Desalination and Water Treatment
www.deswater.com
doi: 10.5004/dwt.2020.26008

198 (2020) 272–283
September

Evaluation of the acute toxicity on *Daphnia magna* throughout the catalytic wet peroxide oxidation of dissolved natural organic matter

Camilo L. Guerrero-Romero[a], Dolly Revelo[b], Alejandra Caicedo[a], Marcela Botina[a], Ana M. García-Mora[c,d], Ricardo A. Torres-Palma[d], Luis-Alejandro Galeano[c], Iván A. Sánchez-Ortiz[a,c,*]

[a]*Departamento de Recursos Hidrobiológicos, Universidad de Nariño, 520002 Pasto, Colombia, Tel. (+57) 3016436357; emails: ivansaor@hotmail.com/iaso@udenar.edu.co (I.A. Sánchez-Ortiz), camiloguerrero.romero@gmail.com (C.L. Guerrero-Romero), alejandra61033@hotmail.com (A. Caicedo), marcela_9410_@hotmail.com (M. Botina)*
[b]*Grupo de Investigación en Bioelectroquímica (BEQ), Departamento de Biología, Universidad de Nariño, 520002 Pasto, Colombia, email: margo.revelo@gmail.com (D. Revelo)*
[c]*Grupo de Investigación en Materiales Funcionales y Catálisis (GIMFC), Departamento de Química, Universidad de Nariño, 520002 Pasto, Colombia, emails: anamariagarcia@udenar.edu.co (A.M. García-Mora), alejandrogaleano@udenar.edu.co (L.-A. Galeano)*
[d]*Grupo de Investigación en Remediación Ambiental y Biocatálisis (GIRAB), Universidad de Antioquia UdeA, 050010 Medellín, Colombia, email: ricardo.torres@udea.edu.co (R.A. Torres-Palma)*

Received 26 December 2019; Accepted 18 April 2020

ABSTRACT

This research was aimed at evaluating the evolution of the acute toxicity (effective concentration measured in terms of immobilization, EC_{50}) on *Daphnia magna* neonates at 48 h of exposure during the catalytic wet peroxide oxidation (CWPO) of dissolved natural organic matter (NOM), activated by an Al/Fe-pillared clay catalyst. The CWPO-degradation of NOM was assessed throughout 240 min (180 min of peroxide addition) as a function of time on samples of either raw surface water (supplies of rural aqueducts) or effluents of conventional physicochemical treatment units at drinking water treatment plants (DWTPs). Total organic carbon (TOC), non-reacted H_2O_2 concentration, and decolorization were the measured response variables as a function of the reaction time. Starting TOC concentrations in the targeted water supplies varied between 1.54 and 4.65 mg/L. The CWPO treatment achieved depletions of the initial immobility ranging between 97.5% and 100% showing this technology as a very promising alternative assisting DWTPs to improve the overall safety of drinking water. The immobilization of the *D. magna* neonates showed to be also affected by the conductivity, whose Spearman correlations varied between 0.69 and 0.96, and then this parameter must be carefully taken into account in measurements of toxicity based on this bio-indicator.

Keywords: Advanced oxidation processes; Drinking water treatment; Catalytic wet peroxide oxidation; Mineralization; Pollution removal; Toxicity test

1. Introduction

For urban and rural communities, contaminated water resources, as a result of inadequate wastewater management, represent one of the largest health challenges, restricting development and increasing poverty through the costs of medical care and lost productivity [1]. Despite the stringent environmental laws recently been implemented all around the world, which have caused a significant drop in the pollution discharged into natural bodies of water, there are a growing fraction of toxic and refractory compounds

---

* Corresponding author.

whose biological or physicochemical treatment is quite difficult to be effectively carried out [2]. This, alongside a lack of sanitation, makes the presence of such compounds, various forms of solids, organic matter and nutrients, as well as a variety of emerging pollutants, prevalent in natural bodies of water intended for human consumption. Natural organic matter (NOM) is a complex matrix of organic substances commonly found in surface and groundwater, as a result of different hydrological, geological, and biological interaction schemes. In general, NOM can be generated in water through biological activity (mainly algal and microbial/native NOM), or introduced to the body of water through drainage of river basins including substances generated during the decomposition of terrestrial organisms (allochthonous NOM) [3].

On the other hand, disinfection is widely recognized as necessary to guarantee the supply of safe drinking water [4]. Advanced oxidation processes (AOPs) could be included among the alternatives to conventional disinfection processes in the short-term. These processes destroy organic molecules, including the most stable and difficult to degrade compounds such as carcinogens and mutagens, by generating highly reactive species. Therefore, AOPs should become of great interest for public health, as well as a promising field of study due to their potential for almost full degradation of dissolved organic pollutants in water, in some cases, under ambient conditions of temperature and pressure [5]. Catalytic wet peroxide oxidation (CWPO) is one of the AOPs that may lead to high mineralization levels of the contaminants towards CO_2 and H_2O; in the case of the partial degradation of the non-biodegradable compounds, fairly less refractory intermediates and by-products are formed. In the CWPO process, H_2O_2 is activated by transition metals (mainly Fe or Cu) retained in the structure or over the surface of different functional materials [Eqs. (1) and (2)] [6]. To be used successfully over several catalytic cycles, these solids must simultaneously guarantee high catalytic activity and chemical stability against leaching in the aggressive environment of the oxidizing reaction [7]. It is important to highlight some great advantages of using H_2O_2 as the oxidizing agent in the CWPO: its decomposition generates harmless species (H_2O and O_2) as major by-products, whereas it is not very expensive [8].

$$\left[\mathrm{Fe}^{3+}-\mathrm{catalyst}_{(s)}\right]+\mathrm{H_2O_2}\rightarrow\mathrm{HO_2^{\bullet}}+\left[\mathrm{Fe}^{2+}-\mathrm{catalyst}_{(s)}\right]+\mathrm{H}^{+} \quad (1)$$

$$\left[\mathrm{Fe}^{2+}-\mathrm{catalyst}_{(s)}\right]+\mathrm{H_2O_2}\rightarrow\mathrm{HO}^{\bullet}+\left[\mathrm{Fe}^{3+}-\mathrm{catalyst}_{(s)}\right]+\mathrm{HO}^{-} \quad (2)$$

Our previous research has depicted the efficiency of CWPO in the degradation of NOM dissolved in surface waters, as well as in the inactivation of model microorganisms, for instance, enteric viruses. In a first study [8], 96% removal of chemical oxygen demand (COD) was achieved in just 4 h and total color removal in less than 45 min of reaction employing an aluminum/iron pillared clay (Al/Fe-PILC) catalyst. More recently, Ordoñez et al. [9] reached 92.9% of MS2 inactivation and 100% of reacted H_2O_2 at close to neutral pH (7.07) in the presence of the same kind of clay catalyst, by using pretty low stoichiometric concentrations of the oxidizing agent.

In this sense, regarding its potential application in drinking water treatment plants (DWTPs), it is necessary to demonstrate that the overall performance of the CWPO process in the treatment of surface water promotes not only the degradation of the starting pollutants but also decreasing their toxicity in a great extent. This is particularly important assessing the application of any technology in the production of water for human consumption, but also in the degradation of highly refractory pollutants whose oxidation by-products (OBPs) might be still hazardous even after full conversion of the starting contaminants, just the case in a lot of wastewaters.

In fact, some works have focused on the possible formation of hazardous by-products through oxidations [10,11]. It has been reported that removal of parent compounds from drinking water by conventional ozonation and AOPs such as UV, photocatalysis (TiO_2/UV, ZnO/UV), UV/chlorine, UV/H_2O_2, O_3/OH^-, O_3/UV, Fenton reaction (Fe(II)/H_2O_2), occurs via deep transformation of their chemical structure leading to new sources of toxicity; they reported different types of toxics usually present in drinking water such as halogenated compounds, olefins, nitro compounds, ethers, alcohols and phenols, nitrogen-containing compounds, organophosphorus compounds, amides, and N-nitroso compounds [12].

A recent study [13] explored the formation of toxic OBPs from the degradation of dissolved organic matter by O_3^-, $H_2O_2^-$ and sulfate radical – based AOPs. The study reported that in the presence of halide (X^-) at a high concentration relative to the dissolved organic matter (DOM), organic halogenated-OBPs can be formed. The principal halogenating agent is HOX/XO^- generated from the reaction of $X^{\bullet}$ with X^-. The accumulation of HOX/XO^- can be avoided by ample concentrations of H_2O_2 in H_2O_2-based AOPs. Although Fenton like or CWPO has been typically associated with the insignificant formation of halogenated-OBPs, the formation of these species can be favored for waters with high Cl^- or Br^- concentrations.

On the other hand, the formation of aldehydes and carboxylic acids as by-products has been reported in the application of ozonation (O_3/UV and/or iron ions) to remove NOM; in this case, total mineralization of NOM cannot be reached because the by-products do not properly react with ozone [14]. In the case of CWPO, it is known that the main by-products that avoid complete mineralization are light carboxylic acids.

In this context, toxicity tests could be appropriate diagnostic tools to determine the effect of physical and chemical agents on varied test organisms under specific and controlled experimental conditions. These effects could be either inhibition or magnification, evaluated by the reaction over the organisms, such as death, growth, proliferation, multiplication, and morphological, physiological or histological changes [15]. Acute toxicity tests are used to estimate the dose or concentration of a toxic agent capable of producing a specific measurable response in a test organism or population, in a relatively short time (usually 24–96 h). In acute toxicity tests are usually recorded mortalities and abnormalities related to appearance and fish behavior, such as swimming capacity [16]. The 50% effective concentration (EC_{50}) is the concentration of the toxic that produces appreciable negative effects in 50% of the population, such as loss of balance or motor function, swimming or respiratory rate in a given time. Meanwhile, the 50% lethal concentration (LC_{50}) is the concentration of a substance that can be expected to cause the death of 50% of the organisms exposed to the toxic substance during a certain period, during the exposure or within a defined period after it [17]. In toxicity tests for evaluation of effluents, the freshwater microcrustacean *Daphnia magna* has been widely used as a biological indicator in studies about water quality control. Toxicity tests with *D. magna* determine the potential lethality of pure chemical substances, domestic and industrial effluents, leachates, surface or underground water, drinking water, among others. Likewise, authors such as Chiva et al. [18], highlighted the use of this bio-indicator in AOP studies.

Based on the above, the objective of the present investigation was to evaluate the evolution of the acute toxicity on *D. magna*, expressed in terms of EC_{50} (loss of mobility), during the CWPO degradation of NOM present in surface waters to improve water quality for human consumption. Besides, critical aspects that should be taken into account in the application of this technique to follow the evolution of toxicity in the frame of the AOPs, in general, are highlighted. For this purpose, the International Organization for Standardization (ISO) standard of the Spanish Association for Standardization and Certification [17] ISO 6341 was used, which established the methodology for the determination of EC_{50} characteristics, as well as conditions of crop management of this bio-indicator.

2. Materials and methods

2.1. *D. magna* maintenance, cultivation, feeding and sensitivity determination

Toxicity tests were conducted using 6360 neonates of the *D. magna* species, each with less than 24 h. The cultivation and maintenance of the *D. magna* were carried out in an acrylic vessel measuring 60 cm in length, 66 cm in height, and 77 cm in depth, following the ISO 6341 recommendations with the modifications described below. The cladoceran was fed by adding *Chlorella* sp. microalgae into reconstituted water at a rate of 4.5×10^6 cells per *D. magna* every day. The algae were grown in 0.1–3.0 L glass and plastic reactors to scale the crop. To obtain better algal growth, KENT A and B Marine Pro-culture® (Milwaukee, Wisconsin, USA) commercial culture mediums were used. All cultures were maintained by a semi-continuous system under the following conditions: 20°C to 25°C, a light intensity of 1,000 to 3,000 lx, constant aeration and 3 days of incubation [19]. The purity of the cultures was verified by direct observation of samples under a microscope (Olympus CX 21 RFS1). Maintenance of the *D. magna* culture was carried out under the following conditions 20°C–25°C, pH 7.0–9.0, photoperiod of 16 light h and 8 h of darkness (timer photoperiod regulator, 220 V 24 h EXCELITE® clock, Washington, D.C., USA), the light intensity of 1,000 lx (Digital Lux Meter LX1330B 0.1–20,000 lx, California, USA), separating breeders from neonatal individuals in the middle of the fresh crop. To avoid loss in the population, daily monitoring of the crops was carried out, taking into account the daily cleaning to eliminate traces of seaweed lodged in the bottom, molts and dead individuals, population density (≤1 individual per 10 mL), total hardness (160–210 mg/L), and dissolved oxygen (≥2.0 mg/L) (YSI 550A oximeter, accuracy 0–20 ± 0.3 mg/L, Yellow Springs, Ohio, USA).

As recommended by the standard ISO 6341 [17], before starting toxicity tests, and periodically during the study, the sensitivity of the individuals of *D. magna* was verified and guaranteed by applying the viability test in the presence of $K_2Cr_2O_7$ (99%, Panreac, Barcelona, Spain) to determine EC_{50} – 24 h. The EC_{50} values of $K_2Cr_2O_7$ after 24 h of treatment obtained in the 10 viability tests, for which 50% of the individuals of *D. magna* were immobilized, varied from 0.67 to 1.51 mg/L. The results, therefore, indicated that the *D. magna* individuals employed in the investigation were viable for the development of the different toxicity tests since the confidence limits of each test were found within the concentration ranges established in the ISO 6341 standard for $K_2Cr_2O_7$, whose values ranging between 0.6 and 2.1 mg/L (Table S1), indicating a high level of confidence for the results obtained.

2.2. Toxicity test of NOM, intermediates, and by-products of the CWPO degradation

The intermediaries' compounds are those formed along the course of the oxidizing reaction by changes of the parent compounds, in this case, NOM, that can either (i) change to form other species or (ii) be mineralized toward H_2O and CO_2. The intermediaries that do not get mineralized and are transformed in any other compounds, which remain until the end of the reaction, are called by-products.

The toxicity tests were carried out using 4 replicates with 10, 24 h-old individuals, deposited in 10 mL of the solution of each compound to be tested [17]. The LC_{50} was determined for a model of dissolved NOM, using a synthetic standard prepared from polyacrylic acid (Analytical, Fluka®, St. Louis, MO, USA), polygalacturonic acid (90%, SIGMA® life science, St. Louis, MO, USA), polystyrene sulfonate (PSS1: 1.0 MDa average molecular weight distribution (MWD), 25%, and PSS2: 200 kDa average MWD, 30%, Aldrich® solution, St. Louis, MO, USA), and humic acids (Aldrich® chemistry, St. Louis, MO, USA), H_2O_2 (50%, Bio Pharchem®, Hatfield, Arkansas, USA), and Na_2SO_3 (98%, Panreac, Barcelona, Spain). To determine the LC_{50} – 48 h for NOM, expressed in mg/L of total organic carbon (TOC), 2 preliminary tests (53–500 mg/L and 10–50 mg/L of TOC) and 1 conclusive test (3–12 mg/L of TOC) were performed.

The NOM surrogate was needed to modulate the TOC concentrations to enable establishing a range of NOM concentrations under which there was toxicity on *D. magna*. To determine the LC_{50} – 48 h of H_2O_2 (1.6–8.4) and Na_2SO_3 (42–126 mg/L), used as an oxidizing agent in the CWPO process and a reducing agent to remove any excess remaining of free peroxide, respectively, toxicological tests were carried out simultaneously on each substance (42, 63, 84, and 126 mg/L for Na_2SO_3; and 1.6, 5.4, and 8.4 mg/L for H_2O_2) and the combined substances (3.5, 6.6, and 10.5 mg/L of H_2O_2 with 63 mg/L of Na_2SO_3 and 5.6, 9.7, and 10.3 mg/L of H_2O_2 with 42 mg/L of Na_2SO_3) in order to avoid interferences of those substances with the toxicity test results.

2.3. Collection and characterization of water samples for CWPO treatment

Water samples were collected according to the guidelines for taking and conserving samples reported in current regulations [20] from 6 different supplies of rural or DWTPs in Nariño, Colombia. These sources corresponded to Chaguaipe rural aqueduct and DWTP, both at the municipality of Ipiales, the DWTP of Túquerres municipality, Jagua and Pueblo Nuevo rural aqueducts of the municipality of Tumaco (on the pacific coast), and the DWTP of the municipality of Pasto. In the case of the samples taken in DWTPs, the sampling point was always the effluent of the conventional physicochemical treatment unit (flocculation–sedimentation), whereas, in the case of the rural aqueducts, fully raw water was collected.

The pH levels, electrical conductivity, and temperature of the collected waters were determined on-site using a portable multiparameter (SensoDirect 150, Dortmund, Germany). Also, the following analyses were performed with standard methodologies [21]: chlorides, phosphates, nitrates, nitrites, sulfates, apparent color, turbidity, fluorides and total hardness (supplementary material contains detailed information).

2.4. CWPO experiments on surface waters

The CWPO treatment on the six surface water samples and an additional artificial water sample spiked with a synthetic NOM surrogate above described were conducted in a 1.5 L stirred tank reactor with mechanical agitation at 600 rpm by an electronic head agitator and aeration at ~2 L/h (Fig. 1), containing a working volume of 1.0 L of the sample of water to be treated, previously filtered (qualitative filter 15–20 µm, BOECO brand, Hamburg, Germany). The semi-batch reaction (stepwise, constant addition of hydrogen peroxide) was carried out under conditions previously optimized by García et al. [22] on the synthetic surrogate of dissolved NOM, as briefly described here: 5.97 g of catalyst (Al/Fe-PILC) containing 0.62% w/w of active Fe (incorporated via Al/Fe intercalation-pillaring of a natural bentonite) and 100 mL of H_2O_2 prepared at the necessary concentration for providing (0.45 mg H_2O_2/mg TOC) according to the TOC concentration in every starting water sample, fed by a peristaltic pump at 0.56 mL/min through 180 min; a 30 min of equilibrium period between water and the solid catalyst was kept before starting the addition of peroxide, whereas further 30 min were still measured once finished the peroxide addition allowing reaction of the oxidizing reagent in the higher possible extent. Experiments were performed under the atmospheric pressure of Pasto city (564 mm Hg), a controlled temperature of 25.0°C+/–0.1°C, initial pH of 7.0 (pH was monitored throughout the entire CWPO treatments, but not modified).

2.5. Analytical methods

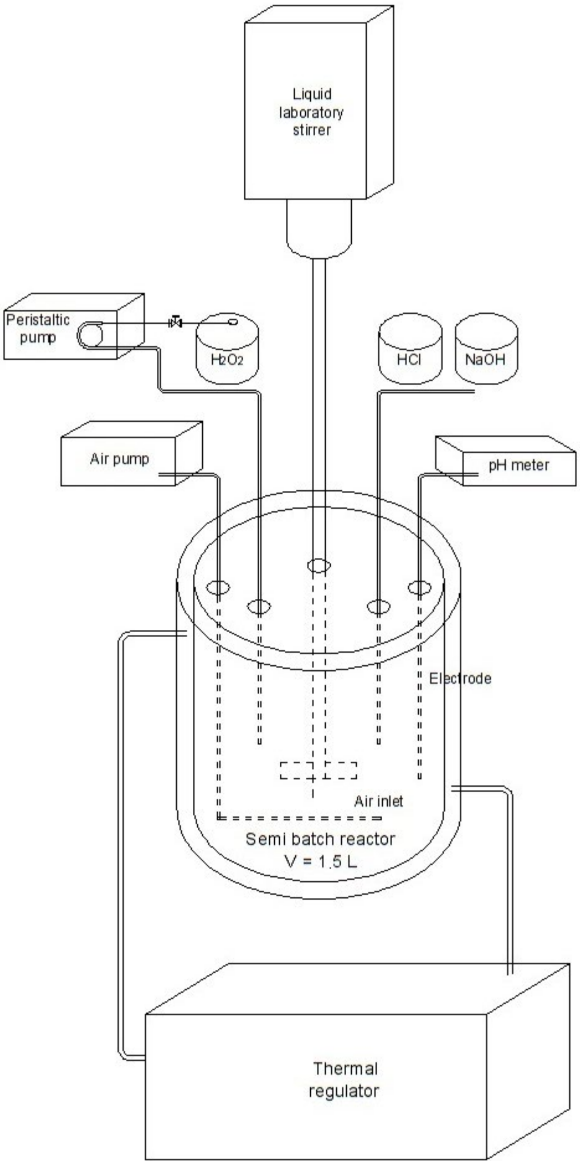

Fig. 1. The experimental setup used in CWPO tests for NOM degradation.

During the whole CWPO reaction time (240 min), 60 mL samples were taken to carry out the toxicity tests at the following times: t_1 (0 min), t_2 (30 min), t_3 (75 min), t_4 (120 min), t_5 (165 min), t_6 (210 min), and t_7 (240 min). One catalytic experiment was performed for every time of reaction, preventing significant effect of dilution. All samples were filtered using a 0.45 µm filter (polyvinylidene fluoride, hydrophilic, QLS®, Miami, USA) to remove any residue of suspended catalyst. Besides, the following analyses were performed on each filtered sample: (i) dissolved organic carbon, measured from 15 mL of a sample using a TOC-L CPH Shimadzu analyzer (Columbia, Maryland, USA), to establish the degree of mineralization of the NOM during the treatment (TOC); (ii) free H_2O_2 (molecular), determined from 0.5 mL samples by the household method, previously described by Pupo et al. [23]. For this analysis, a calibration curve was performed using a standard solution prepared with H_2O_2 ($r^2 = 0.998$, LOD = 0.014 mg/L, LOQ = 0.040 mg/L); (iii) finally, the decolorization of the solution was determined by absorbance at 456 nm from 2.0 mL of sample in a Shimadzu 1280 UV spectrophotometer (Columbia, Maryland, USA) using the Standard Single Wavelength 2120C Method [21]. The concentration is expressed in platinum-cobalt units (PCU) according to the calibration curve prepared from a platinum-cobalt standard solution ($r^2 = 0.994$, LOD = 12 PCU, LOQ = 48 PCU).

2.6. Toxicity tests on D. magna during CWPO treatment

The toxicity EC_{50} – 48 h tests were applied to the effluent samples of the CWPO treatment at previously mentioned times of reaction. The toxicity tests were carried out according to ISO 6341 from 4 replicates, each one including ten *D. magna* neonates, less than 24 h old. At the beginning and the end of each toxicity test, pH and electrical conductivity were also measured. Such water quality parameters were treated as covariates during the statistical analysis, due to their condition of not being easily controllable under practical conditions of treatment at a real scale.

2.7. Statistical analysis

The targeted response variable evaluated was EC_{50}, for which the number of immobilized individuals in each replica was counted. The percentage of immobility was expressed as the ratio between the number of immobilized animals and the total population, multiplied by 100. All the data obtained from the toxicity tests were evaluated with the Statgraphics Centurion Probit statistical program (Centurion XV 16.1.15.), by which the EC_{50} or LC_{50} values were calculated for 24 or 48 h of exposure, according to the case. A Spearman correlation of both the pH and the electrical conductivity against the immobility of *D. magna* was made using the average value of every parameter from the measured at the beginning and at the end of the toxicity tests applied at every time of reaction. As they did not fit a normal distribution pattern, this correlation coefficient was calculated using the same statistical package.

3. Results and discussion

3.1. Toxicity tests of the synthetic NOM surrogate on D. magna

The first trial yielded a LC_{50} of 15.6 mg/L of TOC (Table 1), with lower and upper confidence limits clearly below the evaluated concentrations. As a result, the second test was carried out with narrower ranges of concentration, using TOC values closer to those present in real surface waters at the tropic. In this case, the LC_{50} was 7.94 mg/L of TOC, with lower and upper confidence limits still below or sharply close to the assessed concentrations. Finally, to further refine the value of LC_{50}, a third trial was carried out that yielded a value of 2.18 mg/L of TOC, with the upper confidence limit within the range of NOM concentrations tested.

The TOC content is broadly used in this type of research to represent the NOM concentration in water. However, it does not determine which specific compounds are present in the sample [2], or which could affect human health and aquatic organisms. Non-biodegradable organic matter may have different effects on animals, such as accumulation in tissues, toxic effects for different organisms manifesting their toxicity acutely and chronically, behavior disorders, embryonic development and the viability of individuals. According to the conclusive trial, it was determined that a dose of 2.18 mg/L of TOC has a toxic effect on *D. magna* individuals, which is probably a consequence of the effect exerted by the different components of the synthetic NOM surrogate on *D. magna*.

3.2. Toxicological tests of Na_2SO_3 and H_2O_2 on D. magna

H_2O_2 is mainly used for disinfection in the production of drinking water and aquaculture. Its use for wastewater treatment has increased, but a uncontrolled application may induce harmful effects in sensitive aquatic animals, such as microcrustaceans [24]. H_2O_2 is used in all Fenton processes for the production of $HO^{\bullet}$; even used in very low doses, there would be a remnant of the unreacted oxidizing agent at the end of the process that might affect the measurements of toxicity using *D. magna*, thus it must be efficiently inactivated in advance.

In tests conducted to determine the toxicity of H_2O_2, the following concentrations were evaluated: 1.6, 5.4, and 8.4 mg/L resulting in a LC_{50}–H_2O_2 at 48 h of 7.3 mg/L. According to Bownik and Stepniewska [25], the toxic action of H_2O_2 is associated with the induction of oxidative stress since it is more active than molecular oxygen. When evaluating the protective effects of Ectoin on the behavioral, physiological and biochemical parameters of *D. magna* subjected to H_2O_2, these authors found an LC_{50}–48 h of 5.0 mg/L of H_2O_2, a value close to that obtained in our current study.

To establish the toxicity ranges of the Na_2SO_3 and H_2O_2 on *D. magna*, independent toxicity tests were performed in the presence of each compound, as well as a simultaneous test. The experiments were conducted with H_2O_2 doses used for mineralization of the typical concentrations of NOM during the CWPO polishing of surface waters. Na_2SO_3 was

Table 1
Concentrations and LC_{50} – 48 h exposed to synthetic NOM surrogate (in mg/L of TOC)

Preliminary test 1			Preliminary test 2			Final test		
NOM	LC_{50} – 48 h	Confidence limits lower – upper	NOM	LC_{50} – 48 h	Confidence limits lower – upper	NOM	LC_{50} – 48 h	Confidence limits lower – upper
53	15.6	9.10–24.4	10	7.94	5.59–10.1	3	2.18	1.01–3.10
107			20			6		
214			30			9		
322			40			12		
429			50			–		
500			–			–		

also important because it was used for deactivation of free H_2O_2 in every sample just after being taken. In determining the toxicity of Na_2SO_3 on *D. magna* neonates, the following doses were used: 42, 63, 84, and 126 mg/L. As a result, an LC_{50}–Na_2SO_3 of 35 mg/L was obtained at 48 h. This value demonstrates the need to use the correct dosage of sulfite, taking into account that 1.0 mg of H_2O_2 just requires 3.7 mg of Na_2SO_3 for its full deactivation according to the stoichiometric ratio [26]. Then, at the end of each catalytic test, the amount of H_2O_2 remaining in each sample was quickly measured and just the stoichiometric amount of Na_2SO_3 needed to deactivate it was added. It was made to ensure the measured toxicity to be the result of the NOM intermediates and by-products generated during the oxidation process or even unreacted NOM itself and not by the indirect effect of the Na_2SO_3 in excess.

3.3. Physicochemical parameters of water treated with the CWPO technology

The physicochemical parameters of the water sources for human consumption analyzed in this investigation are presented in Table 2. The pH, chloride, phosphate, nitrate, nitrite, sulfate, fluoride and total hardness values measured were found within the limits allowed by the Colombian regulation on drinking water [27]. In the case of the apparent color, only the sources with previous treatment (Pasto, Ipiales, and Túquerres) registered values within the permissible limits. For turbidity, the samples of the Ipiales and Túquerres DWTPs fitted within the recommended ranges, while others exceeded the permissible limit.

3.4. TOC degradation and discoloration during the CWPO treatment of real surface waters

Table 3 shows initial concentrations, the reaction time of 0 min (t_1), and the final reaction time of 240 min (t_7) of TOC, as well as the apparent color values recorded during CWPO treatment of the different water samples. According to Tapia et al. [28], watercolor and the presence of humic substances from humic and fulvic acids depends on NOM presence, which represents between 50% and 75% of the TOC contained in surface waters. Based on the results obtained, it can be seen that the TOC concentration in the treated samples would not significantly affect mobility in *D. magna*, except for the water from the Chaguaipe rural aqueduct and the synthetic NOM standard that exceeded the LC_{50} – 48 h of 2.18 mg/L of TOC determined in this investigation.

Table 2
Physicochemical parameters of the water sources analyzed

Sampling points	Chaguaipe	Pueblo Nuevo	Bajo Jagua	Pasto DWTP	Túquerres DWTP	Ipiales DWTP	Synthetic NOM pattern	Limit value[c]
pH	6.16	6.76	7.62	6.40	6.44	6.87	–	6.5–9.0
Temperature	15.4	26.5	23.5	17.7	12.8	12.9	18.9	–
Chloride (mg/L)	4.25	14.25	3.25	10.75	3.00	20.24	<0.70	250
Phosphate (mg/L)	0.17	0.18	<0.10	<0.10	<0.10	<0.10	<0.10	0.5
Nitrate (mg/L)	<0.20	<0.20	<0.20	1.04	<0.20	0.39	0.40	10
Nitrite (mg/L)	0.020	0.037	0.032	<0.005	<0.005	<0.005	0.006	0.1
Sulfate (mg/L)	12	8.0	<5	<5	81	82	6	250
Apparent color (PCU)[a]	108	70	31	<8	<8	<8	42	15
Turbidity (NTU)[b]	43.3	28.8	17.5	6.90	1.30	1.30	2.90	2.00
Fluoride (mg/L)	0.24	0.13	0.03	0.05	0.09	0.20	0.01	1.00
Total hardness (mg/L)	30	84	24	23	55	89	–	300

[a]PCU: platinum-cobalt units; [b]NTU: nephelometric turbidity units; [c]Reference limit values according to Colombian drinking water regulation [27].

Table 3
TOC and color removal efficiencies through the CWPO treatment of real water supplies

Evaluated surface water	TOC initial (mg/L)	TOC final (mg/L)	TOC removal efficiency (%)	Initial apparent color (PCU)	Final apparent color (PCU)	Color removal efficiency (%)
Chaguaipe	4.65	3.60	22.6	9.4	9.2	1.8
Pueblo Nuevo	2.40	2.24	6.70	2.5	1.7	31
Bajo Jagua	2.57	2.06	19.8	3.2	1.9	41
DWTP Pasto	2.56	2.09	18.4	1.3	0.9	30
DWTP Túquerres	1.54	1.26	18.2	1.6	1.0	39
DWTP Ipiales	2.12	1.92	9.40	1.3	1.0	17
Synthetic NOM pattern	10.2	7.68	24.7	7.0	5.7	18

Also, considering that in all cases the initial NOM concentration was low, except that registered for the synthetic NOM pattern, only based in the TOC contents, all water samples were suitable for human consumption in Colombia, as they did not exceed the maximum acceptable value of 5.0 mg/L [27]. However, the formation of disinfection by-products upon chlorination of such organic loads could still be problematic in terms of safety. Such side products are not properly considered by many of the regulations about the quality of drinking water all around the world, which is also the case in Colombia.

Munoz et al. [29] evaluated the combination between catalytic hydrodechlorination (HDC) and CWPO under close to stoichiometric H_2O_2 doses to avoid the formation of toxic and strongly inhibitory intermediates in the treatment of chlorophenols; they proposed to use HDC (~30 min) as a pretreatment to remove chlorine followed by CWPO at pH = 3.0. This combined process improved carbon mineralization from 30% to 70% after 120 min of reaction at 30°C (HDC stage) and 50°C (CWPO stage). Meanwhile, in the current work, the pH of the reaction was close to neutral (natural of the water supplies), the ambient temperatures of the water supplies (Table 2) below 20°C in most of the cases, and the H_2O_2 dosing was very low (0.45 mg H_2O_2/mg TOC), corresponding to around 13.0% of the theoretical amount of H_2O_2 assumed to be required for full oxidation of the initial TOC loading, calculated as follows: 1,000 mg H_2O_2/L achieves to remove up to 470.6 mg/L of COD in iron-activated Fenton reactions [9]. Altogether explain the relatively low TOC and color removals obtained (Table 3), but it must be emphasized our central interest in studying the evolution of toxicity instead of improving other physicochemical parameters of the output streams.

Pizarro et al. [30] also studied HDC and CWPO combined for the removal of p-chloro-m-cresol from water under ambient like-conditions. Cresols have been classified by the United States Environmental Protection Agency as persistent and toxic and can be used as model substrates of NOM [31]; in this case, it has attracted much attention because the oxidizing agent may react with the methyl or hydroxyl groups theoretically improving the reactivity towards the aromatic ring. Pizarro et al. [30] achieved full dechlorination and a relatively high mineralization level (55%) in 4 h for the HDC (0.5 h, 25°C, 1.0 atm, pH 10.0) – CWPO (3.5 h, 25°C, 1.0 atm, pH 3.5) combined processes and stoichiometric dosing of H_2O_2; short-chain organic acids were obtained as by-products, meaningless in terms of ecotoxicity, in close correlation with the results here displayed.

3.5. Correlation between the EC_{50} (D. magna) and the TOC depletion through the CWPO treatment

Table 4 shows the CWPO reaction times for which the EC_{50} – 48 h values were reached and the corresponding starting and depleted TOC concentrations. Additionally, the initial TOC of each sample is presented as a reference value. The shortest times required to reach EC_{50} during the CWPO reaction were observed for samples with the partial treatment of potabilization (post-physicochemical, before chlorination) from Pasto and Túquerres DWTPs, as well as the raw water of the Chaguaipe rural aqueduct. This is owed to the complex interaction between the components of each water matrix with the dynamics of the reaction. The negative value of the time at which the EC_{50} would be reached for the synthetic NOM pattern is due to its high initial concentration and difficult mineralization during the reaction time evaluated in the present investigation. This situation can be corroborated in Fig. 2, where LC_{50} values were not reached at any point in the process. According to Kommineni et al. [32], the presence of high NOM concentrations can result in the formation of by-products that can cause water quality to deteriorate beyond its initial state of contamination. This applies to the presence of nitrates and carbonates, as well as other substances that can interfere with the destruction of target contaminants, ultimately reducing the effectiveness of treatment technology.

Although the TOC values for which the EC_{50} was reached were below the maximum recommended value for use in drinking water in the Colombian regulations, it is observed that the cladoceran still displayed sensitivity, which is useful measuring toxicity over drinking water for human consumption.

On the other hand, surface waters from Pueblo Nuevo and DWTP Ipiales, which exhibited the highest concentrations of chlorides and sulfates, respectively (Table 2), showed that the content of both anions exerted a clear inhibitory effect on the performance of the CWPO process (Table 3) in terms of carbon mineralization and decolorization.

Table 4
CWPO reaction times at which 50% of the *D. magna* population was immobilized (EC_{50} – 48 h)

Evaluated surface water	CWPO *t* (min)	TOC at EC_{50} – 48 h (mg/L)	Starting TOC (mg/L)
Chaguaipe	71	3.98	4.65
Pueblo Nuevo	96	1.98	2.40
Bajo Jagua	92	2.30	2.57
Pasto DWTP	69	2.21	2.56
Túquerres DWTP	72	1.40	1.54
Ipiales DWTP	86	2.04	2.12
Synthetic NOM pattern	NA	6.31	10.20

NA: not applicable, since the 50% of the *D. magna* immobilization (EC_{50} – 48 h) was not reached within the assessed CWPO reaction times.

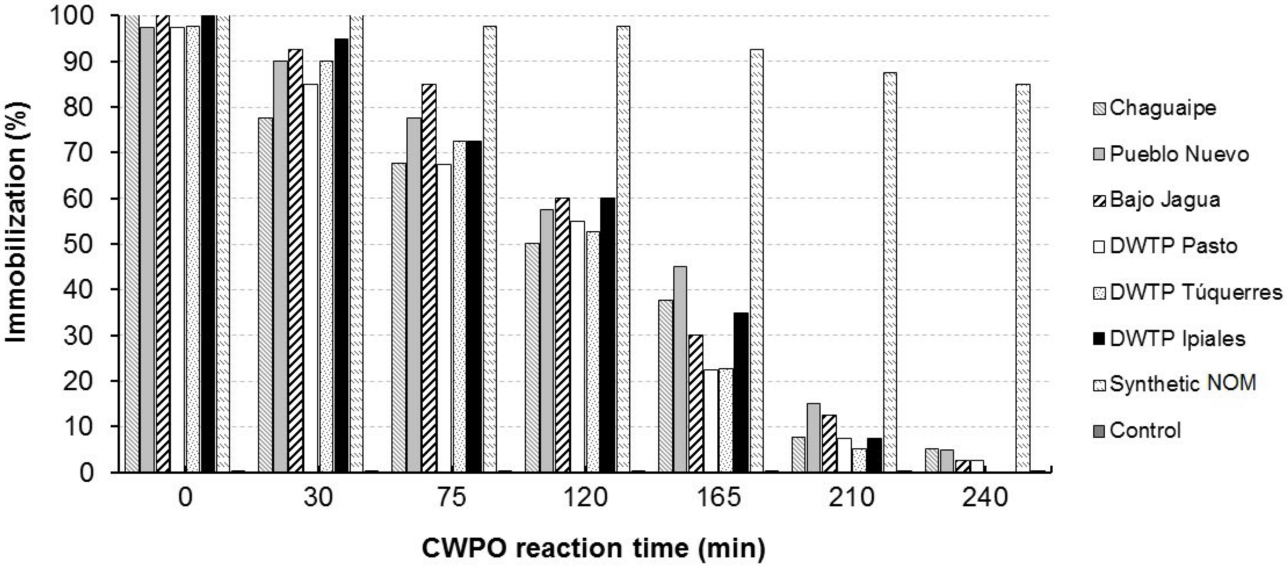

Fig. 2. Immobilization of *D. magna* as a function of the CWPO reaction time.

Nevertheless, it apparently did not affect the levels of *D. magna* immobilization (Table 4) through the course of the reaction. In the rural aqueducts, the CWPO unit could be coupled with a physicochemical pretreatment, for instance, employing membrane filters to remove inorganic anions; in urban DWTPs, adequate dosing of coagulant should be fitted so that the concentration of anions (chlorides and sulfates) do not significantly affect the CWPO performance or increases too much the peroxide dose required to keep proper TOC removal and decolorization.

When comparing the TOC at EC_{50} – 48 h between raw waters of rural aqueducts (Chaguaipe, Pueblo Nuevo and Bajo Jagua) and treated waters of urban aqueducts (Túquerres, Pasto and Ipiales DWTP), it can notice than the EC_{50} for the treated waters (1.40–2.21 mg/L) is slightly smaller than raw waters (1.98–3.98 mg/L), this indicates that the more refractories fraction of NOM remaining upon the physicochemical treatment can be compared with the toxicity of NOM present in raw waters. In this case, it has been reported that the hydrophilic and transphylic fraction are more difficult fractions to be removed by the physicochemical treatment, which could suggest that these types of fractions are of the main responsible of the EC_{50} values obtained.

3.6. Immobilization of D. magna in water samples at different CWPO reaction times

The evolution of the measured *D. magna* immobilization as a function of the CWPO reaction time for the analyzed water samples is plotted in Fig. 2. It can be clearly seen that as the CWPO reaction time increased the toxicity on *D. magna* decreased. It should be noted that in all samples, despite the relatively low starting organic loads (in terms of TOC concentration), there were final levels of immobility between 97.5% and 100%. This demonstrates the high sensitivity of the selected bio-indicator and its huge utility in assessing the toxicity of drinking water. In real surface waters, typically low concentrations of pollutants are present, but not necessarily safe upon the later disinfection stage, especially if chlorination is used. In all cases, a steady decrease in the immobility of *D. magna* was observed as the course of the CWPO reaction took place, demonstrating the efficiency of such AOP for this purpose.

It is noteworthy that in the samples from the Túquerres and Ipiales DWTPs, a final zero immobilization was achieved; full removal of toxicity within the sensitivity range of the bio-indicator *D. magna* was accomplished in these water samples. Meanwhile, in the samples from Bajo Jagua, Pasto DWTP, Chaguaipe, and Pueblo Nuevo, immobility values between 2.5% and 5.0% were obtained at the final reaction time. This is probably because such samples had a higher initial (2.56–4.65 mg/L) and final (2.06–3.6 mg/L) TOC concentrations as compared with those where full depletion of toxicity was achieved. It should be also considered that such samples at the beginning of the CWPO reaction exhibited apparent color and turbidity above the allowed limit established by Colombian regulations [27]. These parameters could be also related to the presence of NOM, and eventually affect the final level of immobility. Finally, the catalytic test in the presence of the NOM synthetic surrogate still displayed a level of immobility of 85% for the final time of reaction (Fig. 2), probably due to the high concentration of starting TOC (10.2 mg/L). Nevertheless, it seems even more plausible to state that the CWPO treatment activated by Al/Fe-PILCs displayed a higher ability to decrease the toxicity over the studied real waters, which would be very useful in general improving real surface waters.

Regarding such promising results, Bekbolet et al. [33], when applying either coagulation, ozonation or coagulation followed by ozonation and photocatalysis on three natural water sources, recorded immobilization percentages of 35%, 60% and 40% on *D. magna* after the photocatalytic treatment. In another more recent study conducted by Ibarra and Salcedo [34], when determining the acute toxicity of water samples treated by chlorination, they obtained immobilizations of 39.3% and 43.5%. The comparison between this couple of studies, in which other methods of treatment were used, with the results of this research, the CWPO technology showed very interesting results in the removal of toxicity from raw and partially treated waters, potentially useful in the production of human drinking water. However, it also shows the higher toxicity of chlorinated waters even performed over low dissolved TOC concentrations. Furthermore, although the oxidation intermediates during the earlier reaction times still showed relatively high levels of toxicity as compared with the starting water samples, OBPs clearly exhibited non-toxic behavior (0.0%–5.0%). Briefly speaking, the CWPO treatment successfully improved water quality, therefore reducing fate negative effects on human health.

3.7. Effect of pH and electrical conductivity on the immobilization of D. magna

Table 5 shows the correlation coefficients of pH and electrical conductivity against the immobilization of *D. magna*. The initial values correspond to 0 min of the CWPO reaction, and the final values correspond to a time of 240 min of the CWPO treatment. However, the Spearman correlation coefficients were obtained from either the average pH or average electric conductivity measured through the toxicity tests at every time of reaction (it must be emphasized that a single catalytic experiment was carried out per every time of CWPO reaction and every water sample). In general, a slight decrease in the pH due to the CWPO treatment was observed. This behavior suggests the formation of short-chain carboxylic acids as the main by-products of the heterogeneous-Fenton oxidation, which has been widely documented by other authors for NOM degradation through different AOPs [35,36]. pH values of the treated waters, between 6.5 and 9.0, were all within the permissible range established by the Colombian regulations for drinking water [27], and also within 6.0–9.0, the range required for proper cultivation and maintenance of *D. magna* according to ISO 6341 [17].

The samples for which there was no significant correlation between pH and immobility of the cladoceran were Chaguaipe, Pueblo Nuevo, Túquerres DWTP and NOM synthetic pattern. On the other hand, the water samples from Bajo Jagua as well as from the Pasto and Ipiales DWTPs reported a low positive relationship, which suggests that an increase in pH could lead to greater immobility of *D. magna*. According to the above-explained, it can be inferred that the effect of pH on the measured results of toxicity was not significant.

On the other hand, on average a 10-fold depletion was observed in the electrical conductivity upon the CWPO at the final time of reaction. Probably this is why Spearman's coefficients between 0.69 and 0.96 showed a significant positive correlation between electrical conductivity and the cladoceran immobility in all samples. The high initial amount of dissolved inorganic ions but also the content of highly hydrolyzed groups bearing from NOM getting removed after the strong oxidizing reaction seems to explain such a behavior. Thus, in this case, it had a positive effect on the survival of the individuals; according to ISO 6341 [17], the electrical conductivity of the pure water in *D. magna* must be less than 10 μS/cm. In this study, the conductivities obtained after the CWPO were below the level defined by such standard; therefore, this aspect should be carefully taken into account when using *D. magna* in other studies regarding the toxicity of water samples exhibiting significantly different electric conductivity. For instance, in further studies, a good solution to decrease the undesirable effect of this factor on the *D. magna* toxicity results could be the control of the electrical conductivity within a narrow range for a series of samples, by the addition of innocuous inorganic electrolytes.

Kommineni et al. [32] stated that many water quality parameters can affect the effectiveness of the AOPs. For example, basically, all dissolved organic compounds present in a water source will reduce the removal efficiency of a target compound by consuming $HO^{\bullet}$, nitrates and nitrites absorb UV light and, consequently, concentrations of anyone of the above 1.0 mg/L would limit the effectiveness of the photocatalytic AOPs. Likewise, phosphates and sulfates commonly present in low concentrations in water supplies, alongside the remaining contents of chlorides and sulfates from the conventional coagulation-flocculation units, also consume $HO^{\bullet}$ radicals. Moreover, there is still a lack of investigations to elucidate the true chemical nature of the oxidizing species involved in this kind of Heterogeneous-Fenton degradations under circumneutral pH conditions, probably less susceptible to such well-known scavenging species of radicals.

Table 5
Correlation between pH and electrical conductivity

Evaluated surface water	pH		Spearman correlation coefficients	*P*-value	Electrical conductivity (μS/cm)		Spearman correlation coefficients	*P*-value
	Initial	Final			Initial	Final		
Chaguaipe	7.00	6.66	0.30	>0.05	51.2	6.20	0.93	**<0.05**
Pueblo Nuevo	7.65	8.16	0.02	>0.05	28.3	2.47	0.84	
Bajo Jagua	7.39	7.00	0.35	**<0.05**	25.1	2.40	0.89	
Pasto DWTP	7.19	6.90	0.38	**<0.05**	48.3	4.23	0.92	
Túquerres DWTP	7.58	7.32	0.33	>0.05	35.3	3.35	0.93	
Ipiales DWTP	7.89	7.65	0.36	**<0.05**	55.8	4.11	0.96	
Synthetic NOM pattern	7.00	6.85	–0.26	>0.05	32.1	11.9	0.69	

4. Conclusions

In this study, the *D. magna* acute toxicity test showed to be very useful recording evolution of toxicity of the Heterogeneous-Fenton, CWPO process activated by an Al/Fe-PILC clay catalyst; moreover, it may be also easily applied in the study of other AOPs, mainly when used on low-concentrated contaminants in water, including those more concerning in drinking water production. It will be particularly useful in the study of very complex contaminants, whose intermediates and by-products are difficult to be followed by other techniques like the chromatographic or spectroscopic ones. Since concentrations of H_2O_2 used in the CWPO and all other Fenton processes greater than 7.3 mg/L of H_2O_2 showed to produce toxic effects on *D. magna*, it must be carefully deactivated prior to the toxicity measurements. Moreover, such a remaining concentration of peroxide in this and other Fenton-like oxidations is properly deactivated by Na_2SO_3, that nevertheless, should be also added just in stoichiometric amount, because in turn, any excess may also significantly influence the toxicity measurements decreasing its usefulness. *D. magna* toxicity was remarkably reduced by the CWPO treatment in real surface waters currently used as drinking water supplies in Nariño – Colombia, reaching depletions of 97.5%–100% in the immobility through 180 min of reaction, under very mild ambient conditions of temperature (12.8°C–26.5°C) and pressure (~0.71 atm) and pretty low dosage of H_2O_2 (0.45 mg H_2O_2/mg TOC). Concentrations of apparent color and turbidity above the limit allowed by the regulations for drinking water in the influent water samples may explain those cases were no full removal of *D. magna* acute toxicity was achieved through the AOP treatment within the timeframe studied. CWPO reaction times between 69.1 and 96.3 min were required to achieve EC_{50} on the individuals of *D. magna*; greater reduction in immobility was recorded as the reaction time further increased. It was therefore concluded that the compounds generated during the CWPO degradation of NOM in real surface water steadily decreased their toxicity. Electrical conductivity showed a high positive correlation for all the samples, indicating that this parameter affects the immobility of the individuals of *D. magna*, and then should be carefully taken into account in further studies.

Acknowledgments

Financial support for this research was provided by the *CTeI* Fund of *Sistema General de Regalías - Colombia* (BPIN: 2014000100020). The authors gratefully acknowledge Dr. Maria Teresa Reguero Reza from Research Group of Ecotoxicology of the Biotechnology Institute from the National University of Colombia for facilitating the strain of *D. magna* used in this study. A.M. Garcia also wishes to express her gratitude to the Ph.D. scholarship granted from Nariño Department, program "Strengthening of regional capacities in research, technological development, and innovation in Nariño Department" managed by CEIBA Foundation, Colombia.

References

[1] E. Corcoran, C. Nellemann, E. Baker, R. Bos, D. Osborn, H. Savelli, Sick Water: The Central Role of Wastewater Management in Sustainable Development – A Rapid Response Assessment, United Nations Environment Programme, UN-HABITAT, GRID-Arendal, Norway, 2010.

[2] A.-M. García, R.A. Torres-Palma, L.A. Galeano, M.Á. Vicente, A. Gil, Separation and Characterization of NOM Intermediates Along AOP Oxidation, A. Gil, L.A. Galeano, M.À. Vicente, Eds., Applications of Advanced Oxidation Processes (AOPs) in Drinking Water Treatment, Vol. 67, Springer, Switzerland, 2017, pp. 99–132.

[3] M. Sillanpää, M.C. Ncibi, A. Matilainen, M. Vepsäläinen, Removal of natural organic matter in drinking water treatment by coagulation: a comprehensive review, Chemosphere, 190 (2018) 54–71.

[4] A. Grunert, A. Frohnert, H.-C. Selinka, R. Szewzyk, A new approach to testing the efficacy of drinking water disinfectants, Int. J. Hyg. Environ. Health, 221 (2018) 1124–1132.

[5] J.H. Ramírez, L.A. Galeano, Natural Organic Matter Removal by Heterogeneous Catalytic Wet Peroxide Oxidation (CWPO), A. Gil, L.A. Galeano, M.À. Vicente, Eds., Applications of Advanced Oxidation Processes (AOPs) in Drinking Water Treatment, Vol. 67, Springer, Switzerland, 2017, pp. 69–98.

[6] J. Barrault, M. Abdellaoui, C. Bouchoule, A. Majesté, J.M. Tatibouët, A. Louloudi, N. Papayannakos, N.H. Gangas, Catalytic wet peroxide oxidation over mixed (Al-Fe) pillared clays, Stud. Surf. Sci. Catal., 130 (2000) 749–754.

[7] L.A. Galeano, M.Á. Vicente, A. Gil, Catalytic degradation of organic pollutants in aqueous streams by mixed Al/M-pillared clays (M = Fe, Cu, Mn), Catal. Rev., 56 (2014) 239–287.

[8] L.A. Galeano, P.F. Bravo, C.D. Luna, M.Á. Vicente, A. Gil, Removal of natural organic matter for drinking water production by Al/Fe-PILC-catalyzed wet peroxide oxidation: Effect of the catalyst preparation from concentrated precursors, Appl. Catal., B, 111–112 (2012) 527–535.

[9] A. Ordoñez-Ordoñez, D.M. Revelo-Romo, A.M. Garcia-Mora, A. Hidalgo-Troya, L.-A. Galeano, MS2 coliphage inactivation by Al/Fe PILC-activated catalytic wet peroxide oxidation: multiresponse statistical optimization, Heliyon, 5 (2019) e01892 1–11.

[10] J.-K. Kim, K.G. Choi, I.-H. Cho, H.-S. Son, K.-D. Zoh, Application of a microbial toxicity assay for monitoring treatment effectiveness of pentachlorophenol in water using UV photolysis and TiO_2 photocatalysis, J. Hazard. Mater., 148 (2007) 281–286.

[11] D.S. Babu, V. Srivastava, P.V. Nidheesh, M.S. Kumar, Detoxification of water and wastewater by advanced oxidation processes, Sci. Total Environ., 696 (2019) 133961.

[12] A. Sharma, J. Ahmad, S.J.S. Flora, Application of advanced oxidation processes and toxicity assessment of transformation products, Environ. Res., 167 (2018) 223–233.

[13] I.A. Ike, T. Karanfil, J.W. Cho, J. Hur, Oxidation byproducts from the degradation of dissolved organic matter by advanced oxidation processes – a critical review, Water Res., 164 (2019) 114929.

[14] M. Sillanpää, M.C. Ncibi, A. Matilainen, Advanced oxidation processes for the removal of natural organic matter from drinking water sources: a comprehensive review, J. Environ. Manage., 208 (2018) 56–76.

[15] M. Díaz, M. Sobrero, Y. Granados, In: G. Castillo, Ed., Toxicological Tests and Methods of Water Quality Assessment; Standardization, Intercalibration, Results and Applications, IMTA, México, 2004, pp. 125–140 (in Spanish).

[16] K. Cubas, Correlation Between Toxicity Factor and Physicochemical Parameters for Treated Domestic Effluents, Dissertation Universidade Tecnológica Federal do Paraná, 2012 (in Portuguese).

[17] AENOR: Asociación Española de Normalización y Certificación, Water Quality: Determination of Mobility Inhibition of *Daphnia magna* Strains (*Cladocera crustacea*), Acute Toxicity Test, ISO 6341, AENOR, Madrid-España, 2013 (in Spanish).

[18] S. Chiva, J. Berlanga, R. Cuenca, J. Climent, Advanced Oxidation Processes in the Integral Water Cycle, Castellón de la Plana: Publicacions de la Universitat Jaume I, 2017 (in Catalan).

[19] L.H. Sipaúba, O. Rocha, Production of Plankton (Phytoplankton and Zooplankton) for the Feeding of Aquatic Organisms, RiMa, São Carlos-Brazil, 2003 (in Portuguese).
[20] IDEAM – Instituto de Hidrología, Meteorología y Estudios Ambientales, INVEMAR – Instituto de Investigaciones Marinas y Costeras, Water Monitoring Protocol, Bogotá D.C., Colombia, 2017 (in Spanish).
[21] APHA, AWWA, WEF, Standard Methods for the Examination of Water and Wastewater, 22nd ed., American Public Health Association, American Water Works Association, and Water Environment Federation, Washington, D.C., 2012.
[22] A.M. García-Mora, H. García, R. Torres-Palma, A. Hidalgo, L.A. Galeano, Optimization of the removal of natural organic matter from a synthetic surrogate and real surface water by the Al/Fe pillared clay-activated catalytic wet peroxide oxidation, Environ. Sci.: Water Res. Technol., (2020) (in Press).
[23] R.F. Pupo, A.G. Trovó, M.R.A. da Silva, R.D. Villa, M.C. de Oliveira, Fundamentals and environmental applications of the Fenton and photo-Fenton processes, Quim. Nova, 30 (2007) 400–408 (in Portuguese).
[24] J.R. Meinertz, S.L. Greseth, M.P. Gaikowski, L.J. Schmidt, Chronic toxicity of hydrogen peroxide to *Daphnia magna* in a continuous exposure, flow-through test system, Sci. Total Environ., 392 (2008) 225–232.
[25] A. Bownik, Z. Stępniewska, Protective effects of ectoine on behavioral, physiological and biochemical parameters of *Daphnia magna* subjected to hydrogen peroxide, Comp. Biochem. Physiol. C: Toxicol. Pharmacol., 170 (2015) 38–49.
[26] W.J. Liu, S.A. Andrews, M.I. Stefan, J.R. Bolton, Optimal methods for quenching H_2O_2 residuals prior to UFC testing, Water Res., 37 (2003) 3697–3703.
[27] Colombia, Ministerio de la Protección Social, Ministerio de Ambiente, Vivienda y Desarrollo Territorial, Resolution 2115, June 22, 2007, Definition of Characteristics, Basic Instruments and Frequencies of the Control and Surveillance System for Water Quality for Human Consumption, Bogotá, Colombia, 2007 (in Spanish).
[28] A. Tapia, A. Reyes, I. García, Study of the fraction of organic matter of greater removal in the coagulation-flocculation process using surface water, Nexo Revista Científica, 24 (2013) 72–80 (in Spanish).
[29] M. Munoz, Z.M. de Pedro, J.A. Casas, J.J. Rodriguez, Combining efficiently catalytic hydrodechlorination and wet peroxide oxidation (HDC–CWPO) for the abatement of organochlorinated water pollutants, Appl. Catal., B, 150–151 (2014) 197–203.
[30] A.H. Pizarro, C.B. Molina, M. Munoz, Z.M. de Pedro, N. Menendez, J.J. Rodriguez, Combining HDC and CWPO for the removal of p-chloro-m-cresol from water under ambient-like conditions, Appl. Catal., B, 216 (2017) 20–29.
[31] M.C. Valsania, F. Fasano, S.D. Richardson, M. Vincenti, Investigation of the degradation of cresols in the treatments with ozone, Water Res., 46 (2012) 2795–2804.
[32] M. Kavanaugh, Z. Chowdhury, S. Kommineni, Removal of MTBE with Advanced Oxidation Processes, IWA Publishing, London, 2004, pp. 111–208.
[33] M. Bekbolet, C.S. Uyguner, H. Selcuk, L. Rizzo, A.D. Nikolaou, S. Meriç, V. Belgiorno, Application of oxidative removal of NOM to drinking water and formation of disinfection by-products, Desalination, 176 (2005) 155–156.
[34] Y. Ibarra, S. Salcedo, Environmental Toxicological Evaluation of Drinking Water, Associated with the Presence of Trihalomethanes, Dissertation, Universidad Mariana, 2017 (in Spanish).
[35] S. Liu, M. Lim, R. Fabris, C. Chow, M. Drikas, R. Amal, TiO_2 photocatalysis of natural organic matter in surface water: impact on trihalomethane and haloacetic acid formation potential, Environ. Sci. Technol., 42 (2008) 6218–6223.
[36] R. Lamsal, M.E. Walsh, G.A. Gagnon, Comparison of advanced oxidation processes for the removal of natural organic matter, Water Res., 45 (2011) 3263–3269.

Supplementary information

S1. Daphnia magna sensitivity determination results

Table S1
Effective concentration and confidence limits of $K_2Cr_2O_7$ toxicity on *Daphnia magna*

Bioassay number	EC_{50} – 24 h (mg/L)	Lower confidence limit (mg/L)	Upper confidence limit (mg/L)
1	0.97	0.85	1.11
2	1.34	1.23	1.48
3	0.67	0.56	0.79
4	0.87	0.77	0.97
5	1.09	0.94	1.27
6	1.36	1.18	1.62
7	1.07	0.94	1.23
8	1.51	1.28	1.92
9	1.26	1.06	1.57
10	0.99	0.84	1.15

S2. Analytical procedures

S2.1. Collection and characterization of water samples for catalytic wet peroxide oxidation treatment

Water samples were collected from six different supplies of drinking water treatment plants (DWTPs) in Nariño State, Colombia, and then transported following the protocol described by the Colombian Institute of Hydrology, Meteorology and Environmental Studies (Instituto de Hidrología, Meteorología y Estudios Ambientales: IDEAM) and the Colombian Institute of Marine and Coastal Research (Instituto de Investigaciones Marinas y Costeras: INVEMAR) [S1]. These sources corresponded to both Chaguaipe rural aqueduct and DWTP of the municipality of Ipiales, the Túquerres municipality DWTP, Jagua and Pueblo Nuevo rural aqueducts of the municipality of Tumaco (on the pacific coast), and the DWTP of the municipality of Pasto. In the case of the samples taken in DWTPs, the sampling point was the effluent of the conventional physicochemical treatment unit (flocculation–sedimentation), whereas, in the case of the rural aqueducts, fully raw water was collected. The pH levels, electrical conductivity and temperature of the collected waters were determined on-site using a portable multiparameter (SensoDirect 150, Dortmund, Germany).

Once the samples were collected, they were transported and kept at 4°C for analysis of total organic carbon (TOC) and total nitrogen (TN) in TOC-L analyzer equipment coupled with a TN analyzer (Shimadzu, Columbia, Maryland, USA), spectral scan 200–700 nm, to determine UV_{254} and UV_{456}, in a UV 2600 spectrophotometer (Shimadzu, Columbia, Maryland, USA), The other analyses were performed according to the methodology reported in [S2]: chlorides (method 4500Cl – B, colorimetric technique), phosphates (method 4500P – D, colorimetric technique), nitrates (method $4500NO_3$ – B, colorimetric technique), nitrites (method $4500NO_2$ – B, colorimetric technique), sulfates (method 4500SO_4 – E, turbidimetric technique), apparent color (method 2120 – C, colorimetric technique), turbidity (method 2130 – B, nephelometric technique), fluorides (method 4500F – C, colorimetric technique) and total hardness (method 2340 – C, titulometric technique).

References

[S1] IDEAM – Instituto de Hidrología, Meteorología y Estudios Ambientales, INVEMAR – Instituto de Investigaciones Marinas y Costeras, Water Monitoring Protocol, Bogotá D.C., Colombia, 2017 (in Spanish).

[S2] APHA, AWWA, WEF, Standard Methods for the Examination of Water and Wastewater, 22nd ed., American Public Health Association, American Water Works Association, and Water Environment Federation, Washington, D.C., 2012.