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Exploring the biological stability situation of a full scale water distribution system in south China by three biological stability evaluation methods

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HIGHLIGHTS

- Correlation between BRP and AOC was significant in either WTP or DWDS.
- Correlation between BRP and BDOC was more significant in WTP than in DWDS.
- High chlorine residuals largely suppressed bacterial regrowth in DWDS.
- Low chlorine residuals led to a positive correlation between AOC/BRP and HPC.
- Heterotrophic bacteria were limited compared to that when AOC was more than 135 μg/L.

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ABSTRACT

Bacterial regrowth especially opportunistic pathogens regrowth and contamination in drinking water distribution systems (DWDS) have become an emerging threat to public health in the whole world. To explore bacterial regrowth and biological stability, assimilable organic carbon (AOC), biodegradable dissolved organic carbon (BDOC) and bacterial regrowth potential (BRP) were evaluated in a full scale DWDS and bench tests in South China. A significant correlation between BRP and AOC in both water treatment processes (WTP) and DWDS was obtained. For BRP and BDOC, the correlation was more significant in WTP than in DWDS. Both AOC and BRP were significantly correlated with UV_{254}, total organic carbon (TOC), and heterotrophic plate count (HPC) ($p < 0.01$), whereas BDOC was only significantly associated with $UV_{254,}$ temperature and chlorine residual ($p < 0.01$). Through a bench test, when chlorine was higher than 0.5 mg/L, the HPC level was low and AOC concentration almost unchanged. On contrary the HPC level increased quickly and declined slightly, with chlorine lower than 0.15 mg/L, which was in accordance with the large amount of biological stability data obtained from DWDS. Through another bench test, the HPC level was positively correlated to AOC concentration and when AOC was below 135 μg/L, the growth rate of HPC was low, which was verified by the analysis of biological stability data from DWDS.

1. Introduction

Bacteria in drinking water, especially opportunistic pathogens, are of growing concern. Water-borne pathogens in DWDS can cause many types of diseases such as acute gastro-intestinal disorder (Wang et al., 2012, 2014a; Huang et al., 2014). Unwanted or excessive bacterial growth in DWDS can cause deterioration of microbial water quality during storage and transport (Prest et al., 2016). Growth of bacteria on the inner pipe surface leads to the biofilm formation, deteriorate water quality, and induce public health issues (Wingender and Flemming, 2004; Ohkouchi et al., 2011; Park et al., 2012; Liu et al., 2013). Biological stability, also known as the tendency of microbial growth supported by water and/or contact material, has been extensively investigated for years

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(Liu et al., 2013). Greater biological stability could be achieved with fewer nutrients present in water (Lai et al., 2006; Lou et al., 2012; Van der Kooij et al., 1989). One approach to assess biological stability is to quantify bacterial biomass (colony-forming units or CFU), such as bacterial regrowth potential (BRP) (Sathasivan, 1999). Another alternative suggestion is to quantify the changes of nutrients present in water, for instance assimilable organic carbon (AOC[1]) (van der Kooij et al., 1982), biodegradable dissolved organic carbon (BDOC) (Servais et al., 1987) in Table 1. Microbial growth in distribution systems is affected by water quality variations and multiple interactions between water, biofilms and sediments. Number of methods has been developed recently to improve the assessment and monitoring of the biological stability of drinking water (van der Kooij et al., 2015; Prest et al., 2016).

In particular, AOC has been considered as an indicator that is directly related to bacterial regrowth during water distribution (Yu et al., 2011; Lautenschlager et al., 2013; Wang et al., 2014b). AOC typically comprises only a small fraction (0.1%–9.0%) of total organic carbon (TOC) (Escobar and Randall, 2001). This is determined by monitoring the changes of organics using two different bacterial species (*Pseudomonas fluorescens* P17 and *Spirillum* NOX) (van der Kooij et al., 1982; Van der Kooij and Hijnen, 1984), which have different substrate specificities. The AOC measurement is performed with specific organics such as acetate and oxalate as the sole substrate, which may lead to different yield of biomass depending on bacterial strains. The AOC level of 10 μg C/L or less was proposed as the indicator for biologically stable drinking water in Netherlands (Van der Kooij, 1992a, 1992b). BDOC is the fraction of DOC that can be assimilated and/or mineralized by heterotrophic bacteria. Biodegradability of DOC is determined through measuring the DOC consumption by microorganisms (Servais et al., 1987). BDOC has been routinely used in water industry laboratories to indicate the quality of drinking water and also to serve as a measure of biological stability (Huck, 1990; Kaplan and Rice, 1994: van der Kooij and van der Wielen, 2013). Some reported values of BDOC and AOC used as measures of biological stability are compared in Table 2.

Despite their prevalence, these indicators are still limited by the assumption that bacterial growth is associated only with organic carbon sources (van der Kooij, 2000). The complex interactions (competition, antagonism, symbiosis, and commensalism) among the diverse bacterial strains in actual environments are not resolved by the AOC assessment (Sathasivan, 1999). Therefore, BDOC and AOC may not sufficiently and efficiently represent the actual organic carbon levels and bacterial growth in water. The BRP method relies on the total bacterial count of a water sample to indicate biological stability (Sathasivan, 1999). BPR is achieved by inoculating water samples with indigenous bacteria (Dixon et al., 2012), followed by the incubation and the measurement of total bacteria counts on R_2A agar. Indigenous bacteria can utilize a much broader and diverse kinds of organic carbon than a single pure culture and it enables a more realistic interpretation of the actual microbial activity (Hammes and Egli, 2005).

Water is generally recognized as relatively biologically stable if its AOC concentration ranges from 50 to 100 μg/L (LeChevallier et al., 1993; LeChevallier et al., 1996). In order to maintain water in DWDS biologically stable, it must have an appropriate level of chlorine residual (LeChevallier et al., 1996) or 10–20 μg/L of AOC concentration without chlorine (van der Kooij, 2000).

Heterotrophic plate count (HPC) was often used as an indicator of the drinking water quality (Uhl and Schaule, 2004; Liu et al., 2015) (Franson, 1995; WHO, 2011). The limit was set to 500 CFU/mL for the HPC (R_2A) 7-day incubation. Escobar et al. (Escobar and Randall, 2001) showed a strong positive association between the HPC and AOC, and a weak positive correlation between the HPC and BDOC. Carter et al (Carter et al., 2000). demonstrated no significant correlations between AOC and HPC; while Zhang and DiGiano (Zhang and DiGiano, 2002) observed weak negative to weak positive correlations among these factors. For which, it indicated that a high residual chloramine (>2 mg/L) could effectively repress microbial activity in waters even at high AOC levels. The reason for different results achieved by different researchers was that the interaction between the promotion of nutrients to support bacterial regrowth and the inhibition of chlorine residual to limit bacterial regrowth need to be considered together.

No existing method is capable of reflecting the biological stability completely. In China, most research studied on AOC levels in WTP and DWDS(W.J. Liu et al., 2000), few studies took BDOC and other indicators to reflect biological stability of drinking water based on a large amount of biological stability continuous monitoring data. Furthermore, few studies compared the interrelationship or correlations between these different evaluation methods of biological stability based on monitoring data in previous research. We performed a comparative study on biological stability of a full scale water distribution system in south China using AOC, BDOC, and BRP indicators and explored the correlations between them. The research is expected to (i) evaluate the biological stability in a full-scale DWDS; (ii) explore the correlation among BRP, AOC, BDOC and water quality; (iii) reveal the interaction mechanism among AOC, HPC and chlorine and disinfectant effect on bacterial regrowth in DWDS.

2. Materials and methods

2.1. Preparation of carbon-free materials

Carbon-free bottles and vials were prepared using the method of Hammes (Hammes et al., 2010). All glassware was first washed with detergent and rinsed three times with deionized water. Then, the glassware was submerged overnight in 0.2 N HCl and subsequently rinsed with deionized water again and then air dried. Finally, the bottles were baked in a Muffle furnace at 500 °C for at least 6 h. Teflon-coated screw caps for the glassware were washed and treated with 0.2 N HCl.

2.2. Layout of full-scale drinking water treatment plant and sampling sites

As is shown in Fig. 1, effluent water samples collected from treatment train of two water treatment plants (WTPs) and twelve sampling sites of the water distribution system in south China were chosen. The first WTP (WTP1) is conventional (i.e., pre-ozonation coagulation, flocculation, filtration, treated water) and the second WTP (WTP2) employs advanced processes (i.e., pre-ozonation coagulation, flocculation, filtration, ozonation and bacterial activated carbon (BAC)-filtration treated water). The operational parameters of two WTPs were shown in Table S1. Twelve sampling sites of the water distribution system were chosen. The sites 1 to 5 were from the conventional WTP1, while the sites 6 to 12 were from the advanced WTP2. Characteristics of sampling sites in DWDS can be seen in Table S2. Raw water and the effluent of the pre-ozonation contact tank (a horizontal sedimentation tank), sand

[1] Abbreviations AOC: assimilable organic carbon; BDOC: biodegradable dissolved organic carbon; BRP: bacterial regrowth potential; HPC: heterotrophic plate count; CFU: colony-forming units; TOC: total organic carbon; DWDS: drinking water distribution system; WTP: water treatment plant; RW: raw water; POCR: pre-ozonation contact reactor; CST: coagulation sedimentation tank; SF: sediment filtration; OCR: ozonation contact reactor; BAC: bacterial activated carbon; FW: finished water.