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# Occurrence and fate of an emerging drug pollutant and its by-products during conventional and advanced wastewater treatment: case study of furosemide

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## Abstract :

Conventional wastewater treatment systems are not designed to remove pharmaceutical compounds from wastewater. These compounds can be degraded into many other transformation products which are hardly, if at all, studied. In this context, we studied the occurrence and degradation of furosemide, a very frequently detected diuretic, along with its known degradation products in several types of wastewater. Influent and effluent from the Seine-Centre Wastewater Treatment Plant (WWTP) (Paris, France) as well as outlet of residential care homes (Dordogne, France) were analyzed by Ultra-Performance Liquid Chromatography-tandem Mass Spectrometry (UPLC-MS/MS) to quantify furosemide and its known degradation products, saluamine and pyridinium of furosemide. Oxidation experiments (chlorination, ozonation and UV photolysis with hydrogen peroxide) were then performed

on furosemide solutions and on water from residential care facilities to study the degradation of furosemide by potential advanced processes, and also to identify unknown oxidation products by high-resolution mass spectrometry. Furosemide was well degraded in Seine-Centre WWTP (>75%) but did not increase the concentrations of its main degradation products. Saluamine and pyridinium of furosemide were already present at similar concentrations to furosemide in the raw wastewater (~2.5-3.5  $\mu\text{g.L}^{-1}$ ), and their removal in the WWTPs were very high (>80%). Despite their removal, the three compounds remained present in treated wastewater effluents at concentrations of hundreds of nanograms per liter. Chlorination degraded furosemide without pyridinium production unlike the other two processes. Chlorination and ozonation were also effective for the removal of furosemide and pyridinium in residential care home water, but they resulted in the production of saluamine. To our knowledge this is the first evidence of saluamine and pyridinium of furosemide in real water samples in either the particulate or dissolved phase.

**Keywords:** Pharmaceutical; wastewater; residential care home; degradation products; oxidation

**Declaration of interest:** none

## 1. Introduction

In the context of a wide consumption of medicines in high income and emerging countries, increasing demographical urban pressure and population aging, hundreds of active substances used for therapeutic purposes and marketed worldwide end up in the wastewater system. Conventional wastewater treatment plant (WWTP) processes based on filtration, sand and grease removal, decantation and biological or physico-chemical treatments are not designed for the elimination of pharmaceutical compounds, so their removal can vary drastically depending on the molecule (e.g., its hydrophobicity) (Chiffre et al. 2016). The active substances of these drugs are discharged with WWTP effluents at concentrations ranging from  $\text{ng.L}^{-1}$  to  $\mu\text{g.L}^{-1}$  and end up in the aquatic environment,

representing a risk for organisms (Cizmas et al. 2015, Obinwanne Okoye et al. 2022). To overcome this problem, the implementation of tertiary or quaternary treatments after conventional treatments is increasingly studied, and several countries (e.g., Switzerland, Germany) already regulate the removal of organic micropollutants from wastewater. The optimization of biological treatments, for example with the addition of fungus (*Trametes versicolor*) in bioreactors (Cruz-Morato et al. 2014; Badia-Fabregat et al. 2015), can be effective on potentially biodegradable compounds. However, the most frequent processes used to achieve a satisfactory removal of micropollutants include adsorption on activated carbon (Guillossou et al. 2019), membrane processes and oxidation processes such as ozonation (Guillossou et al. 2020).

Ozone has been used for a long time as both an oxidant and a disinfectant for drinking water production, and is increasingly applied for the treatment of wastewater effluents (Lim et al. 2022). It is a very powerful oxidant that can react directly with organic molecules, mainly by electrophilic reaction, and also indirectly by the production of radicals initiated by hydroxyl or hydroperoxide ions in the aqueous medium (Guo et al. 2012). Ozonation has been shown to be effective in the degradation of hormones, antibiotics, antivirals and pesticides (Lim et al. 2022). Advanced oxidation processes (AOPs) are also very efficient for the degradation of organic molecules (Miklos et al. 2018). These processes are based on the production of highly reactive hydroxyl radicals from one or more primary oxidants (e.g., chlorine, ozone, hydrogen peroxide, etc.) (Sarathy & Mohseni 2006) or by photocatalysis (Prieto-Rodriguez et al. 2012). Advanced oxidation processes in which Ultraviolet (UV) radiation is combined with ozone, chlorine, peroxodisulfate or hydrogen peroxide are efficient to reach extensive degradation. The most common combination is with H<sub>2</sub>O<sub>2</sub> because it is much less energy-consuming and expensive than with O<sub>3</sub>, and it is also less sensitive to matrix changes and dissolved organic matter than sulfates. While advanced treatment processes for organic micropollutants are not widely used, many WWTP use a disinfection step before discharging effluents into the environment (e.g., in bathing areas). Another process commonly used for disinfection and inactivation of pathogenic

microorganisms is based on UV irradiation (Hijnen et al. 2006). Direct UV photolysis with UV-C (100-280 nm) shows good efficiency on pesticides (Sanches et al. 2010) but some pharmaceutical compounds like carbamazepine are more resistant to it (Pereira et al. 2007).

Although oxidation treatments can be quite efficient for the degradation of micropollutants, they often form oxidation by-products. Transformation products (e.g., metabolites) of pharmaceuticals can also be already present in the incoming water or be formed during conventional biological processes. The formation of oxidation by-products depends on the mode of oxidation, the medium (especially the presence of nitrogen, halogens, organic matter) and the type of radical produced in the case of advanced oxidation (Miklos et al. 2018). These degradation by-products present a hazard for health (Stalter et al. 2016) and for the environment (Wang et al. 2018). Apart from the regulated disinfection by-products (e.g., trihalomethanes and haloacetic acids), hundreds of chlorination by-products have been identified (Zhang et al. 2012) from the reaction of chlorine with organic matter and their potential toxicity can be a threat to human health (Stefán et al. 2019, Mazhar et al. 2020). Many products are also generated during UV-H<sub>2</sub>O<sub>2</sub> oxidations depending on the dose of oxidant, the UV fluence and the pH of the medium. Oxidation by-products are often more toxic than the parent compounds on algae, daphnia, and medaka fish (Wang et al. 2018), it is thus of critical importance to assess the toxicity of degradation products to fully establish the potential impacts of organic micropollutants on the environment.

To address the questions related to the presence of pharmaceuticals in the environment, we selected furosemide from a list of high-risk emerging pollutants (Besse and Garric 2008) and subjected this drug to a predictive study aiming to anticipate the fate of organic contaminants (Laurence et al, 2011, 2014; Olvera Vargas et al 2016). Classified by the World Health Organization as an essential medicine, furosemide is a loop diuretic prescribed all over the world for hypertension and heart, liver, and kidney failure (Abbot and Kovacic 2008). Mostly eliminated unmodified by humans and moderately degraded

in conventional treatment plants (between 8 and 54% in Castiglioni et al. 2006, >65% in Matamoros et al. 2009, 74% in Kasprzyk-Hordern et al. 2009, or between 30-80% according to the WWTP considered in Jelic et al. 2011), furosemide is also widely found in the environment from several tens to few thousand of ng.L<sup>-1</sup> in rivers (Banjac et al. 2015, Baken et al. 2018, Munro et al. 2019, White et al. 2019, Cantwell et al. 2018). Applied to furosemide, our predictive study confirmed that saluamine and furfural are two transformation products (TPs) of furosemide (Laurencé et al. 2014). More importantly, it also identified the pyridinium of furosemide as a TP and a human metabolite of this drug (Laurencé et al. 2019). Whereas furfural presents moderate acute toxicity in freshwater invertebrates (LC50 = 13 mg.L<sup>-1</sup> for 72h exposure in Reed & Kwok 2014), saluamine shows acute toxicity in rats (Al-Omar et al. 2009). We also previously demonstrated that pyridinium of furosemide exhibits neurodegenerative properties in mice (Laurencé et al. 2019). Therefore, the fate of furosemide and its TPs in wastewater and in the environment needs to be assessed to fully characterize the potential impacts of this pharmaceutical on the aquatic ecosystems. Furosemide is also an interesting and highly relevant model to study the fate of pharmaceuticals along with their TPs. Especially, the production of saluamine and pyridinium of furosemide as well as their potential degradation during conventional and advanced wastewater treatment remains unknown to date. The purpose of this work was thus i) to assess the occurrence and removal of furosemide and its known TPs in a large WWTP and in wastewater from an elderly care facility, ii) to study their removal by several advanced oxidation processes (ozonation, UV/H<sub>2</sub>O<sub>2</sub>) and iii) to identify the subsequent formation of potential new TPs following these treatments.

## **2. Material & methods**

### **2.1. Reagents**

The furosemide-d5 standard was purchased from Cluzeau Info Labo. Internal standards (atenolol-d7 and sulfamethoxazole-d4) and saluamine (SAL) were purchased from Sigma-Aldrich. For the extraction

and analysis, MS grade methanol was purchased from Fisher. Ultrapure water was produced by a Milli-Q® IQ 7000 ultrapure water system. Furosemide (FUR) was purchased from Sigma. Pyridinium of furosemide (PYR) was synthesized at ICMPE as previously described (Laurencé et al. 2011), and its structure has been confirmed by NMR with over 96% purity. Hydrogen peroxide 50% was purchased from CarlRoth.

## **2.2. Sampling sites**

To assess the WWP efficiency on furosemide degradation, raw water (RW) and treated effluent water (TW) were collected on 2021-12-07 and 2021-12-08 from the Seine-Centre WWTP (Colombes, France), operated by the Paris public sanitation service (SIAAP) and treating wastewater of about 1,000,000 people equivalent with a flow of 240,000 m<sup>3</sup>/day. The Seine-Centre WWTP is mostly based on biofiltration stages ensuring the treatment of organic carbon as well as nitrification and denitrification processes. For the oxidation tests on raw sewerage water samples, wastewater from residential care homes (RCH) was chosen because of its presumably high concentration of furosemide, thus allowing a better identification of the possible degradation products. Three wastewater outlets of health care institutions were collected in Dordogne, France. The first sample (RCH1) was collected on 2021-05-26 in a RCH of 10,000 m<sup>2</sup> located in Bergerac, in an urban area. It has a capacity of 90 beds plus 15 beds for Alzheimer's patients, 10 places in daycare centers and 5 emergency beds. The sample was taken from a lift station which only collects water from the facility. The second sample (RCH2) was collected on 2021-06-09 from a reeducation center located in a national forest at Antonne-et-Trigonant with 120 places and 40 beds for senior residents. The third sample (RCH3) was collected on 2021-06-08 from a RCH at Lolme with 90 beds and 5 daycare places. Several physico-chemical parameters were recorded: pH, Dissolved Organic Carbon (DOC), Chemical Oxygen Demand (COD) and Biological Oxygen Demand at 5 days (DBO5), Suspended Matter (SM), Total Kjeldahl Nitrogen (TKN), ammoniacal nitrogen and organic nitrogen (table 1). The RCH outlet water samples were filtered on GF/D filters (2.7 µm pore size) and then on GF/F filters (0.7 µm pore size). pH and DOC were measured after

filtration (table 1). Particulate phase samples (i.e., particles collected on filters) were stored at -20 °C and freeze-dried until treatment for analysis. For RCH samples, it is interesting to note the large gap between the Kjeldahl nitrogen and ammonia nitrogen values, which implies a high proportion of organic nitrogen, reflecting the short residence time of the effluent and confirming the relatively fresh effluent sampling

**Table 1.** Physico-chemical parameters of samples from WWTP raw wastewater (RW1 = 2021-12-07, RW2 = 2021-12-08), WWTP treated wastewater (TW1 = 2021-12-07, TW2 = 2021-12-08) and wastewater outlets of residential care homes (RCH). NA = Not available (not measured). (BLACK AND WHITE TABLE)

<Table 1>

### 2.3. Oxidation assays

Furosemide degradation kinetics experiments were performed in ultrapure water spiked with 1 mg.L<sup>-1</sup> of furosemide. UV/H<sub>2</sub>O<sub>2</sub> and ozonation were applied at high doses or exposure as described below, and chlorination was also conducted at high dose (35 mgCl<sub>2</sub>.L<sup>-1</sup>) to maximize the formation of degradation products (see Text S1). Oxidation processes were also applied on 200 mL filtered wastewater samples from RCH facilities.

**Photodegradation under UV light with H<sub>2</sub>O<sub>2</sub>.** The sample was placed in a beaker, under a UV lamp at 254 nm (UV-C ; 0.34 mW/cm<sup>2</sup>) with stirring. To establish degradation kinetics of furosemide in ultrapure water, 1 mg.L<sup>-1</sup> of H<sub>2</sub>O<sub>2</sub> was added and aliquots of 1 mL were taken at 2, 5, 10, 15, 20, 30, 45, 60, 90, 120, 210 and 600 minutes. For the oxidation of wastewater from RCH, the H<sub>2</sub>O<sub>2</sub> dose was determined according to the DOC value by using 0.375 mg of H<sub>2</sub>O<sub>2</sub> per mg of DOC, and the total reaction time under UV irradiation was 5 hours.



**Ozonation.** Ozone was produced by a generator with an external source (O<sub>2</sub> gas cylinder). The bottle containing the sample to be oxidized was immersed in an ice bath throughout the experiment. For the ozonation of furosemide solutions in ultrapure water, a concentrated ozone solution was first produced (33 mg/L O<sub>3</sub>) and diluted in the furosemide samples at the required ozone concentrations, determined according to the DOC concentration of the sample (Equation 1).

$$[O_3](mgO_3.L^{-1}) = 2 \times DOC (mgC.L^{-1}) \quad (Eq. 1)$$

Aliquots of 1 mL were collected at 1, 2, 4, 5, 7, 10, 15, 20, 30, 45 and 60 minutes. For the oxidation of RCH wastewater, ozone was bubbled directly into the samples for 1.5 hours with an O<sub>2</sub> flow to the ozonator of 2 L.h<sup>-1</sup>. The residual O<sub>3</sub> was measured by colorimetry with indigo carmine using a double beam spectrometer (UV6300 PC, VWR) at 600 nm.

#### **2.4. Analytical procedures for quantification**

After filtration, the residential care home samples are split into several aliquots of which 5 are spiked. Furosemide and its TPs present in dissolved phases of wastewater samples were extracted through an automated extraction system (Thermo AutoTrace 280 SPE Instrument) on OASIS HLB cartridges (200 mg, 6 cc). After conditioning with methanol and ultrapure water, samples (200 mL) were loaded onto cartridges and eluted by 10 mL of methanol. The particulate phases of RCH1 and RCH2 samples were also analyzed. The filters (GF/D and GF/F) were frozen and freeze-dried to remove water and determine their dry mass. Furosemide and its TPs were extracted by assisted microwave extraction (Antoon Paar, Multiwave 3000). Two cycles of extractions of 30 minutes in a mixture of 60% methanol-40% dichloromethane (v/v) were performed (100 °C, 800 W). The extracts were filtered on folded filters washed with dichloromethane, evaporated to 1 mL under rotary evaporator and diluted in 100 mL ultrapure water to carry out a purification step on OASIS HLB cartridges. Cartridges were eluted with 10 mL of methanol, internal standards (Atenolol-d7 and Sulfamethoxazole-d4) were added to the

purified extracts and the final volume was adjusted to 1 mL by evaporation. Extract concentrations were evaluated using internal calibration. Internal standards were chosen based on their similar retention time to the one of the targeted molecule (Table S2). An evaluation of matrix effects revealed their low variability for all molecules across 16 urine samples. Spiked samples were also extracted, leading to the evaluation of extraction yields of furosemide, saluamine and pyridinium. These extraction yields were used for the correction of concentrations in the samples. The detection and quantification limit of the instrument for each compound is presented in Table S3. Analyses were performed using Ultra Performance Liquid Chromatography coupled with a triple quadrupole detector (Acquity-TQD, Waters). Separation was carried out on an ACQUITY UPLC BEH C18 column (1.7  $\mu\text{m}$ , 2.1 x 100 mm) with a 15 min gradient elution from 90:10 ultra-pure water (A) and methanol (B) both acidified with 0.1% formic acid, to 0:100 (A:B) maintained for 5 min, before reequilibration of the column (0.4 mL.min<sup>-1</sup>). For the detection, furosemide was ionized in negative electrospray mode, and saluamine and pyridinium of furosemide were ionized in positive mode. MS/MS acquisition was used (Table S2).

The phase distribution ( $K_d$  value) was calculated according to equation (2) (Park et al. 2017), where  $K_d$  is the solid-water distribution coefficient in L.kg<sup>-1</sup>,  $C_s$  is the concentration of furosemide, saluamine or pyridinium of furosemide adsorbed onto MES ng.L<sup>-1</sup> and  $C_w$  is the concentration on furosemide, saluamine or pyridinium of furosemide in the liquid phase in ng.L<sup>-1</sup>, and SS is that of suspended matter in the mixture in mg.L<sup>-1</sup>.

$$K_d = \frac{C_s}{SS \times C_w} \times 10^6 \quad (\text{Eq. 2})$$

## 2.5. Analytical procedures for the identification of TPs

Degradation kinetics experiments were analysed by ultra-performance liquid-chromatography coupled to ion-mobility time-of-flight mass spectrometry (UPLC-IMS-QTOF, Vion, Waters) to enable

the tentative identification of degradation products. The separation was carried out on an ACQUITY UPLC BEH C18 column (1.7  $\mu\text{m}$ , 2.1 x 100 mm) with a 25 min gradient from 98:2 ultra-pure water (A) and acetonitrile (B) both acidified with 0.1% formic acid, to 2:98 (A:B) maintained for 5 min, before reequilibration of the column. Ionization was performed by an electrospray source in both positive and negative mode, in low energy (6 V) and high energy ramp (20 to 56 V). Data was acquired and analyzed with the UNIFI software (Waters). Briefly, after 4D peak detection, the chromatograms at successive kinetics points were compared to spot potential TPs. Molecular formula attribution was performed with a restricted list of atoms (C, H, N, O, P, S and Cl) and several online libraries, as a part of Chemspider in UNIFI, were interrogated (Sigma-Aldrich, Drugbank, NIST, MassBank and LGC Standard) to tentatively identify the detected products. In order to ensure mass precision lockspray infusion was used during all the injections.

### **3. Results & discussion**

#### **3.1. Occurrence and removal in wastewater treatment plants**

First, the concentrations of furosemide were evaluated in the Seine-Centre WWTP, along with the possible presence of its known degradation products saluamine and pyridinium of furosemide. This WWTP was chosen because it is located directly downstream of the city of Paris, thus receiving most of Parisian wastewaters.

The two collected raw water samples (2021-12-07 and 2021-12-08 samples) exhibited substantial concentrations of furosemide of 3351 and 2819  $\text{ng.L}^{-1}$  respectively (table 2). These concentrations were consistent with those found in recent literature for WWTPs raw water in other European countries. Concentrations between 1 to 5  $\mu\text{g.L}^{-1}$  were found: 1491  $\text{ng.L}^{-1}$  in Sweden (Baresel et al. 2019), 2625  $\text{ng.L}^{-1}$  in Poland (Kot-Wasik et al. 2016), 2601  $\text{ng.L}^{-1}$  in Greece (Papageorgiou et al. 2019),

1901 ng.L<sup>-1</sup> and 3410 ng.L<sup>-1</sup> in Spain (Collado et al. 2014; Celic et al. 2019), 4577 ng.L<sup>-1</sup> in Portugal (Santos et al. 2013), 1652 ng.L<sup>-1</sup> in Italy (Feo et al. 2020), 2916 ng.L<sup>-1</sup> and in Switzerland (Lee et al. 2014).

Our samples concentrations at 24 hour interval were of the same order of magnitude, indicating little variation over this period. This result is in contrast to Gomez-Canela et al. (2019), who observed significant variations in furosemide concentrations over 5 consecutive days, but on nursing home effluents. However, for furosemide, other studies usually reported seasonal rather than weekly variations with lower concentrations in summer (Kot-Wasik et al. 2016; Delli Compagni et al. 2020).

The known degradation products of furosemide, saluamine, furfural and pyridinium of furosemide, have been searched for in wastewater. Furfural could not be analyzed with our method, the compound being possibly too small and/or too polar to be extracted and analyzed with the same method as the other three. Although described as a furosemide transformation product since long ago (Andreasen et al. 1942), reports on saluamine toxicity and occurrence in the environment are scarce. Nonetheless, we found saluamine and pyridinium of furosemide in WWTP raw water both at concentrations of a few thousands of ng.L<sup>-1</sup> (table 2).

**Table 2.** Concentrations (ng.L<sup>-1</sup>) of Furosemide, Pyridinium of Furosemide and Saluamine in WWTP water samples (RW = raw wastewater, TW = treated wastewater) (BLACK AND WHITE TABLE)

**<Table 2>**

Interestingly, the distribution between the three compounds in raw water was about one third for each, which means that the degradation products are formed upstream of the WWTP, probably during the journey through the water system as commonly observed. Indeed, other drug TPs have already been found in WWTP influents such as N4-Acetylsulfamethoxazole, 2-hydroxycarbamazepine, O-desmethylvenlafaxine, 4-hydroxydiclofenac which are TPs of sulfamethoxazole, carbamazepine,

venlafaxine and diclofenac, respectively (Aymerich et al. 2016). Furosemide could have been hydrolyzed or biodegraded in the wastewater transport system. The hydrolysis of furosemide has been known since the 70's and leads to the formation of saluamine in an acidic environment (Bundgaard et al. 1988; Andreasen et al. 1982) and some studies showed that saluamine and pyridinium of furosemide can be produced by biotransformation of furosemide by several microorganisms (Hezari & Davis 1993; Laurencé et al. 2014; Olvera-vargas et al. 2016). These three compounds are still present in the two WWTP effluents samples at several hundred nanograms per liter (table 2). Saluamine can be formed in water sediments (Li et al. 2014) and by photolysis (Bundgaard et al. 1988) but to our knowledge, this is the first evidence of saluamine and pyridinium of furosemide presence in wastewater.

#### <Figure 1>

**Figure 1.** Removal percentage of furosemide, saluamine and pyridinium of furosemide in Seine-Centre WWTP. Solid bars represent concentrations in raw samples and hatched bars represent concentrations in samples after treatment in WWTP. The percentages shown on the hatched bars indicate the percent removal for each compound. Error bars represent standard error. (BLACK AND WHITE FIGURE)

All three compounds exhibited some removal in the WWTP. Yields of removal for each compound were very similar between the two sampling dates (figure 1). For furosemide, a reduction of over 70% was observed for both samples, which ranked in the higher range of furosemide removal by comparison with data from the literature. Matamoros et al. (2009) and Kasprzyk-Hordern et al. (2009) found similar furosemide removal rates (65% and 74%, respectively), but other studies found variable removals, with the lowest around 25% (Park et al. 2016), or 40 to 50% removal for WWTP with conventional mechanical-biological treatment (Kot-Wasik et al. 2016, Gros et al. 2010). In some cases, the sampling period could explain the differences in removal; Matamoros et al. (2009) and Kasprzyk-Hordern et al. (2009) samples were collected in spring and summer while Kot-Wasik et al. (2016) collected theirs in

winter. Castiglioni et al. (2006) showed that the elimination rate of furosemide was much lower in winter than in summer (8% versus 54%). These differences could be related to the lower temperatures in winter, which possibly attenuated the biological activity. However, in our case, the samples were collected in winter. Thus, the differences in efficiency of the treatment plants for furosemide removal could be explained by the different processes used in the plants. For example, the plant studied in Matamoros et al. (2009) included several types of wetland constructs that were shown to be relatively efficient for the elimination of furosemide in recent literature (Ahmed et al. 2017; Machado et al. 2017). In the case of Seine-Centre, the purification process, which includes biofiltration, nitrification and denitrification processes, also promoted an efficient elimination of furosemide.

**Table 3.** Concentrations (in ng.L<sup>-1</sup>) of furosemide in WWTP effluents. (BLACK AND WHITE TABLE)

<Table 3>

*Tracol & Duchemin 2007<sup>1</sup>; Celic et al. 2019<sup>2</sup>; Santos et al. 2013<sup>3</sup>; Castiglioni et al. 2018<sup>4</sup>; Giebułtowski et al. 2016<sup>5</sup>; Huber et al. 2016<sup>6</sup>; Frieberg 2018<sup>7</sup>; Diaz-Sosa et al. 2020<sup>8</sup>; Papageorgiou et al. 2016<sup>9</sup>; Kleywegt et al. 2016<sup>10</sup>; Estrada-Arriaga et al. 2016<sup>11</sup>; Afssa et al. 2020<sup>12</sup>; Hanamoto et al. 2018<sup>13</sup>; Al-Odaini et al. 2013<sup>14</sup>.*

In spite of a good elimination rate of furosemide and its TPs saluamine and pyridinium of furosemide (70 to 90% removal), several hundreds of nanograms per liter still remained in the wastewater effluent. Many authors reported significant concentrations of furosemide at the outlet of wastewater treatment plants (table 3). Concentrations exceeding 10 µg.L<sup>-1</sup> were even found in a few studies (22300 ng.L<sup>-1</sup> in De vieno et al. (2017), 26000 ng.L<sup>-1</sup> in Vymazal et al. (2017), and 11000 ng.L<sup>-1</sup> in Rozman et al. (2017)).

WWTP effluents concentrations not only depend on the influent concentrations, but also on the treatment processes used within each WWTP. In some cases, furosemide concentrations can be higher at the WWTP outlet than at the inlet (Kleywegt et al. 2016). Due to the fact that an important fraction of furosemide is excreted as a glucuro-conjugated form (Yang et al. 2006), and thus not taken into account during the analysis, this difference could be assigned to the deconjugation occurring during

the WWTP treatment, leading to an increased concentration of the native, unchanged form of furosemide at the outlet. It cannot be excluded that in our raw samples, furosemide concentrations could be underestimated for similar reasons, and consequently, the removal percentage as well. Overall, the WWTP appeared to be relatively effective in removing furosemide as well as its TPs, thus showing that there was no additional production of the targeted TPs during the biological process.

### 3.2. Occurrence in residential care home wastewater

The concentration of furosemide and its TPs was then evaluated in RCH wastewater samples (Table 4). We anticipated this type of sample to be highly-charged in furosemide, as this medication is frequently prescribed at high doses to elderly people affected by chronic and age-related diseases such as hypertension and heart failure. Effluents from three different RCH were analyzed. In all three samples, significant amounts of furosemide were found, and to a lesser extent, of pyridinium of furosemide and saluamine, but their concentrations and proportions relative to each other were variable.

**Table 4.** Concentration of furosemide, saluamine and pyridinium of furosemide in residential care homes wastewater in dissolved and particulate phases. (BLACK AND WHITE TABLE).

<Table 4>

Furosemide concentration variation across the three sites could be directly related to their hosting capacity. The highest furosemide concentration was found in RCH2 which also had the highest hosting capacity and on the contrary, the lowest concentration was observed in RCH3 which also has the lowest number of beds. On the other hand, Gomez-Canela et al. (2019) showed that there were quite large furosemide concentration variations (of a few thousand nanograms per liter) within the same institution over several samples, thus making it more relevant in this case to discuss the orders of magnitude. The concentrations of furosemide in the RCH1 and RCH2 samples were quite close, but the

concentration of saluamine was 2-fold higher in RCH2. The higher proportion of saluamine in RCH2 could be explained by a greater biological activity which could lead to a more important biodegradation of furosemide (Laurencé et al. 2014; Olvera-Vargas et al. 2016). The proportion of pyridinium of furosemide was also much lower in RCH2, implying a preferential transformation of furosemide to saluamine in this case. This difference could be explained by the neutral pH (7.10 at 20°C) indeed the formation of saluamine is promoted in acidic conditions (Bundgaard et al. 1988; Andreasen et al. 1982).

Few studies have investigated the presence of furosemide in non-treated effluents of RCH. Gomez-Canela et al. (2019) reported concentrations up to 3200 ng.L<sup>-1</sup> and 3400 ng.L<sup>-1</sup> in two different senior residence effluents in Spain with a capacity of 103 and 96 beds, respectively, and Nagarnaik et al. (2010) found 1031 ng.L<sup>-1</sup> in nursing care facility effluents which were considerably lower than the concentrations in this study for an equivalent hosting capacity. Slightly higher concentrations (median 6120 ng.L<sup>-1</sup> and maximum 25100 ng.L<sup>-1</sup>) were found by Kleywegt et al. (2016) in a long care center for senior residents and other clients in Canada but with a much larger capacity (289 beds).

Furosemide and its major degradation products were also analyzed in the solid fraction of RCH1 and RCH2 samples (Table 4). Although the three compounds were difficult to analyze because of an important matrix effect, significant concentrations (several hundreds of nanograms per gram) were found.

Saluamine has only been detected once in natural sediments doped with furosemide (Li et al. 2014) but our present work uncovers for the first time the presence of pyridinium of furosemide in the solid fraction. In sediments, furosemide concentrations of 7, 98, and 350 ng.g<sup>-1</sup> were reported (Ferreira Da Silva et al. (2011), Ferrari et al. (2011), and Björklund et al. (2016) respectively). The concentrations obtained in our study are closer to those obtained in WWTP sludge, where the concentrations reached up to 686 ng.g<sup>-1</sup> (Huber et al. 2016) and 3602 ng.g<sup>-1</sup> (Salgado et al. 2011). These values indicate that a



significant fraction of furosemide is adsorbed onto the particulate phase. The partition coefficients between the liquid and solid phase ( $K_d$ ) are good indicators of the compounds affinity with the solid phase. The  $K_d$  values of the suspended matter of the samples RCH1 and RCH2 were 7.7 and 5.4 L.kg<sup>-1</sup> respectively. Jelic et al. (2012) found a  $K_d$  of 43 L.kg<sup>-1</sup> for furosemide in the solid phase of WWTP effluent which is 5 to 8 fold higher than the values found in this study. The  $K_d$  value is very dependent on the matrix considered. For example, in sediments, Björklund et al. (2016) found a  $K_d$  of 2517 L.kg<sup>-1</sup> which shows a very good affinity of furosemide for sediments whereas Jelic et al. (2012) found a lower  $K_d$  in thickened sludge (127 L.kg<sup>-1</sup>) and digested sludge (110 L.kg<sup>-1</sup>). In a Membrane BioReactor (MBR) or Anaerobic-Anoxic-Aerobic system (A2O) -treated sludge, Park et al. (2017) obtained a  $K_d$  of 2.1, which is quite low in comparison. Narumya et al. (2013) showed a decrease of the  $K_d$  for furosemide before and after sludge digestion. In sample RCH1, the  $K_d$  value of pyridinium of furosemide (12.5 L.kg<sup>-1</sup>) was quite close to the one of furosemide, but was much lower in RCH2 (0.36 L.kg<sup>-1</sup>), which shows a much better affinity of furosemide for the solid phase in sample 1.  $K_d$  values for saluamine were more important (103.7 and 18.3 L.kg<sup>-1</sup>), meaning that a significant fraction of saluamine was adsorbed on the solid phase. Thus, total saluamine load in wastewater could be largely underestimated when only performing the conventional analysis of the dissolved phase.

### **3.3. Advanced oxidation in pure water and in residential care houses wastewater**

#### **3.3.1. Oxidation of furosemide in ultrapure water**

The partial resistance of hundreds of compounds to conventional wastewater treatments highlights the need to go further in the degradation processes. We chose two AOPs which have the potential to upgrade WWTPs worldwide and are already used in some countries like Japan or Switzerland (Prasse et al, 2015). Chlorination oxidation experiments were also performed (Figures S3, S4 and Table S1). Both oxidation processes have been used for a long time for disinfection and the oxidants produced are likely to react with furosemide. The photodegradation of furosemide has been known for several

decades. UV irradiation leads to the substitution of chlorine by a hydroxyl group, to the hydrolysis of the furfuryl group, that results in the production of furfural and saluamine, or the oxidation of the sulfamoyl group (Bundgaard et al. 1988; Moore & Burt 1981). Furosemide is also degraded in sunlight or artificial laboratory light which makes it a good candidate for UV degradation (Starling et al. 2019). The use of ozonation has increased in recent years for the treatment of wastewaters due to its effectiveness in removing organic compounds. As an electrophilic agent, the presence of electron-rich moieties such as aromatic compounds determines the reactivity with ozone (Lim et al. 2022). According to Zoumpouli et al. (2021), furosemide is expected to present an important reactivity with ozone due to the presence of the furan ring and the aniline group. However, the potential formation of furosemide TPs, especially pyridinium of furosemide, remains unknown for AOPs. AOPs are known to generate a variety of TPs that are potentially hazardous to ecosystems, and whose identification is a challenge due to the lack of analytical standards. Furosemide degradation was thus first investigated in ultrapure water spiked with furosemide. Pyridinium of furosemide was quantified after the oxidation, and aliquots were also taken to monitor the formation of unknown TPs by high-resolution mass spectrometry.

As anticipated, the two AOPs were effective in degrading furosemide under our experimental conditions. Ozonation was the most efficient (see supplementary information Figures S1, S2).

**Table 5.** Pyridinium of furosemide concentration and furosemide percentage of conversion during oxidation experiments. (BLACK AND WHITE TABLE)

<Table 5>

The pyridinium of furosemide was produced during both oxidation experiments (table 5). During the UV/H<sub>2</sub>O<sub>2</sub> oxidation, the aliquot taken at 210 minutes already presented a few micrograms per liter of pyridinium with 1.5% conversion rate of furosemide. This rate reached 5.8% after 300 minutes.

However, during this 5h-time lapse, less than 10% of furosemide was degraded, which means that pyridinium formation is slow, probably because it involves reaction intermediates (Olvera-vargas et al. 2016; Zoumpouli et al. 2021). Pyridinium of furosemide was also produced by ozonation within a few minutes. Its production started immediately after ozone addition, and its concentration increased gradually until 4 minutes, with a furosemide conversion rate of 3.2% which remained unchanged after 10 minutes, in conjunction with a total consumption of ozone and furosemide concentration reaching a plateau. As in the UV/H<sub>2</sub>O<sub>2</sub> experiment, pyridinium of furosemide did not seem to be degraded by the ozone treatment. Pyridinium of furosemide was also detected after chlorination (35 mgCl<sub>2</sub>.L<sup>-1</sup>) of furosemide (3 mg.L<sup>-1</sup>), with a maximum conversion rate of 1.4% after only 2 min of reaction time, but contrary to the other oxidation processes, its concentration quickly decreased after 5 min, probably because of the formation of chlorinated derivatives (Table S1). This result might be of importance for the quality of drinking water produced from resources impacted by furosemide, even though the fast degradation of pyridinium of furosemide seems to indicate a low risk of exposure.

### **3.3.2. Oxidation of residential care houses wastewater**

In order to get insights on the AOPs efficiencies with furosemide heavily-loaded real samples and on the formation of TPs in the presence of wastewater matrix components, we next subjected the effluents from the three RCH samples to the two AOPs (figure 2). The overall abatement was quite variable from one sample to another and for the different compounds, depending on the water parameters and the oxidation process used. Ozonation was very effective at removing the three pollutants, with an important reduction (below quantification limit) of furosemide and pyridinium of furosemide for the three tested samples (figure 2). As for chlorination (supplementary data, figure S4, SI), abatement rates of almost 100% were observed for furosemide and pyridinium. These results were thus consistent with the experiments conducted with ultrapure water. Although the lifetime of ozone in WWTP effluent is shorter because of background absorptions (Lim et al. 2022), ozone was injected continuously in the samples and therefore furosemide reached higher removals than in the ultra-pure

water experiment (figure S2, SI). This could also explain why pyridinium of furosemide was not observed in the ozonated samples, a higher ozone dose probably leading to its decomposition.

### <Figure 2>

**Figure 2.** Effectiveness of a) UV/H<sub>2</sub>O<sub>2</sub> and b) ozonation on residential care home (RCH) discharge water. The bars represent removal efficiency in percentage calculated from initial concentration for each compound (dark grey = furosemide, medium grey = saluamine, light grey = Pyridinium of furosemide). Error bars represent standard error. For Ozonation, the samples were exposed for 1.5h with 2L.h<sup>-1</sup> O<sub>3</sub>. For UV/H<sub>2</sub>O<sub>2</sub>, the samples were exposed for 5H to (0.375xCOD value)mg.L<sup>-1</sup> H<sub>2</sub>O<sub>2</sub>. (BLACK AND WHITE FIGURE)

Our results are in accordance with those reported in the litterature. Munoz et al. (2009) showed a 100% removal of furosemide and Gomez et al. (2008) a removal of 99% for both O<sub>3</sub> and O<sub>3</sub> with H<sub>2</sub>O<sub>2</sub> treatments. Ikonen et al. (2021) obtained a reduction of 99.7% of furosemide in wastewater effluents and Lee et al. (2012) observed a reduction to below their detection limits with a dose of 2 mg O<sub>3</sub>.L<sup>-1</sup>. Experiments combining ozonation with other treatments such as biological activated carbon (Reungoat et al. 2012) or ultrasonication (Ibanez et al. 2013) did not significantly improve the removal efficiency and showed that the ozone treatment is responsible for most of the elimination of furosemide.

Contrary to furosemide and pyridinium, saluamine was less removed and its removal rate was variable depending on the sample. Saluamine was well degraded in the RCH3 sample, but higher concentrations of saluamine were observed from RCH1 and RCH2 samples after ozonation. This production of saluamine could be explained by the conversion of both furosemide and pyridinium to saluamine in the conditions of these samples (e.g., different initial concentrations, competition reactions of ozone with wastewater constituents such as nitrites or organic matter leading to a decrease in saluamine

decomposition). Contrary to experiments in ultrapure water, no additional formation of pyridinium was observed, and all pyridinium already present in the RCH samples was degraded by ozone.

Furosemide removal by UV/H<sub>2</sub>O<sub>2</sub> was between 70 and 80% for all the samples, which is very close to the 82% removal found by Jie (2012) in hospital wastewater. However, the effectiveness of UV/H<sub>2</sub>O<sub>2</sub> on furosemide elimination is still uncertain. Park et al. (2017) described the use of UV irradiation alone as not very effective on pharmaceuticals and personal care products, and only achieved 23% of removal of furosemide in WWTP post-treatment. However, as the treatment was performed during a purification process in a WWTP, the suspended particles can interfere and reduce the effectiveness of UV as compared to laboratory experiments performed with filtered samples. With UV/H<sub>2</sub>O<sub>2</sub>, Ikonen et al. (2021) found no removal of furosemide while Singh et al. (2015) observed a complete degradation, but with a lamp 8 times more powerful (Joule/cm<sup>2</sup>). The efficiency of furosemide degradation by UV/H<sub>2</sub>O<sub>2</sub> thus seems to be strongly dependent on the UV irradiation intensity. Similarly to ozonation and chlorination, large differences of removal were observed between the three samples for the two degradation products.

### **3.4. Generation of furosemide new transformation by-products**

As these advanced oxidation processes have been reported to generate numerous by-products by reaction with organic contaminants, many of them being often potentially more toxic than their parent molecules (reviewed in Prasse et al. 2015), the formation of other transformation products of furosemide was investigated after the oxidation experiments by high-resolution mass spectrometry (HRMS). Noteworthy, two other by-products of furosemide were identified: one during chlorination (Figure 2 e.) and one during UV/H<sub>2</sub>O<sub>2</sub> treatment (Figure 3f.).

<Figure 3>

**Figure 3.** Molecular Structure of a. furosemide (m/z 329.74) and its transformation products. b. Pyridinium of furosemide (m/z 328.73), c. Furfural (m/z 96.08), d. Saluamine (m/z 250.66). Transformation products identified by high-resolution mass spectrometry analysis: e. Chlorination by product m/z 276.99; f. UV/H<sub>2</sub>O<sub>2</sub> by product m/z 311.03. (BLACK AND WHITE FIGURE).

To our knowledge, the product 3.e. was not clearly mentioned in the literature. However, Aalizadeh et al. (2018) found a by-product of furosemide of similar mass (m/z 276.99) after ozonation but they did not identify the structure. On the other hand, the compound 3.f. from UV/H<sub>2</sub>O<sub>2</sub> oxidation has already been observed as a photodegradation product of furosemide after exposure to fluorescent lamps, and its structure was identified with <sup>1</sup>H-NMR (Katsura et al. 2015). Its structure resembles the one of furosemide with m/z 311.03, except that the chlorine has been replaced by a hydroxyl group. In Jakimska et al. (2014), this product has been found after photodegradation experiments carried out on river water samples and was considered as one of the most persistent transformation products. These authors then found it in WWTP influents and effluents. In our study, the 3.f product formation kinetics was studied (see supplementary data, figure S4), showing that its degradation occurred after one hour.

Jakimska et al. (2014) and Katsura et al. (2015) also identified saluamine and other degradation products (m/z 352, m/z 555, m/z 231, m/z 295, m/z 215) which not only corresponded to losses or substitution of Cl groups on furosemide or saluamine but also sometimes to recombination of fragments as in the study of Della-Greca et al. (2004), which identified a dimer (m/z 623) after photodegradation of furosemide. These by-products were not found in our oxidation experiments. The Della-Greca et al. (2004) dimer is larger than the other molecules and very polar: it is possible that our extraction method did not retain it. Moreover, these degradation products were observed after UV irradiation alone, thus it is conceivable that by combining UV with H<sub>2</sub>O<sub>2</sub> treatment, they were further degraded as well, as described by Starling et al. (2017). Indeed, these authors found similar

degradation products of furosemide after UV and UV/H<sub>2</sub>O<sub>2</sub> treatment, but they did not remain stable after H<sub>2</sub>O<sub>2</sub> addition. If these compounds were present in our experiments, they may be below the limit of detection.

The m/z 276 and m/z 288 transformation products obtained after ozonation have also been mentioned in the literature but their structure could not be elucidated (Aalizadeh et al. 2019). In Zoumpouli et al. (2021), a molecule with a pyridinium structure (m/z 328) and a saluamine-like molecule (m/z 265) were also obtained by ozonation of the furan ring.

Taken together, our data show that ozonation, UV/H<sub>2</sub>O<sub>2</sub> and chlorination degrade furosemide, generating several different by-products. High oxidant doses were chosen to maximize the formation of transformation products in order to enhance their analytical detection and identification, so their presence should still be investigated at realistic doses (e.g., at a specific ozone dose < 1 mgO<sub>3</sub>/mgDOC) and at the industrial scale (Guillossou et al, 2020). Besides saluamine production, the pyridinium of furosemide was formed under all three conditions but was only degraded by chlorination. Furthermore, chlorination also generated a product (m/z 276) never described before, but which was degraded after a few hours. Another product (m/z 311), resulting from photodegradation and previously reported (Katsura et al. 2015), was also identified.

#### **4. Conclusion**

This study provides new findings on the occurrence and fate of furosemide from its origin to wastewater treatment plants. The known degradation products of furosemide, saluamine and pyridinium of furosemide were searched and quantified for the first time in real samples (WWTP inlets and outlets and RCH wastewaters). The Seine-Centre WWTP showed a good removal of furosemide (>70%), saluamine (>80%) and pyridinium (>90%). However, concentrations of several hundred ng.L<sup>-1</sup>

were found after treatment and are released into the aquatic environment. These concentrations sometimes exceed 10  $\mu\text{g.L}^{-1}$  in the literature for furosemide and may pose a risk to ecosystems.

To limit the release of these pharmaceutical compounds in the environment, we investigated AOPs and the possible production of TPs. UV/H<sub>2</sub>O<sub>2</sub>, chlorination and ozonation were first shown to be effective in degrading furosemide in ultrapure water. In RCH wastewater, ozonation and chlorination showed complete degradation of furosemide and pyridinium, but saluamine was still present in the samples after treatment due to a possible production. UV/H<sub>2</sub>O<sub>2</sub> showed variable removal rates depending on the compounds. Although these treatments appeared quite effective, new TPs were identified following the chlorination (m/z 276) and ozonation (m/z 311) processes. The elimination of pollutants is used as a criterion for the evaluation of AOPs, but the presence of TPs should also be considered. Before concluding on the effectiveness of these advanced treatments, the toxicity and persistence of the end-products should be investigated. Thus, our study underlines the necessity and the relevance to use approaches such as non-targeted analysis for the detection of emerging pollutants, to unveil new TPs, combined with toxicity assessments in order to improve the characterization and design of wastewater treatment processes. Keeping in mind that these data alone do not provide information on their toxic risk and cocktail effects, our study highlights the need to develop an integrative strategy coupling state-of-the art chemical analysis techniques with ecotoxicological tests with defined end-points.

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## References

- Aalizadeh, R., Nika, M.-C., Thomaidis, N.S., 2019. Development and application of retention time prediction models in the suspect and non-target screening of emerging contaminants. *Journal of Hazardous Materials* 363, 277–285. <https://doi.org/10.1016/j.jhazmat.2018.09.047>
- Abbott, L.M., Kovacic, J., 2008. The pharmacologic spectrum of furosemide. *Journal of Veterinary Emergency and Critical Care* 18, 26–39. <https://doi.org/10.1111/j.1476-4431.2007.00267.x>
- Afsa, S., Hamden, K., Lara Martin, P.A., Mansour, H.B., 2020. Occurrence of 40 pharmaceutically active compounds in hospital and urban wastewaters and their contribution to Mahdia coastal seawater contamination. *Environ Sci Pollut Res* 27, 1941–1955. <https://doi.org/10.1007/s11356-019-06866-5>
- Ahmed, M.B., Zhou, J.L., Ngo, H.H., Guo, W., Thomaidis, N.S., Xu, J., 2017. Progress in the biological and chemical treatment technologies for emerging contaminant removal from wastewater: A critical review. *Journal of Hazardous Materials* 323, 274–298. <https://doi.org/10.1016/j.jhazmat.2016.04.045>

622 Al-Odaini, N.A., Zakaria, M.P., Yaziz, M.I., Surif, S., Abdulghani, M., 2013. The occurrence of human  
623 pharmaceuticals in wastewater effluents and surface water of Langat River and its tributaries,  
624 Malaysia. *International Journal of Environmental Analytical Chemistry* 93, 245–264.  
625 <https://doi.org/10.1080/03067319.2011.592949>

626 Al-Omar, I.A., Al-Ashban, R.M., Shah, A.H., 2009. Toxicity Studies on 4-Chloro-5-Sulfamoylanthranilic  
627 Acid the Degradation Product of a Loop Diuretic Furosemide. *Research Journal of Pharmacology* 63–77, 4.

628 Andreasen, F., Botker, H., Lorentzen, K., 1982. In vitro studies on the hydrolysis of frusemide in  
629 gastrointestinal juices. *British Journal of Clinical Pharmacology* 14, 306–309.  
630 <https://doi.org/10.1111/j.1365-2125.1982.tb01984.x>

631 Aymerich, I., Acuña, V., Barceló, D., García, M.J., Petrovic, M., Poch, M., Rodriguez-Mozaz, S., Rodríguez-  
632 Roda, I., Sabater, S., von Schiller, D., Corominas, Ll., 2016. Attenuation of pharmaceuticals and their  
633 transformation products in a wastewater treatment plant and its receiving river ecosystem. *Water*  
634 *Research* 100, 126–136. <https://doi.org/10.1016/j.watres.2016.04.022>

635 Badia-Fabregat, M., Lucas, D., Gros, M., Rodríguez-Mozaz, S., Barceló, D., Caminal, G., Vicent, T., 2015.  
636 Identification of some factors affecting pharmaceutical active compounds (PhACs) removal in real  
637 wastewater. Case study of fungal treatment of reverse osmosis concentrate. *Journal of Hazardous*  
638 *Materials* 283, 663–671. <https://doi.org/10.1016/j.jhazmat.2014.10.007>

639 Baken, K.A., Sjerps, R.M.A., Schriks, M., van Wezel, A.P., 2018. Toxicological risk assessment and  
640 prioritization of drinking water relevant contaminants of emerging concern. *Environment*  
641 *International* 118, 293–303. <https://doi.org/10.1016/j.envint.2018.05.006>

642 Banjac, Z., Ginebreda, A., Kuzmanovic, M., Marcé, R., Nadal, M., Riera, J.M., Barceló, D., 2015. Emission  
643 factor estimation of ca. 160 emerging organic microcontaminants by inverse modeling in a  
644 Mediterranean river basin (Llobregat, NE Spain). *Science of The Total Environment* 520, 241–252.  
645 <https://doi.org/10.1016/j.scitotenv.2015.03.055>

646 Baresel, C., Harding, M., Junestedt, C., 2019. Removal of pharmaceutical residues from municipal  
647 wastewater using UV/H<sub>2</sub>O<sub>2</sub>. Report from Swedish Environmental Research Institute 2019 28

648 Besse, J.-P., Garric, J., 2008. Human pharmaceuticals in surface waters. *Toxicology Letters* 176, 104–123.  
649 <https://doi.org/10.1016/j.toxlet.2007.10.012>

650 Björklund, E., Svahn, O., Bak, S., Bekoe, S.O., Hansen, M., 2016. Pharmaceutical Residues Affecting the  
651 UNESCO Biosphere Reserve Kristianstads Vattenrike Wetlands: Sources and Sinks. *Archives of*  
652 *Environmental Contamination and Toxicology* 71, 423–436. <https://doi.org/10.1007/s00244-016-0303-7>

653 Bundgaard, H., Nørgaard, T., Nielsen, N.M., 1988. Photodegradation and hydrolysis of furosemide and  
654 furosemide esters in aqueous solutions. *International Journal of Pharmaceutics* 42, 217–224.  
655 [https://doi.org/10.1016/0378-5173\(88\)90178-0](https://doi.org/10.1016/0378-5173(88)90178-0)

656 Cantwell, M.G., Katz, D.R., Sullivan, J.C., Shapley, D., Lipscomb, J., Epstein, J., Juhl, A.R., Knudson, C.,  
657 O'Mullan, G.D., 2018. Spatial patterns of pharmaceuticals and wastewater tracers in the Hudson  
658 River Estuary. *Water Research* 137, 335–343. <https://doi.org/10.1016/j.watres.2017.12.044>

659 Castiglioni, S., Bagnati, R., Fanelli, R., Pomati, F., Calamari, D., Zuccato, E., 2006. Removal of  
660 Pharmaceuticals in Sewage Treatment Plants in Italy. *Environmental Science & Technology* 40,  
661 357–363. <https://doi.org/10.1021/es050991m>

662 Castiglioni, S., Davoli, E., Riva, F., Palmiotto, M., Camporini, P., Manenti, A., Zuccato, E., 2018. Mass  
663 balance of emerging contaminants in the water cycle of a highly urbanized and industrialized area  
664 of Italy, *Water Research* 131 287-298. <https://doi.org/10.1016/j.watres.2017.12.047>

665 Čelić, M., Gros, M., Farré, M., Barceló, D., Petrović, M., 2019. Pharmaceuticals as chemical markers of  
666 wastewater contamination in the vulnerable area of the Ebro Delta (Spain). *Science of The Total*  
667 *Environment* 652, 952–963. <https://doi.org/10.1016/j.scitotenv.2018.10.290>

668 Chiffre, A., Degiorgi, F., Buleté, A., Spinner, L., Badot, P.-M., 2016. Occurrence of pharmaceuticals in  
669 WWTP effluents and their impact in a karstic rural catchment of Eastern France. *Environmental*  
670 *Science and Pollution Research* 23, 25427–25441. <https://doi.org/10.1007/s11356-016-7751-5>

671 Cizmas, L., Sharma, V.K., Gray, C.M., McDonald, T.J., 2015. Pharmaceuticals and personal care products  
672 in waters: occurrence, toxicity, and risk. *Environmental Chemistry Letters* 13, 381–394.  
673 <https://doi.org/10.1007/s10311-015-0524-4>

674 Collado, N., Rodriguez-Mozaz, S., Gros, M., Rubirola, A., Barceló, D., Comas, J., Rodriguez-Roda, I.,  
675 Buttiglieri, G., 2014. Pharmaceuticals occurrence in a WWTP with significant industrial contribution  
676 and its input into the river system. *Environmental Pollution* 185, 202–212.  
677 <https://doi.org/10.1016/j.envpol.2013.10.040>

678 Cruz-Morató, C., Lucas, D., Llorca, M., Rodriguez-Mozaz, S., Gorga, M., Petrovic, M., Barceló, D., Vicent,  
679 T., Sarrà, M., Marco-Urrea, E., 2014. Hospital wastewater treatment by fungal bioreactor: Removal  
680 efficiency for pharmaceuticals and endocrine disruptor compounds. *Science of The Total*  
681 *Environment* 493, 365–376. <https://doi.org/10.1016/j.scitotenv.2014.05.117>

682 Della Greca, M., Iesce, M.R., Previtera, L., Rubino, M., Temussi, F., 2004. A new photoproduct of the  
683 drug furosemide in aqueous media. *Environmental Chemistry Letters* 2, 155–158.  
684 <https://doi.org/10.1007/s10311-004-0080-9>

685 Delli Compagni, R., Gabrielli, M., Polesel, F., Turolla, A., Trapp, S., Vezzaro, L., Antonelli, M., 2020. Risk  
686 assessment of contaminants of emerging concern in the context of wastewater reuse for irrigation:  
687 An integrated modelling approach. *Chemosphere* 242, 125185.  
688 <https://doi.org/10.1016/j.chemosphere.2019.125185>

689 Diaz-Sosa, V.R., Tapia-Salazar, M., Wanner, J., Cardenas-Chavez, D.L., 2020. Monitoring and Ecotoxicity  
690 Assessment of Emerging Contaminants in Wastewater Discharge in the City of Prague (Czech  
691 Republic). *Water* 12, 1079. <https://doi.org/10.3390/w12041079>

692 Estrada-Arriaga, E.B., Cortés-Muñoz, J.E., González-Herrera, A., Calderón-Mólgora, C.G., de Lourdes  
693 Rivera-Huerta, Ma., Ramírez-Camperos, E., Montellano-Palacios, L., Gelover-Santiago, S.L., Pérez-  
694 Castrejón, S., Cardoso-Vigueros, L., Martín-Domínguez, A., García-Sánchez, L., 2016. Assessment  
695 of full-scale biological nutrient removal systems upgraded with physico-chemical processes for the  
696 removal of emerging pollutants present in wastewaters from Mexico. *Science of The Total*  
697 *Environment* 571, 1172–1182. <https://doi.org/10.1016/j.scitotenv.2016.07.118>

698 Feo, M.L., Bagnati, R., Passoni, A., Riva, F., Salvagio Manta, D., Sprovieri, M., Traina, A., Zuccato, E.,  
699 Castiglioni, S., 2020. Pharmaceuticals and other contaminants in waters and sediments from

700 Augusta Bay (southern Italy). *Science of The Total Environment* 739, 139827.  
 701 <https://doi.org/10.1016/j.scitotenv.2020.139827>

702 Ferrari, F., Gallipoli, A., Balderacchi, M., Ulaszewska, M.M., Capri, E., Trevisan, M., 2011. Exposure of the  
 703 Main Italian River Basin to Pharmaceuticals. *Journal of Toxicology* 2011, 1–11.  
 704 <https://doi.org/10.1155/2011/989270>

705 Ferreira da Silva, B., Jelic, A., López-Serna, R., Mozeto, A.A., Petrovic, M., Barceló, D., 2011. Occurrence  
 706 and distribution of pharmaceuticals in surface water, suspended solids and sediments of the Ebro  
 707 river basin, Spain. *Chemosphere* 85, 1331–1339.  
 708 <https://doi.org/10.1016/j.chemosphere.2011.07.051>

709 Frieberg, K., 2018. Effects of effluent wastewater in developing zebrafish (*Danio rerio*). Report from  
 710 Uppsala University.

711 Giebułtowicz, J., Stankiewicz, A., Wroczyński, P., 2016. Occurrence of cardiovascular drugs in the  
 712 sewage-impacted Vistula River and in tap water in the Warsaw region (Poland). *Environmental*  
 713 *Science and Pollution Research* 23, 24337–24349. <https://doi.org/10.1007/s11356-016-7668-z>

714 Gómez, M.J., Martínez Bueno, M.J., Agüera, A., Hernando, M.D., Fernández-Alba, A.R., Mezcua, M.,  
 715 2008. Evaluation of ozone-based treatment processes for wastewater containing  
 716 microcontaminants using LC-QTRAP-MS and LC-TOF/MS. *Water Science and Technology* 57, 41–  
 717 48. <https://doi.org/10.2166/wst.2008.809>

718 Gómez-Canela, C., Sala-Comorera, T., Pueyo, V., Barata, C., Lacorte, S., 2019. Analysis of 44  
 719 pharmaceuticals consumed by elderly using liquid chromatography coupled to tandem mass  
 720 spectrometry. *Journal of Pharmaceutical and Biomedical Analysis* 168, 55–63.  
 721 <https://doi.org/10.1016/j.jpba.2019.02.016>

722 Gros, M., Petrović, M., Ginebreda, A., Barceló, D., 2010. Removal of pharmaceuticals during wastewater  
 723 treatment and environmental risk assessment using hazard indexes. *Environment International* 36,  
 724 15–26. <https://doi.org/10.1016/j.envint.2009.09.002>

725 Guillosoy, R., Le Roux, J., Brosillon, S., Mailler, R., Vulliet, E., Morlay, C., Nauleau, F., Rocher, V., Gaspéri,  
 726 J., 2020. Benefits of ozonation before activated carbon adsorption for the removal of organic  
 727 micropollutants from wastewater effluents. *Chemosphere* 245, 125530.  
 728 <https://doi.org/10.1016/j.chemosphere.2019.125530>  
 729 Guillosoy, R., Le Roux, J., Mailler, R., Vulliet, E., Morlay, C., Nauleau, F., Gasperi, J., Rocher, V., 2019.  
 730 Organic micropollutants in a large wastewater treatment plant: What are the benefits of an  
 731 advanced treatment by activated carbon adsorption in comparison to conventional treatment?  
 732 *Chemosphere* 218, 1050–1060. <https://doi.org/10.1016/j.chemosphere.2018.11.182>  
 733 Guo, Y., Yang, L., Wang, X., 2012. The Application and Reaction Mechanism of Catalytic Ozonation in  
 734 Water Treatment. *Journal of Environmental & Analytical Toxicology* 02.  
 735 <https://doi.org/10.4172/2161-0525.1000150>  
 736 Hanamoto, S., Nakada, N., Yamashita, N., Tanaka, H., 2018. Source estimation of pharmaceuticals based  
 737 on catchment population and in-stream attenuation in Yodo River watershed, Japan. *Science of*  
 738 *The Total Environment* 615, 964–971. <https://doi.org/10.1016/j.scitotenv.2017.10.013>  
 739 Hezari, M., Davis, P.J., 1992. N-dealkylation of Furosemide to yield the mammalian metabolite CSA using  
 740 *Cunninghamella elegans*. *Drug Metabolism and Disposition* 20, 92.  
 741 Hijnen, W.A.M., Beerendonk, E.F., Medema, G.J., 2006. Inactivation credit of UV radiation for viruses,  
 742 bacteria and protozoan (oo)cysts in water: A review. *Water Research* 40, 3–22.  
 743 <https://doi.org/10.1016/j.watres.2005.10.030>  
 744 Huber, S., Remberger, M., Kaj, L., Schlabach, M., Jörundsdóttir, H.Ó., Vester, J., Arnórsson, M.,  
 745 Mortensen, I., Schwartzon, R., Dam, M., 2016. A first screening and risk assessment of  
 746 pharmaceuticals and additives in personal care products in waste water, sludge, recipient water  
 747 and sediment from Faroe Islands, Iceland and Greenland. *Science of The Total Environment* 562,  
 748 13–25. <https://doi.org/10.1016/j.scitotenv.2016.03.063>

749 Huerta-Fontela, M., Galceran, M.T., Ventura, F., 2011. Occurrence and removal of pharmaceuticals and  
 750 hormones through drinking water treatment. *Water Research* 45, 1432–1442.  
 751 <https://doi.org/10.1016/j.watres.2010.10.036>  
 752 Ibáñez, M., Gracia-Lor, E., Bijlsma, L., Morales, E., Pastor, L., Hernández, F., 2013. Removal of emerging  
 753 contaminants in sewage water subjected to advanced oxidation with ozone. *Journal of Hazardous*  
 754 *Materials* 260, 389–398. <https://doi.org/10.1016/j.jhazmat.2013.05.023>  
 755 Ikonen, J., Nuutinen, I., Niittynen, M., Hokajärvi, A.-M., Pitkänen, T., Antikainen, E., Miettinen, I.T., 2021.  
 756 Presence and Reduction of Anthropogenic Substances with UV Light and Oxidizing Disinfectants in  
 757 Wastewater—A Case Study at Kuopio, Finland. *Water* 13, 360.  
 758 <https://doi.org/10.3390/w13030360>  
 759 Jakimska, A., Śliwka-Kaszyńska, M., Reszczyńska, J., Namieśnik, J., Kot-Wasik, A., 2014. Elucidation of  
 760 transformation pathway of ketoprofen, ibuprofen, and furosemide in surface water and their  
 761 occurrence in the aqueous environment using UHPLC-QTOF-MS. *Analytical and Bioanalytical*  
 762 *Chemistry* 406, 3667–3680. <https://doi.org/10.1007/s00216-014-7614-1>  
 763 Jelic, A., Fatone, F., Di Fabio, S., Petrovic, M., Cecchi, F., Barcelo, D., 2012. Tracing pharmaceuticals in a  
 764 municipal plant for integrated wastewater and organic solid waste treatment. *Science of The Total*  
 765 *Environment* 433, 352–361. <https://doi.org/10.1016/j.scitotenv.2012.06.059>  
 766 Jelic, A., Gros, M., Ginebreda, A., Cespedes-Sánchez, R., Ventura, F., Petrovic, M., Barcelo, D., 2011.  
 767 Occurrence, partition and removal of pharmaceuticals in sewage water and sludge during  
 768 wastewater treatment. *Water Research* 45, 1165–1176.  
 769 <https://doi.org/10.1016/j.watres.2010.11.010>  
 770 Jie, L., 2012. Potential of advanced wastewater treatment processes to remove pharmaceutical  
 771 compounds under PILLS/SLIK Project.  
 772 Kasprzyk-Hordern, B., Dinsdale, R.M., Guwy, A.J., 2009. The removal of pharmaceuticals, personal care  
 773 products, endocrine disruptors and illicit drugs during wastewater treatment and its impact on the

774 quality of receiving waters. Water Research 43, 363–380.  
 775 <https://doi.org/10.1016/j.watres.2008.10.047>

776 Katsura, S., Yamada, N., Nakashima, A., Shiraishi, S., Furuishi, T., Ueda, H., 2015. Identification of  
 777 Furosemide Photodegradation Products in Water–Acetonitrile Mixture. Chemical and  
 778 Pharmaceutical Bulletin 63, 617–627. <https://doi.org/10.1248/cpb.c15-00122>

779 Kleywegt, S., Pileggi, V., Lam, Y.M., Elises, A., Puddicomb, A., Purba, G., Di Caro, J., Fletcher, T., 2016.  
 780 The contribution of pharmaceutically active compounds from healthcare facilities to a receiving  
 781 sewage treatment plant in Canada: Contribution of pharmaceutically active compounds to STP  
 782 effluent. Environmental Toxicology and Chemistry 35, 850–862. <https://doi.org/10.1002/etc.3124>

783 Kot-Wasik, A., Jakimska, A., Śliwka-Kaszyńska, M., 2016. Occurrence and seasonal variations of 25  
 784 pharmaceutical residues in wastewater and drinking water treatment plants. Environmental  
 785 Monitoring and Assessment 188, 661. <https://doi.org/10.1007/s10661-016-5637-0>

786 Laurencé, C., Rivard, M., Lachaise, I., Bensemhoun, J., Martens, T., 2011. Preparative access to  
 787 transformation products (TPs) of furosemide: a versatile application of anodic oxidation.  
 788 Tetrahedron 67, 9518–9521. <https://doi.org/10.1016/j.tet.2011.10.006>

789 Laurencé, C., Rivard, M., Martens, T., Morin, C., Buisson, D., Bourcier, S., Sablier, M., Oturan, M.A., 2014.  
 790 Anticipating the fate and impact of organic environmental contaminants: A new approach applied  
 791 to the pharmaceutical furosemide. Chemosphere 113, 193–199.  
 792 <https://doi.org/10.1016/j.chemosphere.2014.05.036>

793 Laurencé, C., Zeghib, N., Rivard, M., Lehri-Boufala, S., Lachaise, I., Barau, C., Le Corvoisier, P., Martens,  
 794 T., Garrigue-Antar, L., Morin, C., 2019. A new human pyridinium metabolite of furosemide,  
 795 inhibitor of mitochondrial complex I, is a candidate inducer of neurodegeneration. Biochemical  
 796 Pharmacology 160, 14–23. <https://doi.org/10.1016/j.bcp.2018.12.007>

797 Lee, C.O., Howe, K.J., Thomson, B.M., 2012. Ozone and biofiltration as an alternative to reverse osmosis  
 798 for removing PPCPs and micropollutants from treated wastewater. Water Research 46, 1005–  
 799 1014. <https://doi.org/10.1016/j.watres.2011.11.069>



800 Lee, Y., Kovalova, L., McArdell, C.S., von Gunten, U., 2014. Prediction of micropollutant elimination  
 801 during ozonation of a hospital wastewater effluent. *Water Research* 64, 134–148.  
 802 <https://doi.org/10.1016/j.watres.2014.06.027>  
 803 Li, Z., Maier, M.P., Radke, M., 2014. Screening for pharmaceutical transformation products formed in  
 804 river sediment by combining ultrahigh performance liquid chromatography/high resolution mass  
 805 spectrometry with a rapid data-processing method. *Analytica Chimica Acta* 810, 61–70.  
 806 <https://doi.org/10.1016/j.aca.2013.12.012>  
 807 Lim, S., Shi, J.L., von Gunten, U., McCurry, D.L., 2022. Ozonation of organic compounds in water and  
 808 wastewater: A critical review. *Water Research* 213, 118053.  
 809 <https://doi.org/10.1016/j.watres.2022.118053>  
 810 Machado, A.I., Dordio, A., Fragoso, R., Leitão, A.E., Duarte, E., 2017. Furosemide removal in constructed  
 811 wetlands: Comparative efficiency of LECA and Cork granulates as support matrix. *Journal of*  
 812 *Environmental Management* 203, 422–428. <https://doi.org/10.1016/j.jenvman.2017.08.002>  
 813 Matamoros, V., Arias, C., Brix, H., Bayona, J.M., 2009. Preliminary screening of small-scale domestic  
 814 wastewater treatment systems for removal of pharmaceutical and personal care products. *Water*  
 815 *Research* 43, 55–62. <https://doi.org/10.1016/j.watres.2008.10.005>  
 816 Mazhar, M.A., Khan, N.A., Ahmed, S., Khan, A.H., Hussain, A., Rahisuddin, Changani, F., Yousefi, M.,  
 817 Ahmadi, S., Vambol, V., 2020. Chlorination disinfection by-products in municipal drinking water –  
 818 A review. *Journal of Cleaner Production* 273, 123159.  
 819 <https://doi.org/10.1016/j.jclepro.2020.123159>  
 820 Miklos, D.B., Remy, C., Jekel, M., Linden, K.G., Drewes, J.E., Hübner, U., 2018. Evaluation of advanced  
 821 oxidation processes for water and wastewater treatment – A critical review. *Water Research* 139,  
 822 118–131. <https://doi.org/10.1016/j.watres.2018.03.042>  
 823 Moore, D., Burt, C., 1981. Photosensitization by drugs in surfactant solutions. *Photochemistry and*  
 824 *Photobiology* 34, 431–439.

825 Muñoz, I., Rodríguez, A., Rosal, R., Fernández-Alba, A.R., 2009. Life Cycle Assessment of urban  
 826 wastewater reuse with ozonation as tertiary treatment. *Science of The Total Environment* 407,  
 827 1245–1256. <https://doi.org/10.1016/j.scitotenv.2008.09.029>

828 Munro, K., Martins, C.P.B., Loewenthal, M., Comber, S., Cowan, D.A., Pereira, L., Barron, L.P., 2019.  
 829 Evaluation of combined sewer overflow impacts on short-term pharmaceutical and illicit drug  
 830 occurrence in a heavily urbanised tidal river catchment (London, UK). *Science of The Total*  
 831 *Environment* 657, 1099–1111. <https://doi.org/10.1016/j.scitotenv.2018.12.108>

832 Nagarnaik, P., Batt, A., Boulanger, B., 2010. Concentrations and mass loadings of cardiovascular  
 833 pharmaceuticals in healthcare facility wastewaters. *Journal of Environmental Monitoring* 12, 2112.  
 834 <https://doi.org/10.1039/c0em00216j>

835 Narumiya, M., Nakada, N., Yamashita, N., Tanaka, H., 2013. Phase distribution and removal of  
 836 pharmaceuticals and personal care products during anaerobic sludge digestion. *Journal of*  
 837 *Hazardous Materials* 260, 305–312. <https://doi.org/10.1016/j.jhazmat.2013.05.032>

838 Okoye, C.O., Okeke, E.S., Okoye, K.C., Echude, D., Andong, F.A., Chukwudozie, K.I., Okoye, H.U.,  
 839 Ezeonyejiaku, C.D., 2022. Occurrence and fate of pharmaceuticals, personal care products (PPCPs)  
 840 and pesticides in African water systems: A need for timely intervention. *Heliyon* 8, e09143.  
 841 <https://doi.org/10.1016/j.heliyon.2022.e09143>

842 Olvera-Vargas, H., Leroy, S., Rivard, M., Oturan, N., Oturan, M., Buisson, D., 2016. Microbial  
 843 biotransformation of furosemide for environmental risk assessment: identification of metabolites  
 844 and toxicological evaluation. *Environmental Science and Pollution Research* 23, 22691–22700.  
 845 <https://doi.org/10.1007/s11356-016-7398-2>

846 Papageorgiou, M., Kosma, C., Lambropoulou, D., 2016. Seasonal occurrence, removal, mass loading and  
 847 environmental risk assessment of 55 pharmaceuticals and personal care products in a municipal  
 848 wastewater treatment plant in Central Greece. *Science of The Total Environment* 543, 547–569.  
 849 <https://doi.org/10.1016/j.scitotenv.2015.11.047>

850 Papageorgiou, M., Zioris, I., Danis, T., Bikiaris, D., Lambropoulou, D., 2019. Comprehensive investigation  
851 of a wide range of pharmaceuticals and personal care products in urban and hospital wastewaters  
852 in Greece. *Science of The Total Environment* 694, 133565.  
853 <https://doi.org/10.1016/j.scitotenv.2019.07.371>

854 Park, J., 2016. Removal Characteristics and Predictive Model of Pharmaceutical and Personal Care  
855 Products (PPCPs) in Membrane Bioreactor (MBR) Process. Thesis from Kyoto University  
856 <https://doi.org/10.14989/doctor.k19984>

857 Park, J., Yamashita, N., Park, C., Shimono, T., Takeuchi, D.M., Tanaka, H., 2017. Removal characteristics  
858 of pharmaceuticals and personal care products: Comparison between membrane bioreactor and  
859 various biological treatment processes. *Chemosphere* 179, 347–358.  
860 <https://doi.org/10.1016/j.chemosphere.2017.03.135>

861 Pereira, V.J., Linden, K.G., Weinberg, H.S., 2007. Evaluation of UV irradiation for photolytic and oxidative  
862 degradation of pharmaceutical compounds in water. *Water Research* 41, 4413–4423.  
863 <https://doi.org/10.1016/j.watres.2007.05.056>

864 Pharmaceuticals in the aquatic environment of the Baltic Sea region a status report, 2017. United  
865 Nations Educational, Scientific and Cultural Organization HELCOM, France, Helsinki.

866 Prasse, C., Stalter, D., Schulte-Oehlmann, U., Oehlmann, J., Ternes, T.A., 2015. Spoilt for choice: A critical  
867 review on the chemical and biological assessment of current wastewater treatment technologies.  
868 *Water Research* 87, 237–270. <https://doi.org/10.1016/j.watres.2015.09.023>

869 Prieto-Rodriguez, L., Miralles-Cuevas, S., Oller, I., Agüera, A., Puma, G.L., Malato, S., 2012. Treatment of  
870 emerging contaminants in wastewater treatment plants (WWTP) effluents by solar photocatalysis  
871 using low TiO<sub>2</sub> concentrations. *Journal of Hazardous Materials* 211–212, 131–137.  
872 <https://doi.org/10.1016/j.jhazmat.2011.09.008>

873 Reed, N.R., Kwok, E.S.C., 2014. Furfural, in: *Encyclopedia of Toxicology*. Elsevier, pp. 685–688.  
874 <https://doi.org/10.1016/B978-0-12-386454-3.00147-0>

875 Reungoat, J., Escher, B.I., Macova, M., Argaud, F.X., Gernjak, W., Keller, J., 2012. Ozonation and  
876 biological activated carbon filtration of wastewater treatment plant effluents. *Water Research* 46,  
877 863–872. <https://doi.org/10.1016/j.watres.2011.11.064>

878 Rozman, D., Hrkal, Z., Váňa, M., Vymazal, J., Boukalová, Z., 2017. Occurrence of Pharmaceuticals in  
879 Wastewater and Their Interaction with Shallow Aquifers: A Case Study of Horní Beřkovice, Czech  
880 Republic. *Water* 9, 218. <https://doi.org/10.3390/w9030218>

881 Salgado, R., Marques, R., Noronha, J.P., Mexia, J.T., Carvalho, G., Oehmen, A., Reis, M.A.M., 2011.  
882 Assessing the diurnal variability of pharmaceutical and personal care products in a full-scale  
883 activated sludge plant. *Environmental Pollution* 159, 2359–2367.  
884 <https://doi.org/10.1016/j.envpol.2011.07.004>

885 Sanches, S., Barreto Crespo, M.T., Pereira, V.J., 2010. Drinking water treatment of priority pesticides  
886 using low pressure UV photolysis and advanced oxidation processes. *Water Research* 44, 1809–  
887 1818. <https://doi.org/10.1016/j.watres.2009.12.001>

888 Santos, L.H.M.L.M., Gros, M., Rodriguez-Mozaz, S., Delerue-Matos, C., Pena, A., Barceló, D.,  
889 Montenegro, M.C.B.S.M., 2013. Contribution of hospital effluents to the load of pharmaceuticals  
890 in urban wastewaters: Identification of ecologically relevant pharmaceuticals. *Science of The Total*  
891 *Environment* 461–462, 302–316. <https://doi.org/10.1016/j.scitotenv.2013.04.077>

892 Sarathy, S.R., Mohseni, M., 2006. An Overview of UV-based Advanced Oxidation Processes for Drinking  
893 Water Treatment. *IUVA News* 7, 16–27.

894 Singh, A., Havixbeck, J.J., Smith, M.K., Shu, Z., Tierney, K.B., Barreda, D.R., El-Din, M.G., Belosevic, M.,  
895 2015. UV and hydrogen peroxide treatment restores changes in innate immunity caused by  
896 exposure of fish to reuse water. *Water Research* 71, 257–273.  
897 <https://doi.org/10.1016/j.watres.2015.01.003>

898 Stalter, D., O'Malley, E., von Gunten, U., Escher, B.I., 2016. Fingerprinting the reactive toxicity pathways  
899 of 50 drinking water disinfection by-products. *Water Research* 91, 19–30.  
900 <https://doi.org/10.1016/j.watres.2015.12.047>

901 Starling, M.C., Le Person, A., Amorim, C., Criquet, J., 2017. Degradation of Losartan Potassium and  
 902 Furosemide by UV and UV/H<sub>2</sub>O<sub>2</sub> processes. Environmental Science and Pollution Research.  
 903 Starling, M.C.V.M., Souza, P.P., Le Person, A., Amorim, C.C., Criquet, J., 2019. Intensification of UV-C  
 904 treatment to remove emerging contaminants by UV-C/H<sub>2</sub>O<sub>2</sub> and UV-C/S<sub>2</sub>O<sub>8</sub><sup>2-</sup>: Susceptibility to  
 905 photolysis and investigation of acute toxicity. Chemical Engineering Journal 376, 120856.  
 906 <https://doi.org/10.1016/j.cej.2019.01.135>  
 907 Stefán, D., Erdélyi, N., Izsák, B., Záray, G., Vargha, M., 2019. Formation of chlorination by-products in  
 908 drinking water treatment plants using breakpoint chlorination. Microchemical Journal 149,  
 909 104008. <https://doi.org/10.1016/j.microc.2019.104008>  
 910 Tracol, R., Duchemin, J., 2016. Evaluation de l'occurrence des résidus de médicaments dans un  
 911 échantillon de nappes souterraines vulnérables du bassin seine-normandie utilisées pour la  
 912 production d'eau destinée à la consommation humaine. Rapport DDASS Basse Normandie.  
 913 Venkobachar, C., Iyengar, L., Prabhakara Rao, A.V.S., 1977. Mechanism of disinfection: Effect of chlorine  
 914 on cell membrane functions. Water Research 11, 727–729. [https://doi.org/10.1016/0043-](https://doi.org/10.1016/0043-1354(77)90114-2)  
 915 [1354\(77\)90114-2](https://doi.org/10.1016/0043-1354(77)90114-2)  
 916 Vymazal, J., Dvořáková Březinová, T., Koželuh, M., Kule, L., 2017. Occurrence and removal of  
 917 pharmaceuticals in four full-scale constructed wetlands in the Czech Republic – the first year of  
 918 monitoring. Ecological Engineering 98, 354–364. <https://doi.org/10.1016/j.ecoleng.2016.08.010>  
 919 Wang, W.-L., Wu, Q.-Y., Huang, N., Xu, Z.-B., Lee, M.-Y., Hu, H.-Y., 2018. Potential risks from UV/H<sub>2</sub>O<sub>2</sub>  
 920 oxidation and UV photocatalysis: A review of toxic, assimilable, and sensory-unpleasant  
 921 transformation products. Water Research 141, 109–125.  
 922 <https://doi.org/10.1016/j.watres.2018.05.005>  
 923 White, D., Lapworth, D.J., Civil, W., Williams, P., 2019. Tracking changes in the occurrence and source of  
 924 pharmaceuticals within the River Thames, UK; from source to sea. Environmental Pollution 249,  
 925 257–266. <https://doi.org/10.1016/j.envpol.2019.03.015>

926 Yang, X.-X., Hu, Z.-P., Alex Boelsterli, U., Zhou, S.-F., 2006. Drug Acyl Glucuronides: Reactivity and  
927 Analytical Implication.  
928 Current Pharmaceutical Analysis 2, 259–277. <https://doi.org/10.2174/157341206777934608>  
929 Zhang, H., Zhang, Y., Shi, Q., Hu, J., Chu, M., Yu, J., Yang, M., 2012. Study on Transformation of Natural  
930 Organic Matter in Source Water during Chlorination and Its Chlorinated Products using Ultrahigh  
931 Resolution Mass Spectrometry. Environmental Science & Technology 46, 4396–4402.  
932 <https://doi.org/10.1021/es203587q>  
933 Zoumpouli, G.A., Zhang, Z., Wenk, J., Prasse, C., 2021. Aqueous ozonation of furans: Kinetics and  
934 transformation mechanisms leading to the formation of  $\alpha,\beta$ -unsaturated dicarbonyl compounds.  
935 Water Research 203, 117487. <https://doi.org/10.1016/j.watres.2021.117487>