Environmental Nanotechnology, Monitoring & Management 20 (2023) 100850

Contents lists available at ScienceDirect

Environmental Nanotechnology, Monitoring & Management

journal homepage: www.elsevier.com/locate/enmm

A decade of water monitoring in a Mediterranean region: Pharmaceutical prioritisation for an upgraded analytical methodology

M. Castaño-Trias [a,b], S. Rodríguez-Mozaz [a,b,*], G. Buttiglieri [a,b]

[a] Catalan Institute for Water Research (ICRA-CERCA), C/Emili Grahit 101, 17003 Girona, Spain
[b] University of Girona, Spain

ARTICLE INFO

Keywords:
Water scarcity
PhACs
Transformation products
Catalonia
Greece
Spain

ABSTRACT

In order to evaluate the performance of wastewater treatment strategies and to foster water reclamation it is necessary to evaluate the removal of water pollutants such as pharmaceutical active compounds (PhACs). Therefore, the development and application of appropriate analytical methods for the monitoring of the most relevant ones is crucial. In this work, the occurrence data of 157 PhACs in different water bodies and wastewaters in Catalonia (Spain) in the last 10 years, as reported in circa 30 publications, was gathered and reviewed to select the 50 most relevant compounds, including 12 transformation products and metabolites. Historical data confirmed the high consumption and prescription of analgesic and anti-inflammatory drugs, together with psychiatric drugs and antibiotics. An analytical protocol based on solid-phase extraction preconcentration and further measurement by UHPLC-MS/MS was upgraded for the selected 50 compounds and applied to the characterisation of water samples from Catalonia and the Greek Islands, both areas suffering from water scarcity. Results showed similar PhAC concentration profiles in both countries except in the case of some compounds such as iopromide, venlafaxine, and gemfibrozil.

1. Introduction

In a world suffering the consequences of overpopulation and climate change, the necessity of finding new sources of water to fight water scarcity is a major scientific challenge (Nikolaou et al., 2020). Water reuse is a promising option to solve the lack of fresh water and wastewater. For instance, treated wastewater is one of the most studied sources for irrigation purposes (Siegrist & Joss, 2012). However, emerging contaminants can be present in the water after its treatment (Rosal et al., 2010). These compounds are unregulated, anthropogenic or natural substances detected in the environment that are suspected to pose a risk to wildlife and human health. Among emerging pollutants, pharmaceutically active compounds (PhACs) have been detected in wastewater (Barcellos et al., 2022), (Rodriguez-Mozaz et al., 2020), (Verlicchi et al., 2012) and, since wastewater treatment plants (WWTP) do not remove them completely (Martins et al., 2021), also in different natural water bodies such as surface water (Čelić et al., 2019), (Kim & Zoh, 2016) and marine water (Arpin-Pont et al., 2016), (Borecka et al., 2015). Their presence in the aquatic environment may pose a high risk for the ecosystem as they can generate unexpected responses in non-target organisms even at low concentrations (Arpin-Pont et al., 2016); (Ginebreda et al., 2010). Ecosystems exposed to this continuous presence could suffer a reduction of their biodiversity (Kümmerer, 2009) and some studies also have reported a reduction in organisms mobility (Raldua et al., 2013). Furthermore, the full trophic chain can be affected since some of these compounds tend to be bioaccumulated in aquatic organisms (Previšić et al., 2019), (Valdés et al., 2016). Additionally, the continuous exposure of the aquatic microbial communities to antibiotics can cause the spread of antibiotic resistance (Pantanella et al., 2020). To assess the environmental impact of PhACs in the aquatic environment, a continuous monitoring of their presence is necessary. Additionally, transformation products (TPs) of these compounds can be generated during water treatment and also in the natural environment (M. Ibáñez et al., 2021). These can sometimes be more toxic (Majewsky et al., 2015) and/or recalcitrant than the parent compounds (Jaén-Gil et al., 2019) and so even more environmentally relevant (Escudero-Oñate et al., 2020). The development of appropriate analytical methods to monitor the presence of PhACs and their TPs in the environment is therefore critically important.

Traditionally, these analyses are performed using high pressure liquid chromatography (HPLC) and a mass spectrometry detector to fulfil the sensitivity and specificity requirements. More recently, ultra-

* Corresponding author.
E-mail address: srodriguez@icra.cat (S. Rodríguez-Mozaz).

https://doi.org/10.1016/j.enmm.2023.100850
Received 24 April 2023; Received in revised form 21 June 2023; Accepted 10 July 2023
Available online 13 July 2023

high pressure liquid chromatography (UHPLC), with shorter analyses times and better peak resolution, is substituting for the traditional HPLC systems. Different approaches can also be considered in the development of analytical methodologies. On the one hand, non-target and suspect screening methodologies have the advantage that a broad range of compounds can be analysed at the same time and that no chemical standards are required until the compound confirmation step (Gago-Ferrero et al., 2018). However, information about the structures and identity of the detected compounds are only tentative. In addition, a proper quantification cannot be performed, nor can an accurate assessment of the risk they pose to the environment be made. On the other hand, target methodologies provide reliable information about the detected compounds and their concentration, even at low levels (Bloszies & Fiehn, 2018). A major drawback of target analysis is that the analytes must be selected in advance. As a result, they may not always be present in the samples, whereas non-preselected analytes will be unobserved (Blum et al., 2017).

Therefore, it is vital to select the most relevant analytes to monitor and careful prioritisation is required before the development of the target analytical method. The prioritisation of target compounds can follow different approaches but usually a wide list of analytes is first preselected (*e.g.*, Kuzmanovic et al., 2013), according to available real monitoring data, legislation or other specific interest and criteria. The occurrence data of the contaminants can be obtained from monitoring studies, *i.e.*, measured environmental data (MEC) (Gros et al., 2017), or calculated based on pharmaceuticals prescription (Dong et al., 2013), consumption (Besse & Garric, 2008), frequency of release or pharmacokinetic considerations. Actually, upon consumption, only a fraction of the prescribed pharmaceutical is excreted in its unchanged form with values ranging from 0.5% up to almost 100% (Escolà Casas et al., 2021). For a proper determination of such predicted environmental data (PEC), it is essential to consider also the location and geographical area of the study as well as temporal factors (seasonal changes) (Riva et al., 2015). Additionally, pharmaceutical occurrence is correlated with prescription preferences among countries; numerous studies in sampling two or more different countries in the same year have identified differences in terms of PhACs usage and also in the selection of the preferred PhAC to treat diseases (Rodriguez-Mozaz et al., 2020), (Patel et al., 2019), (aus der Beek et al., 2016).

In this work, PhACs occurrence data generated by our group in Catalonia (NE Spain) in the last 10 years was gathered and used to select a shorter list of the most ubiquitous and abundant PhACs. Data came not from one matrix but five: hospital wastewater, urban (influent and effluent wastewater), river water and sea water. Then, an upgraded multi-residue analytical method was developed for the selected PhACs. For the evaluation of the developed analytical methodology a sampling campaign was performed in an urban site of Catalonia (Girona). In order to assess the transferability of the methodology, a second sampling campaign was carried on in another Mediterranean area that also suffers from water scarcity (Greek Islands). Even if both countries have similar characteristics (Benett et al., 1998) and water consumption habits (Reynauld, 2016), different consumption patterns might be observed. The selection of the most relevant pharmaceuticals in terms of occurrence could be useful for the development of further technologies aiming to wastewater treatment and fostering reclaimed wastewater employment.

2. Materials and methods

2.1. Gathering 10 years of PhACs monitoring in water

The studies performed by our research group in the last 10 years (since 2012) on PhACs presence in water matrices of Catalonia were examined, generating a comprehensive database of PhACs occurrence (Table 1). A total of 30 peer-reviewed articles were gathered regarding different water matrices: hospital wastewater (HWW), urban influent wastewater (IWW), treated wastewater or effluent (EWW), surface water and sea water. Information about the occurrence of a total of 157 pharmaceuticals and TPs was collected from these articles for their evaluation (Table S1).

Table 1
Studies performed by ICRA from 2012 to 2021 in different water matrices. HWW, IWW, EWW stand for hospital wastewater, influent wastewater, and effluent wastewater, respectively. All the analytical methods are based on HPLC/MS-MS.

	Paper	HWW	IWW	EWW	River	Sea	Number of analysed PhACs	Analytical method
1	Cruz-Morató et al., 2014	x					99	Gros et al., 2012
2	Badia-Fabregat et al., 2016	x					76	Gros et al., 2012
3	Mir-Tutusaus et al., 2017	x					40	Gros et al., 2012
4	Mir-Tutusaus et al., 2019	x					74	Gros et al., 2012
5	Mir-Tutusaus et al., 2021	x					19	Gros et al., 2012
6	Cruz-Morató et al., 2013		x				80	Gros et al., 2012
7	Jelic et al., 2015		x				45	Gros et al., 2012
8	Badia-Fabregat et al., 2015		x				56	Gros et al., 2012
9	Badia-Fabregat et al., 2017		x				72	Gros et al., 2012
10	Hom-Diaz et al., 2017		x				81	Gros et al., 2012
11	Collado et al., 2014		x	x			81	Gros et al., 2012
12	Mandaric et al., 2018		x	x	x		68	Gros et al., 2012
13	Gros et al., 2012		x	x	x	x	81	Gros et al., 2012
14	Čelić et al., 2019		x	x	x	x	74	Gros et al., 2012
15	Osorio et al., 2012				x		73	Gros et al., 2012
16	Acuña et al., 2015			x	x		62	Gros et al., 2012
17	Lucas et al., 2016	x					47	Gros et al., 2013
18	Gros et al., 2013	x	x	x	x		53	Gros et al., 2013
19	Rodriguez-Mozaz et al., 2015	x	x	x	x		62	Gros et al., 2013
20	Ávila et al., 2021		x	x			51	Gros et al., 2013
21	Auguet et al., 2017		x				53	Gros et al., 2013
22	Becker et al., 2017		x				38	Gros et al., 2013
23	Rodriguez-Mozaz et al., 2020			x			53	Gros et al., 2013
24	Huerta et al., 2013				x		53	Gros et al., 2013
25	Mamo et al., 2018		x				30	García-Galán et al., 2016
26	García-Galán et al., 2020		x				80	García-Galán et al., 2016
27	García-Galán et al., 2016		x	x			32	García-Galán et al., 2016
28	Aymerich et al., 2016		x	x	x		19	García-Galán et al., 2016
29	Rubirola et al., 2014		x	x			9	Rubirola et al., 2014
30	Ferrando-Climent et al., 2012		x	x			4	Ferrando-Climent et al., 2012

2.2. PhACs prioritisation

The concentration of each of the PhACs was obtained from the selected studies and its average was calculated for each study (in cases where more than one sampling campaign was provided in the corresponding publication). Reported values below the quantification or detection limits were considered as zero for the calculations. The data are collected in Table S1 (in-house database). Then, the overall average concentration, considering all the studies, was calculated for each pharmaceutical compound in each of the five water matrices (Table S2).

The 157 compounds analysed in the last 10 years by our group were ranked according to their occurrence using their average concentration (the first position corresponding to the highest concentration) in each of four matrices (hospital wastewater, influent wastewater, effluent wastewater, river water). Sea water data was not considered for prioritisation as only two publications were available and were considered insufficient to spot trends. Ranking positions of a pharmaceutical in each of the matrix were averaged to obtain a final general score. When the

Table 2
Selected compounds included in the analytical method with their retention time (RT), internal standard used for quantification purposes, MRM transitions, and ionization conditions in the mass analyzer.

Therapeutic class	PhAC	Retention Time	Internal standard	CAS	Precursor ion (*m/z*)	Q3	DP/CE/CXP	Q3	DP/CE/CXP
Analgesics and anti-inflammatories	Acetaminophen	0.61	Acetaminophen-d4	103–90-2	150	107	−45/-24/-15	n/a	n/a
	Codeine	1.24	Carbamazepine-d10	76–57-3	300	152	61/87/12	115	61/101/16
	Diclofenac	1.29	Ibuprofen-d3	15362–40-0	294	250	−65/-16/-11	214	−65/-28/-11
	Ibuprofen (IBU)	1.26	Ibuprofen-d3	15687–27-1	205	161	−60/-10/-13		
	*1-OH-IBU**	0.87	Ibuprofen-d3	53949–53-4	221	177	−55/-10/-9	159	−55/-16/-13
	*2-OH-IBU**	0.77	Ibuprofen-d3	51146-55-5	221	177	−75/-12/-11	n/a	n/a
	*CBX-IBU**	0.40	Ibuprofen-d3	15935-54-3	235	191	−20/-12/-7	73	−20/–22/-13
	Ketoprofen	1.05	Ibuprofen-d3	22071–15-4	253	209	−30/-12/-11	n/a	n/a
	Meloxicam	1.09	Meloxicam-d3	71125–38-7	350	146	−65/-28/-7	286	−65/-18/-15
	Naproxen	1.00	Naproxen-d3	22204–53-1	229	170	30/20/9	185	30/10/2007
	*Salicylic acid**	0.65	Acetaminophen-(ring-d4)	69–72-7	137	93	−50/-20/-1	n/a	n/a
Antibiotics	Azithromycin	2.48	Azithromycin-d3	83905–01-5	749	591	121/41/22	116	121/48/13
	Ciprofloxacin	1.85	Ciprofloxacin-d8	85721–33-1	332	288	66/27/12	245	66/33/16
	Clarithromycin	3.86	Clarithromycin-d3	81103–11-9	748	158	96/37/14	590	96/27/22
	Erythromycin	3.22	Norfloxacin-d5	59319–72-1	734	576	116/27/22	158	116/39/14
	Metronidazole	1.08	Metronidazole-d4	443–48-1	172	128	56/21/10	82	56/35/10
	*Metronidazole-OH**	0.90	Metronidazole-OH-d4	443–48-1	188	126	51/23/18	123	51/19/16
	Ofloxacin	1.80	Ofloxacin-d3	82419–36-1	362	318	86/27/12	261	86/39/12
	Norfloxacin	1.76	Norfloxacin-d5	70458–96-7	320	276	106/25/12	233	106/35/34
	Sulfamethoxazole	2.54	Sulfamethoxazole-d4	723–46-6	254	156	81/23/12	92	81/37/12
	Trimethoprim	1.64	Trimethoprim-d3	738–70-5	291	230	91/33/12	261	91/35/10
Anthelmintics	Levamisole	3.33		16595–80-5	205	178	41/31/14	91	41/59/14
Antihypertensives	Irbesartan	1.31	Valsartan-d8	138402–11-6	427	193	−95/-34/-11	399	−95/-26/-19
	Losartan	1.20	Valsartan-d8	114798–26-4	421	127	−105/-40/-5	179	−105/-34/-11
	Valsartan	1.00	Valsartan-d8	137862–53-4	434	179	−105/-30/-9	350	−105/-26/-13
Calcium channel blockers	Diltiazem	3.33	Carbamazepine-d10	42399–41-7	415	178	91/35/10	109	91/35/10
Lipid regulators	Gemfibrozil	1.61	Gemfibrozil-d6	25812–30-0	249	121	65/24/7	127	65/14/9
Diuretics	Furosemide	1.02	Furosemide-d5	54–31-9	329	285	−95/-20/-11	204	−95/-30/-15
	Hydrochlorothiazide	0.76	Hydrochlorothiazide-d2	58–93-5	296	269	−125/-28/-9	205	−125/–32/-7
Histamine receptor antagonists	Ranitidine	0.96	Cimetidine-d3	66357–59-3	315	176	66/25/24	130	66/35/12
Psychiatric drugs	Carbamazepine (CBZ)	3.55	Carbamazepine-d10	298–46-4	237	194	61/29/28	193	61/49/14
	*2-OH-CBZ**	2.72	Carbamazepine-d10	68011–66-5	253	210	76/27/12	208	76/33/12
	*10,11-epoxy-CBZ**	2.91	Carbamazepine-d10	36507–30-9	253	180	66/43/28	236	66/17/8
X-ray contrast agents	Iopromide	1.07	Sulfamethoxazole-d4	73334–07-3	792	573	156/35/20	300	156/83/10
β-Blocking agents	Atenolol	0.96	Atenolol-d7	29122–68-7	267	145	91/37/14	190	91/27/18
	Metoprolol (MTP)	2.09	Atenolol-d7	37350–58-6	268	133	86/35/12	121	11/33/18
	*MTP acid**	1.45	Atenolol-d7	56392–14-4	268	145	91/31/26	165	91/31/26
	Sotalol	0.97	Atenolol-d7	959–24-0	273	255	51/17/12	133	51/37/12

*Transformation product.

compound was not analysed in one matrix, the average was performed considering the remaining ones. The 50 compounds with the lowest average numbers (highest scores) were considered the most relevant ones and selected for the analytical method upgrade (Table S2). Table 2 reports the compounds, together with the parameters for their analysis with the new method.

2.3. Chemicals and reagents

All chemical standards were of high purity grade (less than90%). All compounds were purchased from Sigma-Aldrich and Toronto Chemicals. The full list of the analytes can be found in Table 2.

The individual stock standards, the isotopically labelled internal standards and the surrogate standard solutions were prepared at 1,000 mg/L by dissolving 10 mg of the analyte in 10 mL of the correspondent solvent. Cefuroxime was dissolved in water; the rest of compounds were dissolved in methanol (MeOH). Nonetheless, 100 μL of NaOH 1 M had to be added to fluoroquinolones and quinolones antibiotics to successfully dissolve the compounds, as described in (Ibáñez et al., 2009). Stock solutions were stored at −20 °C. Three different working standard mixture solutions were prepared in MeOH at a concentration of 0.5 mg/L: the first for the analytes, the second for the internal standard compounds and the third for the surrogate standards.

The cartridges used for the solid phase extraction were Oasis HLB (60 mg, 3 mL) for river and wastewater samples (HWW, IWW, EWW) and Oasis HLB (200 mg, 6 mL) for marine water samples (Waters Corporation; Milford, MA, U.S.A.). Glass fibre filters (1 μm) and PVDF membrane filters (0.45 μm) were purchased from Whatman (U.K.). HPLC grade MeOH, acetonitrile, and water (Lichrosolv) were supplied by Merck (Darmstadt, Germany). Ammonium hydroxide, hydrochloric acid 37% and ethylenediaminetetraacetic acid disodium salt solution (Na_2EDTA) at 0.1 mol/L were from Panreac. Formic acid 98% was from Merck (Darmstadt, Germany). Nitrogen for drying was from Abelló Linde S.A. (Spain) with 99.999% purity. A Milli-Q-Advantage system from Millipore Ibérica S.A. (Spain) was used to obtain HPLC-grade water.

2.4. Analytical methodology

2.4.1. Sample treatment

The extraction was performed as in (Gros et al., 2012). The water samples were first filtered (1 μm and 0.45 μm PVDF) and a suitable volume of a 0.1 M Na_2EDTA solution was added to achieve a final concentration of 0.1% ($g_{solute}/g_{solution}$). This chelating agent improves the recoveries for the compounds that tend to form complexes with calcium, like tetracyclines (Eichhorn & Aga, 2004). Moreover, the samples were spiked with a mixture containing the surrogate standards (Ketoprofen-d3 and Sulfadoxine-d3) to obtain a concentration of 10 ng/mL in the final extract.

Solid phase extraction cartridges were conditioned with 6 mL of MeOH followed by 6 mL of water at 1 mL/min, then the sample was loaded into the cartridges at the same flux: 25 mL for IWW, 50 mL for EWW, 100 mL for river, and 200 mL for sea samples. The cartridges were then washed with 6 mL of water and dried under vacuum conditions.

The elution was performed with 6 mL of MeOH. Then, the solvent was removed under a gentle nitrogen flow and samples were redissolved in a mixture of 2:8 MeOH/Water (v/v) and spiked with a mix containing all the isotopically labelled compounds to a concentration of 10,000 ng/L.

The recoveries were performed at three different levels, 1, 10, and 100 ng/L. When the spiked concentration of a compound was lower than the background concentration in the sample, the recovery value was discarded and labelled as “NA” (Non Applicable). The nearest recovery level to the detected concentration was used for the correction of the sample. Detailed information about the method performance (recoveries, limits of quantification (LOQ) and limits of detection (LOD)) can be found in Tables S3-6. LOD was determined as the minimum detectable concentration with a signal-to-noise ratio of 3, while LOQ was set at a signal-to-noise ratio of 10.

2.4.2. Instrumental analysis

The chromatographic separation was carried out with a Waters Acquity Ultra-Performance™ liquid chromatography system equipped with two binary pumps systems (Milford, MA, USA). Two different separation chromatographic gradients were optimized depending on the polarity of the compound. The method for positive ionized compounds consisted in an adaptation of (Gros et al., 2013) with an Acquity HSS T3 column (50 mm × 2.1 mm i.d., 1.8 μm particle size, Waters Corporation). Acetonitrile and 0.1% formic acid were selected as mobile phases at a flow of 0.5 mL/min (see Table S7 for gradient details). The method for the negative ionized compounds consisted in an adaptation of (Gros et al., 2012) with an Acquity BEH C18 column (50 mm × 2.1 mm i.d., 1.7 m particle size). The mobile phases were acetonitrile and an aqueous solution of 5 mM ammonium acetate/ammonia at pH 8 at a flow of 0.6 mL/min (see Table S8 for gradient details).

The sample volume injected was set at 5 μL for positive (PI) and negative (NI) ionization modes. The UHPLC instrument was coupled to a 5500 QTRAP hybrid triple quadrupole-linear ion trap mass spectrometer (Applied Biosystems, Foster City, CA, USA) with a turbo ion spray source. Compound dependent MS parameters (declustering potential, collision energy, and collision cell exit potential) were optimized for each compound. A summary of the optimum values and selected reaction monitoring (SRM) transitions is available in Table 2. All transitions were recorded by using the Scheduled MRM™ algorithm. In PI mode, target scan time (TST) was set at 0.25 s, with an SRM detection window of 20 s, whereas for NI mode, TST was set at 0.25 s with an SRM detection window of 30 s. In both cases, the resolution at the first quadrupole (Q1) was set at unit, and at the third quadrupole (Q3), it was set at low and the pause between mass ranges was 5 ms. The settings for source-dependent parameters were set-up as follows. For compounds analysed under PI the parameters were: curtain gas, 30 V; Nitrogen collision gas medium; source temperature of 650 °C; ion spray voltage at 5500 V; ion source gases GS1 and GS2 set at 60 and 50 V, respectively. For compounds analysed under NI, such parameters were: curtain gas, 30 V; Nitrogen collision gas medium; source temperature of 650 °C; ion spray voltage at − 3500 V; ion source gases GS1and GS2 set at 60 and 70. The entrance potential was set at 10. All data were acquired and processed using Analyst 1.5.1 software. For each compound, two mass transitions were selected, one at higher intensity for quantification purposes and a second for compound confirmation.

2.5. Sampling campaigns

The method was validated using river, sea, and wastewater samples from regions in Europe subject to water scarcity: in Catalonia (Girona province, NE Spain) and the islands of Lesbos and Tinos (Greece). Both countries have similar annual precipitations (600–900 mm in Greece Click or tap here to enter text.and slightly lower, 480–770 mm, in Spain (Llasat et al., 2021)), and water consumption habits (81.9 and 52.4 m^3 per capita in Greece and Spain, respectively, in 2010 according to Reynauld, 2016).

In Spain, WWTP influent and effluent samples (24 h composite) for one day were collected from Girona WWTP in February 2020, considering WWTP hydraulic retention time of 27 h. Girona WWTP serves a population of 120,000 person equivalent (PE) (Rodriguez-Mozaz et al., 2015) with an important hospital contribution (from Josep Trueta Hospital, 400 beds). Three water samples were collected from the river Ter: upstream, 3 km downstream of the WWTP, and before its mouth. The average flow of the studied river in the season of the sampling campaign (from 1st of January to 31st of March) was 37.28 m^3/s, much higher than 8.84 m^3/s, the average flow from the last 10 years (1st of January 2013 to 1st of January 2023) (values provided by the Catalan

Agency of Water, https://aplicacions.aca.gencat.cat/sdim21/inici.do). Two samples were also taken from the sea: close to the Ter River mouth and at L'Estartit (3 km distance from the mouth of the Ter). The location of the sampling points is shown in Fig. 1.

In Antissa (Lesbos, Greek Islands), which serves a population of 1,700 PE, 24 h composite influent wastewater samples were collected on three consecutive days in September 2019, once per day. One river and one sea samples were grabbed downstream of the WWTP in the Voulgaris river and from the Mediterranean Sea at Tinos Island, respectively, in September 2019.

All water samples were filtered through 1 and 0.45 μm PVDF filters and stored at −20 °C in amber PET bottles until sample processing and analysis. Samples collected in Catalonia were analysed in triplicate. In the case of Greece, wastewater samples were analysed in triplicate; river and sea samples were analysed in duplicate.

3. Results and discussion

3.1. Ten years of PhACs monitoring in water samples in Catalonia

There are several studies on pharmaceutical occurrence worldwide (Demirbas, 2011; Anand et al., 2022; Couto et al., 2019; Khan et al., 2020) and very recently, a comprehensive database has been built as (The UBA database – "Pharmaceuticals in the environment" | Umweltbundesamt, 2023) collecting information from 7,353 papers worldwide. These review papers and database allow to identify overall trends and differences across the countries and even selected regions. On the other hand, other review papers rather focus on selected areas such as Latin America (Valdez-Carrillo et al., 2020) or the Lower Danube Basin and Northwest Black Sea Region (Chiţescu et al., 2021). In the case of Catalonia, no review studies are available gathering pharmaceutical occurrence data except for the study by (Gómez-Canela et al., 2019), in which environmental concentrations are predicted from prescription data. Furthermore, most studies on the presence of PhACs in the natural environment or in wastewater treatment focus on the fate and/or removal of PhACs in their unchanged form and do not consider their transformation products. Nonetheless, a portion of the ingested compound can be excreted as a metabolite, the percentage depends on the compound of interest, but in some cases, the excretion rate of the metabolite can be up to 70% (*e.g.*, for N-acetyl-sulfamethoxazole with a parent compound excretion rate of 20%, Escolà Casas et al., 2021). Water treatment processes can also cause the generation of TPs that may or may not coincide with the excreted metabolites, in some cases at concentration of the same order of magnitude than the parent compounds.

The collection of published data on the presence of contaminants in a particular country or region allows assessing the contamination status of their water bodies and designing appropriate pollution remediation measures, *e.g.*, dedicated wastewater treatment or advanced water treatment. Therefore, occurrence data of 157 PhACs, including some of their TPs, in five water types (HWW, IWW, EWW, river and sea water) in Catalonia were gathered in this work. All data (in-house database) are related to the information reported in 30 peer-reviewed articles by our group between 2012 and 2021 (Table S1). The average concentration per compound and water matrix is also reported in Table S2. The average concentrations ranged from low ng/L up to more than 131,000 ng/L in the different matrices. The highest PhAC concentrations were usually encountered in HWW, followed by IWW, EWW, river, and sea water. Average concentrations higher than 10,000 ng/L were detected in HWW and IWW for 10,11-carbamazepine, acetaminophen, ibuprofen (IBU) and its metabolites 2-OH-IBU and CBX-IBU; salicylic acid, and iopromide (Table S1). In contrast, other PhACs, such as warfarin and tamsulosin, were never found at concentrations higher than 10 ng/L.

Not all the compounds were analysed in every matrix of the study, *e.g.*, only 2 publications were reported for sea water and covering a limited number of pharmaceuticals. The pharmaceuticals analysed in the 5 matrices (HWW, IWW, EWW, river, and sea) are summarized in Table S9 and the sum of the compounds grouped by therapeutical classes shown in Fig. 2. A detailed discussion about the findings in each of the matrix for each therapeutic class having into account only the compounds analysed in all matrices is provided below.

The X-ray contrast agent iopromide was the compound detected at the highest concentration (131,078 ng/L, on average, in hospital wastewater). Other studies have also raised awareness on the high concentrations of this compound in hospital effluent (*e.g.*, up to 164,000 ng/L of iopromide in Coimbra hospital, Portugal; Santos et al., 2013). The concentration in urban wastewater was much lower, with 952 ng/L average concentration in our data pool, close to other reported values in literature (*e.g.*, 1,200 ng/L in Sweden; Knopp et al., 2016). Conversely, other studies have reported much higher concentrations (*e.g.*, 18,100 ng/L in Germany, Altmann et al., 2014).

The therapeutical class with the highest concentrations was analgesics and anti-inflammatories, in particular for 4 compounds: ibuprofen (Fig. 2), acetaminophen, naproxen, and salicylic acid. These trends are in line with those observed by Verlicchi et al., 2012, who evaluated the removal of multiple PhACs in conventional WWTPs in Europe.

Psychiatric drugs were also present at very high concentrations in HWW (total concentration of 59,793 ng/L across eight articles and according to Table 2) and circa one order of magnitude lower in IWW (3,834 ng/L, Table S9). This may be due to the fact that within the eight

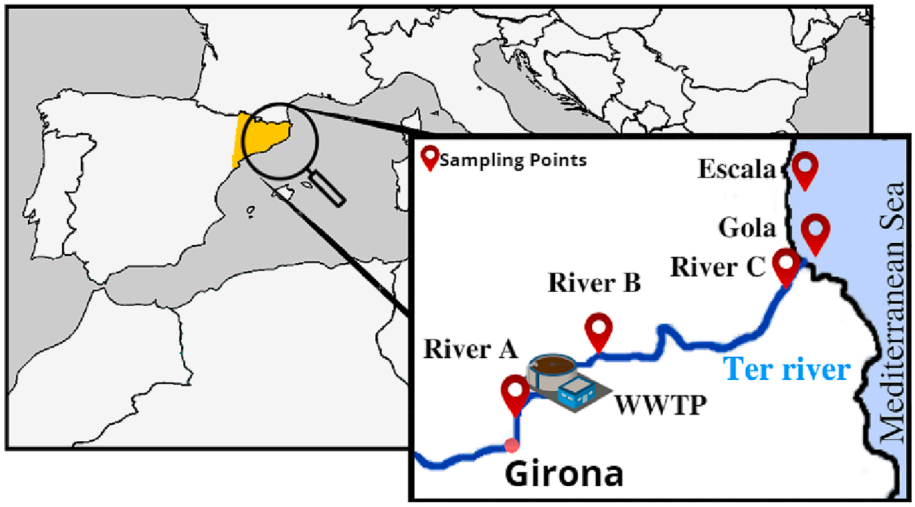

Fig. 1. Sampling points of the campaign performed in Catalonia in February 2020.

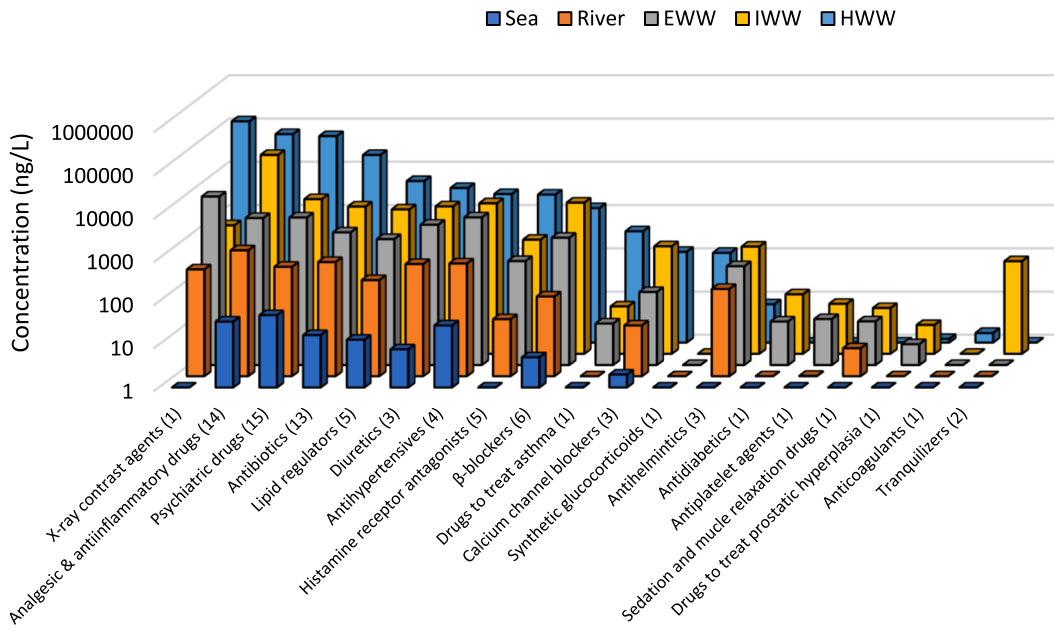

Fig. 2. Sum of the average concentration (ng/L, in logarithmic scale) of the PhACs, grouped by therapeutical class obtained from literature between 2012 and 2021 in each of the 5 water types. In brackets the number of analysed compounds within each therapeutic class. IWW, EWW, and HWW stand for urban influent wastewater, treated wastewater or effluent, and hospital wastewater, respectively.

publications about hospitals considered, three correspond to a hospital with a large psychiatric ward, St. Joan de Déu, Barcelona (J. A. Mir-Tutusaus et al., 2017; Josep Anton Mir-Tutusaus et al., 2019, 2021). Compounds at the highest concentration in IWW were venlafaxine (771 ng/L) and paroxetine (451 ng/L, Table S2). After them, carbamazepine TP, 10,11-epoxy-CBZ (Heye et al., 2016), and carbamazepine itself were found in all the matrices, with the TP at higher concentration than the parent compound (*e.g.*, 784.74 and 205.17 ng/L, respectively, in urban wastewater) except river water (31 and 53.1 ng/L, respectively). These findings are in line with the ones indicated by other authors, for example, (Paíga et al., 2017), who reported an average concentration of 95 ng/L of carbamazepine in Portuguese urban wastewater.

Antibiotics were the therapeutic class with the largest number of monitored compounds (67). Nonetheless, many of them were seldom detected and only 31 of them at a concentration higher than 50 ng/L in at least one of the four matrices. Considering only the antibiotics analysed in the 5 matrices the list shortens to 13. These compounds (Table S9) were consistently detected in all water matrices except by sea water. Highest concentration was observed for ciprofloxacin, up to 7,487 ng/L in HWW.

β-blocking agents, antihypertensives and diuretics were at the same order of magnitude in HWW and IWW as they are probably equally used in both hospitals and urban areas. Regular consumption of these pharmaceuticals is often observed since they are prescribed for long periods or even for the whole life (mainly in elderly population). β-blockers were at much lower concentration in river water than in EWW, compared with other therapeutic classes, suggesting that they are, on average, less persistent. The β-blocking agent at the highest concentration in IWW was atenolol (2,651 ng/L, Table S9), in agreement with the literature (*i.e.*, 3,733 and 25,000 ng/L, as reported by Patel et al., 2019; Verlicchi et al., 2012, respectively). Among antihypertensives, valsartan was at the highest concentration (2,159 in HWW, 2,298 in IWW and 1,392 ng/L in EWW, respectively). In the case of diuretics, furosemide was the most concentrated in HWW and IWW (3,207 and 1,501 ng/L, respectively) and hydrochlorothiazide in EWW and river water (1,043 and 232 ng/L, respectively).

Lipid regulators were present at high concentrations in most matrices (up to 5,488 ng/L in HWW, Table S9) with a high contribution of gemfibrozil (5,315 ng/L in HWW), in line with the very high concentrations reported in raw urban wastewater in other studies, up to 17,100 ng/L (Luo et al., 2014). Among the histamine receptor antagonist group, ranitidine was the compound with the highest concentration in HWW and IWW (2,246 and 403 ng/L, respectively).

Other considered therapeutic classes (anthelmintics, calcium channel blockers, and tranquilizers) were reported at concentrations lower than 400 ng/L in IWW, at less than 200 ng/L in EWW, and barely detected in river or sea water. Salbutamol was detected mostly in HWW (379 ng/L). This pharmaceutical compound is widely prescribed in hospitals to treat asthma and respiratory diseases at a higher dose than for domestic use (Naveen Kumar & Jafrin Antony, 2021, Connett et al., 1994), as well as the synthetic glucocorticoid, dexamethasone, detected only in the same kind of water (120 ng/L).

Relevant information can be extracted from the database, especially for the transformation products, even though some compounds were not analysed in all water matrices. For instance, the ibuprofen metabolite 2-OH-IBU was found at concentrations 2.28 times higher than its parent compound ibuprofen in IWW (Table S2, figure S1). This metabolite is also generated as an ibuprofen TP during wastewater treatment (Ferrando-Climent et al., 2012). Fig. 3 shows the concentration of selected parent compounds and some of their TPs and metabolites extracted from our database (Table S2) corresponding to different points of the urban water system from the WWTP to the river (additional parent/TPs can be found in Figure S2). A representative example is MTP Acid (metoprolol acid) which was found after the treatment up to 30 times higher than the parent compound itself, metoprolol. MTP Acid is not only a TP of metoprolol but also of atenolol and, therefore, its increase needs to be carefully correlated to the degradation of both compounds. Some TPs were found at similar concentration in the different sample types (IWW, effluent and river): O-desmethyl-VLF, ibuprofen TPs, carbamazepine TPs and 4-OH-DCF (Fig. 3). The SMX metabolite N4-acetyl-sulfamethoxazole, present in IWW at high concentration (783 ng/L), is not detected in the secondary effluent (Figure S2) as it may be deconjugating into its parent compound during wastewater treatment. Sulfamethoxazole generation might thus counteract its actual elimination, leading to moderate overall SMX removal values (from 230 to 110 ng/L mean values in influent and effluent wastewater respectively, as seen in Table S2) and even negative removal observed in some cases (Brown et al., 2020).

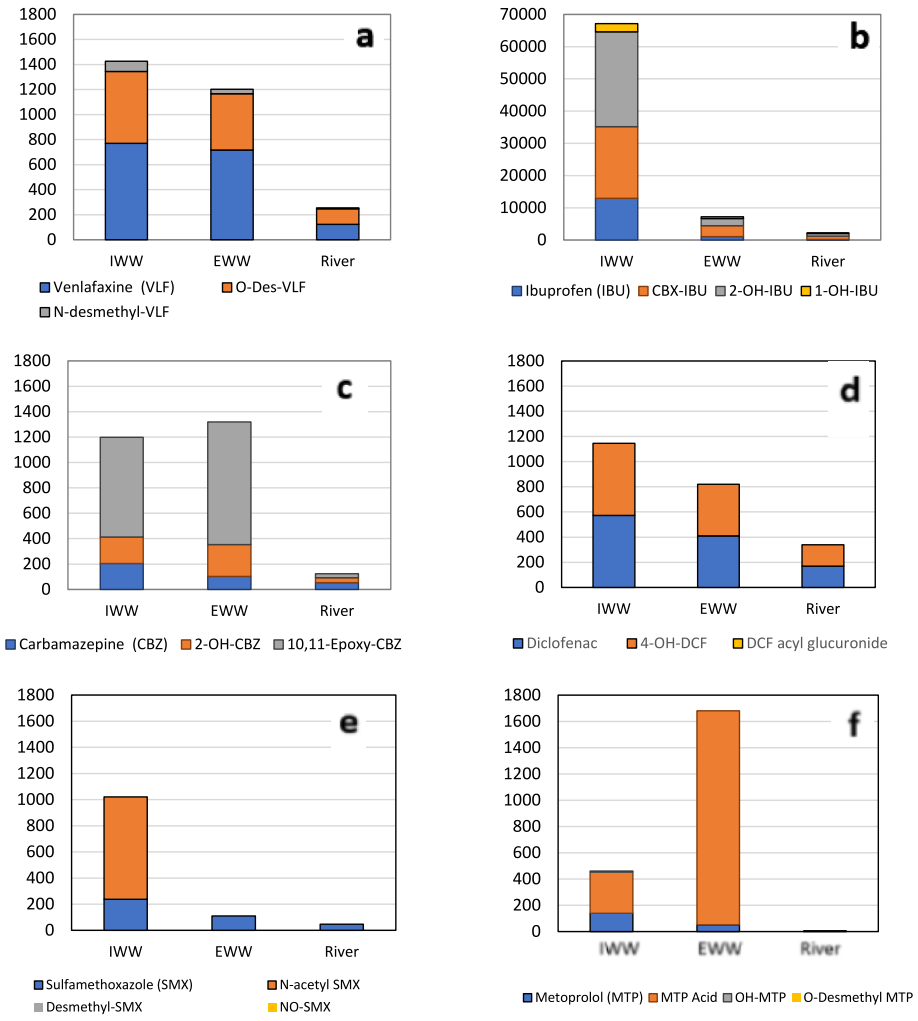

Fig. 3. Average concentration (ng/L) of the parent compounds (in blue) venlafaxine (a), ibuprofen (b), carbamazepine (c), diclofenac (d), sulfamethoxazole I and metoprolol (f), and their corresponding TPs (in orange, grey, and yellow) in IWW, EWW, and river water. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Furthermore, comparing antibiotics findings with other studies in Europe, it is confirmed that the use of antibiotics is highly dependent on the specific geographical area. For example, Verlicchi et al 2012, who reported concentration data of PhACs in influent wastewater all over Europe, reported values of roxithromycin up to 17,000 ng/L, while in our study in Catalonia this antibiotic was only at 19 ng/L. Similarly, sulfapyridine was detected between 914 and 4,971 ng/L (IWW) in the UK (Petrie et al., 2015) while in Catalonia our findings ranged between 12 and 235 ng/L.

Median values are generally close or at the same order of magnitude than the average values (Table S1, Fig. 4). Occurrence data in hospital wastewater exhibited more dispersion than in wastewater influent, effluent, or river water. This can be partially related to the smaller number of collected articles regarding hospital wastewater (8) compared to influent wastewater (20). In addition, larger populations (*e.g.*, those served by large WWTPs) lead to smaller uncertainty in average calculations compared to smaller population (*e.g.*, population in hospitals monitored in this work did not exceed 400 beds). For instance, (Castiglioni et al., 2013) evaluated the uncertainty in biomarkers measurement in influent wastewater associated to population sizes, with a 38% of RSD in a population censed with 120,000 inhabitants and 18% in a one having 1,250,000.

3.2. Occurrence-based prioritisation

The 157 compounds analysed by our group in the last ten years were ranked based on their occurrence in each of the four matrices: *i.e.*, the average of the position assigned to each compound in each of the four matrices based on the concentration levels (Table S2). A final list of 50 compounds with the highest score was selected as the most relevant; the first 20 compounds are presented in Figure S1.

The x-ray contrast agent iopromide was the compound with the highest concentration in HWW and EWW, and the third highest in river water. Antibiotics were more represented in HWW (eighteen compounds in the first fifty) than in IWW (twelve compounds). Higher concentrations of antibiotics were previously detected in hospital wastewater compared to wastewater influent (Rodriguez-Mozaz et al., 2015). Analgesics and anti-inflammatories were the therapeutical group with the most compounds in the final list of prioritised compounds due to their massive consumption and six of them (parent compounds) were prioritised within the 50 most relevant compounds in all water matrices: acetaminophen, ibuprofen, naproxen, ketoprofen, diclofenac (DCF) and codeine. Moreover, five analgesic TPs were prioritised in top positions in several matrices: salicylic acid, 1-OH-IBU, 2-OH-IBU, CBX-IBU and 4-OH-DCF (Figure S1). Also, several TPs from psychiatric drugs (*e.g.*, 10,11-epoxy-CBZ, O-desmethyl-VLF and N-desmethyl-VLF) were prioritised in the selected water matrices. For instance, 10,11 epoxy-CBZ was ranked second in HWW. Fluoxetine was excluded from the final

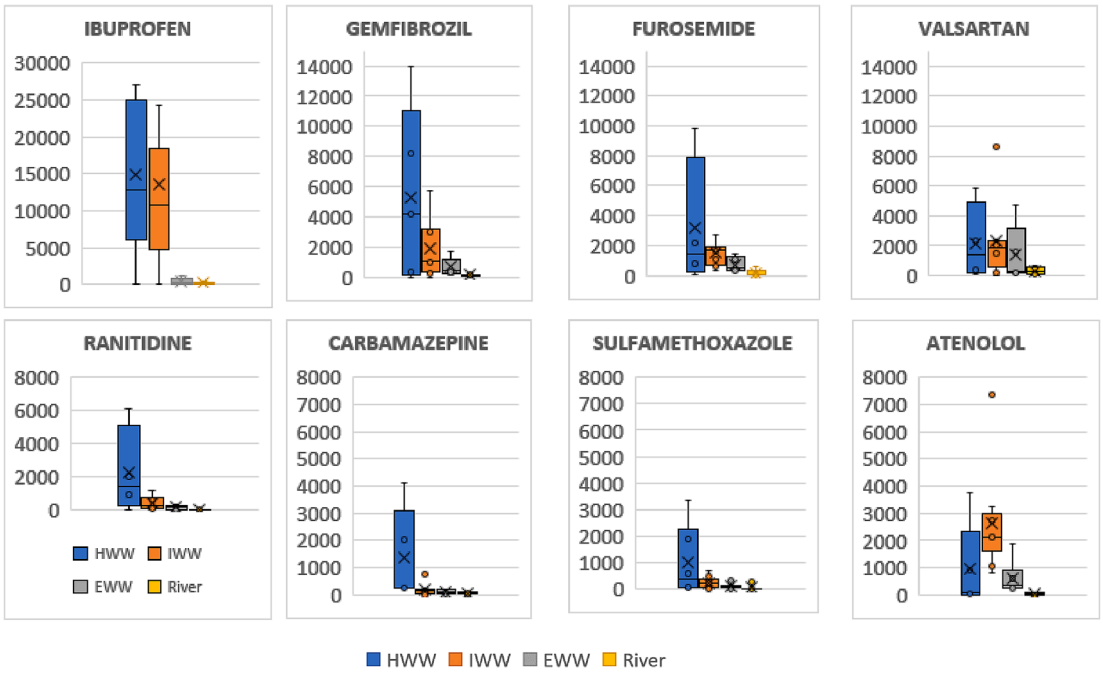

Fig. 4. Average concentrations, median and error dispersion (ng/L) of selected compounds of interest in IWW, EWW and river water. Ibuprofen (analgesic and anti-inflammatory), gemfibrozil (lipid regulator), furosemide (diuretic), valsartan (antihypertensive), ranitidine (histamine receptor antagonist), carbamazepine (psychiatric drug), sulfamethoxazole (antibiotic) and atenolol (β-blocker).

prioritisation list while one of its TPs, norfluoxetine, was detected at concentrations higher than its own parent compound in EWW and prioritised. Regarding the lipid regulators, out of the seven compounds studied by the group, only gemfibrozil was finally selected.

Gómez-Canela also performed a prioritisation of PhACs in Catalonian rivers in 2019 (Gómez-Canela et al., 2019) and out of 165 prioritised in their work only 10 are in common with the 50 of the present study: ciprofloxacin, valsartan, ibuprofen, propranolol, diclofenac, ranitidine, diazepam, norfloxacin, citalopram and venlafaxine. The prioritisation approach by Gómez-Canela et al., 2019 was based on prescription-based predicted concentrations in the environment rather than real monitoring data, as in the present work. Therefore, for example amoxicillin, which is highly prescribed, was in second position in their prediction-based prioritisation list, whereas it was placed only ninety-ninth in our study, probably due to its low stability in water (Kosińska et al., 2016)).

In comparison, (Park et al., 2018), prioritised 38 from an initial list of 189 PhACs based on real monitoring data of Korean rivers. Out of their 38 selected compounds, 11 are shared with our river prioritisation list: carbamazepine and its TP 10,11-epoxy-carbamazepine, naproxen, valsartan, irbesartan, sulfamethoxazole, losartan, venlafaxine, ranitidine, atenolol, and trimethoprim. However, other compounds were selected in their list but not in ours: cimetidine, amoxicillin, lincomycin, sulfadiazine, sulfomerazine, sulfamethazine, sulfadimethoxine, propranolol, fluoxetine, and metoprolol. This difference might be related to regional variations, *i.e.*, different national consumption patterns. In fact, prioritisation results might not be directly comparable in many cases and care must be taken because the initial list of compounds to be prioritised can differ enormously. In the recently published database from the German Environmental Agency (PHARMS-UBA database, 2022, updated in 2021) which collects pharmaceutical levels information from 7353 papers worldwide, 18 of the 31 more frequently detected ones are also considered within the top 50 in the present work. However, this database took only into account 4 articles among the 30 included in our database (Table 1) and did not include hospital wastewater data, whereas 9 articles reporting concentrations in that type of matrix are considered in our in-house database. Despite such variations in the compounds prioritised, these lists are necessary to raise awareness of compounds to be included in future monitoring campaigns that might otherwise be overlooked. It also helps to avoid investing resources in monitoring compounds that are barely detected in the environment.

The objective of the present work was to select the most relevant compounds overall (in terms of occurrence) for a whole set of water types (HWW, IWW, EWW and surface water). However, selected compounds in one matrix can be different than in another (Table S2). For instance, the HWW prioritisation list contains a higher number of antibiotics than the river list or the combined list of the four matrices because a larger number of them were frequently detected in hospital wastewater. Additionally, although beyond the objective of our work, in many occasions the risk posed by the contaminants is taken into consideration for prioritisation approaches. In those cases, the impact of contaminants in the environment is considered using environmental concentrations of the pollutants together with their predicted no-effect-concentration (PNEC), which can be calculated through acute or chronic toxicities in three different trophic levels (Kandie et al., 2020) (Tsaboula et al., 2016), (Dong et al., 2013). Commonly, other factors are considered such as the persistence of the compound in the environment or its potential to bioaccumulate through the trophic chain. Prioritisation approaches considering all the previous aspects are called PBT (persistence, bioaccumulation, and toxicity) prioritisation approaches (Sangion & Gramatica, 2016).

3.3. Method performance

The analytical method based on solid-phase extraction (SPE) for sample preparation followed by UHPLC-MS/MS analysis was upgraded and validated for the prioritised compounds in section 3.2. Four out of the prioritised compounds were not included in the methodology because no reference standards were available: ceftiofur, cefotaxime, tenoxicam and hydrocodone. Furthermore, at the time of the analysis 4-OH-DCF was not analysed due to a contamination in the mass detector. Despite being prioritised, 5-OH-DCF was not added to the methodology since it was only measured once (García-Galán et al., 2020). Norfloxacin was analysed in the sampling campaign of the Greek Islands but could not be analysed in Catalonia. The method thus allowed the successful

detection and quantification of a total of 45 compounds.

The method quality parameters (recoveries, LODs and LOQs) for each compound and matrix, are reported in detail in Table S3 and Table S4 for Catalonia samples, and in Table S5 and Table S6 for Greek Islands samples. Most of the recoveries ranged between 70% and 130% in all the matrices and the LOD reached values as low as 3.93, 2.13, 1.12, and 0.12 ng/L, and the LOQ of 13.09, 7.09, 3.73, and 0.39 ng/L for IWW, EWW, river and sea water, respectively.

3.4. PhACs monitoring in the field sites in Catalonia and Greek Islands.

The upgraded method with the 45 prioritised compounds was validated with real samples from two different areas (Girona province in Catalonia, and the Greek Islands of Lesbos and Tinos). Detailed data of both sampling campaigns are reported in Table S10 (Catalonia) and Table S11 (Greek Islands). The total PhAC concentration for each matrix is reported in Fig. 5.

3.4.1. Monitoring in Catalonia (Girona province, NE Spain).

The findings related to the PhACs in the sampling campaign in Catalonia are shown in Fig. 5, grouped by therapeutic class. Around 96% of the total content in IWW corresponds to only 3 therapeutic classes: analgesics and anti-inflammatory compounds (75%), the x-ray contrast agent iopromide, and diuretics (hydrochlorothiazide and furosemide).

The compounds at the highest concentration in IWW were acetaminophen (105,012 ng/L) and CBX-IBU (91,420 ng/L, Table S11), followed by the contrast agent iopromide at a remarkably high concentration (50,146 ng/L), and the diuretic hydrochlorothiazide at 14,123 ng/L (Table S10). EWW presented a different distribution of therapeutic classes than IWW (Fig. 5). Analgesics and anti-inflammatories, in fact, were largely removed (99%) by the WWTP, as frequently reported in the literature on conventional WWTP (Castaño-Trias et al., 2020), (Margot et al., 2015). The other two therapeutic classes with relevant concentrations in IWW also decreased their concentrations in EWW, but to a different degree: iopromide (96%) and diuretics (67%). The diuretic hydrochlorothiazide was, in fact, the compound at the highest concentration in EWW (4,466 ng/L, Table S10).

As it was expected, the concentration of most PhACs strongly decreased after wastewater treatment. However, the opposite phenomenon was observed for some compounds: ketoprofen concentration increased its concentration from 614 to 1,116 ng/L, azithromycin from 97 to 376 ng/L, venlafaxine (VLF) from 287 to 350 ng/L, and the TPs O-desmethyl-VLF and MTP acid from 685 to 1,358 ng/L and from 373 to 976 ng/L, respectively. The increase in the concentrations may be due to the cleavage of the conjugates and/or re-transformation of metabolites by microbial community present in wastewater (Jelic et al., 2015). For instance, carbamazepine concentration in EWW (100 ng/L, Table S10) was higher than in IWW (77 ng/L, table S10) while its transformation product 10,11-epoxy-carbamazepine, even detected in IWW (30 ng/L, table S10) it was not detected in EWW, which might be due to back transformation to the parent compound (Bahlmann et al. 2014). Another cause of negative removals is the partitioning to the aqueous phase of compounds sorbed in the solid phase during water treatment (Jelic et al., 2011).

The dilution factor of the WWTP effluent in the river (1:65) was calculated based on the WWTP discharge flow of 0.58 m^3/s (corresponding to 50,000 m^2/day, Ferrando-Climent et al., 2012) and the average river flow of the Ter River (37.28 m^3/s) in winter 2020 (January-March). The total concentration of analgesics and anti-inflammatories increased from 208 ng/L to 1,127 ng/L before and after the WWTP, respectively (Table S10), highlighting the impact of the WWTP. A further decrease for all the compounds is observed at the mouth of the river, which is attributed to attenuation processes in the river such as dilution, photo, and biodegradation, as well as sorption processes (Fig. 6). Although other WWTPs of smaller size are also discharging into the Ter River between the sampled WWTP and the river mouth, their contribution to the river contamination was not sufficient to increase the PhACs levels and so these levels are their lowest at the river mouth.

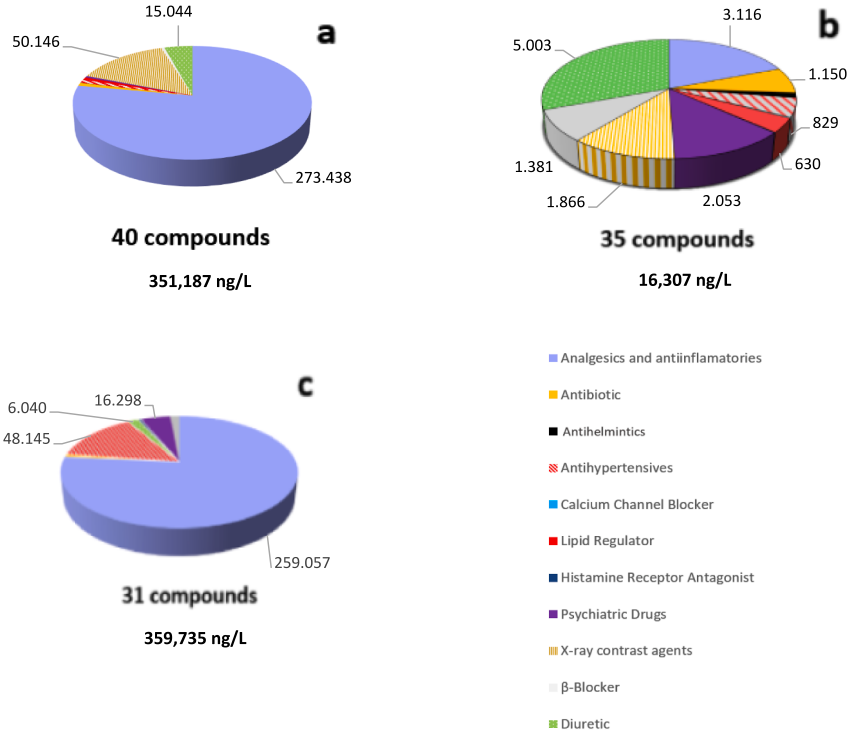

Fig. 5. PhACs in Catalonia IWW (a), Catalonia EWW (b), and Greek Islands IWW (c). Total concentration (ng/L) for the most abundant therapeutic class are indicated. Samples were analysed in triplicate.

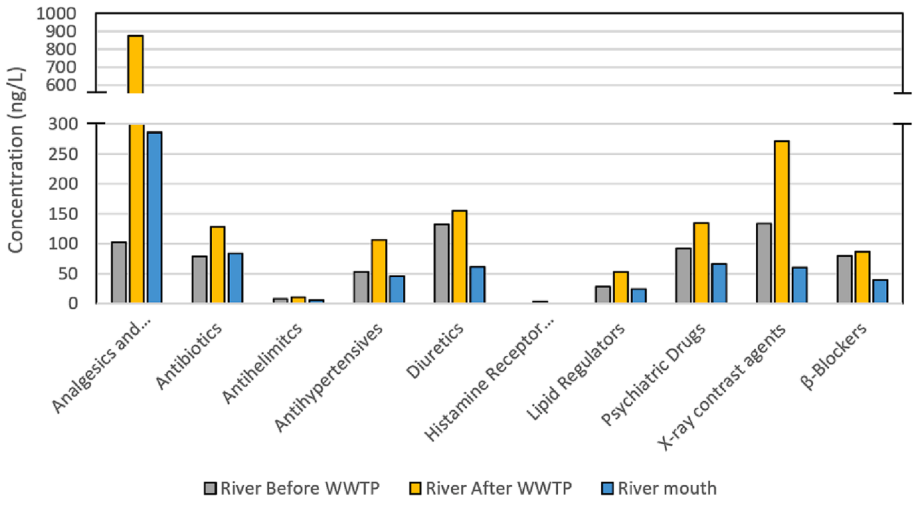

Fig. 6. Concentrations of PhACs and TPs, grouped by therapeutic classes, in the three sampled points of the river Ter in Catalonia, February 2020.

Very few compounds were detected in sea water: six in the site closest to the river mouth, and three at the one located 3.4 km from the river mouth (l'Estartit). Most of them were at concentrations below 5 ng/L, except for clarithromycin (35 ng/L) and salicylic acid (27 ng/L), which were also the only two compounds present at both sea sites (Table S10). As it was expected, concentration in sea water samples were in general lower than those found in the river water: *e.g.*, clarithromycin at 79 ng/L at the river mouth and 35 and 32 ng/L at the sea sites. Iopromide concentration was 60 ng/L at the river mouth and 5 ng/L in the sea close to the river mouth whereas it was not detected in the furthest site in the sea (l'Estartit), relating its presence in the sea to the river. Salicylic acid, on the other hand, was detected at higher concentration at the sea sites (27 and 154 ng/L; Table S10) compared to the river mouth (<LOQ).

Comparing the 2020 sampling campaign with the historical in-house database, several differences could be observed (Table S12). Ibuprofen concentration in IWW of Girona in 2020 was in the range of the in-house database. Ibuprofen TPs 1-OH and 2-OH IBU, on the other hand, were found at concentrations more than twice the historical average IWW values but not too much higher compared to other studies of the group (*e.g.*, 1-OH-IBU at 4,764 ng/L, Garcia-Galan et al., 2020; 2-OH IBU at even higher concentrations of 54,316 ng/L (Ferrando-Climent et al., 2012). Hydrochlorothiazide concentration was 10 times higher than the average historical value in IWW (4,466 ng/L). Also, its concentration in EWW was very high (1,670 ng/L). Overall, the observed concentrations in EWW in 2020 were lower than the historical values (except for hydrochlorothiazide and ketoprofen, and very slightly lower for acetaminophen, azithromycin, citalopram, clarithromycin, and trimethoprim). The total amount in 2020 was half of the historical value. Compounds having larger differences from the historical values were hydroxy ibuprofen TPs (under quantification limits), naproxen and iopromide. The 2020 sampling campaign was carried out one week after the floods caused by Storm Gloria (an extratropical cyclone with rainfall accumulation of 450 mm, (Palau et al., 2022)). In fact, mean flow of the river from 1st of January to 1st of March was 37.28 m^3/s which is higher than the average flow calculated from 2013 to 2023 (8.84 m^3/s). The storm could have affected not only the river flow (thus increasing the dilution factor of WWTP discharges into the river) but also have diluted the WWTP influent and affected the WWTP operation itself (*e.g.*, reduced degradation). On the other hand, sewer overflows could have happened as well, increasing the input of non-treated wastewater in the river. All these factors may explain the differences between the 2020 sampling campaign and the in-house historical database. For example, gemfibrozil and carbamazepine were on average 111 and 105 ng/L in the Ter River based on the in-house database, and only up to 52 and 10 ng/L respectively in the 2020 sampling campaign. In fact, total concentration of pharmaceuticals in the 2020 river sampling (after the WWTP effluent discharge) was just 33% of the total average concentration in the historical database. This effect was also observed in sea water samples, returning much lower values than the expected ones.

3.4.2. *Monitoring in the Greek Islands*

PhACs total concentrations were very similar in the IWW of both countries: 351,187 and 359,735 ng/L in Catalonia and the Greek Islands, respectively. Nonetheless, some relevant differences were observed. Analgesics and anti-inflammatories were the most abundant in the Greek Islands (77% of the total concentration), followed by antihypertensives (14%), and psychiatric drugs (5%) (Table S11 and Fig. 5). Nevertheless, the x-ray contrast agent iopromide was at very low concentration (25 ng/L) in the Greek Islands and extremely high in Catalonia (50,146 ng/L). Iopromide is commonly used in hospitals, therefore it is found in Girona (with hospitals discharging in the sewage system) but not in Antissa (without hospitals). The highest concentration observed was of the analgesic acetaminophen (212,647 ng/L, Table S11), as it was in Catalonia too. Ibuprofen was at much lower concentration than in Catalonia (2,351 vs. 12,866 ng/L, respectively), as were ibuprofen TPs. On the other hand, naproxen and valsartan were more abundant in the Greek Islands (28,651 ng/L and 39,922 ng/L, respectively) than in Catalonia (5,234 and 2,523 ng/L, respectively, Table S10 and Table S11). These differences in PhACs levels in IWW between the two countries can be attributed to prescription and/or consumption patterns of analgesics and anti-inflammatories in the two areas.

While in the Ter River in Catalonia up to 30 PhACs were detected, only seven PhACs were quantified in the Greek river: irbesartan (64 ng/L), salicylic acid (53 ng/L), ketoprofen (50 ng/L), hydrochlorothiazide (44 ng/L), iopromide (9 ng/L), O-desmethyl-VLF (9 ng/L) and azithromycin (7 ng/L, Table S11). In line with these results, the total concentration was almost one order of magnitude lower in the Greek River (240 ng/L) than in the Ter River in Catalonia (2,098 ng/L). Further investigations are required to determine if these differences are consistent.

Finally, with regards to seawater samples, concentrations were low both in Catalonia and the Greek Islands (avg. 141 ng/L of the two sampling points in Catalonia, and 184 ng/L in the Greek Islands). In Greece, all the compounds were below 20 ng/L, except ketoprofen, which was detected at a concentration of 125 ng/L. In Catalonia, the only compound with a concentration higher than 20 ng/L was salicylic acid (27 ng/L at Ter's mouth and 154 ng/L at l'Estartit). It should be noted that the Greek seawater sample was not taken close to the river

mouth as it was at the sea sites in Catalonia and therefore a proper study of the possible contamination source of seawater in this sampling point in Greece is lacking.

Historical data about PhACs occurrence in Greek water bodies is scarcer than that available for Catalonia. Table S12 compares literature values with the 2019 sampling campaign in the Greek Islands. The findings of the sampling campaign in the Greek islands were in line with the few reported in previous studies (Table 3). The largest differences were observed in IWW, with concentrations above the historical data for acetaminophen, irbesartan, naproxen and valsartan. For instance, acetaminophen levels in the 2019 sampling campaign (212,647 ng/L), were much higher than in the literature (41 ng/L, Desbiollesa et al., 2018). Naproxen was at an even higher concentration (28,651 ng/L) than previously described values, which ranged between 127 and 10,000 ng/L (Kosma et al., 2010, Kosma et al., 2014, Papageorgiou et al., 2016, Papageorgiou et al., 2019, Borova et al., 2014).

Table 3
Previous studies on the presence of PhACs in Greek water bodies.

Study	IWW	River	Sea	Number of Analysed Compounds
(Kosma et al., 2010)	x			6
(Kosma et al., 2014)	x			9
(Papageorgiou et al., 2016)	x			22
(Papageorgiou et al., 2019)	x			31
(Borova et al., 2014)	x			6
(Stasinakis et al., 2012)		x		4
(Bellos & Sawidis, 2005)		x		5
(Nannou et al., 2015)		x		12
(Mandaric et al., 2019)		x		30
(Stamatis et al., 2013)		x		7
(Alygizakis et al., 2016)			x	13
(Nödler et al., 2014)			x	7
(Desbiolles et al., 2018)	x		x	25

4. Conclusions

Data about the presence of 157 pharmaceutical compounds in water bodies in Catalonia in the last 10 years (2012–2021) have been compiled and discussed in this work. Different concentration profiles were measured in the water matrices considered: hospital, influent and effluent wastewater, as well as river and sea water. A list of 50 compounds belonging to 11 therapeutic classes was elaborated based on the most abundant compounds in each of the matrices, with analgesics, anti-inflammatories, psychiatric drugs, and antibiotics being the most relevant PhACs, the latter particularly so in hospital wastewater.

Tracking the presence of selected compounds can provide general view of the contamination degree of a water body and therefore support the design of appropriate pollution remediation measures when required, *e.g.*, dedicated wastewater treatment or advanced water treatment. In fact, the performance of such alternative water treatment strategies should also be evaluated based on the removal achieved for these pollutants beyond other parameters (*e.g.*, water quality in terms of standard parameters, greenhouse gases emission, energy requirements, footprint).

An analytical method was upgraded to target the 45 selected PhACs and was applied to monitor their presence in two different European regions in Spain and Greece suffering from water stress. Though they exhibited similar concentration profiles, several differences were spotted regarding some compounds: valsartan and acetaminophen (higher in the Greek Islands); iopromide, and ibuprofen and its TPs (higher in Catalonia).

Transformation products and metabolites, not always considered in target analytical methods and in monitoring programs, proved to be relevant compounds in terms of occurrence (12 out of 50 prioritized PhACs) and cannot be neglected in future investigations.

The target contaminants in monitoring studies need to be continuously updated considering occurrence data from other contaminants pinpointed in exploratory approaches (such as those based on suspect and non-target screening) as well as prescription data. Ecotoxicity parameters are recommended to be included in further prioritization approaches, so that the risk that chemical pollution poses to the environment is properly assessed. Also, the impact of tourism and seasonality on the occurrence of PhACs should be carefully considered in future studies.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Acknowledgements

Research funded by the Spanish State Research Agency of the Spanish Ministry of Science and Innovation (project code: PID2020-115456RB-I00/MCIN/AEI/10.13039/501100011033; ReUseMP3). This study has received funding from the European Union's Horizon 2020 Research and Innovation programme (HYDROUSA, grant agreement N° 776643). M. Castaño-Trias acknowledges his PhD scholarship from AGAUR 2020FI_B00711. Gianluigi Buttiglieri acknowledges Spanish State Research Agency of the Spanish Ministry of Science, Innovation and Universities for the Grant to the Creation of a permanent position Ramon y Cajal 2014 [REF: RYC-2014-16754]. The Ultra-Performance Liquid Chromatography triple Quadrupole Mass spectrometry (UPLC-MS) hybrid Linear Ion Trap (LIT), Acquity UPLC-MS QTRAP 5500, Waters-SCIEX. facility received support from the CERCA Institute through the CERCAGINYS programme, funded by the Spanish Ministry of Science and Innovation.The authors acknowledge the support from the Economy and Knowledge Department of the Catalan Government through a Consolidated Research Group (ICRA-TECH - 2021 SGR 01283 and ICRA-ENV - 2021 SGR 01282). Open Access funding provided thanks to the CRUE-CSIC agreement with Elsevier. The authors are grateful to TRARGISA, SA, Nuria Cáceres, and Dr Rubén Gil Solsona for the sampling in Catalonia; Nicolas Bedau (Tinos Ecolodge, https://www.tinosecolodge.gr/) and the staff of National Technical University of Athens for the sampling in Tinos and Lesbos islands (Greece), respectively.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.enmm.2023.100850.

References

Altmann, J., Ruhl, A.S., Zietzschmann, F., Jekel, M., 2014. Direct comparison of ozonation and adsorption onto powdered activated carbon for micropollutant removal in advanced wastewater treatment. Water Research 55, 185–193. https://doi.org/10.1016/J.WATRES.2014.02.025.

Alygizakis, N.A., Gago-Ferrero, P., Borova, V.L., Pavlidou, A., Hatzianestis, I., Thomaidis, N.S., 2016. Occurrence and spatial distribution of 158 pharmaceuticals, drugs of abuse and related metabolites in offshore seawater. Science of the Total Environment 541, 1097–1105. https://doi.org/10.1016/j.scitotenv.2015.09.145.

Anand, U., Adelodun, B., Cabreros, C., Kumar, P., Suresh, S., Dey, A., Ballesteros, F., & Bontempi, E. (2022). Occurrence, transformation, bioaccumulation, risk and analysis of pharmaceutical and personal care products from wastewater: a review.

Environmental Chemistry Letters 2022 20:6, *20*(6), 3883–3904. 10.1007/S10311-022-01498-7.

Arpin-Pont, L., Martínez-Bueno, M.J., Gomez, E., Fenet, H., 2016. Occurrence of PPCPs in the marine environment: a review. Environmental Science and Pollution Research 23 (6), 4978–4991. https://doi.org/10.1007/s11356-014-3617-x.

Auguet, O., Pijuan, M., Borrego, C.M., Rodriguez-Mozaz, S., Triadó-Margarit, X., Giustina, S.V., Della, & Gutierrez, O., 2017. Sewers as potential reservoirs of antibiotic resistance. Science of the Total Environment 605–606, 1047–1054. https://doi.org/10.1016/j.scitotenv.2017.06.153.

aus der Beek, T., Weber, F. A., Bergmann, A., Hickmann, S., Ebert, I., Hein, A., & Küster, A., 2016. Pharmaceuticals in the environment-Global occurrences and perspectives. Environmental Toxicology and Chemistry 35 (4), 823–835. https://doi.org/10.1002/etc.3339.

Ávila, C., García-Galán, M.J., Borrego, C.M., Rodríguez-Mozaz, S., García, J., Barceló, D., 2021. New insights on the combined removal of antibiotics and ARGs in urban wastewater through the use of two configurations of vertical subsurface flow constructed wetlands. Science of the Total Environment 755, 142554. https://doi.org/10.1016/j.scitotenv.2020.142554.

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

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

Badia-Fabregat, M., Lucas, D., Pereira, M.A., Alves, M., Pennanen, T., Fritze, H., Rodríguez-Mozaz, S., Barceló, D., Vicent, T., Caminal, G., 2016. Continuous fungal treatment of non-sterile veterinary hospital effluent: pharmaceuticals removal and microbial community assessment. Applied Microbiology and Biotechnology 100 (5), 2401–2415. https://doi.org/10.1007/s00253-015-7105-0.

Badia-Fabregat, M., Lucas, D., Tuomivirta, T., Fritze, H., Pennanen, T., Rodríguez-Mozaz, S., Barceló, D., Caminal, G., Vicent, T., 2017. Study of the effect of the bacterial and fungal communities present in real wastewater effluents on the performance of fungal treatments. Science of the Total Environment 579, 366–377. https://doi.org/10.1016/j.scitotenv.2016.11.088.

Barcellos, D.d.S., Procopiuck, M., Bollmann, H.A., 2022. Management of pharmaceutical micropollutants discharged in urban waters: 30 years of systematic review looking at opportunities for developing countries. Science of the Total Environment 809, 151128.

Becker, D., Rodriguez-Mozaz, S., Insa, S., Schoevaart, R., Barceló, D., De Cazes, M., Belleville, M.P., Sanchez-Marcano, J., Misovic, A., Oehlmann, J., Wagner, M., 2017. Removal of Endocrine Disrupting Chemicals in Wastewater by Enzymatic Treatment with Fungal Laccases. Organic Process Research and Development 21 (4), 480–491. https://doi.org/10.1021/acs.oprd.6b00361.

Bellos, D., Sawidis, T., 2005. Chemical pollution monitoring of the River Pinios (Thessalia - Greece). Journal of Environmental Management 76 (4), 282–292. https://doi.org/10.1016/j.jenvman.2005.01.027.

Besse, J.P., Garric, J., 2008. Human pharmaceuticals in surface waters. Implementation of a prioritization methodology and application to the French situation. Toxicology Letters 176 (2), 104–123. https://doi.org/10.1016/j.toxlet.2007.10.012.

Bloszies, C.S., Fiehn, O., 2018. Using untargeted metabolomics for detecting exposome compounds. Current Opinion in Toxicology 8, 87–92. https://doi.org/10.1016/J.COTOX.2018.03.002.

Blum, K.M., Andersson, P.L., Renman, G., Ahrens, L., Gros, M., Wiberg, K., Haglund, P., 2017. Non-target screening and prioritization of potentially persistent, bioaccumulating and toxic domestic wastewater contaminants and their removal in on-site and large-scale sewage treatment plants. Science of the Total Environment 575, 265–275. https://doi.org/10.1016/j.scitotenv.2016.09.135.

Borecka, M., Siedlewicz, G., Haliński, Ł.P., Sikora, K., Pazdro, K., Stepnowski, P., Białk-Bielińska, A., 2015. Contamination of the southern Baltic Sea waters by the residues of selected pharmaceuticals: Method development and field studies. Marine Pollution Bulletin 94 (1–2), 62–71. https://doi.org/10.1016/j.marpolbul.2015.03.008.

Borova, V.L., Maragou, N.C., Gago-Ferrero, P., Pistos, C., Thomaidis, N.S., 2014. Highly sensitive determination of 68 psychoactive pharmaceuticals, illicit drugs, and related human metabolites in wastewater by liquid chromatography-tandem mass spectrometry. Analytical and Bioanalytical Chemistry 406 (17), 4273–4285. https://doi.org/10.1007/s00216-014-7819-3.

Brown, A.K., Ackerman, J., Cicek, N., Wong, C.S., 2020. In situ kinetics of human pharmaceutical conjugates and the impact of transformation, deconjugation, and sorption on persistence in wastewater batch bioreactors. Environmental Pollution 265, 114852. https://doi.org/10.1016/J.ENVPOL.2020.114852.

Castaño-Trias, M., Brienza, M., Tomei, M.C., Buttiglieri, G., 2020. Fate and Removal of Pharmaceuticals in CAS for Water and Sewage Sludge Reuse. Springer, Berlin, Heidelberg, pp. 1–29.

Castiglioni, S., Bijlsma, L., Covaci, A., Emke, E., Hernández, F., Reid, M., Ort, C., Thomas, K.V., Van Nuijs, A.L.N., De Voogt, P., Zuccato, E., 2013. Evaluation of uncertainties associated with the determination of community drug use through the measurement of sewage drug biomarkers. Environmental Science and Technology 47 (3), 1452–1460. https://doi.org/10.1021/ES302722F/SUPPL_FILE/ES302722F_SI_004.XLS.

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

Chiţescu, C. L., Ene, A., Geana, E. I., Vasile, A. M., & Ciucure, C. T. (2021). Emerging and Persistent Pollutants in the Aquatic Ecosystems of the Lower Danube Basin and North West Black Sea Region—A Review. *Applied Sciences 2021, Vol. 11, Page 9721*, *11*(20), 9721. 10.3390/APP11209721.

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

Couto, C.F., Lange, L.C., Amaral, M.C.S., 2019. Occurrence, fate and removal of pharmaceutically active compounds (PhACs) in water and wastewater treatment plants—A review. Journal of Water Process Engineering 32, 100927. https://doi.org/10.1016/J.JWPE.2019.100927.

Cruz-Morató, C., Ferrando-Climent, L., Rodriguez-Mozaz, S., Barceló, D., Marco-Urrea, E., Vicent, T., Sarrà, M., 2013. Degradation of pharmaceuticals in non-sterile urban wastewater by Trametes versicolor in a fluidized bed bioreactor. Water Research 47 (14), 5200–5210. https://doi.org/10.1016/j.watres.2013.06.007.

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

Demirbas, A., 2011. Waste management, waste resource facilities and waste conversion processes. Energy Conversion and Management 52 (2), 1280–1287.

Desbiolles, F., Malleret, L., Tiliacos, C., Wong-Wah-Chung, P., & Laffont-Schwob, I. (2018). Occurrence and ecotoxicological assessment of pharmaceuticals: Is there a risk for the Mediterranean aquatic environment? In *Science of the Total Environment* (Vol. 639, pp. 1334–1348). Elsevier B.V. 10.1016/j.scitotenv.2018.04.351.

Dong, Z., Senn, D.B., Moran, R.E., Shine, J.P., 2013. Prioritizing environmental risk of prescription pharmaceuticals. Regulatory Toxicology and Pharmacology 65 (1), 60–67. https://doi.org/10.1016/j.yrtph.2012.07.003.

Eichhorn, P., Aga, D.S., 2004. Identification of a photooxygenation product of chlortetracycline in hog lagoons using LC/ESI-ion trap-MS and LC/ESI-time-of-flight-MS. Analytical Chemistry 76 (20), 6002–6011. https://doi.org/10.1021/ac0494127.

Escolà Casas, M., Schröter, N.S., Zammit, I., Castaño-Trias, M., Rodriguez-Mozaz, S., Gago-Ferrero, P., Corominas, L., 2021. Showcasing the potential of wastewater-based epidemiology to track pharmaceuticals consumption in cities: Comparison against prescription data collected at fine spatial resolution. Environment International 150, 106404. https://doi.org/10.1016/J.ENVINT.2021.106404.

Escudero-Oñate, C., Rodríguez-Mozaz, S., Ferrando-Climent, L., 2020. Tamoxifen: Occurrence, fate, transformation products, and non- conventional treatment technologies. In: Heath, E., Isidori, M., Kosjek, T., Filipič, M. (Eds.), Fate and Effects of Anticancer Drugs in the Environment. Springer International Publishing, Cham, pp. 71–86.

Ferrando-Climent, L., Collado, N., Buttiglieri, G., Gros, M., Rodriguez-Roda, I., Rodriguez-Mozaz, S., Barceló, D., 2012. Comprehensive study of ibuprofen and its metabolites in activated sludge batch experiments and aquatic environment. Science of the Total Environment 438, 404–413. https://doi.org/10.1016/j.scitotenv.2012.08.073.

Gago-Ferrero, P., Krettek, A., Fischer, S., Wiberg, K., Ahrens, L., 2018. Suspect Screening and Regulatory Databases: A Powerful Combination to Identify Emerging Micropollutants. Environmental Science and Technology 52 (12), 6881–6894. https://doi.org/10.1021/acs.est.7b06598.

García-Galán, M.J., Petrovic, M., Rodríguez-Mozaz, S., Barceló, D., 2016. Multiresidue trace analysis of pharmaceuticals, their human metabolites and transformation products by fully automated on-line solid-phase extraction-liquid chromatography-tandem mass spectrometry. Talanta 158, 330–341. https://doi.org/10.1016/j.talanta.2016.05.061.

García-Galán, M.J., Arashiro, L., Santos, L.H.M.L.M., Insa, S., Rodríguez-Mozaz, S., Barceló, D., Ferrer, I., Garfí, M., 2020. Fate of priority pharmaceuticals and their main metabolites and transformation products in microalgae-based wastewater treatment systems. Journal of Hazardous Materials 390, 121771. https://doi.org/10.1016/j.jhazmat.2019.121771.

Ginebreda, A., Muñoz, I., de Alda, M.L., Brix, R., López-Doval, J., Barceló, D., 2010. Environmental risk assessment of pharmaceuticals in rivers: Relationships between hazard indexes and aquatic macroinvertebrate diversity indexes in the Llobregat River (NE Spain). Environment International 36 (2), 153–162. https://doi.org/10.1016/j.envint.2009.10.003.

Gómez-Canela, C., Pueyo, V., Barata, C., Lacorte, S., Marcé, R.M., 2019. Development of predicted environmental concentrations to prioritize the occurrence of pharmaceuticals in rivers from Catalonia. Science of the Total Environment 666, 57–67. https://doi.org/10.1016/j.scitotenv.2019.02.078.

Gros, M., Rodríguez-Mozaz, S., Barceló, D., 2012. Fast and comprehensive multi-residue analysis of a broad range of human and veterinary pharmaceuticals and some of their metabolites in surface and treated waters by ultra-high-performance liquid chromatography coupled to quadrupole-linear ion trap tandem. Journal of Chromatography A 1248, 104–121. https://doi.org/10.1016/j.chroma.2012.05.084.

Gros, M., Rodríguez-Mozaz, S., Barceló, D., 2013. Rapid analysis of multiclass antibiotic residues and some of their metabolites in hospital, urban wastewater and river water by ultra-high-performance liquid chromatography coupled to quadrupole-linear ion trap tandem mass spectrometry. Journal of Chromatography A 1292, 173–188. https://doi.org/10.1016/j.chroma.2012.12.072.

Gros, M., Blum, K.M., Jernstedt, H., Renman, G., Rodríguez-Mozaz, S., Haglund, P., Andersson, P.L., Wiberg, K., Ahrens, L., 2017. Screening and prioritization of

micropollutants in wastewaters from on-site sewage treatment facilities. Journal of Hazardous Materials 328, 37–45. https://doi.org/10.1016/j.jhazmat.2016.12.055.
Heye, K., Becker, D., Lütke Eversloh, C., Durmaz, V., Ternes, T.A., Oetken, M., Oehlmann, J., 2016. Effects of carbamazepine and two of its metabolites on the non-biting midge Chironomus riparius in a sediment full life cycle toxicity test. Water Research 98, 19–27. https://doi.org/10.1016/j.watres.2016.03.071.
Hom-Diaz, A., Jaén-Gil, A., Bello-Laserna, I., Rodríguez-Mozaz, S., Vicent, T., Barceló, D., Blánquez, P., 2017. Performance of a microalgal photobioreactor treating toilet wastewater: Pharmaceutically active compound removal and biomass harvesting. Science of the Total Environment 592, 1–11. https://doi.org/10.1016/j.scitotenv.2017.02.224.
Huerta, B., Marti, E., Gros, M., López, P., Pompêo, M., Armengol, J., Barceló, D., Balcázar, J.L., Rodríguez-Mozaz, S., Marcé, R., 2013. Exploring the links between antibiotic occurrence, antibiotic resistance, and bacterial communities in water supply reservoirs. Science of the Total Environment 456–457, 161–170. https://doi.org/10.1016/j.scitotenv.2013.03.071.
Ibáñez, M., Guerrero, C., Sancho, J.V., Hernández, F., 2009. Screening of antibiotics in surface and wastewater samples by ultra-high-pressure liquid chromatography coupled to hybrid quadrupole time-of-flight mass spectrometry. Journal of Chromatography A 1216 (12), 2529–2539. https://doi.org/10.1016/j.chroma.2009.01.073.
Ibáñez, M., Bijlsma, L., Pitarch, E., López, F.J., Hernández, F., 2021. Occurrence of pharmaceutical metabolites and transformation products in the aquatic environment of the Mediterranean area. Trends in Environmental Analytical Chemistry 29, e00118.
Jaén-Gil, A., Castellet-Rovira, F., Llorca, M., Villagrasa, M., Sarrà, M., Rodríguez-Mozaz, S., Barceló, D., 2019. Fungal treatment of metoprolol and its recalcitrant metabolite metoprolol acid in hospital wastewater: Biotransformation, sorption and ecotoxicological impact. Water Research 152, 171–180. https://doi.org/10.1016/j.watres.2018.12.054.
Jelic, A., Rodriguez-Mozaz, S., Barceló, D., Gutierrez, O., 2015. Impact of in-sewer transformation on 43 pharmaceuticals in a pressurized sewer under anaerobic conditions. Water Research 68, 98–108. https://doi.org/10.1016/j.watres.2014.09.033.
Kandie, F.J., Krauss, M., Beckers, L.M., Massei, R., Fillinger, U., Becker, J., Liess, M., Torto, B., Brack, W., 2020. Occurrence and risk assessment of organic micropollutants in freshwater systems within the Lake Victoria South Basin. Kenya. *Science of The Total Environment* 714, 136748. https://doi.org/10.1016/J.SCITOTENV.2020.136748.
Khan, N.A., Ahmed, S., Farooqi, I.H., Ali, I., Vambol, V., Changani, F., Yousefi, M., Vambol, S., Khan, S.U., Khan, A.H., 2020. Occurrence, sources and conventional treatment techniques for various antibiotics present in hospital wastewaters: A critical review. TrAC Trends in Analytical Chemistry 129, 115921. https://doi.org/10.1016/J.TRAC.2020.115921.
Kim, M.K., Zoh, K.D., 2016. Occurrence and removals of micropollutants in water environment. Environmental Engineering Research 21 (4), 319–332. https://doi.org/10.4491/eer.2016.115.
Knopp, G., Prasse, C., Ternes, T.A., Cornel, P., 2016. Elimination of micropollutants and transformation products from a wastewater treatment plant effluent through pilot scale ozonation followed by various activated carbon and biological filters. Water Research 100, 580–592. https://doi.org/10.1016/J.WATRES.2016.04.069.
Kosińska, K., Frański, R., Frańska, M., 2016. Unusually stable adduct between methanolyzed amoxicillin or ampicillin and their diketopiperazine derivatives. Acta Poloniae Pharmaceutica - Drug Research 73 (2), 297–302.
Kosma, C.I., Lambropoulou, D.A., Albanis, T.A., 2010. Occurrence and removal of PPCPs in municipal and hospital wastewaters in Greece. Journal of Hazardous Materials 179 (1–3), 804–817. https://doi.org/10.1016/j.jhazmat.2010.03.075.
Kosma, C.I., Lambropoulou, D.A., Albanis, T.A., 2014. Investigation of PPCPs in wastewater treatment plants in Greece: Occurrence, removal and environmental risk assessment. Science of the Total Environment 466–467, 421–438. https://doi.org/10.1016/j.scitotenv.2013.07.044.
Kümmerer, K., 2009. The presence of pharmaceuticals in the environment due to human use – present knowledge and future challenges. Journal of Environmental Management 90 (8), 2354–2366. https://doi.org/10.1016/J.JENVMAN.2009.01.023.
Kuzmanovic, M., Banjac, Z., Ginebreda, A., Petrovic, M., & Barcelo, D. (2013). Prioritization: Selection of environmentally occurring pharmaceuticals to be monitored. In *Comprehensive Analytical Chemistry* (Vol. 62, pp. 71–90). Elsevier B.V. 10.1016/B978-0-444-62657-8.00003-3.
Llasat, M.C., del Moral, A., Cortès, M., Rigo, T., 2021. Convective precipitation trends in the Spanish Mediterranean region. Atmospheric Research 257, 105581. https://doi.org/10.1016/J.ATMOSRES.2021.105581.
Lucas, D., Badia-Fabregat, M., Vicent, T., Caminal, G., Rodríguez-Mozaz, S., Balcázar, J. L., Barceló, D., 2016. Fungal treatment for the removal of antibiotics and antibiotic resistance genes in veterinary hospital wastewater. Chemosphere 152, 301–308. https://doi.org/10.1016/j.chemosphere.2016.02.113.
Luo, Y., Guo, W., Ngo, H. H., Nghiem, L. D., Hai, F. I., Zhang, J., Liang, S., & Wang, X. C. (2014). A review on the occurrence of micropollutants in the aquatic environment and their fate and removal during wastewater treatment. In *Science of the Total Environment* (Vols. 473–474, pp. 619–641). Elsevier B.V. 10.1016/j.scitotenv.2013.12.065.
Majewsky, M., Glauner, T., Horn, H., 2015. Systematic suspect screening and identification of sulfonamide antibiotic transformation products in the aquatic environment. Analytical and Bioanalytical Chemistry 407 (19), 5707–5717. https://doi.org/10.1007/s00216-015-8748-5.
Mamo, J., García-Galán, M.J., Stefani, M., Rodríguez-Mozaz, S., Barceló, D., Monclús, H., Rodriguez-Roda, I., Comas, J., 2018. Fate of pharmaceuticals and their transformation products in integrated membrane systems for wastewater reclamation. Chemical Engineering Journal 331, 450–461. https://doi.org/10.1016/j.cej.2017.08.050.
Mandaric, L., Mor, J.R., Sabater, S., Petrovic, M., 2018. Impact of urban chemical pollution on water quality in small, rural and effluent-dominated Mediterranean streams and rivers. Science of the Total Environment 613–614, 763–772. https://doi.org/10.1016/j.scitotenv.2017.09.128.
Mandaric, L., Kalogianni, E., Skoulikidis, N., Petrovic, M., Sabater, S., 2019. Contamination patterns and attenuation of pharmaceuticals in a temporary Mediterranean river. Science of the Total Environment 647, 561–569. https://doi.org/10.1016/j.scitotenv.2018.07.308.
Margot, J., Rossi, L., Barry, D.A., Holliger, C., 2015. A review of the fate of micropollutants in wastewater treatment plants. Wiley Interdisciplinary Reviews: Water 2 (5), 457–487. https://doi.org/10.1002/wat2.1090.
Martins, R., Boaventura, R., Paulista, L., Swarnakar, A.K., Bajpai, S., Ahmad, I., Vistanty, H., Crisnaningtyas, F., 2021. Integration of upflow anaerobic sludge blanket and constructed wetlands for pharmaceutical wastewater treatment. IOP Conference Series: Earth and Environmental Science 623 (1), 012082. https://doi.org/10.1088/1755-1315/623/1/012082.
Mir-Tutusaus, J.A., Parladé, E., Llorca, M., Villagrasa, M., Barceló, D., Rodriguez-Mozaz, S., Martinez-Alonso, M., Gaju, N., Caminal, G., Sarrà, M., 2017. Pharmaceuticals removal and microbial community assessment in a continuous fungal treatment of non-sterile real hospital wastewater after a coagulation-flocculation pretreatment. Water Research 116, 65–75. https://doi.org/10.1016/j.watres.2017.03.005.
Mir-Tutusaus, J.A., Parladé, E., Villagrasa, M., Barceló, D., Rodríguez-Mozaz, S., Martínez-Alonso, M., Gaju, N., Sarrà, M., Caminal, G., 2019. Long-term continuous treatment of non-sterile real hospital wastewater by Trametes versicolor. Journal of Biological Engineering 13 (1), 47. https://doi.org/10.1186/s13036-019-0179-y.
Mir-Tutusaus, J.A., Jaén-Gil, A., Barceló, D., Buttiglieri, G., Gonzalez-Olmos, R., Rodriguez-Mozaz, S., Caminal, G., Sarrà, M., 2021. Prospects on coupling UV/H2O2 with activated sludge or a fungal treatment for the removal of pharmaceutically active compounds in real hospital wastewater. Science of the Total Environment 773, 145374. https://doi.org/10.1016/j.scitotenv.2021.145374.
Nannou, C.I., Kosma, C.I., Albanis, T.A., 2015. Occurrence of pharmaceuticals in surface waters: analytical method development and environmental risk assessment. International Journal of Environmental Analytical Chemistry 95 (13), 1242–1262. https://doi.org/10.1080/03067319.2015.1085520.
Naveen Kumar, V.P., Jafrin Antony, L., 2021. Prescription pattern of drugs used in pediatric patients of a tertiary care hospital in Puducherry. National Journal of Physiology, Pharmacy and Pharmacology 380 (2021), 4. https://doi.org/10.5455/njppp.2021.11.11328202006122020.
Nikolaou, G., Neocleous, D., Christou, A., Kitta, E., Katsoulas, N., 2020. Implementing sustainable irrigation in water-scarce regions under the impact of climate change. Agronomy 10 (8). https://doi.org/10.3390/AGRONOMY10081120.
Nödler, K., Voutsa, D., Licha, T., 2014. Polar organic micropollutants in the coastal environment of different marine systems. Marine Pollution Bulletin 85 (1), 50–59. https://doi.org/10.1016/j.marpolbul.2014.06.024.
Osorio, V., Marcé, R., Pérez, S., Ginebreda, A., Cortina, J.L., Barceló, D., 2012. Occurrence and modeling of pharmaceuticals on a sewage-impacted Mediterranean river and their dynamics under different hydrological conditions. Science of the Total Environment 440, 3–13. https://doi.org/10.1016/j.scitotenv.2012.08.040.
Paíga, P., Santos, L.H.M.L.M., Delerue-Matos, C., 2017. Development of a multi-residue method for the determination of human and veterinary pharmaceuticals and some of their metabolites in aqueous environmental matrices by SPE-UHPLC–MS/MS. Journal of Pharmaceutical and Biomedical Analysis 135, 75–86. https://doi.org/10.1016/j.jpba.2016.12.013.
Palau, R.M., Berenguer, M., Hürlimann, M., Sempere-Torres, D., 2022. Application of a fuzzy verification framework for the evaluation of a regional-scale landslide early warning system during the January 2020 Gloria storm in Catalonia (NE Spain). Landslides 19 (7), 1599–1616. https://doi.org/10.1007/S10346-022-01854-2/FIGURES/12.
Pantanella, F., Lekunberri, I., Gagliardi, A., Venuto, G., Sànchez-Melsió, A., Fabiani, M., Balcázar, J.L., Schippa, S., De Giusti, M., Borrego, C., Solimini, A., 2020. Effect of Urban Wastewater Discharge on the Abundance of Antibiotic Resistance Genes and Antibiotic-Resistant Escherichia coli in Two Italian Rivers. International Journal of Environmental Research and Public Health 17 (18), 6813. https://doi.org/10.3390/ijerph17186813.
Papageorgiou, M., Kosma, C., Lambropoulou, D., 2016. Seasonal occurrence, removal, mass loading and environmental risk assessment of 55 pharmaceuticals and personal care products in a municipal wastewater treatment plant in Central Greece. Science of the Total Environment 543, 547–569. https://doi.org/10.1016/j.scitotenv.2015.11.047.
Papageorgiou, M., Zioris, I., Danis, T., Bikiaris, D., Lambropoulou, D., 2019. Comprehensive investigation of a wide range of pharmaceuticals and personal care products in urban and hospital wastewaters in Greece. Science of the Total Environment 694, 133565. https://doi.org/10.1016/j.scitotenv.2019.07.371.
Park, N., Choi, Y., Kim, D., Kim, K., Jeon, J., 2018. Prioritization of highly exposable pharmaceuticals via a suspect/non-target screening approach: A case study for Yeongsan River, Korea. Science of the Total Environment 639, 570–579. https://doi.org/10.1016/j.scitotenv.2018.05.081.
Patel, M., Kumar, R., Kishor, K., Mlsna, T., Pittman, C.U., Mohan, D., 2019. Pharmaceuticals of Emerging Concern in Aquatic Systems: Chemistry. Occurrence, Effects, and Removal Methods. 119 (6), 3510–3673.

Petrie, B., Barden, R., Kasprzyk-Hordern, B., 2015. A review on emerging contaminants in wastewaters and the environment: Current knowledge, understudied areas and recommendations for future monitoring. Water Research 72, 3–27. https://doi.org/10.1016/J.WATRES.2014.08.053.

Previšić, A., Rožman, M., Mor, J.R., Acuña, V., Serra-Compte, A., Petrović, M., Sabater, S., 2019. Aquatic macroinvertebrates under stress: Bioaccumulation of emerging contaminants and metabolomics implications. Science of the Total Environment 704, 135333. https://doi.org/10.1016/j.scitotenv.2019.135333.

Raldua, D., Campos, B., Barata, C., Piña, B., García-Reyero, N., Babin, P.J., 2013. Deciphering Emerging Toxicological Effects of Pharmaceuticals on Aquatic Organisms by Using Daphnia magna and Danio rerio as Model Organisms. Comprehensive Analytical Chemistry 62, 611–647. https://doi.org/10.1016/B978-0-444-62657-8.00017-3.

Riva, F., Zuccato, E., Castiglioni, S., 2015. Prioritization and analysis of pharmaceuticals for human use contaminating the aquatic ecosystem in Italy. Journal of Pharmaceutical and Biomedical Analysis 106, 71–78. https://doi.org/10.1016/j.jpba.2014.10.003.

Rodriguez-Mozaz, S., Chamorro, S., Marti, E., Huerta, B., Gros, M., Sànchez-Melsió, A., Borrego, C.M., Barceló, D., Balcázar, J.L., 2015. Occurrence of antibiotics and antibiotic resistance genes in hospital and urban wastewaters and their impact on the receiving river. Water Research 69, 234–242. https://doi.org/10.1016/j.watres.2014.11.021.

Rodriguez-Mozaz, S., Vaz-Moreira, I., Varela Della Giustina, S., Llorca, M., Barceló, D., Schubert, S., Berendonk, T.U., Michael-Kordatou, I., Fatta-Kassinos, D., Martinez, J. L., Elpers, C., Henriques, I., Jaeger, T., Schwartz, T., Paulshus, E., O'Sullivan, K., Pärnänen, K.M.M., Virta, M., Do, T.T., Walsh, F., Manaia, C.M., 2020. Antibiotic residues in final effluents of European wastewater treatment plants and their impact on the aquatic environment. Environment International 140, 105733.

Rosal, R., Rodríguez, A., Perdigón-Melón, J.A., Petre, A., García-Calvo, E., Gómez, M.J., Agüera, A., Fernández-Alba, A.R., 2010. Occurrence of emerging pollutants in urban wastewater and their removal through biological treatment followed by ozonation. Water Research 44 (2), 578–588. https://doi.org/10.1016/j.watres.2009.07.004.

Rubirola, A., Llorca, M., Rodriguez-Mozaz, S., Casas, N., Rodriguez-Roda, I., Barceló, D., Buttiglieri, G., 2014. Characterization of metoprolol biodegradation and its transformation products generated in activated sludge batch experiments and in full scale WWTPs. Water Research 63, 21–32. https://doi.org/10.1016/j.watres.2014.05.031.

Sangion, A., Gramatica, P., 2016. PBT assessment and prioritization of contaminants of emerging concern: Pharmaceuticals. Environmental Research 147, 297–306. https://doi.org/10.1016/J.ENVRES.2016.02.021.

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

Siegrist, H., Joss, A., 2012. Review on the fate of organic micropollutants in wastewater treatment and water reuse with membranes. Water Science and Technology 66 (6), 1369–1376. https://doi.org/10.2166/WST.2012.285.

Stamatis, N., Triantafyllidis, V., Hela, D., Konstantinou, I., 2013. Occurrence and distribution of selected pharmaceutical compounds on sewage-impacted section of River Acheloos, Western Greece. International Journal of Environmental Analytical Chemistry 93 (15), 1602–1619. https://doi.org/10.1080/03067319.2013.814121.

Stasinakis, A.S., Mermigka, S., Samaras, V.G., Farmaki, E., Thomaidis, N.S., 2012. Occurrence of endocrine disrupters and selected pharmaceuticals in Aisonas River (Greece) and environmental risk assessment using hazard indexes. Environmental Science and Pollution Research 19 (5), 1574–1583. https://doi.org/10.1007/s11356-011-0661-7.

The UBA database – "Pharmaceuticals in the environment" | Umweltbundesamt. (n.d.). Retrieved February 21, 2023, from https://www.umweltbundesamt.de/en/database-pharmaceuticals-in-the-environment-0.

Tsaboula, A., Papadakis, E.N., Vryzas, Z., Kotopoulou, A., Kintzikoglou, K., Papadopoulou-Mourkidou, E., 2016. Environmental and human risk hierarchy of pesticides: A prioritization method, based on monitoring, hazard assessment and environmental fate. Environment International 91, 78–93. https://doi.org/10.1016/J.ENVINT.2016.02.008.

Valdés, M.E., Huerta, B., Wunderlin, D.A., Bistoni, M.A., Barceló, D., Rodriguez-Mozaz, S., 2016. Bioaccumulation and bioconcentration of carbamazepine and other pharmaceuticals in fish under field and controlled laboratory experiments. Evidences of carbamazepine metabolization by fish. Science of the Total Environment 557–558, 58–67. https://doi.org/10.1016/j.scitotenv.2016.03.045.

Valdez-Carrillo, M., Abrell, L., Ramírez-Hernández, J., Reyes-López, J. A., & Carreón-Diazconti, C. (2020). Pharmaceuticals as emerging contaminants in the aquatic environment of Latin America: a review. *Environmental Science and Pollution Research 2020 27:36*, *27*(36), 44863–44891. 10.1007/S11356-020-10842-9.

Verlicchi, P., Al Aukidy, M., Zambello, E., 2012. Occurrence of pharmaceutical compounds in urban wastewater: Removal, mass load and environmental risk after a secondary treatment—A review. Science of The Total Environment 429, 123–155.