

AperTO - Archivio Istituzionale Open Access dell'Università di Torino

THE ROLE OF HUMIC AND FULVIC ACIDS IN THE PHOTOTRASFORMATION OF PHENOLIC COMPOUNDS IN SEAWATER

This is the author's manuscript

Original Citation:

Availability:

This version is available <http://hdl.handle.net/2318/158130> since 2016-10-10T11:10:37Z

Published version:

DOI:10.1016/j.scitotenv.2014.05.145

Terms of use:

Open Access

Anyone can freely access the full text of works made available as "Open Access". Works made available under a Creative Commons license can be used according to the terms and conditions of said license. Use of all other works requires consent of the right holder (author or publisher) if not exempted from copyright protection by the applicable law.

(Article begins on next page)



UNIVERSITÀ DEGLI STUDI DI TORINO

This Accepted Author Manuscript (AAM) is copyrighted and published by Elsevier. It is posted here by agreement between Elsevier and the University of Turin. Changes resulting from the publishing process - such as editing, corrections, structural formatting, and other quality control mechanisms - may not be reflected in this version of the text. The definitive version of the text was subsequently published in *[The role of humic and fulvic acids in the phototransformation of phenolic compounds in seawater, Science of the Total Environment 493 (2014) 411–418, and DOI: <http://dx.doi.org/10.1016/j.scitotenv.2014.05.145>]*.

You may download, copy and otherwise use the AAM for non-commercial purposes provided that your license is limited by the following restrictions:

- (1) You may use this AAM for non-commercial purposes only under the terms of the CC-BY-NC-ND license.
- (2) The integrity of the work and identification of the author, copyright owner, and publisher must be preserved in any copy.
- (3) You must attribute this AAM in the following format: Creative Commons BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/deed.en>), [<http://www.sciencedirect.com/science/article/pii/S0048969714008377>]

**THE ROLE OF HUMIC AND FULVIC ACIDS IN THE PHOTOTRASFORMATION OF
PHENOLIC COMPOUNDS IN SEAWATER**

P. Calza*, D. Vione, C. Minero

Dipartimento di Chimica, Università di Torino, via P. Giuria 5, 10125 Torino, Italy

*corresponding author: Dipartimento di Chimica, Università di Torino, via P. Giuria 5, 10125
Torino, Italy; e-mail: paola.calza@unito.it; phone: +390116705268; fax: +390116705242

Keywords: humic acid, fulvic acid, phenol, photodegradation, seawater

Abstract

Humic substances (HS) are known to act as photosensitizers toward the transformation of pollutants in the surface layer of natural waters. This study focused on the role played by HS toward the transformation of xenobiotics in seawater, with the purpose of assessing the prevailing degradation routes. Phenol was chosen as model xenobiotic and its transformation was investigated under simulated sunlight in the presence of terrestrial or marine humic and fulvic acids, in pure water at pH 8, artificial seawater (ASW) or natural seawater (NSW). The following parameters were determined: (1) phenol degradation rate; (2) the variation in HS concentration with irradiation time; (3) the production of transformation products; (4) the influence of iron species on the transformation process.

Faster transformation of phenol was observed with humic acids (HA) compared to fulvic acids (SRFA), and transformation induced by both HA and SRFA was faster in ASW than in pure water. These observations can be explained by assuming an interplay between different competing and sometimes opposite processes, including the competition between chloride, bromide and dissolved oxygen for reaction with HS triplet states.

The analysis of intermediates formed in the different matrices under study showed the formation of several hydroxylated (hydroquinone, 1,4-benzoquinone, resorcinol) and condensed compounds (2,2'-bisphenol, 4,4'-bisphenol, 4-phenoxyphenol). Although 1,4-benzoquinone was the main transformation product, formation of condensed molecules was significant with both HA and SRFA. Experiments on natural seawater spiked with HS confirmed the favored formation of condensed products, suggesting a key role of humic matter in dimerization reactions occurring in saline water.

1. Introduction

The photo-induced transformation of phenol in seawater is known to proceed through the formation of a wide range of intermediates including mono- and dichlorophenols, nitrophenols, bromophenols and bisphenols (Calza et al., 2008). In an attempt to clarify the role played by different photosensitizers (dissolved organic matter, nitrate, nitrite, H₂O₂, iron species) in the formation of the detected intermediates, each photosensitizer has been separately investigated. This paper is the last of a set where the link between natural photosensitizers and secondary pollutants has been highlighted for the first time in seawater. Previous studies have investigated the photosensitising ability of iron species (Calza et al, 2012a), the role played by hydrogen peroxide and its interaction with iron species (Calza et al, 2012b), as well as transformations induced by nitrate and nitrite (Calza et al, 2012c). In this study, we focused on the ability of humic substances (HS) to act as photosensitizers toward the transformation of organic compounds in the marine surface layer.

The abundant chromophoric moieties in DOM are able to initiate a number of photochemical reactions of high environmental significance (Zepp, et al., 1977; Aguer and Richard, 1999). Such reactions belong to the class of indirect photolytic processes that can promote the abiotic phototransformation of different organic compounds, such as toxic chemicals (Spokes and Liss, 1995; Magi, 1997, Fukushima et al. 2000, Aguer and Richard, 1999). When irradiated by sunlight, HS produce solvated electrons as well as several reactive oxygen species (ROS), such as hydroxyl radicals ($\cdot\text{OH}$), peroxy radicals ($\text{ROO}\cdot$), singlet oxygen ($^1\text{O}_2$), carbon-centred radicals and excited triplet states of DOM (Faust and Hoignè, 1987; Blough and Zepp, 1995, Brezonik and Fulkerson-Brekken, 1998)

Chemical analysis of DOM reveals noteworthy compositional variability between materials from different sources, which affects DOM photochemistry. Terrestrial humic substances (THA) are formed through transformation of organic matter derived from allochthonous sources. In contrast, marine humic (MHA) and fulvic acids (SRFA) are predominantly autochthonous (Nissenbaum and Kaplan, 1972, Stuermer et al. 1978) and their formation and diagenesis is partially mediated by aquatic bacteria and enzymes (Malcom, 1990).

HS are a complex class of anionic biogenic polyelectrolytes able to interact with organic substances and to play an important role as complexing agents for heavy metal ions or metal oxides (Calza et al. 2012a, Gu et al. 1994, Leenheer and Brown, 1998). HS are strongly bound to mineral surfaces by different mechanisms, *e.g.* through specific adsorption upon ligand exchange with protonated surface hydroxyl groups (Calza et al, 2012a, Aguer and Richard, 1999, Christl and Kretzschmar, 2000, Murphy and Zachara, 1995).

HS adsorption on iron oxides can strongly modify the surface properties of the latter and affect the fate, behavior, transport and transformation of environmental pollutants (Fu and Quan, 2006). Humic coatings make hydrophilic oxide surfaces become more hydrophobic, thereby affecting the interaction of these surfaces with hydrophobic organic pollutants (McKnight and Bencala, 1992, Marshall et al. 1998). Considering the photochemical properties of iron oxides, it is interesting to report that surface Fe(III)-humate complexes are efficient photoreactants (Faust and Hoffmann, 1989). HS are also involved in the natural reduction of Fe(III), and the dissolution of iron oxides sensitized by surface Fe(III)-humate complexes is an important process that controls iron concentration and bioavailability in natural environments (Voelker et al. 1997).

The main aim of this study is the assessment of the HS-sensitized phototransformation of phenol in seawater. For this reason, different types of humic matter including commercial compounds (terrestrial humic acid and fulvic acids) and extracted matter from seawater (marine humic acids) were investigated. To take the effect of ionic strength into account, experiments were carried out in pure water at pH 8, in artificial seawater (ASW, pH 8) and in natural seawater (NSW). After the study of the effects of HS toward phenol degradation, attention was focused on more complex systems where organic matter and inorganic constituents such as Fe(III) species coexist and interact. The present study is not aimed at investigating the interaction mechanism between HS and Fe(III) species or Fe(III) oxides, but rather the consequence of such an interaction on phenol disappearance kinetics, type and amount of intermediates formed, and Fe(II) formation from Fe(III), to point out typical transformation routes photoinduced by HS in seawater.

2. EXPERIMENTAL SECTION

2.1. Material and reagents

Suwanee River Fulvic Acids (SRFA) were purchased from IHSS (International Humic Substances Society). Terrestrial humic acids (THA) were purchased from Aldrich, while marine humic acids (MHA) were extracted from Antarctic marine sediments following a procedure described elsewhere (Thurman and Malcolm, 1981).

Hematite (α -Fe₂O₃), goethite (α -FeOOH) and akaganeite (β -FeOOH) were synthesized following literature methods (Bakoyannakis et al. 2003). Phenol, catechol, 1,4-benzoquinone, 2-chlorophenol, 3-chlorophenol, 4-chlorophenol, 4-phenoxyphenol, 2-phenoxyphenol, 2-nitrophenol, 4-nitrophenol, 2-bromophenol, 4-bromophenol, 3,4-dichlorophenol, 2,2'-bisphenol, 4,4'-bisphenol, Fe(ClO₄)₃, NaOH, Na₂SO₄, 4-aminoantipyrine and hydroquinone (all analytical grade) were purchased from Aldrich and used as received. 3,5-Dichlorophenol was purchased from Fluka, FeSO₄ from Carlo Erba and acetonitrile from Scharlau (Supergradient HPLC grade). Eluents for liquid chromatography were prepared in MilliQ water and degassed before use. Solution pH was adjusted by addition of 0.1 M NaOH and measured with a combined glass electrode connected to a Metrohm 713 pH meter. Artificial seawater (ASW) was prepared in MilliQ water by addition of the major sea salts (NaCl, Na₂SO₄, KCl, NaHCO₃, KBr, B(OH)₃, NaF, MgCl₂, CaCl₂ and SrCl₂), in amounts indicated elsewhere (Calza et al., 2012a). A non-purgeable carbon content of 12.5 mg C L⁻¹ and a pH of 7.9 were measured on NSW samples (Calza et al., 2008).

2.2. Irradiation procedures

Irradiation experiments were carried out in cylindrical Pyrex glass cells (4.0 cm diameter, 2.5 cm height) containing 5 mL aqueous sample, under a Solarbox (CO.FO.MEGRA, Milan, Italy) equipped with a 1500 Watt Philips Xenon lamp and a 340 nm cut-off filter, to simulate AM1 solar

light. The cell temperature during irradiation was 38 °C and the solutions were magnetically stirred during irradiation. Experiments were carried out in NSW, ASW (pH 8) or in pure water at pH 8.

2.3. Sample preparation

The phototransformation of phenol (initial concentration 20 mg L⁻¹) was investigated upon addition of THA, SRFA or MHA (50 mg L⁻¹) in pure water, ASW or NSW. NSW was sampled in June 2007 in the Gulf of Trieste, Northern Adriatic Sea (Italy) (see Calza et al. 2012a for NSW analysis). Irradiation experiments were carried out just after sampling. To study the behavior of iron species, solutions were spiked with Fe(ClO₄)₃ or with Fe (hydr)oxides (hematite, goethite or akaganeite). At equal concentration, SRFA showed significantly higher radiation absorption compared to THA or MHA. Above 300 nm, the absorption spectra of all HS samples showed a featureless exponential decay with increasing wavelength, which is typical of these compounds (Del Vecchio and Blough 2004).

2.4. Analytical determinations

The intermediates formed by phenol photo-induced transformation were identified and quantified by HPLC-UV through comparison with standard solutions. Moreover, GC-MS analyses were carried out to confirm the identity of the supposed structures.

The disappearance of phenol and the evolution of intermediates was monitored with a Merck-Hitachi HPLC equipped with L-6200 and L-6000 pumps for high-pressure gradients, a Rheodyne injector (54 µL sample loop), a RP C18 column (LichroCART, Merck, 12.5 cm × 0.4 cm, 5 µm packing) and a UV-Vis detector (Merck Hitachi L-4200) set at 220 nm. Elution was carried out with 70/30 phosphate buffer (10 mM, pH 2.8)/acetonitrile at a flow rate of 1 mL min⁻¹.

The formation of intermediates was confirmed using a GC-MS Agilent 6890, series II, equipped with a 5% phenylmethylpolysiloxane column (Agilent HP-5; 30 m × 0.25 mm). Water samples were extracted in dichloromethane, and the extract was dried with anhydrous Na₂SO₄. The GC

operating parameters were as follows: injector at 300°C, splitless injection (1 min), injection volume 1 µL. Temperature was linearly increased at a rate of 10 °C min⁻¹ from 50 to 300 °C.

The transformation of humic and fulvic acids was monitored as non-Purgeable Organic Carbon (nPOC), using a Shimadzu TOC-5000 analyzer (catalytic oxidation on Pt at 680 C). Calibration was achieved by injecting standards of potassium hydrogen phthalate.

Iron (II) concentration was measured by UV-Vis spectrophotometry (Varian Cary 100 Scan), upon Fe²⁺ complexation with 1,10-phenanthroline (Kester et al., 1967). The absorbance of the complex [(C₁₂H₁₈N₂)₃Fe]²⁺ was measured at 511 nm. The detection limit was 1 µmol L⁻¹.

3. Results

3.1. Photoinduced decomposition of humic acids

Preliminary experiments were run to assess the stability of HS over an extended irradiation time (72 h). Pure water at pH 8 or ASW were spiked with THA, MHA or SRFA and irradiated. Partial degradation occurred within 72 h, as indicated by the decrease of dissolved organic carbon (nPOC, see Figures 1 and 2). These data are not surprising, because humic substances can be photochemically degraded by sunlight into biologically labile and/or volatile organic compounds, carbon monoxide and carbon dioxide. Such reactions involve hydroxyl radicals ([•]OH) and Fe complexes with organic ligands (Mopper et al. 1991; Bertilsson and Tranvik, 2000; Wiszniowski et al., 2002; Brinkmann et al., 2003; Anesio and Graneli, 2003). HS were more easily degraded in pure water than in ASW, which can be attributed to a scavenging effect of bicarbonate/carbonate and most notably bromide ions toward [•]OH radicals (Buxton et al., 1988). When reacting with [•]OH, bromide and carbonate/bicarbonate produce Br₂^{-•} and CO₃^{-•}, respectively, which are oxidizing agents (Br₂^{-•} is also a brominating agent; Vione et al., 2008; Tamtam and Chiron, 2012) but less reactive than [•]OH itself (Neta et al., 1988; Calza et al. 2012a). This means that Br₂^{-•} and CO₃^{-•} can be involved in the transformation of organic compounds (*vide infra*), but hardly so in their mineralization. A similar issue applies to HS

triplet states (Canonica et al. 1995; Vione et al., in press). In both matrices, degradation was more pronounced for MHA than for THA or SRFA.

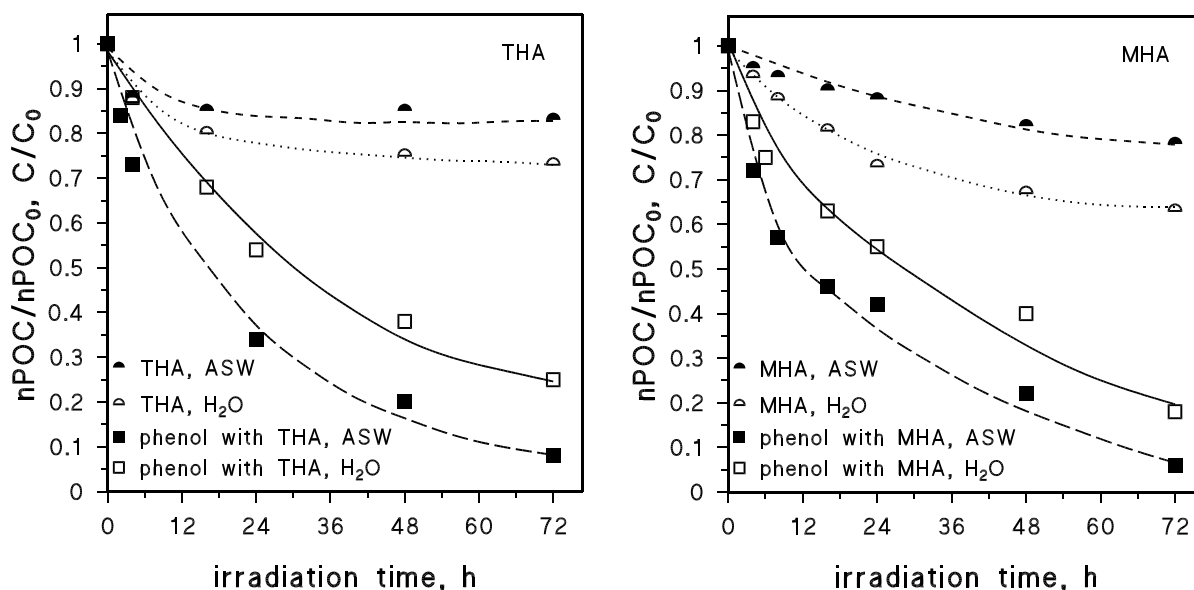


Figure 1. nPOC measurement for terrestrial humic acids (THA, left) and marine humic acids (MHA, right) in pure water (adjusted to pH 8) or in artificial seawater (ASW) as a function of irradiation time. Phenol disappearance ($C_0= 20 \text{ mgL}^{-1}$) as a function of irradiation time in the presence of humic acids (50 mgL^{-1}), in pure water and ASW.

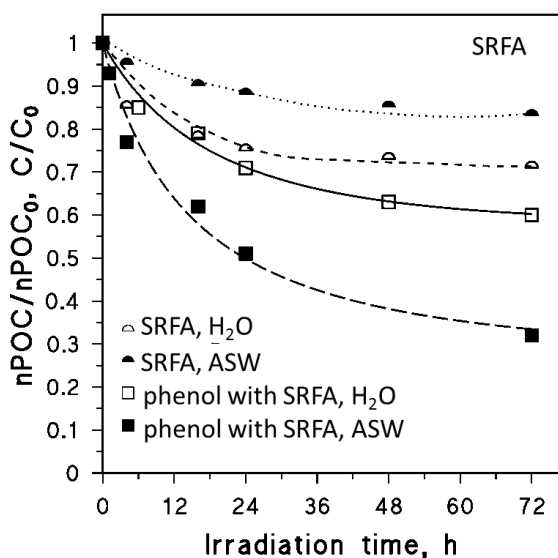


Figure 2. nPOC measurement for Suwannee River fulvic acids (SRFA) in pure water (adjusted to pH 8) and in artificial seawater (ASW) as a function of irradiation time. Phenol disappearance

($C_0 = 20 \text{ mgL}^{-1}$) as a function of irradiation time in the presence of SRFA (50 mgL^{-1}), in pure water and ASW.

3.2. Phenol photodegradation

Pure water or ASW were spiked with HS (THA, MHA or SRFA) and phenol (20 mg L^{-1}) and irradiated under simulated sunlight. Under these experimental conditions HS are the only important light absorbers and the direct photolysis of phenol can, therefore, be neglected (note that phenol does not absorb radiation above 300 nm).

Phenol time profiles are plotted in Figures 1 and 2. Degradation occurred in all cases, although to a different extent. Disappearance rates were higher in ASW than in pure water (see Table 1).

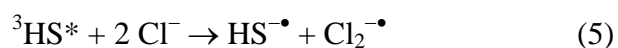
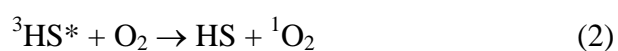
Table 1. Phenol disappearance rates in the presence of humic matter (50 mgL^{-1}).

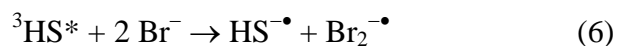
Phenol, rate:	$v \text{ (M} \times 10^5 \text{ h}^{-1}\text{)}$		
Spiked with:	$H_2O, pH 8$	ASW	NSW
THA	0.41	0.74	2.08
MHA	0.42	0.72	1.16
SRFA	0.36	0.15	1.80
THA, hematite	0.18	0.92	-
THA, goethite	0.24	0.74	
THA, akaganeite	0.21	0.54	
THA, Fe(III)	0.28	0.48	
SRFA, Fe(III)	0.16	0.22	0.68
SRFA, goethite	-	-	0.54
SRFA, akaganeite	-	-	0.70
SRFA, hematite	0.27	0.30	-

The photo-transformation of phenol sensitized by HS is a rather complex process and a number of reactive species has been invoked. Literature debate has concerned the type of oxidant that accounts for the degradation of phenolic compounds. Canonica et al. (1995) had shown that phenolic pollutants are readily photo-transformed by HA/SRFA and proposed the involvement of sensitizers in excited triplet states. The possible reaction pathway should involve an electron

transfer from phenol to the excited states of carbonyl groups present in the HS structure, yielding protonated phenoxyl radicals ($\text{PhOH}^{\cdot+}$, readily transformed into $\text{PhO}^{\cdot-}$) and deprotonated ketyl radicals ($\text{R}_1\text{R}_2\text{-C-O}^{\cdot-}$). Further reaction with oxygen causes regeneration of the carbonyl groups and formation of $\text{HO}_2^{\cdot}/\text{O}_2^{\cdot-}$ radicals. Kepczynski et al. (2007) suggested a contribution from photo-oxidants such as hydroxyl radicals, singlet oxygen and peroxy radicals, all formed from excitation of HA. However, recent evidence has provided additional support for the hypothesis that excited triplet states ($^3\text{HS}^*$) play an important role in the transformation of phenolic compounds (Halladja et al., 2007; Cavani et al., 2009; Trubetskoj et al., 2009; De Laurentiis et al., 2013).

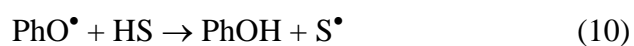
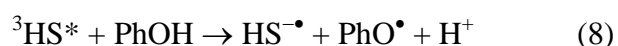
A major obstacle to the transformation of phenol by $^3\text{HS}^*$ in aerated solution is represented by the reaction between the triplet states and oxygen to yield $^1\text{O}_2$ (Canonica et al. 1995). Indeed, $^1\text{O}_2$ is much less reactive than typical $^3\text{HS}^*$ toward phenolic compounds and it would be largely inactivated by collision with water molecules. In the presence of ASW, the $^3\text{HS}^*$ species could undergo an important scavenging by chloride and bromide to produce transients (the radicals $\text{Cl}_2^{\cdot-}$ and $\text{Br}_2^{\cdot-}$; De Laurentiis et al., 2012; Brigante et al., 2014) that are more reactive than $^1\text{O}_2$ towards phenol (Wilkinson and Brummer, 1981; Neta et al., 1988). As far as phenol transformation is concerned, oxygen would thus channel the incoming light energy towards thermal deactivation through $^1\text{O}_2$ generation. In contrast, chloride and bromide contained in ASW would favor light-energy exploitation *via* formation of reactive radicals. The reactions that are proposed to take place in the system, and which explain why phenol transformation is faster in ASW compared to pure water, are the following:





Reaction between phenol and ${}^1\text{O}_2$ could take place but it would be less important. Note that the reactions involving $\text{Cl}_2^{\bullet-}$ and $\text{Br}_2^{\bullet-}$ (as well as ${}^3\text{HS}^*$ and ${}^1\text{O}_2$) would not cause significant mineralization of HS (Vione et al., in press). Therefore, while enhancing phenol transformation, seawater would inhibit HS mineralization because of the scavenging of $\bullet\text{OH}$ by bromide and carbonate/bicarbonate, as discussed in section 3.1.

Comparison between Figure 1 and Figure 2 showed that phenol transformation was more effective with THA and MHA compared to SRFA, despite the higher ability of SRFA to absorb the incident radiation. Several studies have shown that HS structure has an important impact on photoreactivity (Gao and Zepp, 1998; Vialaton and Richard, 2002; Leenheer et al., 2001, These et al., 2004; Chin et al., 2003; Ge et al., 2010). Generally speaking, the low-molecular weight fractions of HS should show higher ability to form triplet states (Minella et al., 2011; Minella et al., 2013). Indeed the larger molecules, as supra-molecular aggregates of smaller compounds (Nebbioso and Piccolo, 2012), are characterized by charge-transfer interactions that favor internal conversion at the expense of photophysical (*e.g.* fluorescence) and photochemical processes (reactivity) (Del Vecchio et al., 2004). From this point of view, SRFA should have higher ability to produce reactive triplet states compared to HA. However, the HS-photosensitized degradation of organic substrates also depends on the occurrence of antioxidant moieties within HS, which inhibit substrate degradation by favoring the back-reduction of partially oxidized species ($\text{PhO}^\bullet =$ phenoxy radical) to the initial compound ($\text{PhOH} =$ phenol) (Canonica and Lubscher, 2008; Wenk et al., 2011):



Therefore, the effectiveness by which THA, MHA and SRFA induce phenol phototransformation would depend on the budget between their ability to produce reactive triplet states under irradiation and the occurrence of the back reactions (10). Note that different functional groups would be responsible for triplet photogeneration and for the occurrence of back-reactions, which could explain why different HS samples did not behave in the same way.

3.3. Intermediate formation

As far as intermediates are concerned, no compound detectable by HPLC-UV was found upon light exposure of THA, MHA or SRFA alone in pure water at pH 8 or in ASW. Conversely, in the presence of HS and phenol under irradiation, several hydroxylated and condensed intermediates were detected. The main intermediate was always 1,4-Benzoquinone and, among the hydroxyderivatives, hydroquinone and resorcinol were also identified (see Figure 3 for the time evolution profiles of intermediates in ASW and Figure 4 for their maximum concentration values in the different cases).

A parallel pathway brings about the formation of condensed products. The compounds 4,4'-bisphenol and 2,2'-bisphenol were formed in all cases, while 4-phenoxyphenol was only detected in SRFA-spiked samples. Condensed products formation would most likely involve abstraction of an hydrogen atom from phenol with the formation of a phenoxy radical, in equilibrium with its tautomeric form.

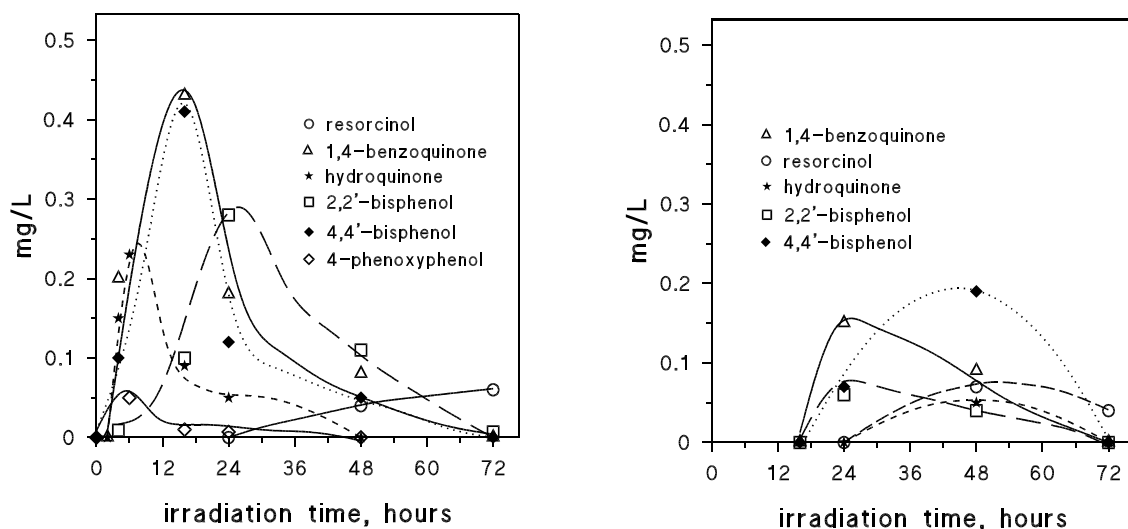
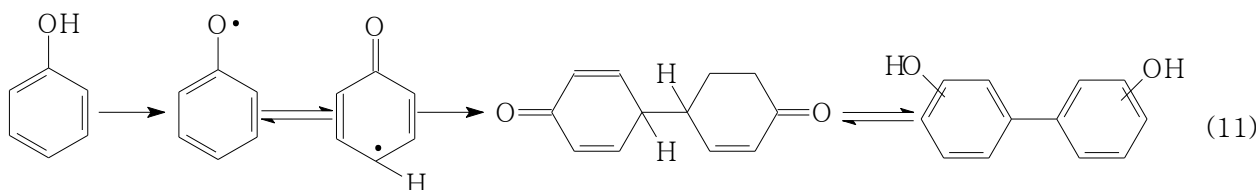
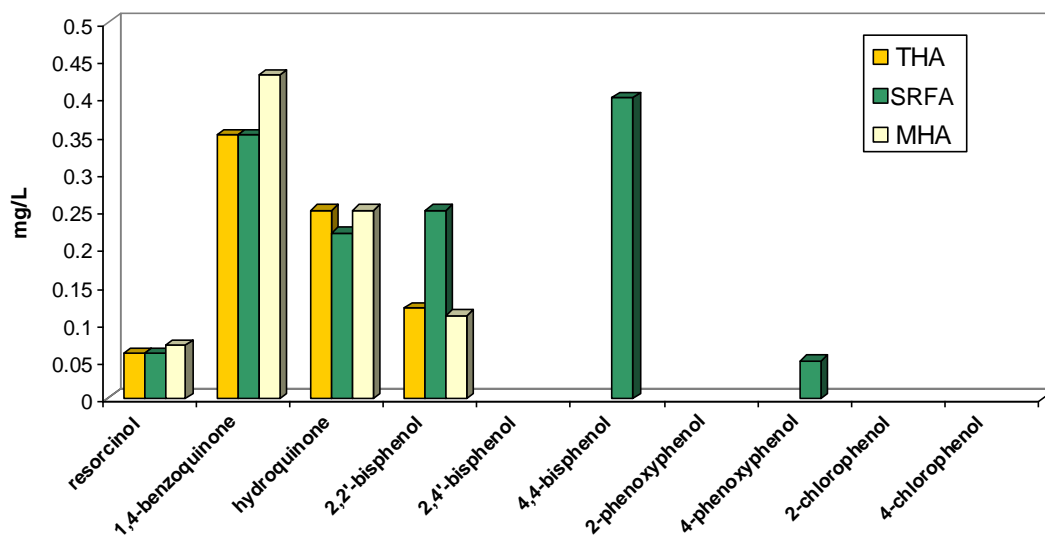


Figure 3. Formation of transformation products from phenol degradation in the presence of SRFA (left) or THA (right) in ASW.

Dimerization of phenoxy would then yield bisphenols and phenoxyphenols



In ASW in the presence of MHA, also 2- and 4-chlorophenol were formed in detectable amounts. Note that chlorophenol formation from phenol is accounted for by reaction between phenoxy radicals and Cl_2^- (a process that usually has low yields; Vione et al., 2005). Because phenoxy radicals may be consumed by HS anti-oxidant sites (reaction 10), one could explain why the formation of halogenoderivatives was not observed with all the HS samples.



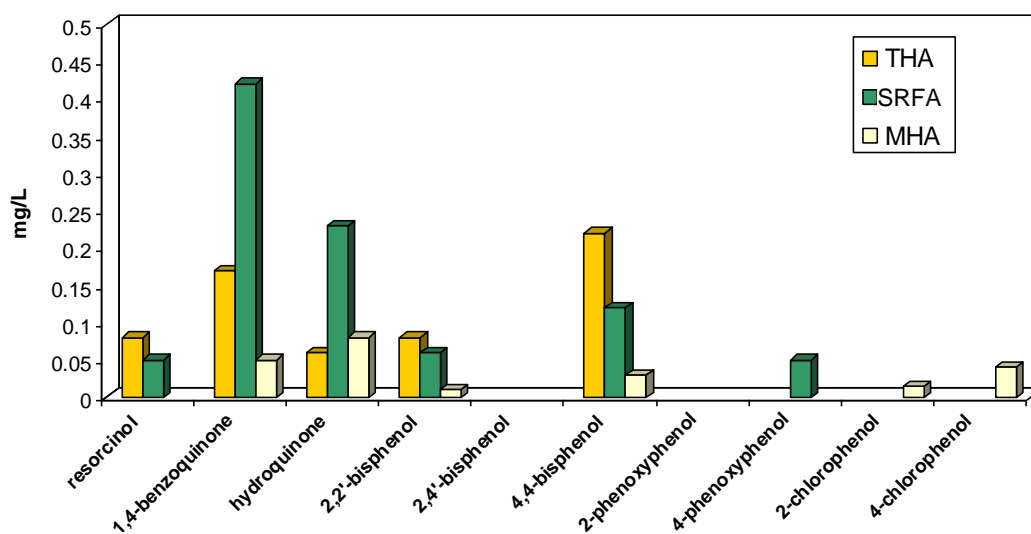


Figure 4. Maximum concentration of intermediate compounds formed during phenol photo-transformation in H₂O (top) and in ASW (bottom) spiked with THA, MHA and SRFA.

3.4. Influence of iron species on phenol and humic acids photodegradation

The THA solution was spiked with Fe(ClO₄)₃ or different iron oxides (hematite, goethite, akaganeite) and irradiated. The degradation curves plotted in Figure 5 showed that THA were more easily degraded when spiked with iron species. In ASW, the highest enhancement of degradation was observed with the Fe(ClO₄)₃-spiked solution. An explanation may lie in the improved oxidation of humic substances in the presence of Fe species under irradiation (Voelker et al., 1997). The increased efficiency highlights a synergic effect of the two photosensitizers toward the production of active species, and most notably of •OH. The Fe effect is particularly marked in pure water, while the lower effect observed in ASW medium could be attributed to iron complexation with chloride ions or other ASW salts (Vione et al., 2005).

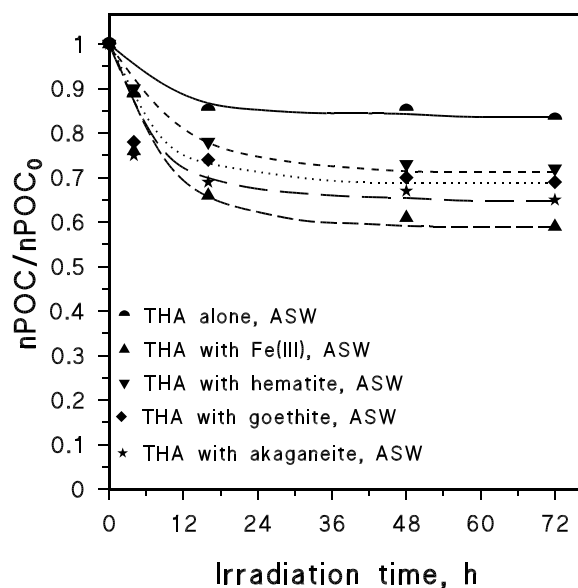


Figure 5. THA disappearance as a function of irradiation time in the presence of Fe(III) or Fe(III)-oxides in ASW.

Pure water or ASW containing phenol and THA were then spiked with selected iron species and irradiated, monitoring phenol disappearance. The relevant rate constants are reported in Table 1. Addition of iron species decreased phenol disappearance rates. This effect could be partially attributed to enhanced humic acids photodecomposition by Fe compounds, which would decrease the photosensitizing ability of THA. In ASW, the adsorption of ions (for example, chloride) on the iron-oxide surface could also inhibit photoactivity (Al- Rasheed and Cardin, 2003, Abdullah et al. 1990, Bekbolet et al., 1998). Moreover, THA would scavenge the reactive species produced by Fe compounds. It is true that such transient species would not be produced at all in the absence of Fe, but Fe compounds and THA would mutually compete for the incident irradiance. Light absorption by Fe compounds would decrease the photoactivity of THA in THA+Fe systems compared to THA alone, thereby decreasing the ability of THA to photodegrade phenol. Such a decrease of THA photoactivity would not be compensated for by the photoactivity of Fe compounds, because of the THA-mediated scavenging of Fe-photogenerated reactive species.

Upon exposure of Fe species to simulated sunlight, formation of Fe(II) took place as shown in Figure 6. Fe(II) was formed at higher concentration in pure water than in ASW. A first issue is that Fe(III) (hydr)oxides are less soluble in seawater compared to freshwater (Wells et al, 1991; Millero, 1998). In natural seawater the formation of dissolved Fe(II) is thought to take place upon iron photoreduction by humic matter, *via* a mechanism similar to that observed for Fe(III)-carboxylate complexes. Light absorption induces ligand-to-metal charge transfer (LMCT), with Fe(II) production and ligand oxidation upon dissociation of the complex. The photoinduced LMCT reactions of Fe(III)-fulvate complexes are known to occur both in solution and on the surface of iron oxides, and they cause the reduction of Fe(III) (Voelker et al. 1997).

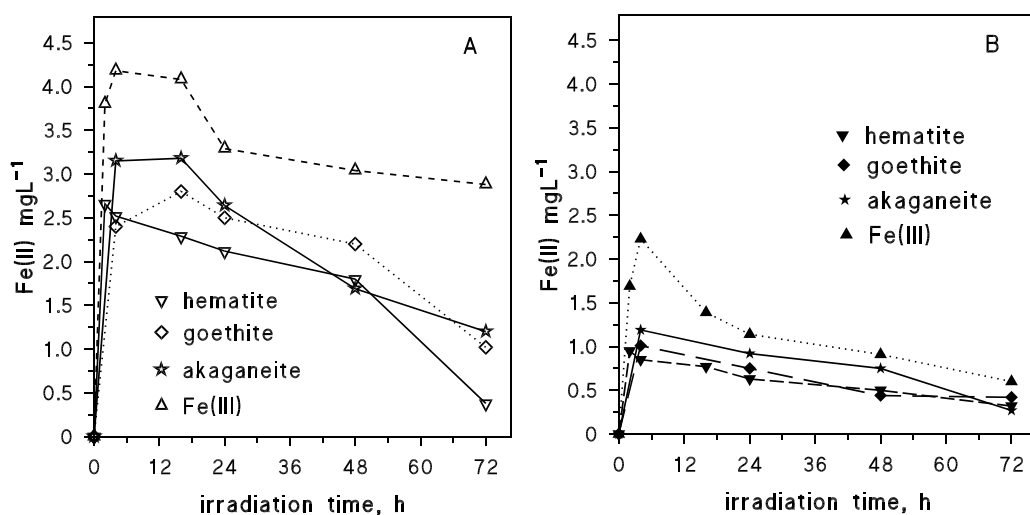


Figure 6. Formation of Fe(II) from Fe(III), hematite (Fe₂O₃), goethite (α -FeOOH), akaganeite (β -FeOOH) as a function of irradiation time: (a) pure water; (b) ASW.

An additional issue is that one expects faster Fe(II) oxidation to Fe(III) in ASW compared to pure water, which could also explain the results reported in Figure 6. The reason is that light absorption by HS in pure water would give ³HS*, which would mainly be scavenged by O₂ to produce ¹O₂. Singlet oxygen is poorly reactive towards Fe(II) (Wilkinson and Brummer, 1981). In contrast, ³HS* in ASW would be significantly scavenged by chloride and bromide to produce Cl₂^{•-}

and $\text{Br}_2^{\bullet-}$, respectively (De Laurentiis et al., 2012; Brigante et al., 2014), which are involved in Fe(II) oxidation (Neta et al., 1988).

The same TPs already reported for phenol degradation by HS were formed in HS + Fe systems, namely hydroxylated (1,4-benzoquinone, resorcinol, hydroquinone) and condensed compounds (2,2'-bisphenol and 4,4'-bisphenol). The maximum concentration values of intermediates are reported in Figure 7. The 1,4-benzoquinone was still the main product, with hematite as the only exception. In ASW, 2-chlorophenol was also detected in the presence of humic acids and hematite, goethite or akaganeite. These results are in agreement with the occurrence of a chlorination process mediated by iron species in seawater, as previously evidenced (Calza et al., 2012a), and with the detection of chlorophenols in ASW + HS (section 3.3).

3.5. Natural seawater

Natural seawater (NSW) was spiked with phenol and irradiated as described earlier. It was also spiked with HS or HS/iron species. Phenol disappearance rate was higher in NSW+HS compared with ASW+HS ($t_{1/2}$ decreased from 24 h (ASW) to 15 h (NSW)), presumably due to the presence of natural photo-sensitizers in seawater (see Table 1). By contrast, phenol degradation rate was decreased in the presence of NSW+HS+Fe compared to NSW+HS. A likely explanation, already advanced for ASW, is that Fe compounds and HS mutually hamper the respective photochemical activities and do not operate synergistically. The possible reason is competition for light absorption, coupled with the ability of HS to scavenge the reactive species photogenerated by Fe compounds.

Numerous phenol intermediates were identified in NSW (most notably halophenols and nitrophenols) and in NSW + HS, and they are reported in Figure 7. The addition of HS enhanced the formation of condensed compounds already detected in NSW (2,4'-bisphenol, 2-phenoxyphenol and 4-phenoxyphenol), which underlines the key role of HS in inducing the formation of phenol dimers in seawater. Moreover, 2,2'-bisphenol and 4,4'-bisphenol were only formed in NSW+HS.

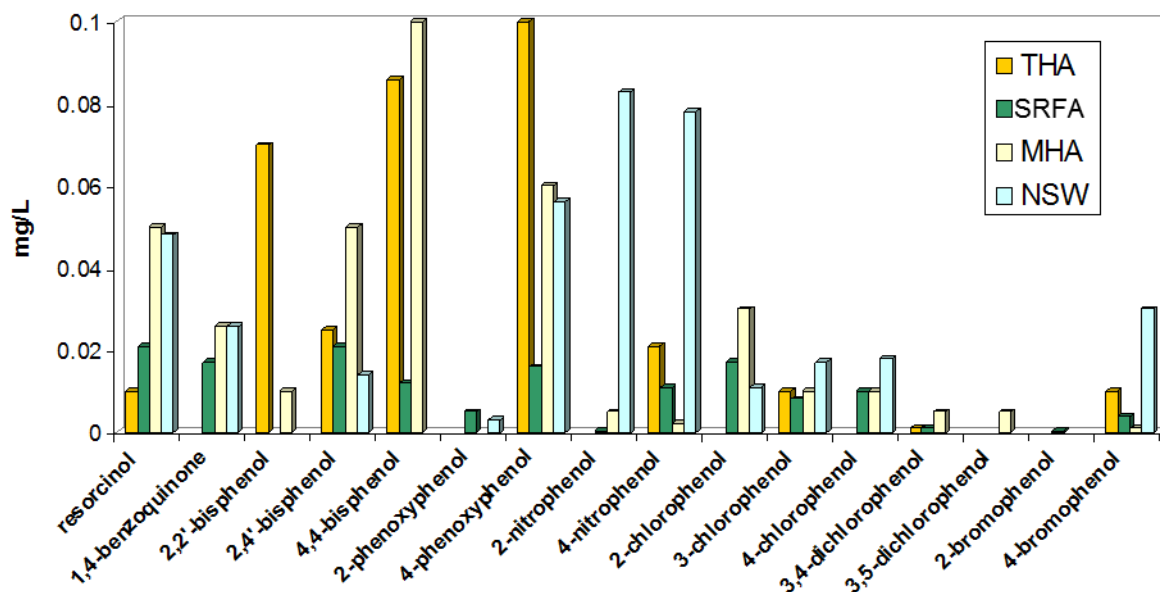


Figure 7. Intermediates from phenol in NSW not spiked or spiked with THA, MHA or SRFA.

These two compounds were also detected in ASW+HS (see Figure 8) and they can be considered typical phenol intermediates, the formation of which is induced by HS. The production of condensed compounds and particularly of 4,4'-bisphenol and 4-phenoxyphenol was particularly enhanced in the presence of THA and MHA.

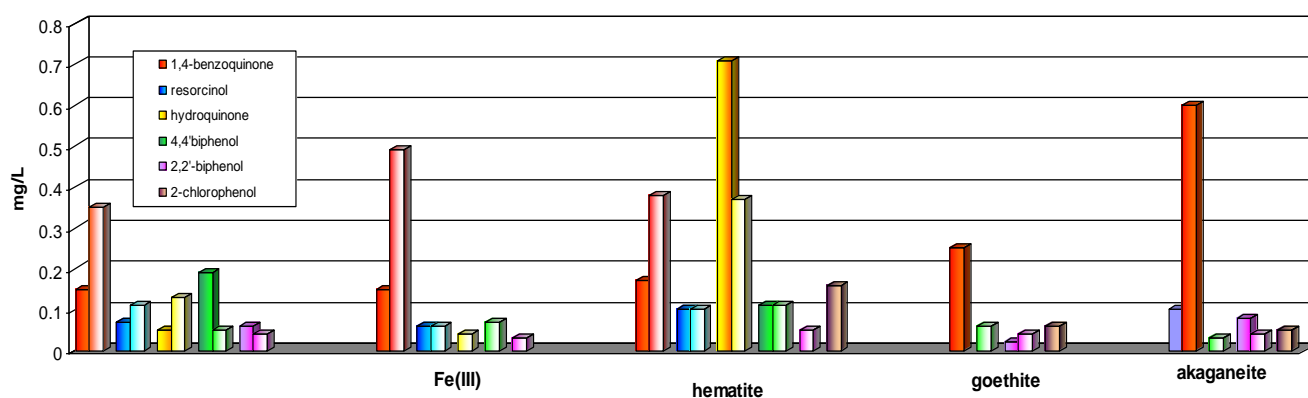


Figure 8. Intermediates from phenol (relative colors are in the legend) in ASW (deep colors) and in H₂O (pale colors).

The formation of nitro- and bromoderivatives was strongly depressed with NSW+HS compared with NSW, while hydroxyderivatives were formed in similar amount. Chloroderivatives formation

seems to be scarcely influenced by the presence of HS, with SRFA as the only exception. In fact, in SRFA-spiked samples, formation of chloroderivatives and most notably of 2-chlorophenol was slightly favored.

Irradiation of systems containing NSW + HS + Fe gave similar results as those observed with NSW + HS (data not shown).

Conclusions

The transformation of phenol in both artificial and natural seawater proceeded through the production of hydroxylated and condensed compounds. While the formation of hydroxy derivatives is non-specific and it is known to occur with all photosensitizers, the formation of condensed products can be considered as a signature of the photoreactivity of humic matter.

Different types of humic substances (HS) are able to act as photo-sensitizers toward the degradation of phenol. Irradiation of natural seawater spiked with phenol and HS enhanced the formation of several condensed products, suggesting a central role played by fulvic and humic acids of different origins on phenol dimerization in marine water. Interestingly, the dimerization and oligomerization of phenol and of other phenolic compounds represents a potential source of humic-like and fulvic-like substances in sunlit natural waters (De Laurentiis et al. 2013; Bianco et al. 2014). This issue might allow the suggestion that HS could undergo some sort of photoinduced auto-replication in the presence of phenols in environmental waters.

The HS-sensitized transformation of phenol was faster in seawater (either natural or artificial) than in pure water. A likely explanation is that the triplet states of HS ($^3\text{HS}^*$) in pure water are mainly scavenged by dissolved O_2 to produce $^1\text{O}_2$, which is poorly reactive towards phenol and mainly undergoes inactivation by collision with the solvent. In sea water, the ions chloride and bromide can effectively scavenge $^3\text{HS}^*$ and undergo oxidation to the radicals $\text{Cl}_2^{\cdot-}$ and $\text{Br}_2^{\cdot-}$, respectively. The latter are considerably more reactive than $^1\text{O}_2$ towards phenol, and they may induce significant substrate transformation. Following addition of iron species, phenol degradation

rate slowed down. This effect could be attributed to an enhanced HS photodecomposition by Fe compounds, which would decrease the HS photosensitizing ability, and to a scavenging by HS of the reactive species produced by Fe compounds.

Acknowledgements

Prof. B.M. Petronio is kindly acknowledged for providing the marine humic acid extracts.

References:

Abdullah M., Low G.K.C., Matthews R.W., Effects of common inorganic anions on rates of photocatalytic oxidation of organic carbon over illuminated titanium dioxide. *J. Phys. Chem.*, 1990; 94(17): 6820-6825

Aguer J. P., Richard C., Influence of the excitation wavelength on the photoinductive properties of humic substances, *Chemosphere*, 1999; 38(10): 2293-2301

Al-Rasheed R., Cardin D. J., Photocatalytic degradation of humic acid in saline waters. Part 1. Artificial seawater: influence of TiO₂, temperature, pH, and air-flow *Chemosphere*, 2003; 51: 925-933

Anesio, A. M., Graneli, W., Increased photoreactivity of DOC by acidification: Implications for the carbon cycle in humic lakes. *Limnol. Oceanogr.* 2003; 48: 735-744.

Bakoyannakis; D.N.; Deliyanni, E.A.; Zouboulis, A.I.; Matis, K.A.; Nalbandian, L.; Kehagias, Th.; Akaganeite and goethite-type nanocrystals: synthesis and characterization *Micropor. Mesopor. Mater.* 2003; 59: 35-42

Baxendale, J.H.; Magee, J. The photochemical oxidation of benzene in aqueous solution by ferric ion. *Trans. Faraday Soc.*, 1955; 51: 205-213

Bekbolet M., Boyacioglu Z., Ozkaraova B., The influence of solution matrix on the photocatalytic removal of color from natural waters *Water Sci. Technol.*, 1998; 38(6): 155-162

Bertilsson S., Tranvik L. J., 2000. Photochemical transformation of dissolved organic matter in lakes. *Limnol. Oceanogr.* 2000; 45: 753-762.

Bianco A., Minella M., De Laurentiis E., Maurino V., Minero C., Vione D., Photochemical generation of photoactive compounds with fulvic-like and humic-like fluorescence in aqueous solution. *Chemosphere* 2014; 111: 529-536

Blough N. V., Zepp R.G., Active oxygen: reactive oxygen species in chemistry, C. Foote, J. Valentine, A. Greenberg J.F. Liebman eds., Chapman and hill, New York, 1995, 280-332

Brezonik P.L., Fulkerson-Brekken J., Nitrate-induced photolysis in natural waters: Controls on concentrations of hydroxyl radical photo-intermediates by natural scavenging agents *Environ. Sci. Technol.*, 1998, 32: 3004-3010

Brigante, M., Minella, M., Mailhot, G., Maurino, V., Minero, C., Vione, D., Formation and reactivity of the dichloride radical ($\text{Cl}_2^{\bullet-}$) in surface waters: A modelling approach. *Chemosphere* 2014; 95: 464-469.

Brinkmann, T., Hörsch, P., Sartorius, D., Frimmel, F. H., Photoformation of low-molecular-weight organic acids from brown water dissolved organic matter. *Environ. Sci. Technol.* 2003; 37: 4190-4198.

Buxton GV, Greenstock CL, Helman WP, Ross AB. Critical review of rate constants for reactions of hydrated electrons, hydrogen atoms and hydroxyl radicals ($\text{OH}^{\bullet}/\text{O}^{\bullet-}$) in aqueous solution. *J Phys Chem Ref Data* 1988;17:513–886.

Calza P., Massolino C., Pelizzetti E., Minero C. Photochemically induced production of toxic halogenated and nitroaromatic compounds in seawater. *Sci. Total Environ.* 2008; 398 (1-3): 196-202

Calza P., Massolino C., Pelizzetti E., Minero C. Role of iron species in the photo-transformation of phenol in artificial and natural seawater, *Sci. Total Environ.* 2012a; 426: 281–288

Calza P., Campra L., Pelizzetti E., Minero C. Role of H_2O_2 in the phototransformation of phenol in artificial and natural seawater, *Sci. Total Environ.* 2012b; 431: 84-91

Calza P., Vione D., Novelli A., Pelizzetti E., Minero C., The role of nitrite and nitrate ions as photosensitizers in the phototransformation of organic matter in seawater, *Sci. Total Environ.* 2012c; 439: 67–75

Canonica S., Jans U., Stemmler K., Hoignè J., Transformation kinetics of phenols in water. Photosensitization by dissolved natural organic material and aromatic ketones. *Environ. Sci. Technol.* 1995; 29: 1822-1831

Canonica S, Laubscher HU. Inhibitory effect of dissolved organic matter on triplet-induced oxidation of aquatic contaminants. *Photochem Photobiol Sci* 2008; 7: 547-51.

Cavani, L., Halladja, S., Ter Halle, A., Guyot, G., Corrado, G., Ciavatta, C., Boulkamh, A., Richard, C., Relationship between photosensitizing and emission properties of peat humic acid fractions obtained by tangential ultrafiltration. *Environ. Sci. Technol.* 2009; 43: 4348–4354.

Chin Y.P., Aiken G., O'loughlin E., Role of dissolved organic matter composition on the photoreduction of Cr(VI) to Cr(III) in the presence of iron *Environ. Sci. Technol.*, 2003; 37: 1130-1136

Christl I., Kretzschmar R., Interaction of copper and fulvic acid at the hematite-water interface *Geochim. Cosmochim. Acta*, 2001; 65(20): 3535-3442

De Laurentiis, E., Minella, M., Maurino, V., Minero, C., Mailhot, G., Sarakha, M., Brigante, M., Vione, D., Assessing the occurrence of the dibromide radical ($\text{Br}_2^{\bullet-}$) in natural waters: Measures of triplet-sensitized formation, reactivity, and modelling. *Sci. Total Environ.* 2012; 439: 299–306.

De Laurentiis, E., Maurino, V., Minero, C., Vione, D., Mailhot, G., Brigante, M., Could triplet-sensitized transformation of phenolic compounds represent a source of fulvic-like substances in natural waters? *Chemosphere* 2013; 90: 881-4.

Del Vecchio, R., Blough, N. V., On the origin of the optical properties of humic substances. *Environ. Sci. Technol.* 2004; 38: 3885-3891.

Faust B., Hoignè J., Srensitized photooxidation of phenols by fulvic acid and in natural waters. *Environ. Sci. Technol.*, 1987; 21: 957-964

Faust B. C., Hoffmann M. R., Bahnemann, D. W. Photocatalytic oxidation of sulfur dioxide in aqueous suspensions of α -Fe₂O₃. *J. Phys. Chem.*, 1989; 93: 6371-6381

Feng, W. Nansheng, D. Photochemistry of hydrolytic iron(III) species and photoinduced degradation of organic compounds. A minireview. *Chemosphere*, 2000; 41: 1137-1147

Fu H., Quan X., Complexes of fulvic acid on the surface of hematite, goethite and akaganeite *Chemosphere*, 2006; 63: 403-410

Fukushima M., Tatsumi K., Morimoto K., Influence of iron(III) and humic acid on the photodegradation of pentachlorophenol. *Environ. Toxicol. Chem.*, 2000; 19(7): 1711-1716

Gao H., Zepp R.G., Factors influencing photoreactions of dissolved organic matter in a coastal river of the southeastern United States. *Environ. Sci. Technol.*, 1998; 32: 2940-2946

Ge L., Chen J., Wei X., Zhang S., Qiao X., Cai X., Xie Q., Aquatic photochemistry of fluoroquinolone antibiotics: kinetics, pathways, and multivariate effects on main water constituents, *Environ. Sci Technol.*, 2010; 44: 2400-2405

Gu B., Schmit J., Chen J., Liang, LY, McCarthy, JF Adsorption and desorption of natural organic matter on iron oxide. Mechanism and models. *Environ. Sci. Technol.*, 1994; 28 : 38-46

Halladja, S., Ter Halle, A., Aguer, J.-P., Boulkamh, A., Richard, C., Inhibition of humic substances mediated photooxygenation of furfuryl alcohol by 2,4,6-trimethylphenol. Evidence for reactivity of the phenol with humic triplet excited states. *Environ. Sci. Technol.* 2007; 41: 6066–6073.

Kepeczynski M., Czosnyka A., Nowakowska M., Photooxidation of phenol in aqueous nanodispersion of humic acid. *J. Photochem. Photobiol. A Chem.*, 2007; 185,: 198-205

Kester D. R.; Duedall I. W.; Connors D. N.; Pytkowicz R. M., Preparation of Artificial Seawater, *Limnol. Oceanogr.*, 1967; 12(1): 176-179

Kim, K, Choi, W, Hoffmann MR, Yoon, HI, Park BK, Photoreductive dissolution of iron oxides trapped in ice and its environmental implications, *Environ. Sci. Technol.*, 2010; 44: 4142-4148

Joseph, J.M.; Varghese, R.; Aravindakumar, C.T. Photoproduction of hydroxyl radicals from Fe(III)-hydroxy complex: a quantitative assessment. *J. Photochem. Photobiol. A: Chem*, 2001; 146: 67–73

Leenheer J. A., Brown G. K., MacCarthy, P, Cabaniss, SE, Models of metal binding structures in fulvic acid from the Suwannee River, Georgia. *Environ. Sci. Technol.*, 1998; 32: 2410-2416

Leenheer J.A., Rostad C.E., Gates P.M., Furlong E.T., Ferrer I., Molecular resolution and fragmentation of fulvic acid by electrospray ionization/multistage tandem mass spectrometry. *Anal. Chem.*, 2001; 73: 1461-1471

Magi E., Determination of trace metals complexed with humic acids in Antarctic marine sediments. *Chem. Spec. Bio.*, 1997; 9(2): 67-70

Malcom L. R., The uniqueness of humic substances in each of soil, stream and environment *Anal. Chim. Acta*, 1990; 232: 19-30

Marshall S. J., House W.A., Russel N. J., White G. F., Comparative adsorption of natural and commercially available humic acids to river sediments. *Colloids Surf. A-Physicochem. Engineer. Asp.* 1998; 144: 127-137

McKnight D. M., Bencala K. E., Sorption of dissolved organic carbon by hydrous aluminium and iron oxides occurring at the confluence of Deer Creek with the Snake river, Summit County, Colorado. *Environ. Sci. Technol.*, 1992; 26: 1388-1396

Millero F.J., Solubility of Fe(III) in seawater *Earth Planetary Sci. Lett.* 1998; 154: 323-329

Minella, M., Romeo, F., Vione, D., Maurino, V., Minero, C., Low to negligible photoactivity of lake-water matter in the size range from 0.1 to 5 μm . *Chemosphere* 2011; 83: 1480-1485.

Minella, M., Merlo, M. P., Maurino, V., Minero, C., Vione, D., Transformation of 2,4,6-trimethylphenol and furfuryl alcohol, photosensitised by Aldrich humic acids subject to different filtration procedures. *Chemosphere* 2013; 90: 306-311.

Mopper K., Zhou X., Kieber R. J., Sikorski R. J., Jones R. D., Photochemical degradation of dissolved organic carbon and its impact on the oceanic carbon cycle, *Nature*, 1991; 353: 60-62

Murphy E. M., Zachara J. M., The role of sorbed humic substances on the distribution of organic and inorganic contaminants in groundwater. *Geoderma*, 1995; 67: 103-124

Nebbioso, A., Piccolo, A., Advances in humeomics: Enhanced structural identification of humic molecules after size fractionation of a soil humic acid. *Anal. Chim. Acta* 2012; 720: 77-90.

Neta P, Huie RE, Ross AB. Rate constants for reactions of inorganic radicals in aqueous solution. *J Phys Chem Ref Data* 1988;17: 1027–284.

Nissenbaum A., Kaplan I. R., Chemical and isotopic evidence for the in situ origin of marine humic substances *Limnol. Oceanogr.*, 1972; 17: 570-582

Stuermer D. H., Peters K. E., Kaplan I. R., Organic Marine Geochemistry *Geochim. Cosmochim. Acta*, 1978; 42: 989-997

Spokes L. J., Liss P. S., Photochemically induced redox reactions in seawater. 1. Cations *Mar. Chem.*, 1995; 19: 201-213

Tamtam F., Chiron S., New insight into photo-bromination processes in saline surface waters: the case of salicylic acid. *Sci. Total Environ.* 2012; 435-436: 345-50.

These A., Winkler M., Thomas C., Reemtsma T., Determination of molecular formulas and structural regularities of low molecular weight fulvic acids by size-exclusion chromatography with electrospray ionization quadrupole time-of-flight mass spectrometry. *Rapid Commun. Mass. Spectrom.*, 2004; 18: 1777-1786

Trubetskoj, O.A., Trubetskaya, O.E., Richard, C., Photochemical activity and fluorescence of electrophoretic fractions of aquatic humic matter. *Water Res.* 2009; 36: 518–524.

E.M. Thurman, R.L. Malcolm, Preparative isolation of aquatic humic substances, *Environ. Sci. Technol.* 1981; 15: 463-466.

Vialaton D., Richard C., Phototransformation of aromatic pollutants in solar light: Photolysis versus photosensitized reactions under natural water conditions *Aquat. Sci.*, 2002; 64: 207-215

Vione D., Maurino V., Minero C., Calza P., Pelizzetti E., Phenol chlorination and photochlorination in the presence of chloride ions in homogeneous aqueous solution. *Environ. Sci. Technol.*, 2005; 39: 5066-5070

Vione D., Maurino V., Cucu Man S., Khanra S., Arsene C., Olariu R. I., Minero C., Formation of organobrominated compounds in the presence of bromide under simulated atmospheric aerosol conditions. *ChemSusChem* 2008; 1: 197-204.

Vione D., Minella M., Maurino V., Minero C., Indirect photochemistry in sunlit surface waters: Photoinduced production of reactive transient species. *Chem. - Eur. J.*, in press, DOI: 10.1002/chem.201400413.

Voelker B. M., Morel M. M., Sulzberger B., Iron Redox Cycling in Surface Waters: Effects of Humic Substances and Light. *Environ. Sci. Technol.* 1997; 31: 1004-1011

Wells M.L., Mayer L.M., Donard O.F.X., de Souza Sierra M.M., Ackelson S.G., *Nature* 1991; 353: 248-250

Wenk J, von Gunten U, Canonica S. Effect of dissolved organic matter on the transformation of contaminants induced by excited triplet states and the hydroxyl radical. *Environ Sci Technol* 2011;45:1334-40.

Wilkinson, F., Brummer, J., Rate constants for the decay and reactions of the lowest electronically excited singlet-state of molecular oxygen in solution. *J. Phys. Chem. Ref. Data* 1981; 10: 809-1000.

Wiszniewski J, Robert, D, Surmacz-Gorska, J, Miksch, K, Weber, JV Photocatalytic decomposition of humic acids on TiO₂ Part I: Discussion of adsorption and mechanism. *J. Photochem. Photobiol. A: Chem.*, 2002; 152: 267-273

Zepp R.G., Wolfe N. L., Baughman G. L., Hollis R.C., Singlet oxygen in natural waters. *Nature*, 1977; 267: 421-423