

Article

Removal of Tetracycline Oxidation Products in the Nanofiltration Process

Renata Żyła ^{1,*} , Stanisław Ledakowicz ², Tomasz Boruta ², Magdalena Olak-Kucharczyk ¹,
Magdalena Foszpańczyk ^{1,2} , Zdzisława Mrozińska ¹ and Jacek Balcerzak ² 

- ¹ Łukasiewicz Research Network—Textile Research Institute, ul. Brzezińska 5/15, 92-103 Łódź, Poland; molak-kucharczyk@iw.lodz.pl (M.O.-K.); mfoszpanczyk@iw.lodz.pl (M.F.); zmrozinska@iw.lodz.pl (Z.M.)
- ² Faculty of Process and Environmental Engineering, Lodz University of Technology, ul. Wólczańska 213, 90-924 Łódź, Poland; stanislaw.ledakowicz@p.lodz.pl (S.L.); tomasz.boruta@p.lodz.pl (T.B.); jacek.balcerzak@p.lodz.pl (J.B.)
- * Correspondence: zylla@iw.lodz.pl

Abstract: The possibility of removing tetracycline (TRC) from water in an integrated advanced oxidation and membrane filtration process was investigated. Ozonation and UV/H₂O₂ photooxidation were applied for the destruction of TRC. Six oxidation products (OPs) retaining the structural core of TRC have been identified. One new TRC oxidation product, not reported so far in the literature, was identified—ethyl 4-ethoxybenzoate. All identified OPs were effectively retained on the membrane in the nanofiltration process. However, chemical oxygen demand (COD) measurements of the filtrates showed that in the case of UV/H₂O₂ oxidation, the OPs passed through the membrane into the filtrate. Various water matrices were used in the research, including the river water untreated and after ozone treatment. It has been shown that organic matter present in surface water can improve pharmaceutical retention, although it contributes to significant membrane fouling. Pre-ozonation of the river water reduced the membrane fouling. The XPS analysis was used to show ozone and H₂O₂ influence on the top polymer layer of the membrane. It was shown that the oxidants can damage the amide bond of the polyamide.

Keywords: tetracycline; advanced oxidation processes; oxidation products; nanofiltration



Citation: Żyła, R.; Ledakowicz, S.; Boruta, T.; Olak-Kucharczyk, M.; Foszpańczyk, M.; Mrozińska, Z.; Balcerzak, J. Removal of Tetracycline Oxidation Products in the Nanofiltration Process. *Water* **2021**, *13*, 555. <https://doi.org/10.3390/w13040555>

Academic Editor: Dionysios (Dion) D. Dionysiou

Received: 7 January 2021
Accepted: 17 February 2021
Published: 22 February 2021

Publisher's Note: MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



Copyright: © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

1. Introduction

The main routes for pharmaceuticals entering the environment are wastewater streams due to their release into surface waters or through ground systems, when the treated wastewater is used for irrigation or when sewage sludge is applied as a fertilizer to improve soil properties on agricultural land [1]. Many reports show that conventional wastewater treatment plants that have been designed and optimized to remove suspensions and compounds responsible for eutrophication (e.g., biodegradable organic matter, nitrogen, and phosphorus) are not sufficient to remove many pharmaceuticals [1–5].

Tetracyclines (TRCs) are antibiotics well known for their broad spectrum of activity, spanning a wide range of Gram-positive and Gram-negative bacteria, spirochetes, obligate intracellular bacteria, as well as protozoan parasites. Therefore, TRCs are among the most frequently used antibiotics, not only for human and animal therapy but also as agricultural feed additives [6]. According to the estimates, the worldwide usage of veterinary antibiotics will increase by about 70% from 2010 to 2030 [7,8]. The group of TRCs includes tetracycline (TRC), chlorotetracycline, oxytetracycline, and doxycycline [6]. TRC is one of the pharmaceuticals commonly found in waters [6,9]. Its presence in the surface water (range of 1.1–110,000 ng/L), groundwater (range of 9.51–29,700 ng/L), drinking water (range of 11.2–632 ng/L), wastewater (range of 0.0019–12,000 µg/L), sediments (range of 0.1–156,000 ng/g), and sludge (range of 14.3–1620 µg/kg) has been documented [6]. TRCs inhibit the growth and development of many aquatic organisms [10–14]. On the other hand,

exposure to TRC leads to the development of antibiotic resistance genes [15–17]. Therefore, studies are undertaken on the application of various TRC decomposition/removal methods from water and wastewaters [18–27].

In the case of pharmaceuticals, methods such as advanced oxidation processes (AOPs) and membrane filtration are highly effective in their removal [3,4]. These methods have been found to be suitable for the disposal of pharmaceuticals, especially when treated water is intended for reuse as a source of drinking water or to supplement the aquifer [1]. Combined membrane filtration and AOPs provides complementary effect. First, AOPs treatment assist in eliminating membrane fouling and remediation of organics in the concentrate via oxidation of the foulants, dissolve organic matters and organic compounds in the concentrate. Secondly, membrane filtration concentrates the pollutants to the level that can be efficiently treated by AOPs, and moreover confines/retains the toxic intermediates within the membrane to ensure prolonged contact with the oxidants [4]. Liu et al. applied the nanofiltration (NF) combined with ozone-based advanced oxidation processes to removal of four antibiotics: norfloxacin, ofloxacin, roxithromycin, and azithromycin from wastewater [3]. The treatment of NF concentrate by UV/O₃ process leads to excellent removal efficiencies of antibiotics (>87%), a partial removal of dissolved organic carbon (DOC) (40%), an increase of five-day biochemical oxygen demand/chemical oxygen demand ratio (BOD₅/COD) (4.6 times), and a reduction of acute toxicity (58%) [3].

AOPs include chemical oxidation processes using ozone or other oxidizing agents such as UV/H₂O₂, O₃/H₂O₂, TiO₂/UV, Fe²⁺/H₂O₂, which partially degrade or provide chemical modification of organic compounds. Due to the rapid development of instrumental analytical methods, knowledge about the mechanisms of oxidation reactions is still expanding and includes compounds, e.g., pharmaceuticals, that appear in wastewater at nanogram concentrations [18,19,28,29].

Ozone as an oxidizing agent is an unstable compound and decomposes by reacting with chemical compounds present in water or wastewater, producing highly reactive hydroxyl radicals (•OH). Interest in ozone as a strong oxidant continues to increase due to the progressive reduction of its production costs. This is evidenced by the constantly growing number of publications devoted to the use of ozone for the decomposition of various organic compounds, including pharmaceuticals [29–36].

Additionally, in the case of TRC, ozonation and AOPs ensure complete transformation of this molecule into intermediate products [5,21,22]. Although the AOPs allow complete degradation of TRC, they do not provide complete mineralization of oxidation products (OPs) [5,23,24]. In the case of antibiotics, total mineralization is very difficult, because the arrangement of aromatic rings in their structure tends to stabilize the oxidation by-products formed during photolysis [25]. The resulting low quantum efficiency values show that the use of UV radiation is inefficient in terms of removing TRC from water, because long exposure times are required to achieve 100% degradation. The use of indirect photolysis, radiolysis, and ozonation confirmed that TRCs can be effectively removed from water, obtaining the highest degree of degradation using ozone and ozone/H₂O₂ [26]. Studies have shown that the low degree of TRC mineralization is associated with the formation of more toxic OPs [26].

Membrane filtration is a technique that ensures effective removal of compounds from a solution [27,31,37–39]. This is a separation process based on the differences in physical and chemical properties of the separated substances, such as the particle size, partial vapor pressure, chemical affinity, electric charge, etc. These processes are widely used in many areas of life, in food and pharmaceutical industries, as well as in environmental protection [40–43]. The cost of these processes, however, is high and is associated, among other things, with the sensitivity of membranes to fouling [40–43].

An effective method of removing TRC is membrane filtration. However, as a result of the filtration process, a concentrate is received that needs to be utilized. TRC retention has been shown to be sufficient in reverse osmosis and nanofiltration (NF) tight membranes, and in most cases, it was observed to be over 80% [9].

AOPs and membrane techniques are now increasingly used in wastewater treatment and water recovery processes [4,44–48]. There are many reports on the phenomena occurring in integrated processes of chemical oxidation and membrane filtration (mainly ultrafiltration) of surface waters leading to the removal of humic compounds [49–52]. In the case of other organic compounds that may be present in water and wastewater as a result of human activity, the phenomena occurring on the surface of membranes, such as fouling reduction, change in membrane charge due to blockage of membrane active centers, adsorption, are poorly understood and described. This applies to, among others, the pharmaceuticals used to treat humans and animals. There are many literature reports concerning the oxidation of pharmaceuticals using ozone or AOPs treated as a unit process [18–21,23–25,27,29–36,53–55]. Such data are also available for the processes of NF or reverse osmosis [4,42,43]. Understanding the phenomena that occur during the combined use of chemical oxidation and membrane filtration will contribute to solving important problems associated with the removal of hazardous substances such as pharmaceuticals and will improve the efficiency and effectiveness of water recovery. The integration of membrane filtration and AOPs combines the advantages and eliminates the problems associated with these two purification approaches.

Surface waters (lakes and rivers) are the main resources of drinking water in many parts of the world. However, they contain a number of pollutants, including natural organic matter (NOM). NOM is a complex matrix of organic matter derived from the decomposition of living terrestrial and aquatic organisms and consists mainly of humic (HA) and fulvic (FA) and non-humic fractions, which are a mixture of carbohydrates, amino acids, and proteins. [19,20]. The presence of organic matter in an aqueous solution may cause severe membrane contamination, especially in the presence of calcium ions [30], which leads to a decrease in the filtrate flux [20] and reduced retention efficiency [32]. The experiments carried out for deionized water may give completely different results than those carried out on the real matrix, e.g., in post-flood water from a river.

This paper presents the results of research on the integrated process of AOPs and NF. The phenomena occurring during the NF of the oxidized TRC solution were analyzed. The work included issues typically omitted in the current literature on the AOP/NF hybrid processes. Previously published works were rather focused on process optimization, obtaining the highest removal efficiency of hazardous compounds and the reduction of fouling phenomenon. In literature, there is a lack of information about integrated AOP/membrane filtration processes from the point of view of OPs and their influence on the membrane filtration effectivity.

2. Materials and Methods

2.1. Materials

Tetracycline hydrochloride (TRC) was purchased from Sigma Aldrich (Poznań, Poland). Catalase (2000–5000 units/mg protein) used as hydrogen peroxide decomposition catalyst, hydrogen peroxide $\geq 30\%$, for trace analysis used to generate hydroxyl radical in UV/H₂O₂ system, as well as Na₂SO₃ p.a used as ozone destructor were purchased from Sigma Aldrich.

The chemical structure of TRC is presented in Figure 1. TRC stock solution at concentration 1000 mg/L was prepared. The target solution at concentration 75 mg/L used in the studies were obtained by dilution of stock solution.

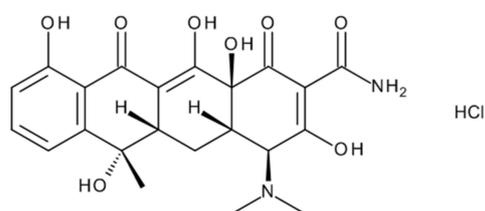


Figure 1. Chemical structure of tetracycline hydrochloride (TRC).

The catalase solution was prepared by pouring 1 g of dry catalase with 25 mL of deionized water to achieved catalase concentration of about 40 g/L, which is about 80×10^3 – 200×10^3 units/L. Prepared solutions were kept in refrigerator. The catalase solution was added to each sample in an amount of 400–1000 units per mL of sample taken for analysis. Na_2SO_3 solution at concentration 24 mM was prepared. The solution was added to the samples after the ozonation process in amount 100 μL /5 mL of samples.

The catalase solution was prepared by pouring about 1 g of dry catalase with 25 mL of deionized water. After saturating the solution with catalase, it was filtered on a soft paper filter and the solution prepared in this way was kept in a refrigerator. To each sample containing unreacted hydrogen peroxide, 50 μL of catalase solution was added per 10 mL of the sample.

Natural water samples were collected in September 2020 from the mouth of the Dobryznka River into the Ner River (RW) and stored at 6 °C. Table 1 presents the characteristic parameters of RW. The total organic carbon (TOC), chemical oxygen demand (COD), and selected ions were measured with using appropriate cuvette tests (HACH LANGE) and photometer DR 3900 (HACH LANGE).

Table 1. Characteristic parameters of natural river water (RW).

Parameter	Value	Standard Deviation
pH	8.06	0.007
Conductivity, $\mu\text{S}/\text{cm}$	570	0.71
COD, mg/L	30.7	0.07
TOC, mg/L	10.2	0.14
K^+ , mg/L	4.66	0.25
PO_4^{3-} , mg/L	0.148	0.01
SO_4^{2-} , mg/L	43	1.56
NO_3^- , mg/L	1.73	0.08
Cl^- , mg/L	31.3	1.77
CO_2 , mg/L	162	2.12

2.2. Research Equipment

Oxidation with UV/ H_2O_2 was carried out in a photoreactor with a capacity of 3.7 L equipped with two UV lamps ($\lambda = 254$ nm) with a total power of 22 W. The scheme of the reactor construction was presented in previous work [56]. A 30% solution of hydrogen peroxide was used in the oxidation with UV/ H_2O_2 . To 3700 mL of the solution, 1.2 mL of H_2O_2 was added (which constituted a molar concentration of 2.86×10^{-3} mol/L). Process of UV/ H_2O_2 oxidation was performed in room temperature. The samples were taken after 5, 15, 30, and 60 min and analyzed in liquid chromatography and UPLC/MS. Then, 50 μL of catalase was added to each sample (10 mL) to remove residual H_2O_2 . Catalase was added only to samples taken for chromatographic and COD, TOC analyzes. No catalase was added to the reaction mixture after the UV/ H_2O_2 oxidation process, which was directed to the membrane module, as it could disturb the nanofiltration process. Hence, the mixture could contain some concentration of unreacted H_2O_2 .

The ozonation processes were performed using carried out in a 20-L bubble column connected to the ozone generator (TOGC8X Degremont Technologies LTD, Dübendorf, Switzerland). The details of the ozonating system are given elsewhere [57]. The ozone dose was 60 mg O_3 /min/L of reaction solution. The O_3 concentration in outlet flux was measured by ozone analyzer BTM 964 OG. The ozonation processes were performed in room temperature. The samples were taken after 5, 10, 20, 40, and 60 min and analyzed in liquid chromatography and UPLC/MS. Next, 100 μL of Na_2SO_3 was added to each sample (5 mL) to remove residual ozone. The mixtures after the ozonation process, which were then directed to the nanofiltration, were previously blown with oxygen for 30 min in order to remove residual ozone and left for 1 h at low temperature.

The process of NF was carried out using the cross-flow method at a constant liquid flow within the system being 2 L/min at 30 °C. The tests were carried out at a pressure of 1.0 MPa. The volume of solution was 3 L, the solution was concentrated to a volume of 1.5 L (1/2). The flat sheet NF membrane HL (GE Osmonics, USA) with an area of about 314 cm² was selected for the tests.

Experimental results are expressed in terms of the retention by the membrane of the compounds used in the present work. Retention is determined by employing the substance concentration in the permeate with reference to a representative value of substance concentration in the retentate solution, as follows:

$$R = \left(1 - \frac{C_P}{C_R}\right) \cdot 100\%, \quad (1)$$

where C_P and C_R are the concentrations of a specific compound in the permeate and retentate, respectively.

The samples were collected for analysis at four fixed measuring points: at the beginning of the process (zero degree concentration), after obtaining 0.5 L of filtrate from 3 L of the initial bath (concentration degree 1/6), and analogously—after obtaining 1 L of filtrate (concentration degree 1/3) and 1.5 L of filtrate (concentration degree 1/2). This parameter was chosen to be more representative than the nanofiltration time, which depends on the membrane parameters and the filtered mixture properties.

2.3. Analytical Methods

The oxidation progress was monitored by determination of TRC concentration and was performed on the Agilent 1290 Infinity rapid resolution liquid chromatography system (RRLC) coupled to an Agilent 6460 triple quadrupole mass spectrometer. The liquid chromatographic system was equipped with a binary pump, thermostated column compartment, auto-sampler, and ultraviolet diode-array detector. Chromatographic separation was achieved on a Poroshell 120 Stable Bond SB-C18 column (2.7 µm, 150 mm × 3 mm) (Agilent Technologies, Santa Clara, CA USA). The column temperature was maintained at 40 °C, and the injection volume was 20 µL. The mobile phase (delivered at 0.9 mL/min) consisted of solvent A, 0.1% (*v/v*) formic acid in water, and solvent B, methanol. The quantitative assessment was made using a nine-point calibration curve ranging from 1 to 250 µg/mL for each compound.

The products of TRC degradation were identified with the use of ultra-performance liquid chromatography system ACQUITY (Waters, Milford, MA, USA) coupled with high resolution mass spectrometer SYNAPT G2 (Waters, Milford, MA, USA). The electrospray ionization method (positive mode, ESI+) was used throughout the study. The UPLC BEH Shield RP18 column (2.1 mm × 100 mm × 1.7 µm) (Waters, Milford, MA, USA) was applied for chromatographic separation, which was conducted under isocratic conditions (85% water/15% acetonitrile; both eluents acidified with formic acid at the concentration of 0.1% *v/v*). Quantification of $[M + H]^+$ peak areas was performed with the use of Target Lynx software (Waters, Milford, MA, USA).

An attempt to identify low-molecular products was made using an Agilent Technologies 7890A GC gas spectrometer with a 5975C mass detector (Agilent Technologies). The detailed parameters of this method are presented in the Supplementary Materials (Table S1). Before this analysis, the samples were subjected to solid phase extraction with the use of Strata[®]SDBL columns (Phenomenex, Aschaffenburg, Germany).

Chemical oxygen demand (COD) and total organic carbon (TOC) were analyzed using the HACH LANGE cuvette tests. The cuvette tests were annealed at 100 or 148 °C in an LT 200 HACH LANGE oven (HACH LANGE Sp. z o.o., Wrocław, Poland). The measurement of COD and TOC concentration was based on the color change of the annealed samples using the DR 3900 HACH LANGE spectrophotometer (HACH LANGE Sp. z o.o., Wrocław, Poland).

The analyses of the surface composition of membranes were carried out with the use of the AXIS Ultra photoelectron spectrometer by Kratos Analytical Ltd. (Manchester, U.K.). The source of photoelectron emission from the sample surface (down to approx. 5 nm deep) was X radiation generated by the Al anode with monochromator ($K\alpha$ line with an energy of 1486.6 eV). All XPS spectra were made using a charge neutralizer due to the insulating nature of the sample material. The analysis was performed on from 3 to 6 fields with an area of $700 \times 300 \mu\text{m}$ each, on the surface of each sample.

The SEM microscopic examination was performed on a TESCAN VEGA3 scanning electron microscope (Brno – Kohoutovice, Czech Republic). A magnification of $20,000\times$ was used to study the surface topography of membranes. The acute toxicity test was performed with *Daphnia magna* Straus. The neonates hatched from dormant eggs of *D. magna* Straus according to the International Standard Procedure ISO 6341. The *D. magna* neonates were cached between 72–76 h of ephippia incubation. The daphnia used for the toxicity test were younger than 24 h. The toxicity results of the samples have been read after 48 h of crustaceans exposure to the toxicant.

3. Results and Discussion

3.1. Ozonation and AOPs—Product Identification

The several previously described oxidation products (OPs) of TRC previously described in the literature [27,54,58] were identified. Based on the results of the samples taken after different times of ozonation and UV/ H_2O_2 oxidation, the dynamics of individual OPs formation was established (Figure 2). The OPs of TRC identified during the ozonation and UV/ H_2O_2 oxidation processes are presented in Table 2.

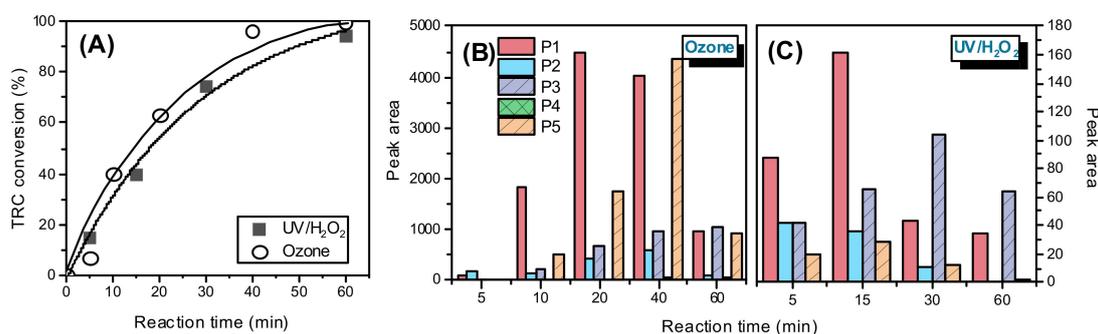


Figure 2. (A) Dependence of TRC conversion on oxidation time (for ozone and UV/ H_2O_2); (B) Peak area of the identified products during ozonation; (C) Peak area of the identified products during UV/ H_2O_2 treatment.

Table 2. Formulae, retention times and m/z values of tetracycline hydrochloride (TRC) and its oxidation products (OPs).

Molecule	Retention Time, min	Formula	Experimental m/z , [M + H] ⁺	Theoretical m/z , [M + H] ⁺	Error ($\Delta m/z$)
TRC	2.3	$\text{C}_{22}\text{H}_{24}\text{N}_2\text{O}_8$	445.1609	445.1605	+0.0004
P1	1.9	$\text{C}_{22}\text{H}_{24}\text{N}_2\text{O}_9$	461.1540	461.1555	−0.0015
P2	3.6	$\text{C}_{22}\text{H}_{24}\text{N}_2\text{O}_9$	461.1540	461.1555	−0.0015
P3	1.6	$\text{C}_{22}\text{H}_{24}\text{N}_2\text{O}_{10}$	477.1526	477.1504	+0.0022
P4	2.9	$\text{C}_{22}\text{H}_{24}\text{N}_2\text{O}_{12}$	509.1401	509.1402	−0.0001
P5	2.6	$\text{C}_{22}\text{H}_{22}\text{N}_2\text{O}_9$	459.1394	459.1398	−0.0004

The identified TRC OPs can be found after both oxidation processes (ozonation and UV/ H_2O_2 oxidation) (Figure 2A). It was observed that the peak areas of OPs marked as P1–P5 were relatively higher for the ozonation process (Figure 2B) compared to the values obtained for the oxidation with UV/ H_2O_2 (Figure 2C). The P1 and P2 molecules represented the isomers of hydroxylated TRC, whereas P3 corresponded to one of the isomers resulting from double hydroxylation (Figure 3). The presence of P4 and P5 could

be attributed to ring opening and dehydrogenation of hydroxylated TRC [27,54], as shown in Figure 3. The experimental mass spectra are presented in the Supplementary Materials (Figures S1–S6).

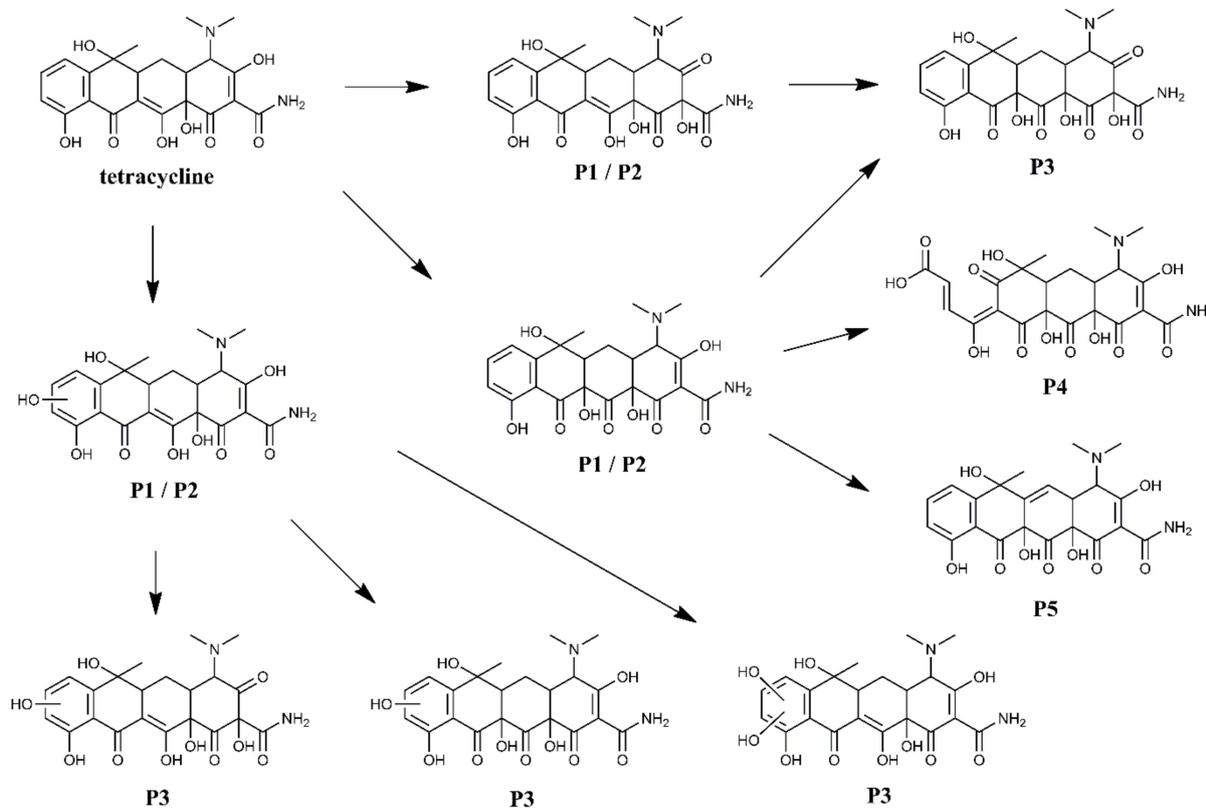


Figure 3. Suggested TRC degradation pathway [27,54,58] including the products identified in the current study.

The tested TRC solutions were acidic (pH = 3.5). Based on our studies (which are not shown here), it was found that increasing the pH or adding H₂O₂ to the TRC solution did not accelerate the TRC decomposition. It is assumed that the ozonation process to a large extent proceeded through the direct ozonolysis mechanism, not excluding the radical mechanism.

On the basis of the obtained results, reaction mixtures (3 L) subjected to oxidation at different times were prepared and then subjected to nanofiltration. The nanofiltration process provided additional information on the reaction mechanism. Therefore, a further discussion is included in the section on nanofiltration.

3.2. Nanofiltration

Figure 4A,B shows the TRC retention coefficients for different concentration ratios (at the process beginning, after collecting 0.5, 1.0, and 1.5 L of filtrate). It was observed that the TRC solution oxidized with UV/H₂O₂ for 5–15 min showed a slightly lower retention coefficient compared to other results. There may be several reasons for this phenomenon: (1) the oxidation products may cause a decrease in the retention coefficients of unreacted starting compound in the reaction mixture [59]. Oxidation products may affect the change in the charge on the membrane surface and its hydrophilicity; (2) residual H₂O₂ can affect the surface properties of the membrane. Our research has shown that some oxidation products can compete with TRC for membrane active centers [59]. We also cannot exclude the influence of H₂O₂. It was not possible to remove residual H₂O₂ from the system without introducing a substance into the solution that could interfere with the nanofiltration process. Strong oxidants such as chlorine or ozone can destroy the

structure of the polyamide, which is the top layer of the composite membrane [60]. Ozone is an unstable substance and it is easy to remove from the system without introducing additional substances into the system. Unfortunately, H_2O_2 is relatively stable and only the introduction of a strong reducer can break it down. The addition of catalase to the mixture could affect the filtration parameters. H_2O_2 is a weak oxidant, its concentration in the reaction mixture was low, hence the tests were performed in the presence of residual H_2O_2 .

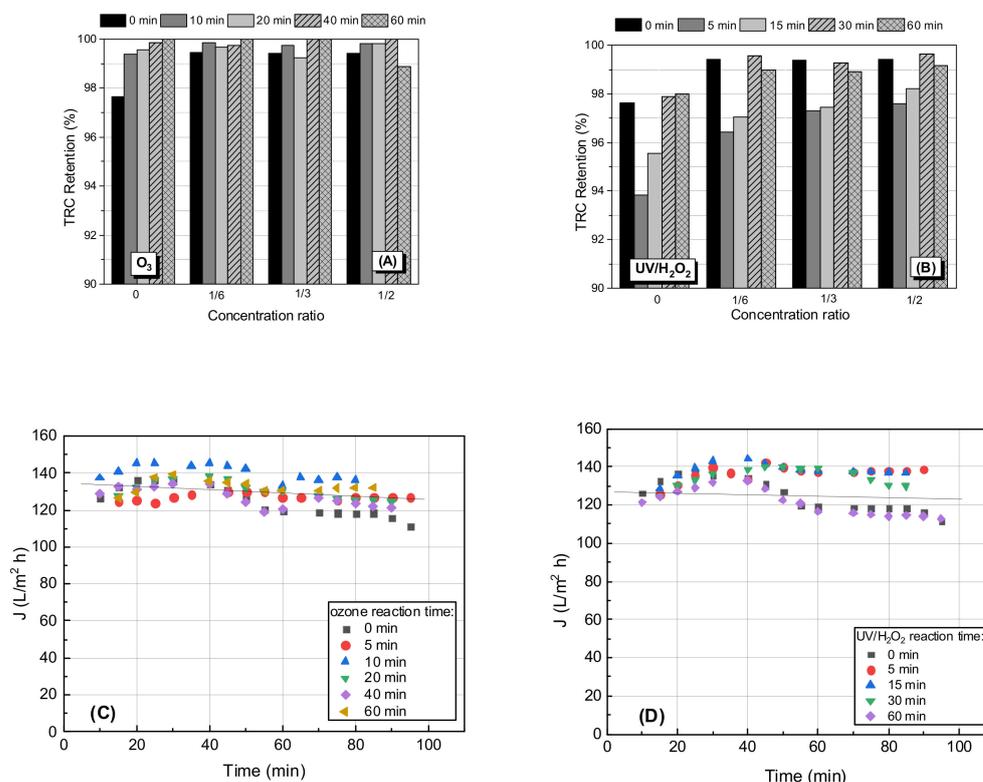


Figure 4. (A) TRC retention for different reaction time of ozonation; (B) TRC retention for different reaction time of UV/ H_2O_2 ; (C) filtrate flux (J) for nanofiltration of ozonated TRC solution; (D) filtrate flux for nanofiltration of UV/ H_2O_2 —oxidized TRC solution; membrane HL; pressure 1.0 MPa; temp. 30 °C.

Figure 4C,D show the filtrate flux (J) for the TRC solution after different oxidation times. The oxidation process did not significantly affect the efficiency of the process, although it appears that the short oxidation time has a positive effect on the efficiency of the filtration process.

Appropriate studies were performed to verify the effects of H_2O_2 and ozone on the membranes stability. Several commercial membranes have been exposed to ozone (ozone-saturated water, exposure to ozone depletion) and H_2O_2 (24–48 h)—studies not shown in this article. Membranes conditioned in this way were used for nanofiltration of various pharmaceuticals. No significant differences were observed in the retention of these compounds.

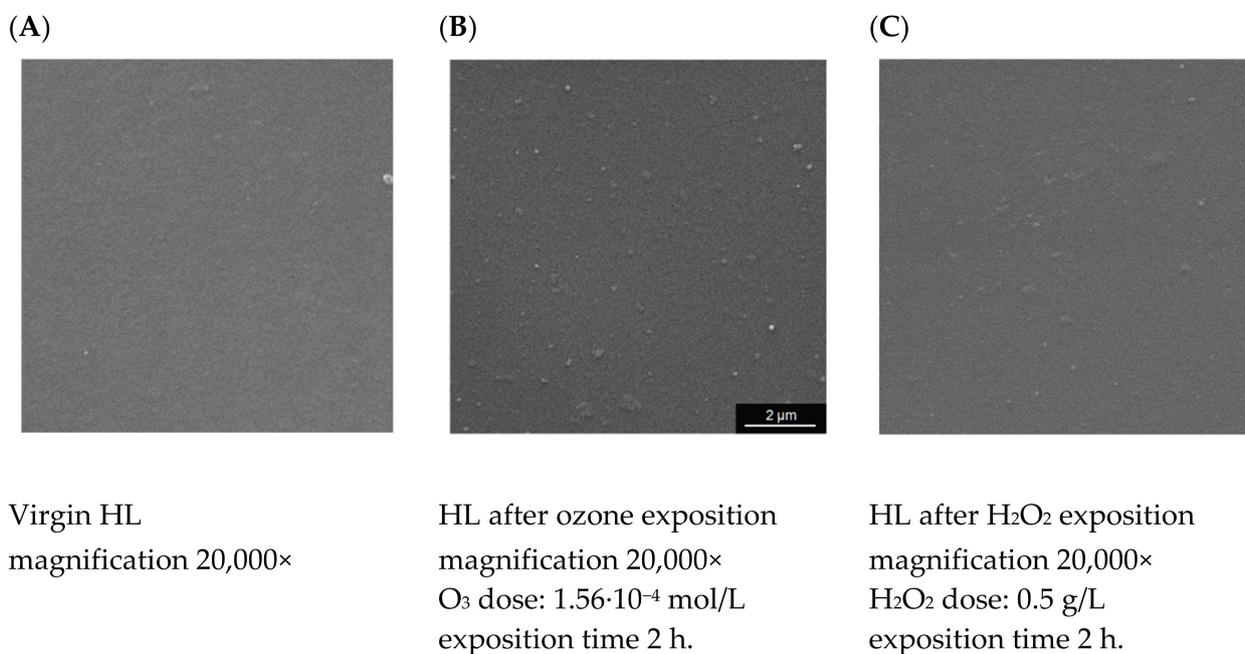
Nevertheless, it has been shown that exposure to oxidizing agents has a destructive effect on the structure of polyamide. There was approximately 1% decrease in nitrogen relative content in the sample after contact of the membrane with hydrogen peroxide and ozone. The cause of polyamide degradation may be the cleavage of amide bonds followed by nitrogen flushing (e.g., in the form of NH_4^+ ammonium residues). The increase in the value of the O/N ratio as a result of exposure to ozone and H_2O_2 may indicate a decrease in the cross-linking of the surface layer of the membrane. It was found (Table 3) that the relative share of the component assigned to the C-N and C-O bonds decreased after exposure of the membrane to both oxidizing agents. This phenomenon may be confirmed

by the previously stated fact of membrane degradation by breaking the C-N bonds and the connections between individual polyamide chains in the polymer network.

Table 3. Quantitative analysis of membrane components of the new membrane HL (GE Osmionics) and after ozone or H₂O₂ exposition, based on the XPS spectroscopy.

	O (% at.)	N (% at.)	C (% at.)	O/N	C-C/C-H	C-O/C-N	C=O
Virgin membrane	16.00	11.29	72.71	1.42	47.8	36.6	15.6
Membrane after O ₃ exposition	19.02	10.37	70.60	1.83	51.1	32.2	16.7
Membrane after H ₂ O ₂ exposition	16.29	10.09	73.61	1.61	54.2	32.1	13.7

In Figure 5, SEM pictures of the pure HL membrane and after exposure to oxidants: ozone and H₂O₂ are presented. There were no significant differences between the membranes. In the case of ozone exposure, increased porosity and loosening of the membrane surface structure can be observed (Figure 5B), although the changes were slight.



Virgin HL
magnification 20,000×

HL after ozone exposition
magnification 20,000×
O₃ dose: 1.56·10⁻⁴ mol/L
exposition time 2 h.

HL after H₂O₂ exposition
magnification 20,000×
H₂O₂ dose: 0.5 g/L
exposition time 2 h.

Figure 5. SEM pictures of the HL membrane: (A) virgin HL; (B) HL after ozone exposure; (C) HL after H₂O₂ exposure.

In conclusion, we cannot exclude the influence of H₂O₂ on TRC retention, especially since the negative influence is visible at the beginning of filtration, when the H₂O₂ concentration is at its highest. Rather, the influence of membrane damage should be excluded, because H₂O₂ in this concentration is a very weak oxidant and damage to amide bonds is visible after long exposure. H₂O₂ has not been reported to have a similar effect on the retention rate of other tested compounds [61]. It should be emphasized that the changes are slight (decrease from 97.5% to 94%).

All identified Ops exhibited very high retention values (Figure 6). Furthermore, they retained the structural core of TRC. However, as shown in Figure 7, COD reduction in the obtained filtrates for the UV/H₂O₂ process was clearly lower than for ozonation. Therefore, it cannot be excluded that the lower COD reduction was caused by the presence of low molecular weight Ops. This suggests that the process of TRC oxidation with UV/H₂O₂ might have followed a different path than the ozonation process. The nanofiltration process was carried out for the reaction mixture after 15 min of UV/H₂O₂ oxidation and 40 min of ozonation. In the case of UV/H₂O₂, one main product was recorded, namely P1, with a retention factor similar to that of TRC. In the case of ozonation within 15 min, two

dominant products, namely P1 and P5, appeared. P1 retention was observed to be nearly 100% for this mixture. Due to the complex water matrix, there can be many reasons for this phenomenon. One possible cause may be the formation of hydrogen bonds between P1 molecules and other Ops as a result of their concentration at the membrane surface. Such a phenomenon may result in improved retention of test compounds. At the same time, there was a slight decrease in the retention of the P3 product in relation to the P3 retention obtained for the UV/H₂O₂ mixture. This may be due to a number of reasons, e.g., increased competition from other Ops in greater concentration to the active sites of the membrane.

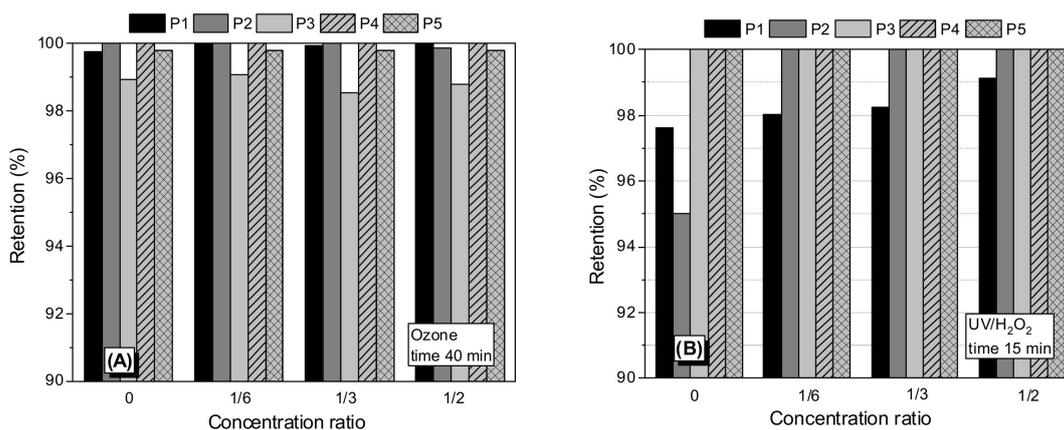


Figure 6. Ops retention (A) for 40 min of ozonation; (B) for 15 min of UV/H₂O₂ oxidation; membrane HL.

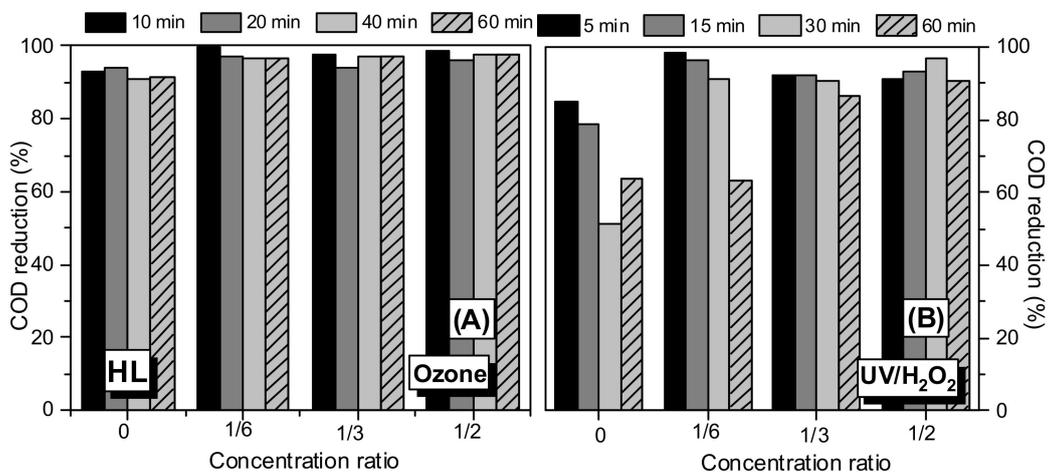


Figure 7. Chemical oxygen demand COD reduction during NF of oxidized TRC solution: (A) after ozonation; (B) after UV/H₂O₂.

3.3. Identification of New TRC Oxidation Product

The filtrate obtained in the process of NF of the oxidized solution with UV/H₂O₂ (time 60 min) was analyzed by gas chromatography with mass spectrometry. Preliminary analysis showed that at least one product (P6) containing only one aromatic ring in its structure (based on the spectral libraries of gas chromatograph) passes into the filtrate. By means of the purchased standard, it was confirmed by GC/MS that the product detected in the filtrate was ethyl 4-ethoxybenzoate (MW = 194 g/mol) (Figure 8).

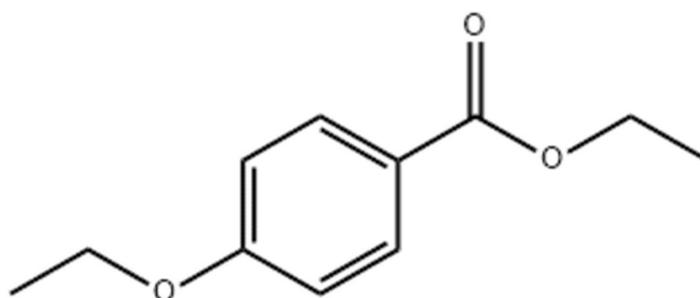


Figure 8. Chemical structure of ethyl 4-ethoxybenzoate (P6).

This is consistent with literature data. Liu [45] analyzed the change in the FTIR spectrum of TRC in the photoelectro-Fenton process. According to the data contained in this work, initially, the spectrum contained three bands (1450 cm^{-1} and 930 cm^{-1} , $750\text{--}800\text{ cm}^{-1}$) corresponding to benzene ring vibrations and benzene ring plane vibrations, respectively. These peaks disappeared after 3 h of oxidation and a new band characteristic of a secondary amine appeared at 1110 cm^{-1} . Meanwhile, the peaks of $750\text{--}800\text{ cm}^{-1}$ and 930 cm^{-1} corresponding to the vibrations of the benzene ring disappeared. These results indicate deamination and destruction of the TRC structure.

The studies did not confirm that the significant decrease in COD (Figure 7B) can be attributed only to the presence of the product P6 in the filtrate. As shown in Figure 9, its retention is approximately 90%. However, the rapid decrease of the retention during filtration suggests a significant influence of adsorption on the separation mechanism of this compound. P6 is a poorly soluble compound of a hydrophobic nature. These types of compounds often have the ability to adsorb to the membrane surface. Adsorption is the result of various interactions between the solute and the membrane. They include electrostatic, hydrophobic and some specific interactions (e.g., hydrogen bonds) [62–64].

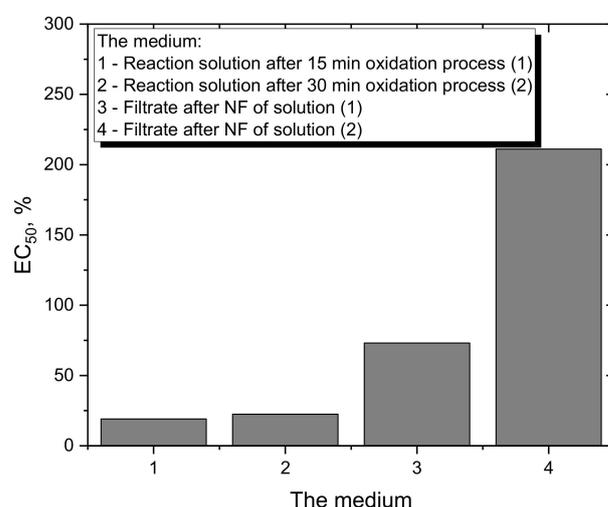


Figure 9. Toxicity tests of solution tetracycline hydrochloride (TRC) and its oxidation products (Ops) during oxidation by UV/H₂O₂ exposition time 48 h.

3.4. Toxicity Tests

The toxicity tests of solutions were performed (Figure 9). Applying bioluminescence test for the inhibition of *Vibrio fischeri* was impossible due to the yellowish color of the TRC solution, therefore the acute toxicity test was performed with neonates hatched from dormant eggs of *Daphnia magna* Straus according to the International Standard Procedure ISO 6341. The 48 h EC₅₀ for the TRC solution before the UV/H₂O₂ reaction was 30.15%.

It was observed that the solution with the products at the 15th minute of the reaction were more toxic than TRC itself. Extending the reaction time has contributed to reducing

the toxicity effect. The use of NF caused the significant removal of toxic oxidation products. Nevertheless, the obtained filtrate after NF of the solution (1) showed a toxic effect at a higher concentration. Due to the fact that the products marked in Table 2, were retained on the membrane, it can be assumed that low molecular weight oxidation products which can be toxic, have entered the filtrate.

López-Peñalver et al. [25] studied the toxicity of three TRCs and their products using a standardized bioluminescence test (DIN/EN/ISO 11348-2) for the inhibition of *V. fischeri* (NRRL B-11177). It was observed that toxicity initially increased and then decreased after 60 min. According to the authors, this was due to the fact that TRC photolysis produces many very stable degradation products that have higher toxicity than that of the parent compound.

3.5. River Water Tests

The tests were carried out in a natural water matrix, water taken from the river and compared with the results obtained for deionized water. In our previous work, the characteristic of natural water collected from the Sulejow Reservoir, situated in central Poland in the middle course of the Pilica River, was presented [65]. The values of main parameters of water from Sulejow Reservoir were similar to the water taken from Dobrznka River (Table 1). In our opinion, relatively higher COD and TOC may be linked to presence in the river water NOM including mainly HA and FA. The river water was pre-ozonized for 15 and 30 min before the standard TRC was dissolved therein. Figure 10 shows the nanofiltration parameters of TRC and its oxidation product—P6, for a different water matrix.

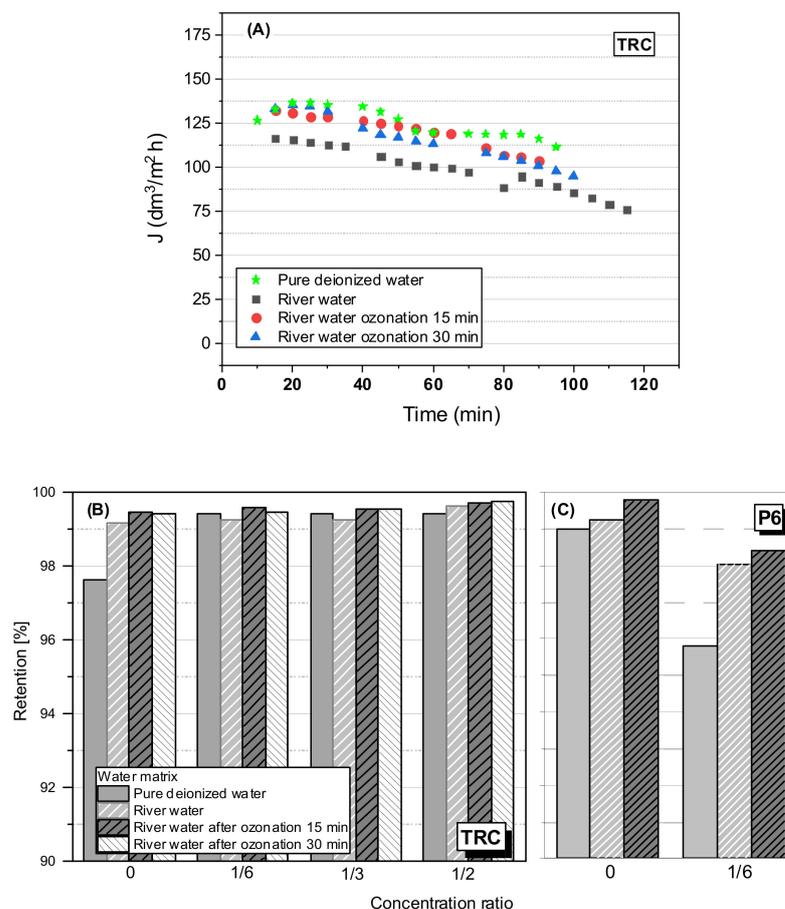


Figure 10. (A) The filtrate flux for a different water matrix with dissolved TRC; (B) TRC retention for different water matrix; (C) P6 retention for different water matrix.

The studies confirmed that the presence of natural organic matter (NOM) reduces the nanofiltration process efficiency, which is consistent with the available literature [56,60] (Figure 10A). Taking into account the tendency of increasing TRC retention with a simultaneous decrease in filtrate flux, it can be assumed that the separation mechanism proceeds by spatial blockade. TRC particles get stuck in the pores of the membrane during the filtration process. The presence of additional organic matter from natural river water increases the blockage of the internal spaces of the membrane. At the same time, the presence of an additional space block in the form of NOM contributes to better retention of pharmaceuticals (Figure 10B) and their OPs on the membrane surface (Figure 10C).

Initial ozonation of the river water contributed to the improvement of filtration parameters. There was a flux increase with a slight increase of the retention ratio. This may be due to the fact that ozonation contributes to the formation of more hydrophilic oxidation products. This may be evidenced by a decrease of the membrane contact angle.

4. Conclusions

Five oxidation products (OPs) have been identified that have retained the structural core of TRC. All identified OPs were effectively retained on the membrane in the nanofiltration process. Process of TRC oxidation with UV/H₂O₂ might follow a different path than the ozonation process. All identified OPs and TRC itself exhibited very high retention values during the NF process. Nevertheless, the lower COD reduction was observed during NF of solution oxidized by UV/H₂O₂. This may be evidence of the formation of low molecular weight OPs that passed through the membrane, which was confirmed by gas chromatography with mass spectrometry. There is possibility to formation of hydrogen bonds between P1 molecules and other OPs as a result of their concentration at the membrane surface. Which may be the reason for the retention improvement during filtration.

One new TRC oxidation product, not reported so far in the literature, was identified—ethyl 4-ethoxybenzoate. The degradation of the stable tetracyclic structure of the TRC is indicated by the presence of a product with one aromatic ring (P6). This proves the possibility of the formation of other low molecular weight degradation products. The confirmation of this may be the low COD reduction during nanofiltration of solutions after H₂O₂/UV oxidation.

The influence of ozone and H₂O₂ on the structure of the HL membrane was investigated. It has been found that H₂O₂ and ozone can damage the membrane surface. Although the changes in the surface components of membrane HL after ozone or H₂O₂ exposition, (based on the XPS spectroscopy) were slight (over 1%). Microscopic analysis did not show any significant differences between the virgin membrane and the one exposed to oxidants. The membrane, under the influence of ozone, was more loosened and rougher. Ozone showed more aggressive oxidizing properties than H₂O₂, although the advantage of this oxidant is its instability. In practice, solutions without residual ozone were directed to the nanofiltration process. Unfortunately, H₂O₂ is difficult to remove from solution without introducing additional chemicals into the system. Its presence may have an impact on the observed phenomena occurring on the membrane surface in the nanofiltration process. Although no such impact has been demonstrated in this study.

It cannot be ruled out that these low molecular weight products are toxic and that the low COD reduction is connected with residual H₂O₂.

The research results indicate the need to pay attention to the transport of oxidation products in the further process of wastewater treatment. Combining different treatment modules requires a broader analysis not only in terms of process optimization, cost reduction, but also the safety of the society. Further research on integrated processes of advanced oxidation and membrane filtration is recommended in terms of transporting the decomposition products of micropollutants, which may pose a serious threat to the environment.

Supplementary Materials: The following are available online at <https://www.mdpi.com/2073-4441/13/4/555/s1>; Figure S1. ESI+ mass spectrum of tetracycline; Figure S2. ESI+ mass spectrum of degradation product P1; Figure S3. ESI+ mass spectrum of degradation product P2; Figure S4. ESI+ mass spectrum of degradation product P3; Figure S5. ESI+ mass spectrum of degradation product P4; Figure S6. ESI+ mass spectrum of degradation product P5; Table S1. GC-MS method parameters of identification of low-molecular products.

Author Contributions: R.Ż. development the research concept, setting the work schedule and supervising the team's work, performing some of the experiments, data analysis and description, preparing the publication, manuscript correction in the review process, S.L. cooperation in writing the manuscript, T.B. chromatographic analysis of oxidation products of TRC; M.O.-K. performing of real water analysis, cooperation in manuscript correction in the review process; M.F. chromatographic analysis, toxicity tests performing; Z.M. gas-chromatography analysis; J.B. XPS analysis. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by Polish National Science Center grant number UMO-2016/21/B/ST8/00982.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Eggen, T.; Vogelsang, C. Occurrence and fate of pharmaceuticals and personal care products in wastewater. *Compr. Anal. Chem.* **2015**, *67*, 245–294.
2. Verlicchi, P.; Zambello, E.; Al Aukidy, M. Removal of pharmaceuticals by conventional wastewater treatment plants. *Compr. Anal. Chem.* **2013**, *62*, 231–286.
3. Liu, P.; Zhang, H.; Feng, Y.; Yang, F.; Zhang, J. Removal of trace antibiotics from wastewater: A systematic study of nanofiltration combined with ozone-based advanced oxidation processes. *Chem. Eng. J.* **2014**, *240*, 211–220. [[CrossRef](#)]
4. Ganiyu, S.O.; van Hullebusch, E.D.; Cretin, M.; Esposito, G.; Oturan, M.A. Coupling of membrane filtration and advanced oxidation processes for removal of pharmaceutical residues: A critical review. *Sep. Purif. Technol.* **2015**, *156*, 891–914. [[CrossRef](#)]
5. Wang, L.; Ben, W.; Li, Y.; Liu, C.; Qiang, Z. Behavior of tetracycline and macroline antibiotics in activated sludge process and their subsequent removal during sludge reduction by ozone. *Chemosphere* **2018**, *206*, 184–191. [[CrossRef](#)]
6. Xu, L.; Zhang, H.; Xiong, P.; Zhu, Q.; Liao, C.; Jiang, G. Occurrence, fate, and risk assessment of typical tetracycline antibiotics in the aquatic environment: A review. *Sci. Total Environ.* **2021**, *753*, 141975. [[CrossRef](#)] [[PubMed](#)]
7. Van Boeckel, T.P.; Brower, C.; Gilbert, M.; Grenfell, B.T.; Levin, S.A.; Robinson, T.P.; Teillant, A.; Laxminarayan, R. Global trends in antimicrobial use in food animals. *Proc. Natl. Acad. Sci. USA* **2015**, *112*, 5649–5654. [[CrossRef](#)] [[PubMed](#)]
8. Alhaji, N.B.; Isola, T.O. Antimicrobial usage by pastoralists in food animals in Northcentral Nigeria: The associated socio-cultural drivers for antimicrobials misuse and public health implications. *One Health* **2018**, *6*, 41–47. [[CrossRef](#)] [[PubMed](#)]
9. Dai, Y.; Liu, M.; Li, J.; Yang, S.; Sun, Y.; Sun, Q.; Wang, W.; Lu, L.; Zhang, K.; Xu, J.; et al. A review on pollution situation and treatment methods of tetracycline in groundwater. *Sep. Sci. Technol.* **2020**, *55*, 1005–1021. [[CrossRef](#)]
10. Babin, M.; Boleas, S.; Tarazona, J. In vitro toxicity of antimicrobials on RTG-2 and RTL-W1 fish cell lines. *Environ. Toxicol. Pharmacol.* **2005**, *20*, 125–134. [[CrossRef](#)]
11. Zhang, Y.; Wang, X.; Yin, X.; Shi, M.; Dahlgren, R.A.; Wang, H. Toxicity assessment of combined fluoroquinolone and tetracycline exposure in zebrafish (*Danio rerio*). *Environ. Toxicol.* **2016**, *31*, 736–750. [[CrossRef](#)]
12. Ding, L.; Zang, L.; Zhang, Y.; Zhang, Y.; Wang, X.; Ai, W.; Ding, N.; Wang, H. Joint toxicity of fluoroquinolone and tetracycline antibiotics to zebrafish (*Danio rerio*) based on biochemical biomarkers and histopathological observation. *J. Toxicol. Sci.* **2017**, *42*, 267–280. [[CrossRef](#)] [[PubMed](#)]
13. Ye, J.; Du, Y.; Wang, L.; Qian, J.; Chen, J.; Wu, Q.; Hu, X. Toxin Release of Cyanobacterium *Microcystis aeruginosa* after Exposure to Typical Tetracycline Antibiotic Contaminants. *Toxins* **2017**, *9*, 53. [[CrossRef](#)] [[PubMed](#)]
14. Xu, D.; Xiao, Y.; Pan, H.; Mei, Y. Toxic effects of tetracycline and its degradation products on freshwater green algae. *Ecotoxicol. Environ. Saf.* **2019**, *174*, 43–47. [[CrossRef](#)]
15. Zhang, M.; Yuan, L.; Li, Z.; Zhang, H.; Sheng, G. Tetracycline exposure shifted microbial communities and enriched antibiotic resistance genes in the aerobic granular sludge. *Environ. Internat.* **2019**, *130*, 104902. [[CrossRef](#)]
16. Jia, J.; Cheng, M.; Xue, X.; Guan, Y.; Wang, Z. Characterization of tetracycline effects on microbial community, antibiotic resistance genes and antibiotic resistance of *Aeromonas* spp. in gut of goldfish *Carassius auratus* Linnaeus. *Ecotoxicol. Environ. Saf.* **2020**, *191*, 110182. [[CrossRef](#)] [[PubMed](#)]

17. Zainab, S.M.; Junaid, M.; Xu, N.; Malik, R.N. Antibiotics and antibiotic resistant genes (ARGs) in groundwater: A global review on dissemination, sources, interactions, environmental and human health risks. *Water Res.* **2020**, *187*, 116455. [[CrossRef](#)] [[PubMed](#)]
18. Klavarioti, M.; Mantzavinos, D.; Kassinos, D. Removal of residual pharmaceuticals from aqueous systems by advanced oxidation processes. *Environ. Int.* **2009**, *35*, 402–417. [[CrossRef](#)] [[PubMed](#)]
19. Yu, X.; Lin, X.; Feng, W.; Li, W. Enhanced catalytic performance of a bio-templated TiO₂ UV-Fenton system on the degradation of tetracycline. *Appl. Surf. Sci.* **2019**, *465*, 223–231. [[CrossRef](#)]
20. Wang, Y.; Zhang, H.; Zhang, J.; Lu, C.; Huang, Q.; Wu, J.; Liu, F. Degradation of tetracycline in aqueous media by ozonation in an internal loop-lift reactor. *J. Hazard. Mater.* **2011**, *192*, 35–43. [[CrossRef](#)] [[PubMed](#)]
21. Chen, Y.Y.; Ma, Y.L.; Yang, J.; Wang, L.Q.; Lv, J.M.; Ren, C.J. Aqueous tetracycline degradation by H₂O₂ alone: Removal and transformation pathway. *Chem. Eng. J.* **2017**, *307*, 15–23. [[CrossRef](#)]
22. Zhi, D.; Wang, J.; Zhou, Y.; Luo, Z.; Sun, Y.; Wan, Z.; Luo, L.; Tsang, D.C.W.; Dionysiou, D.D. Development of ozonation and reactive electrochemical membrane coupled process: Enhanced tetracycline mineralization and toxicity reduction. *Chem. Eng. J.* **2020**, *383*, 123149. [[CrossRef](#)]
23. Dalmázio, I.; Almeida, M.O.; Augusti, R.; Alves, T.M.A. Monitoring the degradation of tetracycline by ozone in aqueous medium via atmospheric pressure ionization mass spectrometry. *J. Am. Soc. Mass Spectrom.* **2007**, *18*, 679–687. [[CrossRef](#)]
24. Li, D.; Niu, Z.; Bao, R.; Wan, D.; Liu, Y.; Zhang, L. Degradation of tetracycline with Fenton reagent in aqueous solution. *Chin. J. Environ. Eng.* **2017**, *11*, 2227–2232.
25. López-Peñalver, J.J.; Sánchez-Polo, M.; Gómez-Pacheco, C.V.; Rivera-Utrilla, J. Photodegradation of tetracyclines in aqueous solution by using UV and UV/H₂O₂ oxidation processes. *J. Chem. Technol. Biotechnol.* **2019**, *85*, 1325–1333. [[CrossRef](#)]
26. Rivera-Utrilla, J.; Ocampo-Pérez, R.; Sánchez-Polo, M.; López-Peñalver, J.J.; Gómez-Pacheco, C.V. Removal of tetracyclines from water by adsorption/bioadsorption and advanced oxidation processes. A short review. *Curr. Org. Chem.* **2018**, *22*, 1005–1021. [[CrossRef](#)]
27. Khan, M.H.; Bae, H.; Jung, J.Y. Tetracycline degradation by ozonation in the aqueous phase: Proposed degradation intermediates and pathway. *J. Hazard. Mater.* **2010**, *181*, 659–665. [[CrossRef](#)] [[PubMed](#)]
28. Fatta-Kassinos, D.; Vasquez, M.I.; Kümmerer, K. Transformation products of pharmaceuticals in surface waters and wastewater formed during photolysis and advanced oxidation processes—Degradation, elucidation of byproducts and assessment of their biological potency. *Chemosphere* **2011**, *85*, 693–709. [[CrossRef](#)] [[PubMed](#)]
29. Miao, H.F.; Cao, M.; Xu, D.Y.; Ren, H.Y.; Zhao, M.X.; Huang, Z.X.; Ruan, W.Q. Degradation of phenazone in aqueous solution with ozone: Influencing factors and degradation pathways. *Chemosphere* **2015**, *119*, 326–333. [[CrossRef](#)]
30. Benitez, F.J.; Acero, J.L.; Real, F.J.; Roldán, G. Ozonation of pharmaceutical compounds: Rate constants and elimination in various water matrices. *Chemosphere* **2009**, *77*, 53–59. [[CrossRef](#)] [[PubMed](#)]
31. Michael, I.; Frontistis, Z.; Fatta-Kassinos, D. Removal of pharmaceutically relevant matrices by advanced oxidation processes (AOPS). *Compr. Anal. Chem.* **2013**, *62*, 345–407.
32. Tizaoui, C.; Grima, N.; Hilal, N. Degradation of the antimicrobial triclocarban (TCC) with ozone. *Chem. Eng. Process. Proc. Intens.* **2011**, *50*, 637–643. [[CrossRef](#)]
33. Rodríguez, E.M.; Márquez, G.; León, E.A.; Álvarez, P.M.; Amat, A.M.; Beltrán, F.J. Mechanism considerations for photocatalytic oxidation, ozonation and photocatalytic ozonation of some pharmaceutical compounds in water. *J. Environ. Manag.* **2013**, *127*, 114–124. [[CrossRef](#)]
34. Antoniou, M.G.; Hey, G.; Vega, S.R.; Spiliotopoulou, A.; Fick, J.; Tysklind, M.; la Jansen, J.C.; Andersen, H.R. Required ozone doses for removing pharmaceuticals from wastewater effluents. *Sci. Total Environ.* **2013**, *456–457*, 42–49. [[CrossRef](#)]
35. Gao, S.; Zhao, Z.; Xu, Y.; Tian, J.; Qi, H.; Lin, W.; Cui, F. Oxidation of sulfamethoxazole (SMX) by chlorine, ozone and permanganate—A comparative study. *J. Hazard. Mater.* **2014**, *274*, 258–269. [[CrossRef](#)]
36. Quero-Pastor, M.; Valenzuela, A.; Quiroga, J.M.; Acevedo, A. Degradation of drugs in water with advanced oxidation processes and ozone. *J. Environ. Manag.* **2014**, *137*, 197–203. [[CrossRef](#)]
37. Kim, S.; Chu, K.H.; Al-Hamadani, Y.A.J.; Park, C.M.; Jang, M.; Kim, D.H.; Yu, M.; Heo, J.; Yoon, Y. Removal of contaminants of emerging concern by membranes in water and wastewater: A review. *Chem. Eng. J.* **2018**, *335*, 896–914. [[CrossRef](#)]
38. Nasirabadi, P.S.; Saljoughi, E.; Mousavi, S.M. Membrane processes used for removal of pharmaceuticals, hormones, endocrine disruptors and their metabolites from wastewaters: A review. *Desalin. Water Treat.* **2016**, *57*, 24146–24175. [[CrossRef](#)]
39. Taheran, M.; Brar, S.K.; Verma, M.; Surampalli, R.Y.; Zhang, T.C.; Valero, J.R. Membrane processes for removal of pharmaceutically active compounds (PhACs) from water and wastewater. *Sci. Total Environ.* **2016**, *547*, 60–77. [[CrossRef](#)] [[PubMed](#)]
40. Kim, S.D.; Jaeweon, C.; Kim, I.S.; Vanderford, B.J.; Snyder, S.A. Occurrence and removal of pharmaceuticals and endocrine disruptors in South Korean surface, drinking, and waste waters. *Water Res.* **2007**, *41*, 1013–1021. [[CrossRef](#)] [[PubMed](#)]
41. Belkacem, M.; Bensadok, K.; Refes, A.; Charvier, P.M.; Nezzal, G. Water produce for pharmaceutical industry: Role of reverse osmosis stage. *Desalination* **2008**, *221*, 298–302. [[CrossRef](#)]
42. Radjenović, J.; Petrović, M.; Ventura, F.; Barceló, D. Rejection of pharmaceuticals in nanofiltration and reverse osmosis membrane drinking water treatment. *Water Res.* **2008**, *42*, 3601–3610. [[CrossRef](#)]
43. Al-Rifai, J.H.; Khabbaz, H.; Schäfer, A.I. Removal of pharmaceuticals and endocrine disrupting compounds in a water recycling process using reverse osmosis systems. *Sep. Purif. Technol.* **2011**, *77*, 60–67. [[CrossRef](#)]

44. Abdelmelek, S.B.; Greaves, J.; Ishida, K.P.; Cooper, W.J.; Song, W. Removal of pharmaceutical and personal care products from reverse osmosis retentate using advanced oxidation processes. *Environ. Sci. Technol.* **2011**, *45*, 3665–3671. [[CrossRef](#)] [[PubMed](#)]
45. Real, F.J.; Benitez, F.J.; Acero, J.L.; Roldan, G. Combined chemical oxidation and membrane filtration techniques applied to the removal of some selected pharmaceuticals from water systems. *J. Environ. Sci. Health Part A* **2012**, *42*, 522–533. [[CrossRef](#)]
46. Justo, A.; Gonz ales, O.; Aceña, J.; P erez, S.; Barcel o, D.; Sans, C.; Esplugas, S. Pharmaceuticals and organic pollution mitigation in reclamation osmosis brines by UV/H₂O₂ and ozone. *J. Hazard. Mater.* **2013**, *263*, 268–274. [[CrossRef](#)]
47. Miralles-Cuevas, S.; Oller, I.; P erez, J.A.; Malato, S. Removal of pharmaceuticals from MWTP effluent by nanofiltration and solar photo-Fenton using two different iron complexes at neutral pH. *Water Res.* **2014**, *64*, 23–31. [[CrossRef](#)] [[PubMed](#)]
48. Iglesias, O.; Rivero, M.J.; Utriaga, A.M.; Ortiz, I. Membrane-based photocatalytic systems for process intensification. *Chem. Eng. J.* **2016**, *305*, 136–148. [[CrossRef](#)]
49. Kim, J.O.; Jung, J.T.; Jung, J.T.; Yeom, I.T.; Aoh, G.H. Effect of fouling reduction by ozone backwashing in a microfiltration system with advanced new membrane material. *Desalination* **2007**, *202*, 361–368. [[CrossRef](#)]
50. Kim, J.; Davies, S.H.R.; Baumann, M.J.; Tarabara, V.V.; Masten, S.J. Effect of ozone dosage and hydrodynamic conditions on the permeate flux on a hybrid ozonation-ceramic ultrafiltration system treating natural waters. *J. Membr. Sci.* **2008**, *311*, 165–172. [[CrossRef](#)]
51. Nguyen, S.T.; Roddick, F.A. Effects of ozonation and biological activated carbon filtration on membrane fouling in ultrafiltration of an activated sludge effluent. *J. Membr. Sci.* **2010**, *363*, 271–277. [[CrossRef](#)]
52. Zhu, H.; Wen, X.; Huang, X. Membrane organic fouling and the effect of pre-ozonation microfiltration of secondary effluent organic matter. *J. Membr. Sci.* **2010**, *352*, 213–221. [[CrossRef](#)]
53. Dong, H.; Qiang, Z.; Lian, J.; Qu, J. Degradation of nitro-based pharmaceuticals by UV photolysis: Kinetics and simultaneous reduction on halonitromethanes formation potential. *Water Res.* **2017**, *119*, 83–90. [[CrossRef](#)] [[PubMed](#)]
54. Li, S.; Hu, J. Photolytic and photocatalytic degradation of tetracycline: Effect of humic acid on degradation kinetics and mechanisms. *J. Hazard. Mater.* **2016**, *318*, 134–144. [[CrossRef](#)]
55. Liu, S.; Zhao, X.R.; Sun, H.; Li, R.; Fang, Y.; Huang, Y. The degradation of tetracycline in a photo-electro-Fenton system. *Chem. Eng. J.* **2014**, *231*, 441–448. [[CrossRef](#)]
56. Żyła, R.; Sójka-Ledakowicz, J.; Michalska, K.; Kos, L.; Ledakowicz, S. Effect of UV/H₂O₂ oxidation on fouling in textile wastewater nanofiltration. *Fibres Text. East. Eur.* **2012**, *20*, 99–104.
57. Bilińska, L.; Blus, K.; Gmurek, M.; Żyła, R.; Ledakowicz, S. Brine recycling from industrial textile wastewater treated by ozone. by-products accumulation. Part 2: Scaling-up. *Water* **2019**, *11*, 233. [[CrossRef](#)]
58. Maroga Mboula, V.; Héquet, V.; Gru, Y.; Colin, R.; Andr es, Y. Assessment of the efficiency of photocatalysis on tetracycline biodegradation. *J. Hazard. Mater.* **2012**, *209–210*, 355–364. [[CrossRef](#)] [[PubMed](#)]
59. Żyła, R.; Milala, R.; Kamińska, I.; Kudzin, M.; Gmurek, M.; Ledakowicz, S. Impact of advanced oxidation products on nanofiltration efficiency. *Water* **2019**, *11*, 541. [[CrossRef](#)]
60. Park, M.; Anumol, T.; Simon, J.; Zraick, F.; Snyder, S.A. Pre-ozonation for high recovery of nanofiltration (NF) membrane system: Membrane fouling reduction and trace organic compound attenuation. *J. Membr. Sci.* **2017**, *523*, 255–263. [[CrossRef](#)]
61. Żyła, R.; Boruta, T.; Gmurek, M.; Milala, R.; Ledakowicz, S. Integration of advanced oxidation and membrane filtration for removal of micropollutants of emerging concern. *Process. Saf. Environ. Prot.* **2019**, *130*, 67–76. [[CrossRef](#)]
62. Comerton, A.M.; Andrews, R.C.; Bagley, D.M.; Yang, P. Membrane adsorption of endocrine disrupting compounds and pharmaceutically active compounds. *J. Membr. Sci.* **2007**, *303*, 267–277. [[CrossRef](#)]
63. Schäfer, A.I.; Akanyeti, I.; Semião, A.J.C. Micropollutant sorption to membrane polymers: A review of mechanisms for estrogens. *Adv. Colloid Interface Sci.* **2011**, *164*, 100–117. [[CrossRef](#)] [[PubMed](#)]
64. Liu, Y.-L.; Wang, X.-M.; Yang, H.-W.; Xie, Y.F. Quantifying the influence of solute-membrane interactions on adsorption and rejection of pharmaceuticals by NF/RO membranes. *J. Membr. Sci.* **2018**, *551*, 37–46. [[CrossRef](#)]
65. Gmurek, M.; Olak-Kucharczyk, M.; Ledakowicz, S. Influence of dissolved organic matter in natural and simulated water on the photochemical decomposition of butylparaben. *J. Environ. Health Sci. Eng.* **2015**, *13*, 28. [[CrossRef](#)]