4	0	0.9
	2014	0.84	1.7	1.3	3.4	0	1.6
MTPA/MTP	*2013*	*no data*	30.8	35.7	39.3	0	25.4
	2014	30.4	28	27	33.7	0	17.7
α-OH-MTP/MTP	*2013*	*no data*	0.8	0.5	0.2	0	0.4
	2014	0.7	0.6	-	0.4	0	0.2

Supporting Information

Figure S1. Concentration (ng L^{-1}) of the studied compounds in the RO and NF concentrates. Gaps in the figure show that the analyte was not detected.

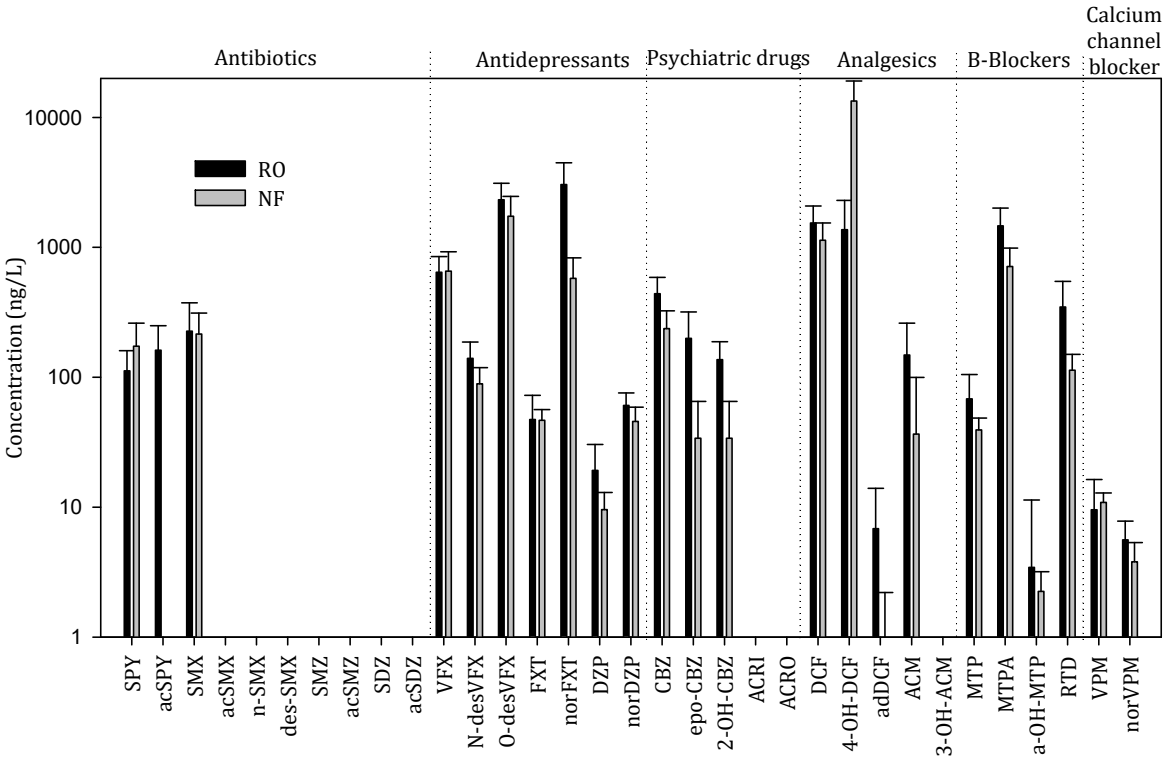

Table S1. MBR, RO and NF membrane processes conditions and membrane properties

MBR Process Conditions	**Units**	
Membrane Area	m^2	8
System flow (Q)	Lh^{-1}	140
Permeate flow	Lh^{-1}	150
System flux	$Lm^{-2}h^{-1}$	18.8
HRT	Hours	16
Solids concentration (MBR tank)	$g\ L^{-1}$	7
SRT	Days	30
Filtration - relaxation cycles	Min	9:1
Air scour flow rate	$m^3 \cdot h^{-1}$	10
DO aerobic set point	$mg\ O_2 \cdot L^{-1}$	0.5

RO and NF Process Conditions and Membrane Properties			
Membrane		ESPA2 LD-4040	NF90 - 4040
Membrane surface area	m^2	7.43	7.62
Permeate flux (average)	$Lm^{-2}h^{-1}$	18	18 (26, 32)
Permeate flow	Lh^{-1}	135	135 (196, 240)
Feed pressure	bar	6.5	3.3 (4.5, 5.7)
System recovery	%	75	75
Membrane Properties			
Material		Polyamide	Polyamide
NaCl Rejection	%	99.6	85 – 95
Surface charge		Negative	Negative
MWCO estimate	Da	≈100	≈200
Surface roughness	nm	30.0	63.9

Table S2: Physico-chemical properties of the studied PhACs, metabolites and TPs (Souce: National Center for Biotechnology Information. PubChem Compound Database)

Therapeutic class	Compound		CAS number	Formula	MW (g/mol)	pK_a	Log K_{ow}	Charge at pH 7
Antibiotics	Sulfapyridine	SPY	144-83-2	$C_{11}H_{11}N_3O_2S$	249.3	Pk1= 8043, Pk2=2.3	0.35	Neut./Neg.
	N^4−acetylsulfapyridine	*acSPY*	19077-98-6	$C_{13}H_{13}N_3O_3S$	291.3		1.25	Neut./Neg.
	Sulfamethoxazole	SMX	723-46-6	$C_{10}H_{11}N_3O_3S$	253.3	5.7	0.89	Neut./Neg.
	N^4−acetylsulfamethoxazole	*acSMX*	21312-10-7	$C_{12}H_{13}N_3O_4S$	295.3		1.21	Neut./Neg.
	Nitrosulfamethoxazole	n-SMX	29699-89-6	$C_{10}H_9N_3O_5S$	283.3	5.62	1.22	
	Desaminosulfamethoxazole	*des-SMX*	-	-	238.3			
	Sulfamethazine	SMZ	57-68-1	$C_{12}H_{14}N_4O_2S$	278.3	2.65	0.89	Neut./Neg.
	N^4-Acetylsulfamethazine	*acSMZ*	100-90-3	$C_{14}H_{16}N_4O_3S$	320.4		1.48	Neut./Neg.
	Sulfadiazine	SDZ	68-35-9	$C_{10}H_{10}N_4O_2S$	250.3	pK1= 6.36 pK2= 2.1	-0.09	Neut./Neg.
	N^4-Acetylsulfadiazine	*acSDZ*	127-74-2	$C_{12}H_{12}N_4O_3S$	292.3		0.39	Neut./Neg.
Anti-depressants	Venlafaxine	VFX	93413-69-5	$C_{17}H_{27}NO_2$	277.4	14.42	3.28	positive
	N−Desmethylvenlafaxine	*N-desVFX*	149289-30-5	$C_{16}H_{25}NO_2$	263.4		3.07	positive
	O−Desmethylvenlafaxine	*O-desVFX*	93413-62-8	$C_{16}H_{25}NO_2$	263.4		2.72	positive
	Fluoxetine	FXT	54910-89-3	$C_{17}H_{18}F_3NO$	309.3	9.5	4.05	positive
	Norfluoxetine	*norFXT*	83891-03-6	$C_{16}H_{16}F_3NO$	295.3	9.05	4.07	
	Diazepam	DZP	439-14-5	$C_{16}H_{13}ClN_2O$	284.7	3.4	2.82	Neutral
	Desmethyldiazepam	*norDZP*	1088-11-5	$C_{15}H_{11}ClN_2O$	270.7		3.89	Neutral
Psychiatric drugs	Carbamazepine	CBZ	298-46-4	$C_{15}H_{12}N_2O$	236.3	13.9	2.45	Neutral
	10,11-Epoxy−Carbamazepine	*epo-CBZ*	36507-30-9	$C_{15}H_{12}N_2O_2$	252.3		0.95	Neutral
	2−Hydroxycarbamazepine	*2-OH-CBZ*	68011-66-5	$C_{15}H_{12}N_2O_2$	252.3		1.42	Neutral
	Acridine	ACRI	260-94-6	$C_{13}H_9N$	179.1	6.15	3.4	
	Acridone	ACRO	578-95-0	$C_{13}H_9NO$	195.2		1.69	Neutral
Analgesics/ anti-inflammatories	Diclofenac	DCF	15307-86-5	$C_{14}H_{11}Cl_2NO_2$	296.2		4.51	Negative
	4-OH-diclofenac	*4-OH-DCF*	64118-84-9	$C_{14}H_{11}Cl_2NO_3$	312.2			
	Diclofenac−amide	*adDCF*	15362-40-0	$C_{14}H_9Cl_2NO$	278.1	13.31	3.11	
	Acetaminophen	ACM	103-90-2	$C_8H_9NO_2$	151.2	9.38	0.46	Neutral
	3-OH-acetaminophen	*3-OH-ACM*	37519-14-5	$C_8H_9NO_3$				
B-Blockers	Metoprolol	MTP	37350-58-6	$C_{15}H_{25}NO_3$	267.4	9.6	1.88	Positive
	Metoprolol−acid	*MTPA*	56392-14-4	$C_{14}H_{21}NO_4$	267.3	3,54; 9,67	-2.34	Neutral
	Alfa−Metoprolol−OH	*α-OH-MTP*	56392-16-6	$C_{15}H_{25}NO_4$				
	Ranitidine	RTD	66357-35-5	$C_{13}H_{22}N_4O_3S$	314.4	2.4	0.27	Positive
Calcium channel blocker	Verapamil	VPM	52-53-9	$C_{27}H_{38}N_2O_4$	454.6	9.68	3.79	
	Norverapamil	norVPM	67018-85-3	$C_{26}H_{36}N_2O_4$	440.6	10.29	4.59	

Table S3: Concentration (ng L^{-1}) of the studied PhACs, metabolites and TPs in the sewer entering the WWTP

Therapeutic class	Compound	8:00 PM	10:00 PM	12:00 AM	2:00 AM	4:00 AM	6:00 AM	8:00 AM	10:00 AM	12:00 PM	2:00 PM	4:00 PM	6:00 PM
Antibiotics	**SPY**	123.20	195.42	195.92	390.69	251.77	112.51	134.93	201.44	214.02	266.10	245.28	320.35
	acSPY	128.52	357.07	235.11	472.44	284.58	92.79	109.31	253.03	170.70	202.81	287.67	223.67
	SMX	88.03	235.13	131.45	361.57	243.41	680.35	349.02	314.13	568.21	723.78	303.22	183.32
	acSMX	116.60	195.16	99.72	336.60	197.90	439.15	185.34	239.01	540.00	473.60	111.14	196.31
	n-SMX	31.5*	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	des-SMX	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	<LOD	n.d.	n.d.	n.d.	n.d.
	SMZ	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	acSMZ	n.d.	n.d.	n.d.	17.50	n.d.	n.d.	n.d.	n.d.	1.96*	4.51	n.d.	n.d.
	SDZ	n.d.	n.d.	<LOD	n.d.	n.d.	n.d.	n.d.	n.d.	175.47	n.d.	n.d.	n.d.
	acSDZ	8.66*	<LOQ	<LOD	n.d.	n.d.	n.d.	<LOD	<LOD	<LOQ	23.32*	n.d.	<LOD
Anti-depressants	**VFX**	268.67	344.62	318.34	390.90	347.93	278.86	210.06	400.02	246.44	288.57	347.78	281.41
	N-desVFX	38.09	46.83	33.88	50.81	44.67	40.79	26.18	83.72	37.03	41.40	38.44	36.46
	O-desVFX	607.46	638.12	640.97	602.04	711.32	640.85	413.59	854.10	592.17	629.73	631.64	554.48
	FXT	n.d.	<LOD	n.d.	33.98	<LOD	21.22	16.95*	24.85	25.25*	31.78	<LOD	n.d.
	norFXT	344.30	336.10	343.00	315.70	264.50	283.30	224.10	607.40	462.30	397.10	404.76	281.60
	DZP	1.55	2.10	4.79	28.59	2.02	1.24	0.50	2.18	3.20	5.94	3.62	<LOQ
	norDZP	13.58	14.00	13.96	19.46	13.51	19.61	9.23	23.15	16.84	26.97	20.51	15.96
Psychiatric drugs	**CBZ**	80.07	106.50	67.62	77.12	116.25	118.42	129.92	80.82	89.88	87.59	211.67	83.58
	epo-CBZ	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	2-OH-CBZ	32.07	32.46	16.13	23.37	39.44	19.94	27.66	37.45	38.58	20.83	27.18	24.92
	ACRI	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	ACRO	0.85	0.62	1.57	3.27	0.86	0.76	0.41	<LOD	<LOQ	<LOQ	<LOQ	0.66*
Analgesics/ anti-inflammatories	**DCF**	519	554	483	359	373	368	135	671	565	770	955	518
	4-OH-DCF	11789	13646	9254	4674	12104	10768	5759	22408	12345	15495	32590	18181
	adDCF	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	ACM	23432	22950	25034	21876	25611	18041	20744	74160	43124	22460	42579	23746
	3-OH-ACM	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
B-Blockers	**MTP**	46.75	n.d.	26.96	37.97	35.63	45.30	39.38	72.79	59.96	64.54	41.15	38.66
	MTPA	1380.44	1404.29	1093.92	1434.19	1237.72	1022.95	835.30	2005.61	1947.00	1551.55	1345.17	1220.74
	α-OH-MTP	35.08	31.45	21.28	48.11	30.00	21.03	17.51	57.73	55.36	42.52	27.72	25.40
	RTD	191.02	306.51	208.71	177.26	175.77	264.12	143.47	1036.89	357.86	524.57	390.98	263.62
Calcium channel blocker	**VPM**	6.59	<LOQ	7.33	44.11	7.18	7.52	4.64	7.18	6.38	13.98	5.99	<LOQ
	norVPM	n.d.	n.d.	3.36	11.93	<LOQ	4.71	2.84	<LOQ	1.75	<LOQ	n.d.	<LOD

Table S4: A comparison of the concentration (ng L^{-1}) of the studied PhACs, metabolites and TPs in the influent of the WWTP

			2013		2014	
Therapeutic class	Compound		Average	Standard Deviation	Average	Standard Deviation
Antibiotics	Sulfapyridine	SPY	96	36	123	42
	N^4−acetylsulfapyridine	*acSPY*	*132*	*47*	*202*	*67*
	Sulfamethoxazole	SMX	120	88	175	38
	N^4−acetylsulfamethoxazole	*acSMX*	*103*	*92*	*126*	*43*
	Nitrosulfamethoxazole	n-SMX	-	-	0	0
	Desaminosulfamethoxazole	*des-SMX*	-	-	*0*	*0*
	Sulfamethazine	SMZ	-	-	0	0
	N^4-Acetylsulfamethazine	*acSMZ*	-	-	*2*	*4*
	Sulfadiazine	SDZ	-	-	950	307
	N^4-Acetylsulfadiazine	*acSDZ*	-	-	*3*	*8*
Anti-depressants	Venlafaxine	VFX	159	50	272	61
	N−Desmethylvenlafaxine	*N-desVFX*	*25*	*8*	*33*	*10*
	O−Desmethylvenlafaxine	*O-desVFX*	*397*	*128*	*595*	*179*
	Fluoxetine	FXT	22	34	15	13
	Norfluoxetine	*norFXT*	*508*	*305*	*377*	*128*
	Diazepam	DZP	3	2	2	2
	Desmethyldiazepam	*norDZP*	*3*	*3*	*17*	*4*
Psychiatric drugs	Carbamazepine	CBZ	65	22	81	16
	10,11-Epoxy−Carbamazepine	*epo-CBZ*	*22*	*15*	*0*	*0*
	2−Hydroxycarbamazepine	*2-OH-CBZ*	*18*	*13*	*17*	*13*
	Acridine	ACRI	0	0	0	0
	Acridone	ACRO	-	-	1	1
Analgesics/ anti-inflammatories	Diclofenac	DCF	431	185	715	200
	4-OH-diclofenac	*4-OH-DCF*	*242*	*114*	*427*	*116*
	Diclofenac−amide	*adDCF*	*0*	*0*	*0*	*0*
B-Blockers	Acetaminophen	ACM	18733	7242	30766	8141
	3-OH-acetaminophen	*3-OH-ACM*	*0*	*0*	*0*	*0*
	Metoprolol	MTP	18	7	40	10
	Metoprolol−acid	*MTPA*	*623*	*195*	*1115*	*236*
	Alfa−Metoprolol−OH	*α-OH-MTP*	*15*	*6*	*24*	*5*
	Ranitidine	RTD	286	118	239	93
Calcium channel blocker	Verapamil	VPM	0	0	3	3
	Norverapamil	norVPM	3	2	0	0

Table S5: Method limits of detection (LOD) and quantification (LOQ) for the studied PhACs, metabolites and TPs

Therapeutic class	Compound		MBR INF (ng L^{-1})		MBR EFF (ng L^{-1})		RO-FEED (ng L^{-1})		RO-PERM (ng L^{-1})		RO-CONC (ng L^{-1})	
			LOD	LOQ	LOD	LOQ	LOD	LOQ	LOD	LOQ	LOD	LOQ
Antibiotics	Sulfapyridine	SPY	1.68	5.60	2.24	7.48	1.54	5.13	0.44	1.45	2.36	7.86
	N^4–acetylsulfapyridine	*acSPY*	1.33	4.44	0.80	2.66	0.52	1.72	0.08	0.25	2.12	7.07
	Sulfamethoxazole	SMX	2.63	8.77	2.24	7.46	1.47	4.89	0.23	0.76	3.76	12.54
	N^4–acetylsulfamethoxazole	*acSMX*	4.99	16.63	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.
	Nitrosulfamethoxazole	n-SMX	-	-	-	-	-	-	-	-	-	-
	Desaminosulfamethoxazole	*des-SMX*	-	-	-	-	-	-	-	-	-	-
	Sulfamethazine	SMZ	-	-	-	-	-	-	-	-	-	-
	N^4-Acetylsulfamethazine	*acSMZ*	-	-	-	-	-	-	-	-	-	-
	Sulfadiazine	SDZ	-	-	-	-	-	-	-	-	-	-
	N^4-Acetylsulfadiazine	*acSDZ*	-	-	-	-	-	-	-	-	-	-
Anti-depressants	Venlafaxine	VFX	0.42	1.41	0.21	0.70	0.22	0.74	0.01	0.03	0.36	1.20
	N–Desmethylvenlafaxine	*N-desVFX*	0.94	3.14	0.28	0.92	0.28	0.94	n.d.	n.d.	0.64	2.13
	O–Desmethylvenlafaxine	*O-desVFX*	0.63	2.10	0.40	1.32	1.07	3.57	0.03	0.11	0.91	3.05
	Fluoxetine	FXT	0.42	1.39	0.10	0.33	0.09	0.29	n.d.	n.d.	0.53	1.77
	Norfluoxetine	*norFXT*	2.06	6.85	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.	3.68	12.27
	Diazepam	DZP	0.09	0.30	0.02	0.07	0.01	0.05	n.d.	n.d.	0.19	0.62
	Desmethyldiazepam	*norDZP*	0.05	0.17	0.15	0.49	0.11	0.36	n.d.	n.d.	0.10	0.34
Psychiatric drugs	Carbamazepine	CBZ	0.10	0.34	0.10	0.33	0.07	0.23	0.00	0.01	0.19	0.64
	10,11-Epoxy–Carbamazepine	*epo-CBZ*	1.24	4.13	19.65	65.49	13.25	44.18	0.01	0.04	11.32	37.72
	2–Hydroxycarbamazepine	*2-OH-CBZ*	0.22	0.74	1.27	4.24	1.52	5.07	n.d.	n.d.	0.29	0.96
	Acridine	ACRI	n.d.	n.d.	0.30	0.99	0.30	0.99	0.18	0.60	n.d.	n.d.
	Acridone	ACRO	-	-	-	-	-	-	-	-	-	-
Analgesics/ anti-inflammatories	Diclofenac	DCF	7.91	26.36	19.41	64.72	10.18	33.94	n.d.	n.d.	2.29	7.63
	4-OH-diclofenac	*4-OH-DCF*	-	-	-	-	-	-	n.d.	n.d.	n.d.	n.d.
	Diclofenac–amide	*adDCF*	6.96	23.21	0.17	0.57	0.12	0.41	n.d.	n.d.	1.46	4.87
	Acetaminophen	ACM	52.69	175.65	43.71	145.68	23.68	78.94	7.36	24.54	65.76	219.19
	3-OH-acetaminophen	*3-OH-ACM*	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
B-Blockers	Metoprolol	MTP	0.41	1.36	0.26	0.87	0.19	0.63	n.d.	n.d.	0.49	1.63
	Metoprolol–acid	*MTPA*	5.15	17.18	4.18	13.92	3.72	12.40	0.52	1.73	2.94	9.80
	Alfa–Metoprolol–OH	*α-OH-MTP*	1.05	3.48	0.82	2.74	0.10	0.32	0.11	0.37	0.46	1.52
	Ranitidine	RTD	1.92	6.39	0.40	1.34	0.45	1.51	0.01	0.04	1.14	3.79
Calcium channel blocker	Verapamil	VPM	0.00	0.00	0.03	0.10	0.02	0.07	n.d.	n.d.	0.06	0.19
	Norverapamil	norVPM	0.27	0.91	0.01	0.05	n.d.	n.d.	n.d.	n.d.	0.14	0.48

Table S6: Concentration (ng L^{-1}) of the studied compounds in the membranes feed, concentrate and permeate samples.

	Compound	NF Feed		NF Permeate			NF Concentrate		RO Feed		RO Permeate		RO Concentrate	
Antibiotics	Sulfapyridine	25,73	4,1	0,79	0,1		173,84	87,2	2,39	2,7	-	-	111,82	48,6
	N4–acetylsulfapyridine	21,98	1,8	-	-		-	-	28,07	14,5	0,25	0,6	161,78	87,8
	Sulfamethoxazole	82,94	12,7	0,52	0,4		215,29	96,5	41,03	21,5	0,91	2,2	226,78	148,2
	N4–acetylsulfamethoxazole	-	-	-	-		-	-	-	-	-	-	-	-
	Nitrosulfamethoxazole	-	-	-	-		-	-	-	-	-	-	-	-
	Sulfamethoxazole Amide	-	-	-	-		-	-	-	-	-	-	-	-
	Sulfamethazine	-	-	-	-		-	-	-	-	-	-	-	-
	N4-Acetylsulfamethazine	-	-	-	-		-	-	-	-	-	-	-	-
	Sulfadiazine	-	-	-	-		-	-	-	-	-	-	-	-
	N4-Acetylsulfadiazine	-	-	-	-		-	-	-	-	-	-	-	-
Antidepressants	Venlafaxine	261,31	42,3	8,37	2,0		657,31	268,4	174,21	38,8	-	-	642,84	206,1
	N–Desmethylvenlafaxine	30,51	7,9	2,23	0,3		88,95	29,5	30,60	7,2	-	-	140,08	46,9
	O–Desmethylvenlafaxine	665,05	126,2	36,21	15,8		1738,66	728,8	538,77	131,8	1,59	1,0	2322,19	793,6
	Fluoxetine	16,99	3,7	0,41	0,7		46,71	9,7	2,85	4,4	-	-	47,44	25,1
	Norfluoxetine	822,10	130,2	11,31	6,1		577,49	253,2	-	-	-	-	3046,71	1442,2
	Diazepam	1,36	0,6	-	-		9,58	3,4	0,15	0,3	-	-	19,18	11,2
	Desmethyldiazepam	17,27	3,7	0,92	0,0		45,73	13,3	14,00	4,9	-	-	60,83	14,9
Psychiatric drugs	Carbamazepine	96,90	19,1	15,47	8,8		237,14	87,5	95,49	30,5	-	-	440,91	146,9
	Epoxy– Carbamazepine	-	-	4,94	5,2		33,94	31,2	80,15	21,6	0,16	0,4	199,34	118,5
	2–Hydroxycarbamazepine	-	-	4,94	5,2		33,94	31,2	61,06	27,5	-	-	136,93	51,2
	Acridine	-	-	-	-		-	-	552,28	1219,3	0,34	0,7	0,00	0,0
Analgesics	Diclofenac	259,78	44,1	-	-		1137,46	404,1	343,36	127,6	-	-	1544,88	539,0
	Diclofenac–OH	884,40	137,0	17,60	20,2		1368,79	582,1	487,89	250,9	-	-	1368,64	929,4
	Diclofenac–amide	-	-	-	-		0,81	1,4	1,64	1,1	-	-	6,84	7,1
	Acetaminophen	39,47	0,0	12,27	0,0		36,53	63,3	24,96	35,3	-	-	148,52	112,3

	Acetaminophen–OH	-	-	-	-		-	-	-	-	-	-	-	-
B-Blockers	**Metoprolol**	46,87	4,2	9,73	0,7		39,33	9,3	22,21	12,4	-	-	68,31	36,7
	Metoprolol–acid	1582,52	259,2	-	-		712,96	274,7	933,37	467,6	5,39	8,8	1466,68	538,3
	Alfa–Metoprolol–OH	0,16	0,0	0,03	0,0		2,25	0,9	2,70	1,1	3,10	3,1	3,44	7,9
	O–Desmethyl–Metoprolol	10,98	0,0	-	-		61,30	27,8	-	-	-	-	19,91	32,1
Other drugs	**Verapamil**	4,00	0,9	0,06	0,1		10,88	2,0	3,76	1,2	-	-	9,56	6,8
	Norverapamil	1,19	1,0	0,14	0,0		3,80	1,5	-	-	-	-	5,61	2,2
	Ranitidine	35,79	9,4	2,83	0,5		113,20	37,2	194,66	139,5	0,01	0,0	346,86	200,3
	Acridone	0,22	0,4	2,50	0,3		-	-	-	-	-	-	-	

Table S7: Water quality parameters measured in the sewer entering the WWTP.

Parameter	Units	8:00 PM	10:00 PM	12:00 AM	2:00 AM	4:00 AM	6:00 AM	8:00 AM	10:00 AM	12:00 PM	2:00 PM	4:00 PM	6:00 PM
Elec. Conductivity	(µS/cm)	1440	1204	1143	1233	1276	1396	1408	1463	1376	1719	1697	1301
COD_T	(mg L^{-1})	542	660	706	889	282	170	157	433	535	606	586	448
COD_S	(mg L^{-1})	97	289	233	199	100	72	22	84	100	123	129	106
TKN_T	(mg N L^{-1})	54	59	58	77	49	50	38	84	77	55	51	49
TKN_S	(mg N L^{-1})	45	48	45	45	41	41	33	71	60	48	41	40
NH_4^+	(mg N L^{-1})	32	37	40	34	32	28	17	60	47	42	35	34
NO_3^-	(mg N L^{-1})	0	0	0	0	0	0	0	0	0	0	0	0
NO_2^-	(mg N L^{-1})	1.1	0	0	10	0	0	0	2.3	0	0.3	12.9	9.1
PO_4^{3-}	(mg L^{-1})	3.9	1.6	0.7	3.2	3.6	3.2	2.5	2.9	6.2	2.1	5.3	5.6