



Suspended TiO₂-assisted photocatalytic degradation of emerging contaminants in a municipal WWTP effluent using a solar pilot plant with CPCs

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H I G H L I G H T S

- ▶ Municipal WWTP effluent contaminated with pharmaceuticals and earthy-musty compounds.
- ▶ CPC pilot plant for TiO₂-solar photocatalysis as a complementary tertiary treatment.
- ▶ Complete removal of 19 out of 22 pharmaceuticals with ca. 32 kJ L⁻¹ solar UV energy.
- ▶ Zahn–Wellens test indicates many pharmaceuticals are refractory to biotreatment.
- ▶ *V. fischeri* bioassay revealed no toxicity increase during photocatalytic treatment.

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Emerging pollutants, such as pharmaceuticals, are widely disseminated in the aquatic media. Though in low concentrations in the environment, they still pose concerns mainly over potential chronic toxicity effects. Consequently, this work reports on the successful attempt to develop a photocatalytic treatment method, using suspended TiO₂ in a concentration of 200 mg L⁻¹ and solar UV radiation as the photon source, firstly designed for lorazepam (Lorenin® pills) degradation and further applied to the treatment of a real municipal WWTP effluent, containing several other emerging contaminants (ECs).

Initial effluent physicochemical characterization revealed the presence of 22 pharmaceutical compounds in moderate concentrations (maximum of 680 ng L⁻¹, except for diclofenac ~24 µg L⁻¹ and hydrochlorothiazide ~3 µg L⁻¹) and a low dissolved organic carbon (DOC) content. Therefore, the main purpose of the work was not to increase the effluent's biodegradability, but to improve the removal rates of the several present ECs.

Pharmaceuticals' degradation kinetics, using a solar pilot plant with CPCs, were thoroughly studied. A pseudo-first order kinetic model was able to successfully predict the experimental data. The overall treatment was considered efficient, with a complete removal of the majority of these micropollutants, except for ciprofloxacin (35%), ketoprofen (61%) and bisoprolol (77%). Nevertheless, a small increase in the reaction time could easily accomplish their total degradation.

Zahn–Wellens biodegradability assay allowed withdrawing some conclusions about which pharmaceuticals could be degraded by means of biotreatment, thus avoiding the need to apply a photocatalytic treatment.

Finally, *Vibrio fischeri* acute toxicity test showed that the effluent itself presented no significant toxicity and that the intermediate oxidation compounds, possibly formed during phototreatment, did not reflect any significant increase of toxicity.

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1. Introduction

Emerging pollutants are defined as compounds that are not currently covered by the existing legislation in the field of water

quality, whose environmental impact is not yet sufficiently studied and which are thought to be potentially harmful to environmental ecosystems and human health. They encompass a wide range of products, including pharmaceuticals, personal care products, fragrances, detergents, plasticizers, flame retardants, pesticides and several other classes [1].

Concerns about the hazards associated with long-term exposure to pharmaceutically active compounds for non-target organisms and for

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human health are rising. One of the best known detrimental effects of these compounds in the environment is the disruption of the endocrine system in wild species, affecting their growth, physiology and reproduction. The selection of multi-resistant strains of pathogenic microorganisms is also a threat to humans and ecosystems, resulting from the uncontrolled use of antibiotics and deficient treatment of effluents from wastewater treatment plants (WWTPs) [2,3].

In fact, WWTPs' effluents are considered a major source of environmental pharmaceutical contamination, since they are not prepared for the complete removal or degradation of such micropollutants, as mentioned in several studies [4–6]. They can, therefore, reach the aquatic media, where they have been found in concentrations ranging from 0.1 to 20 $\mu\text{g L}^{-1}$, among treated wastewaters, surface and groundwater and even drinking water [7–11].

Recently, there has been a research outbreak in the field of advanced water treatment technologies, designed to become part of the solution to this problem. They include the advanced oxidation processes (AOPs) (using the combination of strong oxidants, e.g. O_3 or H_2O_2 , with high energy sources, such as ultraviolet (UV), ultrasound (US) or electron beam (EB), and catalysts, e.g. Fe^{2+} , and photocatalysts, e.g. TiO_2 , to produce hydroxyl radicals), membrane based technologies (microfiltration, ultrafiltration, nanofiltration and reverse osmosis) and adsorption/ion exchange processes (activated carbon, resins), each one presenting their own advantages and disadvantages [12].

Special attention has been given to AOPs, in which hydroxyl radicals ($\cdot\text{OH}$) are the responsible agents for the oxidation and mineralization of almost any organic molecule, yielding CO_2 and inorganic ions as final products, due to their strong unselective oxidative power [13]. Among AOPs, the heterogeneous photocatalysis with UV/ TiO_2 has shown to be a promising technique for wastewater detoxification [12]. In heterogeneous photocatalysis, dispersed solid particles absorb efficiently larger fractions of the UV spectrum and generate chemical oxidants *in situ* from dissolved oxygen or water. Regarding the catalyst choice, TiO_2 has generally been demonstrated to be the most active, whenever tested against other semiconductor materials, under comparable conditions [14]. When illuminated with light energy higher than their bandgap, semiconductors like TiO_2 (BGE = 3.2 eV) produce excited high-energy states of electron and hole pairs (e^-/h^+) [12]. TiO_2 is also technologically interesting due to its chemical and photocorrosion resistance, its safety and low cost and, specially, to its capability of using natural and renewable solar UV energy (since the energetic separation between its valence and conduction bands can be surpassed by the energy content of a solar photon) [14].

The purpose of this work was to study the application of heterogeneous photocatalysis with solar UV/ TiO_2 to the degradation of an anxiolytic drug – lorazepam – which is frequently detected in WWTPs' effluents and surface waters [1]. To the authors' knowledge, never has this drug been studied under accelerated phototransformation processes (photo-induced structural alterations). To do so, a solar pilot plant with compound parabolic collectors (CPCs) designed for solar photo(cata)lytic applications was used, taking profit of solar renewable energy. Besides, lorazepam was tested in one of its most commercialized dosage forms in Portugal – Lorenin[®] pills, 1 mg (Wyeth) – in order to simulate a more realistic scenario. The optimized treatment process was further applied to a real WWTP effluent containing, apart from lorazepam, several other emerging contaminants.

2. Experimental

2.1. Contaminated water samples

TiO_2 (P25 Degussa, 80% anatase and 20% rutile) concentration optimization was performed using a ca. 200 $\mu\text{g L}^{-1}$ lorazepam solution, obtained by dissolving 1 mg Lorenin[®] pills (Wyeth) in dis-

tilled water [15]. Lorazepam's photodegradation (photo-induced structural simplification) was furthermore evaluated on a real wastewater effluent, where several other emerging contaminants were as well present.

Table 1 presents the main characteristics of the effluent sample collected at Febros WWTP. This WWTP discharges its treated effluent into Febros River, a southern bank small tributary of Douro River, located in the north of Portugal. Moreover, a 2 mg L^{-1} lorazepam spiked effluent sample was also submitted to photocatalytic treatment. This concentration was obtained by dissolving 40 Lorenin[®] pills (1 mg of lorazepam each) in 20 L of the real effluent sample. Besides assuring a proper kinetic characterization and possible future identification of some lorazepam's degradation products (LDPs) formed during photodegradation, this procedure also simulates a more realistic scenario, where lorazepam is among several other compounds, also present in pharmaceutical formulations (numerous excipients, including titanium dioxide itself).

2.2. Analytical determinations

pH, temperature and conductivity were measured using a pH meter HANNA HI8424 and a conductivity meter HANNA HI4522; turbidity was determined in a Merck[®] Turbiquant 3000 R; dissolved oxygen was measured using a multi-parametric YSI probe; oxidability was determined according to ISO 8467:1993 [16] and total suspended solids (TSSs) and volatile suspended solids (VSSs) were determined according to the standard methods book [17]. Nitrate, nitrite, bromide, chloride, fluoride and sulphate were quantified by ion chromatography (Dionex DX-120), using a Dionex Ionpac AS9-HC 4 \times 250 mm column. The program for anions determination comprises a 20 min run using 9 mM Na_2CO_3 as eluent at a flow rate of 1.0 mL min^{-1} . Ammonium and phosphate were measured with Merck[®] Spectroquant kits. Metals were analyzed and quantified using an ICP-MS (X-Series, Thermo Elemental). Dissolved organic carbon (DOC) was measured in a TC-TOC-TN analyzer (Shimadzu, model TOC-V_{CSN}) provided with a NDIR detector and calibrated with standard solutions of potassium phthalate. Dissolved nitrogen was measured by thermal decomposition and NO detection by a chemiluminescence method in the same TC-TOC-TN analyzer coupled with a TNM-1 unit (Shimadzu, model TOC-V_{CSN}) calibrated with standard solutions of potassium nitrate. Absorbance at 254 nm was determined using an Unicam spectrophotometer (model Super Sipper), in order to indirectly evaluate the content in aromatic compounds. Finally, the concentration profile of several pharmaceutical compounds was obtained by solid-phase extraction followed by liquid chromatography coupled to mass spectrometry (SPE-LC-tandem MS), according to the procedure thoroughly described by Sousa et al. [1], while some fragrances and earthy–musty compounds were also quantified by gas chromatography–mass spectrometry (scan or MS/MS mode), as detailed in Machado et al. [18].

During the photocatalytic experiment, pH, temperature, absorbance, total nitrogen (TN) and DOC were followed, giving indications about the mineralization state. Finally, all different emerging contaminants detected in the original effluent sample were also continuously followed through the entire photo-treatment process.

2.3. Solar CPC pilot plant

The solar TiO_2 -photocatalytic experiments were performed in a pilot plant with compound parabolic collectors (CPCs) installed in the roof of the Chemical Engineering Department of the Faculty of Engineering, University of Porto (FEUP), Portugal (Fig. 1) [19]. The plant is composed by one CPC unit (0.91 m^2) of four borosilicate tubes (Schott-Duran type 3.3, Germany, cut-off at 280 nm, external diameter 50 mm, length 1500 mm and thickness

Table 1
Physicochemical characterization of the effluent sample collected at Febros WWTP and used for the solar photocatalytic experiment.

Parameter (units)	Febros WWTP effluent (18/03/2011)	ELV ^a Dec. Lei no. 236/98, de 1 de Agosto 1998 (portuguese legislation)
Color	Pale yellow; n.d. ^b at dil.1:20	n.d. For dilution 1:20
Odor	n.d. (dil.1:20)	n.d. For dilution 1:20
pH	7.3	6.0–9.0
Temperature (°C)	20.0	3 °C Increase ^c
Turbidity (NTU)	115	–
Conductivity ($\mu\text{S cm}^{-1}$)	555	–
Dissolved oxygen (mg L^{-1})	2.2	–
Oxidability (mg L^{-1})	56.2	–
Total dissolved carbon (mg L^{-1})	37.5	–
Inorganic carbon (mg L^{-1})	25.4	–
Dissolved organic carbon (mg L^{-1})	12.1	–
Absorbance at 254 nm (AU)	0.07	–
Total suspended solids (mg L^{-1})	363	60
Volatile suspended solids (mg L^{-1})	223	–
Ammonium – NH_4^+ (mg L^{-1})	1.2	10
Nitrate – NO_3^- (mg L^{-1})	23.3	50
Nitrite – NO_2^- (mg L^{-1})	<0.5	–
Bromide – Br^- (mg L^{-1})	<0.5	–
Chloride – Cl^- (mg L^{-1})	78.5	–
Fluoride – F^- (mg L^{-1})	<0.5	–
Phosphate – PO_4^{3-} (mg L^{-1})	7	–
Sulphate – SO_4^{2-} (mg L^{-1})	59.8	2000
Phosphorus – ^{31}P (mg L^{-1})	3.0	10,000
Metals		
Sodium – ^{23}Na (mg L^{-1})	78.8	–
Potassium – ^{39}K (mg L^{-1})	21.1	–
Calcium – ^{44}Ca (mg L^{-1})	274.8	–

^a ELV – Emission limit value.

^b n.d. – Not detected.

^c Comparatively to the receptor medium.

1.8 mm) connected in series by polypropylene junctions with their CPC mirrors in anodized aluminum, supported by an aluminum structure and tilted 41° (local latitude). The pilot plant has also two recirculation tanks (10 L and 20 L), two recirculation pumps (maximum 20 L min⁻¹), two flow rate meters, five polypropylene valves and an electric board for process control. The pilot plant is operated in batch mode and can be used in two ways: using the total CPCs area (0.91 m²) or using 0.455 m² of the CPCs area individually, giving the possibility to carry out two distinct experiments at the same time and at the same solar radiation conditions.

The intensity of the solar UV radiation is measured by a global radiometer (ACADUS 85-PLS), placed at the same inclination angle, which provides the data in terms of instantaneous UV radiation (W m^{-2}). According to Eq. (1), one is able to calculate the accumu-

lated UV energy ($Q_{UV,n}$, kJ L^{-1}) received on any surface in the same position with regard to the sun, per unit of water volume inside the reactor, in a given time interval Δt_n :

$$Q_{UV,n} = Q_{UV,n-1} + \Delta t_n \overline{UV}_{G,n} \frac{A_r}{V_t}; \quad \Delta t_n = t_n - t_{n-1} \quad (1)$$

where t_n is the time corresponding to n -water sample (s), $V_{t,n}$ is the total volume of wastewater at n -water sample (L), A_r is the illuminated collector surface (m²), and $\overline{UV}_{G,n}$ is the average solar UV radiation (W m^{-2}) measured in the time interval Δt_n .

2.4. Experimental set-up

Photocatalytic processes were conducted during sunny days, using the solar CPC pilot plant described in Section 2.3.

Experiments using the WWTP effluent started with the addition of 20 L to one recirculation tank and 25 L to the other recirculation tank of the CPC units, followed by homogenization by turbulent recirculation during 15 min, in the darkness (a first control sample was taken for characterization). The goal was to perform two parallel experiments, one with the real effluent and another with lorazepam spiked effluent, under the same solar irradiation conditions, taking profit of the two independent CPC units (see Section 2.3.). Then, TiO_2 was added to both reservoirs, up to a final concentration of 0.2 g L⁻¹ – optimal determined concentration [15,20]. Next, Lorenin[®] pills were dissolved only in the reservoir supplying the CPC unit containing the 20 L of WWTP effluent, in order to achieve a lorazepam spiking concentration of 2 mg L⁻¹. After 15 min of turbulent recirculation in the dark, another control sample was taken from each tank to evaluate the contaminants' adsorption onto the TiO_2 surface and, finally, the solar collectors were uncovered and the experiments began.

Samples were taken at pre-defined times, from each recirculation tank, with several purposes: degradation kinetics evaluation (by SPE-LC-MS/MS [1] and GC-MS [18]) of the different emerging



Fig. 1. Solar Pilot Plant with CPCs where the TiO_2 -photocatalytic treatment of the WWTP effluent was performed.

contaminants, including lorazepam, DOC, TN and absorbance (254 nm) measurements, biodegradability and toxicity assays.

2.5. Biodegradability test

A 28 days Zahn–Wellens biodegradability test was carried out, according to the OECD standards [21], for biodegradability evaluation of all samples, collected at different phototreatment times, from the experiment with 2 ppm lorazepam spiked effluent. 240 mL of each sample were added to an open glass flask, along with some mineral nutrients (KH_2PO_4 , K_2HPO_4 , Na_2HPO_4 , NH_4Cl , CaCl_2 , MgSO_4 and FeCl_3) and activated sludge from Freixo WWTP (Porto, Portugal), previously centrifuged and added in proportion to the DOC of the sample. A control and a blank experiment were performed in parallel, using glucose as carbon source (highly biodegradable) and distilled water, respectively. All vessels were kept in the dark, inside a thermostatic chamber at 25 °C, under continuous magnetic stirring, for a 28 days period. The percentage of biodegradation (D_t) can be determined by Eq. (2):

$$D_t = \left[1 - \frac{(C_t - C_B)}{(C_A - C_{BA})} \right] \times 100 \quad (2)$$

where C_A and C_{BA} are the DOC (mg L^{-1}) in the mixture and in the blank experiment, respectively, measured 3 h after the beginning of the experiment, C_t and C_B are the DOC (mg L^{-1}) in the mixture and in the blank experiment, measured at the sampling time t . Samples are considered biodegradable only when D_t is higher than 70%. After the 28 days experiment, the levels of lorazepam, as well as of all other pharmaceuticals previously detected, were determined by SPE-LC-MS/MS [1] and compared to those obtained prior to the biodegradability assay.

2.6. Toxicity assessment

The toxicity test herein used for acute toxicity measurements is based on the bioluminescence inhibition of *Vibrio fischeri* NRRL B-111 77 by means of the ToxAlert 100 system from Merck. It is a well-established test, extensively described in literature [21–24] and similar in principle to the better-known Microtox® test.

It is based on the fact that toxic substances will cause changes either in cell structures and/or metabolic pathways of the marine bacteria *V. fischeri*, consequently reflected in a decrease of bioluminescence. This inhibition, caused by the toxic effect of the sample, can be calculated against the response given by a saline control solution, which accounts for the natural decrease in light emission. Data were collected after 15 and 30 min of time exposure of the bacteria to all non-spiked effluent samples, collected over the photocatalytic treatment, after a tonicity adjustment corresponding to 2% NaCl. Phenol (20 mg L^{-1}) and potassium dichromate (40 mg L^{-1}) were used as positive controls.

3. Results and discussion

3.1. Lorazepam and TiO_2 concentrations

Experimental variables were previously optimized in distilled water, including lorazepam/catalyst's concentrations and UV light source [15], in order to achieve a maximal removal efficiency at the least reagents/energy expenses, so that the process becomes economically viable and potentially applicable at a larger scale.

Different criteria were taken into account for decision making regarding which lorazepam's concentration ought to be used. The concentration of ca. $200 \mu\text{g L}^{-1}$ (obtained by dissolving 1 mg Lorenin® pills) was adopted, bearing in mind a compromise between a concentration value simulating real conditions in WWTP effluents and the concentration required for a proper kinetic characteriza-

tion, according to instrumental quantification limits (LC–MS). Under these conditions, and using the Solar Pilot Plant with CPCs described in Section 2.3., the required TiO_2 concentration for optimal performance was ca. 200 mg L^{-1} [15].

3.2. WWTP effluent physicochemical characterization

The performance of the optimized photo-oxidation treatment process was further assessed using a real municipal WWTP effluent. Some physicochemical properties of this grab effluent sample are summarized in Table 1.

Results of the analyzed parameters were compared to the legal emission limit values (ELVs) of wastewaters' discharge into the aquatic environment, as stated in the Law Decree 236/98 [25]. However, these discharge values are only established for monthly averages, considering daily average values of representative 24 h composite samples, collected and analyzed over the respective month.

Nevertheless, all analyzed parameters were within/below the respective legal interval/limit value, with the exception of TSS – 363 mg L^{-1} , far above the legally imposed 60 mg L^{-1} . With regard to the DOC content, it was low (12.1 mg L^{-1}), as expected, since we are dealing with a municipal WWTP effluent. A low DOC indicates that some recalcitrant organic compounds that might still exist, namely pharmaceuticals (see Table 2), shall be in low concentrations.

Regarding metals' composition, apart from those presented in Table 1 (which are below the respective ELV), several others were investigated (including arsenic, lead, mercury, cadmium, iron and aluminum), but all showed to be below the respective method quantification limit (MQL), already in agreement with legal limit values.

Therefore, AOPs' application, and in this particular case TiO_2 -photocatalysis, may be seen as an alternative or complementary method to the already existing/applied technologies, in order to improve the removal rates of some refractory compounds, such as pharmaceuticals. These compounds are usually present at low levels, though which may be high enough to induce several chronic toxicity effects.

3.3. Heterogeneous TiO_2 -photocatalytic treatment

In this work, the photon source used for the phototreatment of the municipal WWTP effluent was the sunlight radiation, a renewable energy source and, therefore, more economically attractive and sustainable.

Samples collected from both experiments, at pre-defined times, were analyzed with several purposes. From DOC analysis (Fig. 2a), it can be deduced that the significant difference between the spiked and non-spiked effluent samples is mainly due to lactose, the only water-soluble excipient present in Lorenin® formulation. The excipients constitute the major portion of each Lorenin® pill (100 mg), in which there is only 1 mg of lorazepam. Fig. 2a also shows that a considerable DOC reduction ($\sim 37\%$ and $\sim 41\%$ for spiked and non-spiked effluents, respectively) took place still in the dark phase of the experiment, due to adsorption on the catalyst surface, as described in literature [26]. Total mineralization percentages achieved at the end of the photocatalytic reaction were approximately 50% for spiked and 59% for non-spiked effluents. These results allowed deriving two main conclusions: on the one hand, and regarding the non-spiked effluent, there was still a significant amount of organic compounds not totally degraded by the phototreatment – though chemical toxicants, including pharmaceuticals, may have ceased to exist, possible oxidation intermediates still remained; on the other hand, regarding the spiked effluent, the major components of the pills (excipients) are also refractory to complete mineralization (i.e. leading to H_2O and CO_2 as sole final products), since the final DOC value was still very distant from the initial DOC of the non-spiked effluent sample.

Table 2

Initial concentration values (C_i) and first order degradation kinetic parameters ($C_t = C_0 \times e^{-kt}$) determined for all emerging contaminants (pharmaceuticals, fragrances and earthy-musty compounds) quantified in the WWTP effluent sample (value \pm standard error).

Emerging Pollutant	MQL ^a (ng L ⁻¹)	C_i^b (ng L ⁻¹)	C_0^c (ng L ⁻¹)	k^d (L kJ ⁻¹)	r_0^e (ng L ⁻¹)	r^2^f	$S_r^2^g$ (ng L ⁻¹)	
Pharmaceutical compounds	Fluoxetine	9	24	29	1.2 \pm 0.3	34 \pm 8	0.8873	14
	Paroxetine	3	29	38	0.8 \pm 0.02	29 \pm 9	0.7331	58
	Diclofenac	79	24,256	10,473	0.48 \pm 0.03	4991 \pm 320	0.9888	217570
	Clotrimazole	7	12	16	0.40 \pm 0.08	6 \pm 1	0.8176	4
	Azithromycin	5	631	680	0.37 \pm 0.03	250 \pm 19	0.9834	1402
	Lorazepam	6	682	640	0.33 \pm 0.05	208 \pm 35	0.9143	6465
	Propranolol	17	52	30	0.26 \pm 0.05	8 \pm 1	0.9371	4
	Furosemide	35	492	448	0.26 \pm 0.03	117 \pm 16	0.9331	1539
	Hydrochlorothiazide	58	3051	2740	0.256 \pm 0.033	701 \pm 92	0.9434	80,508
	Carbamazepine	7	417	399	0.21 \pm 0.02	82 \pm 6	0.9888	642
	Bisoprolol	2	132	119	0.17 \pm 0.01	20 \pm 2	0.9730	59
	Fenofibrate	7	53	39	0.16 \pm 0.02	6.3 \pm 0.9	0.9091	20
	Ofloxacin	33	101	90	0.14 \pm 0.01	13 \pm 1	0.9430	29
	Losartan	6	149	78	0.117 \pm 0.008	9.1 \pm 0.6	0.9598	19
	Ketoprofen	7	410	317	0.10 \pm 0.01	33 \pm 3	0.9569	519
	Norfloxacin	33	138	112	0.10 \pm 0.01	11 \pm 2	0.8364	167
	Lorazepam spk ^h	6	1.21 $\times 10^6$	1.08 $\times 10^6$	0.094 \pm 0.003	1.0 $\times 10^5 \pm 0.4 \times 10^5$	0.9912	1 $\times 10^9$
	Carvedilol	7	95	78	0.06 \pm 0.01	4.5 \pm 0.8	0.8933	75
	Fluconazole	25	110	82	0.049 \pm 0.003	4.0 \pm 0.2	0.9658	3
	Ciprofloxacin	4	254	229	0.040 \pm 0.002	9.2 \pm 0.4	0.9660	33
	Gemfibrozil	36	215	223	0.035 \pm 0.002	7.8 \pm 0.4	0.9675	26
	Alprazolam	4	244	89	0.026 \pm 0.004	2.3 \pm 0.4	0.8917	34
Terbinafine	15	37	34	–	–	–	–	
Fragrances & earthy-musty compounds	2,4,6-Trichloroanisole	7	226	170	0.56 \pm 0.02	95 \pm 4	0.9958	19
	Galaxolide	7	3604	206	0.24 \pm 0.04	49 \pm 9	0.9245	548
	Musk ketone	10	134	33	0.15 \pm 0.01	5.0 \pm 0.4	0.9536	2
	Tonalide	13	703	55	0.12 \pm 0.02	6 \pm 1	0.8285	63

^a MQL – Method quantification limit.

^b C_i – Concentration before the addition of TiO₂.

^c C_0 – Concentration after the addition of TiO₂.

^d k – Reaction rate constant.

^e r_0 – Initial reaction rate.

^f r^2 – Determination coefficient.

^g S_r^2 – Estimation of variance.

^h spk – Spiked.

Concerning total nitrogen content, it did not vary significantly during the photocatalytic process for both experiments.

Temperature and pH variations were also accompanied over the phototreatment experiments (Fig. 2b). Firstly, it should be remarked that the photocatalytic experiments took place mainly during one day – only the last sample (at ca. 32 and 42 kJ L⁻¹ of accumulated UV energy for non-spiked and spiked effluent samples, respectively) was collected at the end of the following day. Bearing in mind that temperature usually rises from morning start-up (15–20 °C) to an almost constant value for several hours until 2 pm, decreasing again over the afternoon, depending on the sunlight intensity, the maximum temperature achieved during the phototreatment was approximately 28 °C, what discards the hypothesis of any thermo-mediated degradation [27]. Since pH is “temperature-dependent”, pH variations accompanied temperature fluctuations over the day. Nevertheless, pH values were in compliance with the Law Decree 236/98 (between 6.0 and 9.0) during the entire experiment.

Finally, the aromatic content followed a similar trend to the DOC, decreasing 94% for the non-spiked effluent, while for the spiked sample it only diminished ca. 64%. Additionally, special attention was given to the degradation kinetics, over the phototreatment, of each emerging contaminant (EC) detected in the initial WWTP effluent sample.

As already pointed out in the Introduction section, conventional WWTPs are not specifically prepared for the complete removal¹ of such micropollutants as pharmaceuticals. These can

still remain in the effluents due to their low tendency to adsorb onto the activated sludge or because their microbial degradation is not fast enough to be completed within the respective WWTP hydraulic retention time [6]. Therefore, this photocatalytic approach was a promising attempt to increase pharmaceuticals' removal rates.

Table 2 presents the initial concentration values (C_i) obtained for the 22 pharmaceuticals, 3 fragrances and 1 earthy-musty compound (2,4,6-Trichloroanisole – 2,4,6-TCA) detected in the effluent sample, prior to phototreatment. As previously mentioned, these C_i values may differ from the concentrations measured at $Q_{UV} = 0$ (C_0) due to adsorption to the catalyst surface (in the dark). Apart from some exceptions (fluoxetine, paroxetine, clotrimazole, norfloxacin, carvedilol, alprazolam and tonalide), the majority of the degradation profiles of the detected ECs follows well a first order kinetic model, with r^2 above 0.90. Table 2 also compiles the kinetic constant (k) and the initial reaction rate (r_0) estimated for each compound, when submitted to the described TiO₂-photocatalytic reaction. In Table 2, all pharmaceutical compounds are ordered from the highest to the lowest rate constant value (k). Interestingly, if we compare the k value of native lorazepam (naturally present in the effluent sample) with that of total lorazepam, achieved after spiking with Lorenin[®] pills, we can observe that the first one is higher (0.33 \pm 0.05 > 0.094 \pm 0.003). Since the only difference relates to the presence of the pills' excipients in the spiked sample, it can be concluded that they might be competing for the hydroxyl radicals, leading to a lower lorazepam degradation rate. Please note that the DOC content in the spiked sample was much higher than the unspiked one.

¹ The term “removal” is always used in this article referring to the conversion of the pollutant to compounds other than the parent compound.

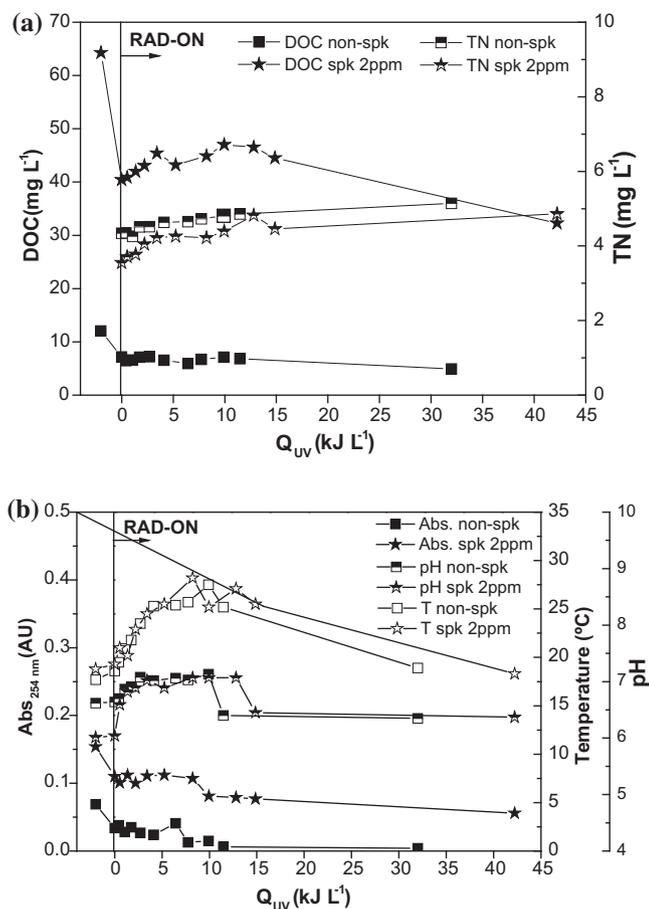


Fig. 2. Physicochemical parameters followed throughout the photocatalytic reaction: (a) DOC (dissolved organic carbon) and TN (total nitrogen) and (b) absorbance at 254 nm, temperature and pH variation represented as a function of the amount of accumulated UV energy per liter of effluent.

Some problems could be anticipated when applying the optimized photocatalytic method to the treatment of a real WWTP effluent, since wastewaters are more complex mixtures. Interference of other oxidizable compounds and scavengers such as carbonates, which typically occur in high concentrations in wastewaters [28,29], could influence the method's efficiency. Nevertheless, except for ciprofloxacin and a small amount of ketoprofen and bisoprolol, all pharmaceuticals initially quantified in the effluent sample decreased to concentration levels below the respective MQLs, attesting therefore the method's practical applicability (Fig. 3). Even lorazepam, a recalcitrant compound whose removal improvement has never been tested by means of accelerated oxidation processes, showed a complete degradation.

Nonetheless, and still regarding the three remaining pharmaceuticals, they presented already much lower concentrations at the end of the phototreatment (corresponding to a total accumulated UV energy of approximately 32 kJ L⁻¹). A small increase in the UV irradiation exposure (time/intensity) could easily allow their complete removal (up to 38 kJ L⁻¹, except for ciprofloxacin – 101 kJ L⁻¹ necessary). Otherwise, and depending on the local weather conditions, a proper management of the combination with a complementary artificial UV [30] source could also be an economically viable solution.

Furthermore, and according to Gros et al. [6], the anti-epileptic carbamazepine usually presents a low removal percentage, or sometimes even a concentration increase, after leaving the WWTP, due to conversion of carbamazepine glucuronides and other

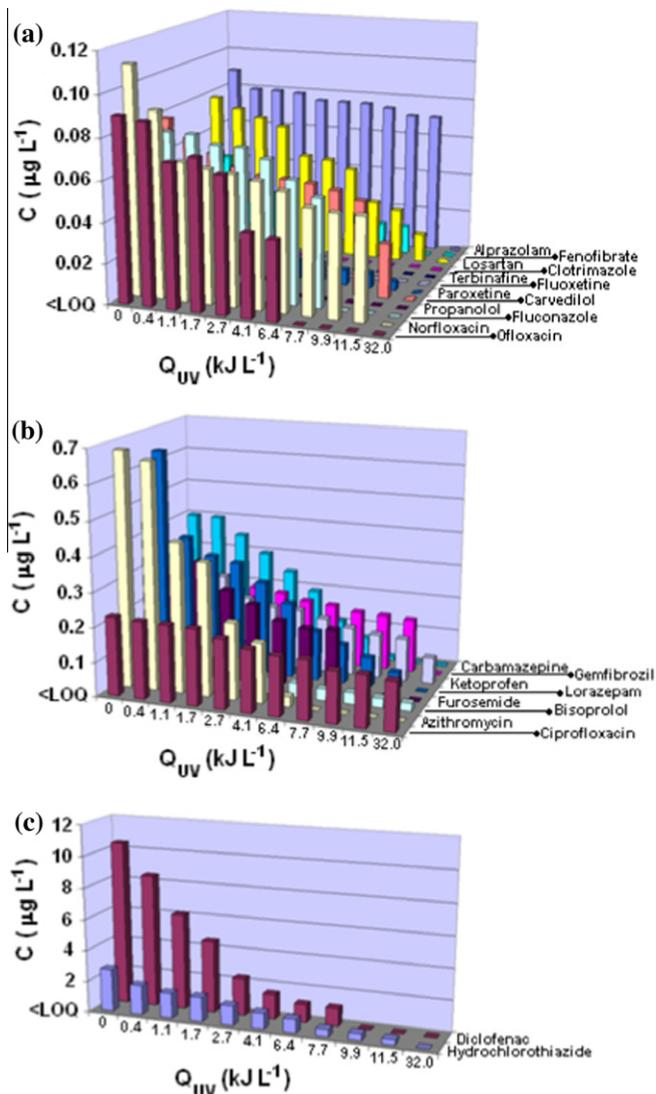


Fig. 3. Graphic representation of the concentration evolution of the 22 pharmaceutical compounds present in the WWTP effluent, over the phototreatment process, as a function of the amount of accumulated UV energy per liter of effluent.

conjugated metabolites to the parent compound by enzymatic reactions occurring over the treatment processes. With the applied photocatalytic treatment, its removal was complete, which is in agreement with previous results obtained by Miranda-García et al. [31] and Rizzo et al. [32]. The same occurred with the moderately removed antibiotics norfloxacin and ofloxacin, the diuretic furosemide and the lipid regulators bezafibrate and gemfibrozil [1,6].

3.4. Biodegradability assay

Zahn–Wellens biodegradability test is considered to be especially accurate, as it involves a long contact period (28 days) between the effluent sample and the sludge, in order to allow some adaptation of the microorganisms.

This assay was performed at different stages of the photocatalytic treatment of the Lorenin[®] spiked effluent, with the purpose of determining the optimal phototreatment time to reach a biodegradable effluent ($D_t > 70\%$). It is however important to highlight that it would not be reasonable to perform such a test over the non-spiked effluent, since it was collected at the exit of the WWTP (itself including the treatment by conventional activated sludge)

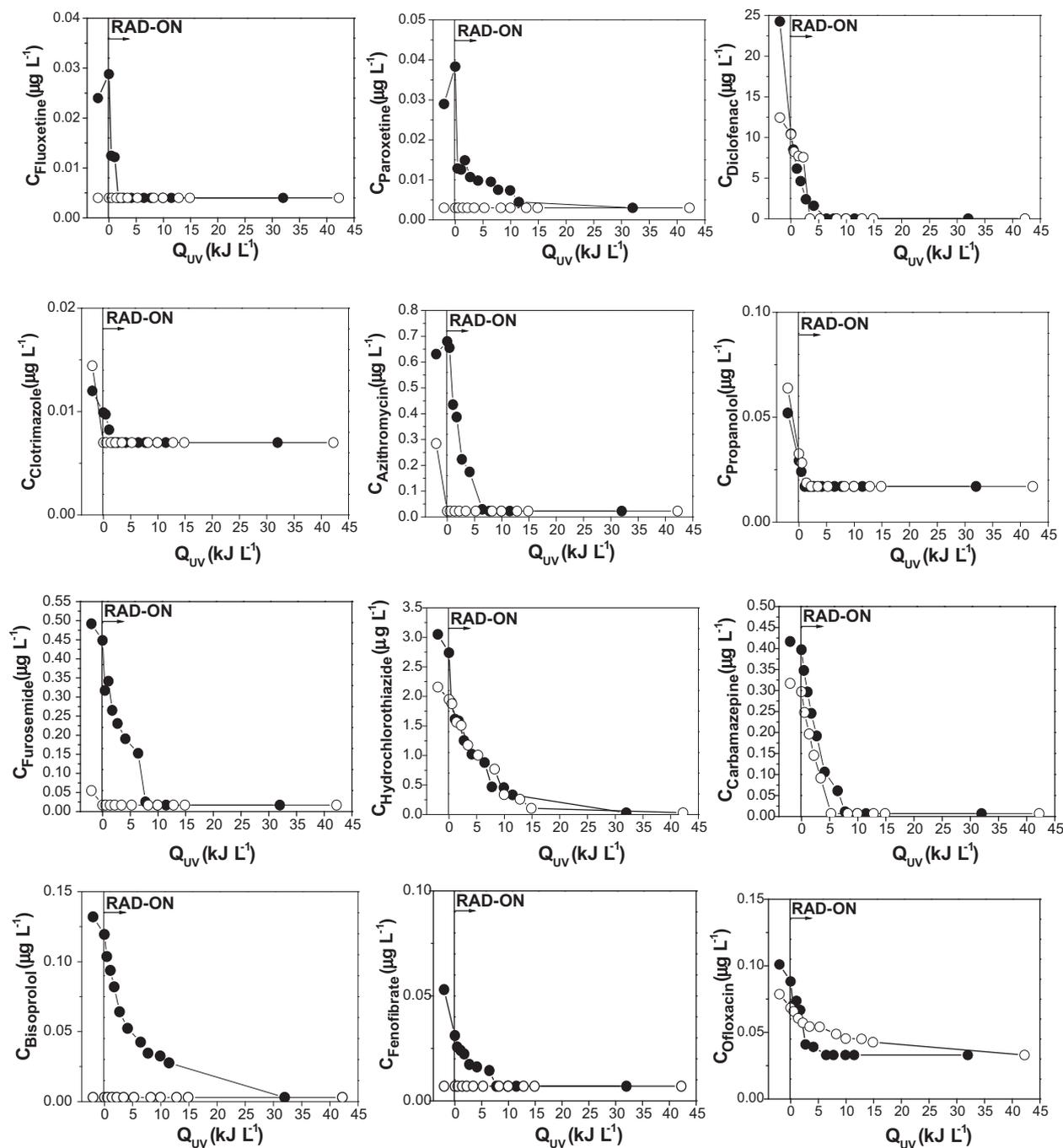


Fig. 4. Representation of the concentration evolution of all pharmaceutical compounds (except for terbinafine) present in the spiked WWTP effluent, over the phototreatment process, as a function of the amount of accumulated UV energy per liter of effluent: ● – before Zahn–Wellens test; ○ – after Zahn–Wellens test; ★ – lorazepam already present in the effluent sample, before spiking with Lorenin® pills.

presenting an already low DOC of ca. 12 mg L^{-1} , which moderately decreased to ca. 5 mg L^{-1} at the end of the phototreatment (refer to Section 3.2.).

Zahn–Wellens' results showed that none of the spiked effluent samples collected over the photocatalytic process presented a D_t value $>70\%$, therefore the phototreatment did not significantly improve the effluent's biodegradability. Nevertheless, all samples were again analyzed in terms of pharmaceutical content, with the aim of understanding which pharmaceutical compounds would already be biodegradable in the first place. This aspect is particularly important considering that biological treatments are usually more interesting than phototreatments (when efficient), due to economic reasons.

Fig. 4 demonstrates that the majority of the analyzed pharmaceuticals seems refractory to biological treatment, since the concentrations before and after Zahn–Wellens test are quite similar. Nonetheless, some pharmaceuticals such as fluoxetine, paroxetine, azithromycin, furosemide, bisoprolol, fenofibrate, losartan, ketoprofen, norfloxacin, carvedilol and gemfibrozil look susceptible to the microorganisms present in the sludge, as their concentrations significantly decreased after the Zahn–Wellens biodegradability test. After thorough analysis of the removal rates of several pharmaceuticals and personal care products by different biological wastewater treatment processes, Sui et al. [33] reported that in the case of gemfibrozil, conventional activated sludge treatment could reach an 80% removal efficiency, which is in agreement with

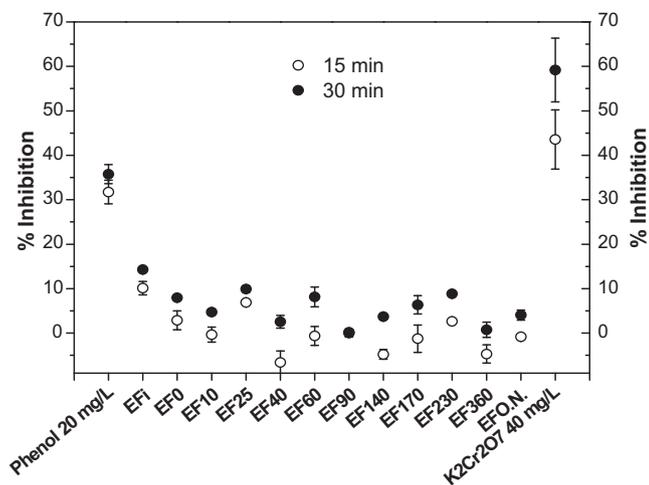


Fig. 5. *Vibrio fischeri* inhibition percentage, after 15 and 30 min, for two positive controls (phenol 20 mg L⁻¹ and potassium dichromate 40 mg L⁻¹) and all non-spiked effluent samples collected over the phototreatment process; EF – effluent; *i* – initial (before TiO₂ addition); 0, 10, 25, 40, 60, 90, 140, 170, 230, 360 and O.N. (overnight) correspond to the sunlight exposure time (following TiO₂ addition) after which each sample was collected, during the photocatalytic reaction.

the results herein obtained. Also Laera et al. [34] observed that the refractory compound carbamazepine, despite not being biodegradable, presented a ca. 95% removal rate after photocatalytic treatment with TiO₂, which again reinforces our results/conclusions.

3.5. Toxicity outcome

According to Rizzo [35], despite the fact that AOPs are being widely used in wastewater treatment for the removal of both organic and inorganic contaminants, as well as to increase effluents' biodegradability, a partial oxidation may result in the formation of intermediates more toxic than parent compounds.

Therefore, and to prevent this potential drawback in cases where complete mineralization is not achieved, AOPs are expected to be carefully monitored and, consequently, toxicity tests must be used to evaluate whether effluents' detoxification is accomplished.

In this work, the acute toxicity of the non-spiked effluent sample was followed over the photocatalytic treatment, by means of % inhibition of *V. fischeri*'s bioluminescence, after 15 and 30 min. Two positive controls (phenol and potassium dichromate) were used to assure bacteria viability.

Results given in Fig. 5 show that inhibition percentages were already low (<15%) from the beginning of the experiment, therefore indicating that the effluent presented no significant toxicity. Nevertheless, these results are still of great importance to attest that no toxicity increase occurred over the phototreatment process, contrarily to the examples reported by Klammer et al. [36].

Although acute toxicity tests give a fast and preliminary information on the hazard of the sample, these tests may not always be the most suitable to evaluate the ecotoxicological hazard of micropollutants such as pharmaceuticals, due to their usually low environmental concentrations [37]. Thus, studies on chronic effects should be used in environmental studies, but due to their duration they are not appropriate to be used as toxicity checks in photocatalytic experiments.

4. Conclusions

The optimized TiO₂-photocatalytic treatment, using a solar pilot plant with CPCs, proved to fit its purpose of removal/degradation of

several ECs, among which are lorazepam and other pharmaceuticals, present in a real WWTP effluent. With a total accumulated UV energy of approximately 32 kJ L⁻¹, 19 out of the 22 pharmaceutical compounds present in the effluent sample were completely removed. Regarding the three remaining pharmaceuticals, ciprofloxacin, ketoprofen and bisoprolol, their removal percentages were 35%, 61% and 77%, respectively.

In addition to the almost complete pharmaceuticals' removal through phototreatment, a Zahn–Wellens biodegradability test allowed to distinguish, among the present pharmaceutical compounds, between the ones that could be removed through biological treatment and those probably refractory to biotreatment, therefore requesting the photocatalytic treatment.

The acute toxicity assay using the marine bacteria *V. fischeri* revealed that, despite the initial effluent itself presented no significant toxicity, it did not increase over the photocatalytic process, thus discarding the possibility of formation of more toxic intermediate oxidized compounds.

In conclusion, this photocatalytic treatment proved to be a promising tool with practical future applicability, since it requires the use of solar renewable energy.

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