



# Effects of chlorination/chlorine dioxide disinfection on biofilm bacterial community and corrosion process in a reclaimed water distribution system

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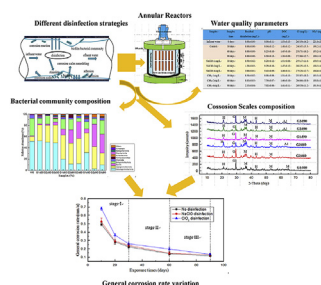
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## HIGHLIGHTS

- 1 mg/L NaClO or ClO<sub>2</sub> caused OTUs reduction but had little effect on biofilm composition.
- *Firmicutes* presented resistance to 4 mg/L NaClO and ClO<sub>2</sub> dosage as corrosion scales accumulation.
- Disinfection enhanced corrosion, the promoting effect of ClO<sub>2</sub> was more pronounced than NaClO.
- Strong oxidization of disinfection was predominant factor promoting corrosion in first 30 days.
- Bacterial community also played a crucial role in corrosion as scales accumulation after 30 days.

## GRAPHICAL ABSTRACT



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## ABSTRACT

In this work, reclaimed water treated with sodium hypochlorite (NaClO) or chlorine dioxide (ClO<sub>2</sub>) at 1, 2, and 4 mg/L was operated successively for 30 days respectively, in annular reactors with new cast iron coupons, corresponding to stages I (days 0–30), II (days 31–60), and III (days 61–90). The Illumina HiSeq 2500 sequencing platform was used to analyze the bacterial community composition, scanning electron microscopy and X-ray diffraction analyses were conducted to characterize corrosion scales, and the weight loss method was served to determine the general corrosion rate. Results reveal the precise disinfection effect on biofilm bacteria to be dose dependent and species specific. In stage I, disinfection caused a reduction in the number of operational taxonomic units, but, had little effect on biofilm composition. In stage II, NaClO and ClO<sub>2</sub> induced a reduction of *Proteobacteria* proportion, but increased the dominance of *Firmicutes*; the diminished *Proteobacteria* in NaClO test mainly included *Gammaproteobacteria*, while, that in ClO<sub>2</sub> test mainly included the *Gammaproteobacteria* and *Betaproteobacteria*. In stage III, *Firmicutes* presented a certain resistance to NaClO and ClO<sub>2</sub> as the accumulation of corrosion scales. Results also indicated that disinfection enhanced the corrosion process, and the promoting effect of ClO<sub>2</sub> was more pronounced than that of NaClO. Moreover, this promoting effect was more obvious in stage I than that in the latter two stages. The strong oxidization effect associated with disinfection in

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stage I was the dominant factor promoting corrosion, whereas, the bacterial community also played a crucial role in stages II and III.

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## 1. Introduction

The proliferation of microorganisms in water distribution systems has long been acknowledged as a serious issue because many associated problems occur, such as pathogen growth, biofilm formation, water quality deterioration, corrosion, and bad taste and odors generation (Boe-Hansen et al., 2002; Gomes et al., 2016). Consequently, it is extremely important to control the growth of microorganisms within water distribution systems.

According to the different bacterial characteristics within such system, the microbial phases therein have been defined as either bulk water bacteria, which flow through the water mains, or pipe wall biofilm, which form on the inner pipe surface (Gomes et al., 2016). It is reported that the pipe wall biofilm accounts for 95% of the overall biomass in water distribution systems, while the water phase accounts for only 5% (Flemming et al., 2002). Generally, maintaining a disinfectant residual is a common practice for controlling suspended bacteria in bulk water (Gagnon et al., 2005). However, the biofilm bacteria are more resistant to the disinfectant than the bacteria suspended in water because of bacterial community abundance, interspecies relationships, and extracellular polymeric substance formation (Behnke and Camper, 2012; Douterelo et al., 2014; Emtiazi et al., 2004). In this respect, some research find that different disinfection methods (chloramination,  $\text{ClO}_2$ , and  $\text{ClO}_2^-$ ) induce significant reduction in biofilm biomass and thickness within plastic pipes, but they cannot prevent biofilm growth (Gagnon et al., 2005; Ling and Liu, 2013). It is also of note that different types of pipes affect the biofilm structure and community composition obviously. Cast iron pipes have been widely used for over 500 years and account for a large proportion of the pipe materials in the water distribution systems (Gerke et al., 2008). Meanwhile, the use of disinfectants in metal pipe systems may also affect the corrosion process and iron oxides formation substantially (Zhu et al., 2014). The iron oxides formation can not only provide favorable conditions for biofilm attachment but also cause the undesirable disinfectant attenuation. In addition, disinfectant attenuation can modulate the effect of the disinfectant on actual biofilm microorganisms close to the pipe surface (Wang et al., 2012a). Li et al. reported that nitrate-dependent Fe(II) oxidation proceeds to a greater extent with chloramines than with chlorine; and this phenomenon could also favor the formation of denser corrosion scales (Li et al., 2015). Moreover, when chlorine was substituted with chloramines as a disinfection, there was a significant decrease in the number of corrosion-related bacteria, which lead to less bio-corrosion (Wang et al., 2012b). To the best of our knowledge, however, only a few studies have been conducted in relation to iron pipes in water distribution systems to investigate the effect of different disinfectants on corrosion and the bacterial community. However, there is rarely any discussion relating to systematic research and comparative analyses of different disinfection strategies, as well as the microorganisms to the corrosion process, particularly with respect to cast iron pipes in a reclaimed water distribution system (RWDS).

Therefore, it is necessary to conduct further research to gain a better understanding of this ecosystem in RWDS, including the disinfection, biofilm microorganisms and corrosion process, as such data would greatly assist in ensuring a safe and high-quality water

supply. The aims of the current study were to (1) investigate the effects of different disinfection strategies ( $\text{NaClO}$  and  $\text{ClO}_2$ ) on the development of biofilm bacterial community (abundance, diversity, and composition) in a RWDS using the Illumina HiSeq 2500 sequencing platform; (2) elucidate the evolutionary details of corrosion-related bacteria with respect to different disinfectant compounds and concentrations; (3) examine the composition and structure of corrosion scales using scanning electron microscopy (SEM) and X-ray powder diffraction (XRD) analysis; and (4) discuss the corrosion mechanisms driven by the bacterial community as well as water quality under the influence of different disinfection strategies.

## 2. Materials and methods

### 2.1. Materials and water preparation

The cast iron coupons used in this study were manufactured by Yangzhou Xiangwei Machinery Manufacturing Co., Ltd, Yangzhou, China, according to standard method (Chinese Standard, 2008), and they were comprised of C 19.08%, O 6.09%, Si 2.06%, Ca 0.58%, P 0.65%, S 1.60%, Fe 65%, Cu 1.98%, Mn 0.92%, and Zn 2.04%. The exposed surface area of each coupon was  $20.00 \text{ cm}^2$ , and the corresponding dimensions of each coupon were length  $\times$  width  $\times$  thickness =  $(72.4 \pm 0.1) \text{ mm} \times (11.5 \pm 0.1) \text{ mm} \times (2.0 \pm 0.1) \text{ mm}$ , respectively. Prior to the experiment, the coupons were polished using a series of waterproof-abrasive-paper pieces (#200, #400, #600, #800, #1000, #1200, and #1500), degreased in acetone, dehydrated in absolute ethanol, dried in a laminar flow cabinet, and weighed using high-precision scales (Sartorius, Germany, readability 0.01 mg).

The reclaimed water treatment process can be seen in Supporting Information 1. The parameters of effluent water used in the present research are summarized in Table 1. Water quality parameters such as pH, dissolved organic carbon (DOC), dissolved oxygen (DO), chloride ion ( $\text{Cl}^-$ ) and sulfate ion ( $\text{SO}_4^{2-}$ ) were measured according to the Analysis Method for Water and Waste Water (Wei and Q, 2002). The concentration of DOC was measured as "mg/L of C". All measurements were conducted in triplicates.

### 2.2. Preparation of disinfectant solution and experimental procedure

Annular reactors (ARs) were used to simulate the RWDS, and each was comprised of a rotating inner cylinder driven by a brushless direct current motor. The cast iron coupons were fixed on the rotating inner cylinder to imitate new distribution pipes. The internal structure of AR can be seen in Supporting Information 2, and the scheme figure of the whole operation system can be seen in Supporting Information 3. In a control experiment, reclaimed water without disinfectant was pumped into the ARs by a peristaltic pump lasting 90-days. The flow rate of the peristaltic pump was set to 4.17 mL/min so that hydraulic retention time in the ARs was 6 h and the rotation speed of the brushless direct current motor was 50 rpm to imitate a shear stress of  $0.25 \text{ N/m}^2$ . In disinfection experiments, two ARs with reclaimed water treated with  $\text{NaClO}$  or  $\text{ClO}_2$  at 1, 2, and 4 mg/L were operated successively for 30 days

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