Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/watres

# Anti-estrogenic activity formation potential assessment and precursor analysis in reclaimed water during chlorination

Xin Tang [a], Qian Yuan Wu [b], Ye Du [a], Yang Yang [a], Hong Ying Hu [a,b,*]

[a] Environmental Simulation and Pollution Control State Key Joint Laboratory, and State Environmental Protection Key Laboratory of Microorganism Application and Risk Control (MARC), School of Environment, Tsinghua University, Beijing 100084, PR China

[b] Shenzhen Laboratory of Microorganism Application and Risk Control, Graduate School at Shenzhen, Tsinghua University, Shenzhen 518055, PR China

ARTICLE INFO

*Article history:*
Received 13 July 2013
Received in revised form
1 October 2013
Accepted 2 October 2013
Available online 23 October 2013

*Keywords:*
Chlorination
Reclaimed water
Anti-estrogenic activity formation potential
Precursor

ABSTRACT

Chlorination was reported to increase the anti-estrogenic activity in reclaimed water from domestic wastewater treatment plants, which may add to the risk of reclaimed water reuse. In order to assess the anti-estrogenic disinfection by-product (DBP) precursors, the anti-estrogenic activity formation potential (AEAFP) during chlorination was studied. Firstly, the conditions for the experimental measurement of AEAFP were determined. A 24-h chlorination experiment was applied for AEAFP measurement. After chlorination, dechlorination using reductive reagents led to significant loss of anti-estrogenic activity formation. In addition, as the presence of ammonia nitrogen and other major chlorine consumers would result in lower anti-estrogenic activity formation, a basic chlorine dose of 3× DOC (mg-$Cl_2$ $L^{-1}$) was adequate for completely transforming the anti-estrogenic DBP precursors while an extra chlorine dose of 8× ammonia-nitrogen + 5× nitrite-nitrogen (mg-$Cl_2$ $L^{-1}$) should be added when there was a high level of ammonia nitrogen and nitrite nitrogen in the reclaimed water. Therefore, 24-h chlorination without dechlorination or using only non-reductive quenching reagents (e.g. ammonium) for dechlorination and a total chlorine dose of 3× DOC + 8× ammonia nitrogen + 5× nitrite nitrogen (mg-$Cl_2$ $L^{-1}$) should be fulfilled for the AEAFP measurement. Moreover, the AEAFP (0.2–2.1 mg-TAM $L^{-1}$) of the reclaimed water samples ($n$ = 20) were further analyzed. The AEAFP was highly correlated to $UV_{254}$ and the fluorescence volume in excitation emission matrix fluorescence spectrum which can be used as surrogates to indicate the level of the AEAFP and assess the precursors in reclaimed water.



* *Corresponding author*. Environmental Simulation and Pollution Control State Key Joint Laboratory, and State Environmental Protection Key Laboratory of Microorganism Application and Risk Control (MARC), School of Environment, Tsinghua University, Beijing 100084, PR China. Tel.: +86 10 6279 4005; fax: +86 10 6279 7265.
E-mail address: hyhu@tsinghua.edu.cn (H.Y. Hu).

0043-1354/$ – see front matter © 2013 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.watres.2013.10.009

## 1. Introduction

In order to eliminate the pathogenic bacteria and guarantee the safety of reclaimed water, disinfection using chlorine is becoming a necessity in the reclamation treatment process. Nevertheless, the addition of chlorine into the reclaimed water often results in the formation of disinfection by-products (DBPs) which are often with higher toxicity than that in surface and drinking water (Sirivedhin and Gray, 2005) and increase the potential risk of the wastewater reuse. In previous studies, kinds of DBPs exerting genotoxicity, mutagenicity, carcinogenicity, endocrine disrupting effects and other negative biological effects were reported (Yang and Shang, 2004; Richardson et al., 2007; Wu et al., 2009; Huang et al., 2013).

Among those DBPs, the endocrine disrupting DBPs may lead to reproductive defects and other potential harms to wildlife (Colborn et al., 1993; Windham et al., 2003). Recently, the anti-estrogenic activity, a typical endocrine-disrupting effect, was found to increase significantly after chlorination of reclaimed water (Wu et al., 2009, 2010). Chemicals with anti-estrogenic activity are defined as estrogen receptor antagonists who bind to estrogen receptor but do not activate the receptor. Rather, these chemicals inhibit receptor activity by preventing the endogenous estrogen from binding to and activating the receptor (Hodgson, 2004). In laboratory, some anti-estrogenic chemicals have been shown in females to disrupt estrous cycles, impair fertility, increase preimplantation loss, and cause embryolethality (Hodgson, 2004). Therefore, it is urgently needed to assess the formation potential of the anti-estrogenic DBPs during chlorination of reclaimed water.

In order to access the DBPs and precursors, researchers generated several experimental methods to measure the formation potential of the identified DBPs such as trihalomethane (THMs), haloacetic acids (HAAs) and nitrogenous DBPs, and long contact time (generally 24 h for chlorination) and high dose of chlorine versus dissolved organic carbon (DOC) were considered. Besides, ammonia nitrogen, nitrite and other chlorine consumers should also be taken into consideration. Krasner et al. (2004) has developed DBP formation potential tests for treated wastewater, reclaimed water and drinking water, and the chlorine dose is based on DOC, ammonia nitrogen and nitrite nitrogen. Both detailed DBP formation potential experimental conditions (APHA, 1998; Krasner et al., 2004) and DBP analytical methods such as USEPA methods 551.1 and 552.2 were previously reported.

For those regular DBPs, previous studies revealed the correlations between the DBPs and their precursors. Chen and Westerhoff (2010) generated a mathematical approach using a multivariable power equation and concluded that functions with the water quality parameters such as DOC, UV absorbance, bromide and nitrogenous species can be used to predict the formation potential of DBPs. In addition, the effects of wastewater treatment on the DBP formation potential were also reported. Musikavong et al. (2005) reported that tertiary treatment process can reduce the formation potential of THMs in reclaimed water.

However, when we come for the anti-estrogenic DBPs during chlorination, seldom identified chemicals were in the list except for one called 2, 4-diphenylcrotonaldehyde deriving from the phenylalanine during chlorination (Wu et al., 2010), nor did the formation potential of the anti-estrogenic DBPs be assessed.

In order to get more important information about the anti-estrogenic DBPs and the precursors, the formation potential of the anti-estrogenic activity, instead of the individual DBPs, should be assessed during chlorination of reclaimed water. The formation potential of the anti-estrogenic activity can be used to indicate the total amount of its precursors in the reclaimed water.

To assess the formation potential of the anti-estrogenic activity, we suppose that the precursors can be completely converted to the anti-estrogenic DBPs if appropriate chlorination conditions are defined, and a maximum value of the anti-estrogenic activity formation during chlorination can be observed.

Therefore, the purpose of this paper is to establish the experimental conditions for the anti-estrogenic activity formation potential (AEAFP) measurement and assess the precursors during chlorination in reclaimed water.

## 2. Materials and methods

### 2.1. Sampling and water quality analysis

The reclaimed water samples used in this study were secondary and tertiary effluents from 6 municipal wastewater treatment plants (WWTPs) in north China. Samples were kept at 3–6 °C with ice and immediately delivered to the laboratory for water quality measurement and chlorination within 24 h.

Before analysis and chlorination, water samples were firstly filtered with glass fiber filters (0.45 μm) to eliminate suspended solids. Afterward, the dissolved organic carbon (DOC) was measured on a TOC-5000A analyzer (Shimadzu, Japan). UV absorbance was measured on a UV-2401PC UV-VIS recording spectrophotometer (Shimadzu, Japan). The pH value was measured on a FE-20 detector (Mettler Toledo, Switzerland). The ammonia nitrogen concentration was measured on a HI-96715 ammonia medium range analyzer (Hanna, USA). The nitrate concentration was measured on an ICS-1000 ion chromatography system (Dionex, USA).

Particularly, the secondary effluent samples (Table S1 in Supplementary Information) from the 6 WWTPs were mainly used to study the changes of the anti-estrogenic activity during chlorination and determine the experimental conditions for AEAFP measurement. The water quality of the secondary effluent samples and the main treatment process of each WWTP were listed in Table S2 in Supplementary Information.

In addition, more reclaimed water samples (secondary and tertiary effluents) from the 6 WWTPs were used for further AEAFP and precursor analysis.

### 2.2. Chlorination experiments

The filtrates (280 ml) of the reclaimed water samples were prepared in 300-ml glass bottles for chlorination. Sodium hy-