Insights into the photo-induced formation of reactive intermediates from effluent organic matter: The role of chemical constituents

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A B S T R A C T

In the present study, the formation of triplet states of organic matters ($^3OM^*$) from effluent organic matter (EfOM) under simulated solar irradiation was investigated. EfOM was separated into hydrophobic (HPO), transphilic (TPI), and hydrophilic (HPI) components. The quantum yield coefficients (f_{TMP}) of $^3OM^*$ were measured for each component and compared to those of reference natural organic matter (NOM). $NaBH_4$ reduction was performed on the EfOM, and the effect of aromatic ketones moieties on triplet formation was also determined. Furthermore, the apparent quantum yield of $^1O_2$ (Φ_{1O2}) and O_2^- ($\Phi_{O2^{\bullet-}}$) was measured. Our results suggested that the HPI fraction acted as a sink for $^3OM^*$. A linear correlation was observed between f_{TMP} and Φ_{1O2} for NOM/EfOM, except for $NaBH_4$-reduced effluent and HPI components. Both f_{TMP} and Φ_{1O2} were positively correlated with the contribution rates of $NaBH_4$-reducible moieties (aromatic ketones) toward $^3OM^*$. Aromatic ketones were primarily responsible for the production of $^3OM^*$ from EfOM, whereas quinone moieties played a key role in the production of $^3OM^*$ in NOM-enriched solutions. Understanding the role of chemical constituents on the photo activity of EfOM/NOM is essential for providing useful insights on their photochemical effects in aquatic systems.

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1. Introduction

Municipal wastewater treatment plants (WWTPs) discharge effluent organic matter (EfOM) into surface water, and EfOM has received increasing attention due to its environmental impacts and potential threat to human health (Dong et al., 2010; Yan and Song, 2014; Zheng et al., 2014). In general, EfOM is composed of three groups: natural organic matter (NOM) originating from drinking water sources, soluble microbial products (SMPs) produced during biological treatment processes, and trace organic contaminants (TrOCs) and their degradation metabolites (Shon et al., 2006). In contrast to NOM, EfOM is relatively hydrophilic, with a high proportion of protein-like substances and exhibits high fluorescence peak intensities due to its production from biological sources (Coble, 1996; Filloux et al., 2012; Leenheer et al., 2000; Vakondios et al., 2014). Moreover, the molecular sizes of EfOM are variable, ranging from 10^2 to 10^5 Da (Fattahi and Solouki, 2003; Her et al., 2003; Lee et al., 2013). The unique compositions of EfOM and NOM lead to differences in their physical and chemical behavior (Bodhipaksha et al., 2015; Dong et al., 2010), including potentially dissimilar photochemical transformations in aquatic systems.

In the past three decades, the photochemistry of NOM has been extensively studied (Andrew et al., 2013; Gonsior et al., 2009; Janssen et al., 2014; Niu et al., 2014; Sun et al., 2015; Wenk et al., 2015; Zepp et al., 1985). NOM are collections of diverse, relatively low molecular mass components forming dynamic associations stabilized by hydrophobic interactions and hydrogen bonds (Sutton and Sposito, 2005), playing a key role in the photodegradation of dissolved organic pollutants in natural aquatic systems (Chen et al., 2009; Li et al., 2016; Ryan et al., 2011; Wang et al., 2012; Yang et al., 2013). Solar irradiation of NOM produces a range of reactive intermediates (RI), including singlet oxygen ($^1O_2$), superoxide (O_2^-), hydrogen peroxide (H_2O_2), hydroxyl radical ($HO^\bullet$), carbonate radical ($CO_3^{\bullet-}$), etc. (Garg et al., 2011; Glover and Rosario-Ortiz, 2013; Vione et al., 2014; Zhang et al., 2014). In many cases, these photochemical processes were known to drive from excited triplet states of NOM ($^3NOM^*$). Upon illumination, a ground-state NOM chromophore (^1NOM) is promoted to an excited singlet state ($^1NOM^*$), which may undergo nonradiative relaxation processes, such as internal conversion and vibrational relaxation. $^1NOM^*$ can also rapidly undergo intersystem crossing (ISC) to $^3NOM^*$. $^3NOM^*$ is a spin-forbidden transition that exhibits a longer lifetime. The

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relatively long lifetime of $^3NOM^*$ enables it to play a significant role in photochemical reactions (Canonica et al., 2006; Grebel et al., 2012; Parker et al., 2013; Wenk et al., 2011).

Despite the numerous studies on NOM, the photochemistry of EfOM is not fully understood. Compared to NOM, EfOM usually exhibits lower $SUVA_{254}$ (absorbance at 254 nm per mg-C) values, greater E_2/E_3 ratios (the ratio of absorbance at 254 nm–365 nm), higher hydrophilic organic matter concentrations and polysaccharide contents, and a clear protein-like peak in its excitation emission matrix (EEM) fluorescence spectra (Nam et al., 2008; Shon et al., 2006). The photo-reactivity of EfOM and NOM was assessed in relation to their optical properties (e.g., $SUVA_{254}$ and E_2/E_3 ratio) (Dalrymple et al., 2010; Mostafa et al., 2014; Mostafa and Rosario-Ortiz, 2013; Sharpless, 2012). Recent studies have reported that the E_2/E_3 ratios are positively correlated to the quantum yield coefficients of excited triplet states of EfOM ($^3EfOM^*$) and apparent quantum yields of $^1O_2$ (Φ_{1O2}). Similar trends have also been observed for humic and fulvic acids (Bodhipaksha et al., 2015). EfOM has been shown to alter the photochemical production of reactive intermediates in wastewater-impacted waters. Rosario-Ortiz and coworkers studied the effects of optical characteristics and apparent molecular weight (AMW) on the apparent quantum yields of $^1O_2$ and $HO^\bullet$ from EfOM (Lee et al., 2013; Mostafa and Rosario-Ortiz, 2013). In addition to the positive correlation between the quantum yield of $^1O_2$ and the E_2/E_3 ratio, the lower AMW humic fraction of EfOM was found to be highly efficient for the removal of refractory TrOCs via photochemical processes, producing more $HO^\bullet$. In our previous study (Zhang et al., 2014), EfOM was separated into hydrophobic (HPO), transphilic (TPI), and hydrophilic (HPI) portions. The HPI component had a greater quantum yield of $^1O_2$ than the HPO and TPI fractions. A positive correlation was observed between the phenolic concentrations and steady-state $O_2^{\bullet-}$ concentrations. McKay et al. recently studied the photochemical formation of RI from DOM, particularly focused on the effects of variable molecular weight and chemical reduction on the formation of $^3DOM^*$, $^1O_2$, and $HO^\bullet$ (McKay et al., 2016). They discovered that decreasing molecular weight and borohydride reduction work in opposition regarding quantum efficiencies for $^1O_2$ and $^3DOM^*$ production (McKay et al., 2016). However there is lack of information regarding the effect of chemical constitutions on triplet formation based on the hydrophobicity of EfOM. The isolation of EfOM based on the hydrophobicity will be useful for additional studies that aim to determine the specific roles of different functional groups during the generation of RI. Due to the difference in chemical compositions of EfOM and NOM, it is necessary to explore RI production in EfOM systems, which may have enormous influence on the environmental transformation and fate of trace organic contaminates.

To describe the effects of chemical constituents on the photochemical production of RI, EfOM was separated based on hydrophobicity. Furthermore, $NaBH_4$ reduction was employed to explore the photochemical role of aromatic ketones. In general, quinones and aromatic ketones are two typical proxies frequently employed to study the photochemical properties of $^3NOM^*$ (Canonica et al. 1995, 2006; Cory and McKnight, 2005; Sharpless et al., 2014). $NaBH_4$ is known to selectively reduce carbonyl-containing compounds to hydroxyl-containing compounds, such as quinones and aromatic ketones, under anaerobic conditions. Upon introduction of molecular oxygen, any quinones previously reduced to hydroquinones are reoxidized to quinones (Ma et al., 2010). Thus, the contribution rates of aromatic ketones to the formation of $^3EfOM^*$ (defined as $\frac{k_{untreated} - k_{NaBH4\_treated}}{k_{untreated}}$) can be determined using the $NaBH_4$ reduction technique. Simultaneously, the relationship between the contribution rate of aromatic ketones and RI production ($^3OM^*$, $^1O_2$

and $O_2^{\bullet-}$) was investigated. Using these results, we attempted to explore the internal correlations of the chemical constituents of EfOM and its photochemical behavior and provide useful insights into its photochemical effects on aquatic systems.

2. Experimental section

2.1. Chemicals

Suwannee river natural organic matter (SRNOM, cat.# 1R101N), humic acid (SRHA, cat. 2S101H), fulvic acid (SRFA, cat. # 2S101F), and Pony lake fulvic acid (PLFA, cat. # 1R109F) were obtained from the International Humic Substances Society (IHSS). Furfuryl alcohol (FFA), trifluoroacetic acid (TFA), sodium borohydride ($NaBH_4$), DAX-8, XAD-4, and MSH-1 resins, 2,4,6-trimethylphenol (TMP), glucose, tryptophan, tyrosine, phenylalanine, 1,4-benzoquinone, and 3-methoxyacetophenone were purchased from Sigma-Aldrich. 2-Methyl-6-[p-methoxyphenyl]-3, 7-dihydroimidazo [1,2-a] pyrazine-3-one, which is a methyl cypridina luciferin analog (MCLA), was obtained from TCI chemicals. Tryptone was purchased from OXOID. All chemicals were used as received.

2.2. EfOM isolation and sample preparation

Two wastewater effluents were obtained from municipal sewage plants located in Jiangsu and Shanghai, China. These two sewage plants treat domestic sewage using a circulatory activated sludge treatment system. The effluents were adjusted to pH 2 and were separated into hydrophobic (HPO), transphilic (TPI), and hydrophilic (HPI) fractions using XAD resins, in accordance with the previously established procedure (Zhang et al., 2014). The water quality characteristics of the wastewater effluents are listed in Table S1 of the Supporting Information (SI). All solutions were prepared using Milli-Q water. Stock solutions (~200 mg-C L^{-1}) of OM were prepared in phosphate buffer (5.0 mM, pH 7.3) and were stored at 4 °C. The experimental solutions were prepared by diluting the stock solutions with a phosphate buffer to a final concentration of 5.0 mg-C L^{-1}. Dissolved organic carbon (DOC) was measured using a TOC analyzer (Shimadzu® L-CPH).

2.3. $NaBH_4$ reduction

Sodium borohydride ($NaBH_4$) was employed to reduce the OM, as previously reported (Golanoski et al., 2012; Ma et al., 2010; Thorn et al., 1996). Stock solutions (~200 mg-C L^{-1}, 20 mL) of OM were adjusted to pH 8.0 with a solution of NaOH (1.0 M) and were purged with high-purity nitrogen for 15 min to remove molecular oxygen. To obtain the required $NaBH_4$ solution, 1.0 g of solid $NaBH_4$ was dissolved into 1 mL of 0.1 M NaOH, which was subsequently added dropwise to the OM solutions. Complete homogeneity within the mixture was achieved via magnetic stirring under a flow of nitrogen. The reduction was considered to be complete when further changes in the optical properties were not observed as presented in Fig. S1 (approximately 100 mg $NaBH_4$ was used for reducing the OM solution). The resulting solutions were passed through an H^+-saturated-cation exchange resin (MSC-H) to remove the residual $NaBH_4$, bubbled with dioxygen for 5 min to regenerate the quinone moieties within the OM and then freeze-dried to obtain the reduced OMs.

2.4. Photochemical experiments

Photochemical experiments were performed using a solar simulator (Suntest XLS$^+$, Atlas) equipped with a 1700-W xenon lamp, and a solar filter was employed to prevent irradiation at