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Q3 Characterization of bacterial community dynamics in a 2 full-scale drinking water treatment plant

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ABSTRACT

Understanding the spatial and temporal dynamics of microbial communities in drinking water systems is vital to securing the microbial safety of drinking water. The objective of this study was to comprehensively characterize the dynamics of microbial biomass and bacterial communities at each step of a full-scale drinking water treatment plant in Beijing, China. Both bulk water and biofilm samples on granular activated carbon (GAC) were collected over 9 months. The proportion of cultivable cells decreased during the treatment processes, and this proportion was higher in warm season than cool season, suggesting that treatment processes and water temperature probably had considerable impact on the R2A cultivability of total bacteria. 16s rRNA gene based 454 pyrosequencing analysis of the bacterial community revealed that *Proteobacteria* predominated in all samples. The GAC biofilm harbored a distinct population with a much higher relative abundance of *Acidobacteria* than water samples. Principle coordinate analysis and one-way analysis of similarity indicated that the dynamics of the microbial communities in bulk water and biofilm samples were better explained by the treatment processes rather than by sampling time, and distinctive changes of the microbial communities in water occurred after GAC filtration. Furthermore, 20 distinct OTUs contributing most to the dissimilarity among samples of different sampling locations and 6 persistent OTUs present in the entire treatment process flow were identified. Overall, our findings demonstrate the significant effects that treatment processes have on the microbial biomass and community fluctuation and provide implications for further targeted investigation on particular bacteria populations.

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Introduction

Drinking water treatment plants (DWTPs) produce potable water that meets public health regulations from natural water sources by a series of treatment processes, typically including coagulation, sedimentation, filtration and disinfection. These

treatment processes result in profound changes in the physicochemical and biological profiles of the raw water (Au, 2004; Chen et al., 2007). The stable performance of the treatment processes is crucial to the safety of the treated water and the microbial communities in drinking water are particularly important for public health because it is directly

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linked to the occurrence of pathogens. As microbial quantity and composition varied spatially and temporally in drinking water systems (DWSs) and sometimes the temporal patterns were striking in both treated and untreated water (Sharp et al., 2006; Henne et al., 2013; Isaac-Renton et al., 1996), end-product monitoring alone is inadequate to keep the high level of confidence in drinking water safety. Holistic characterization of microbial features in DWTPs is necessary.

Treatment processes such as coagulation, sedimentation and filtration can physically remove some of the microorganisms. Disinfection inactivates most of the microorganisms, but those that can endure the disinfection stress are then transported to distribution networks and proliferated there even at a low organic nutrient concentration (Boe-Hansen et al., 2002; Liu et al., 2002; Lu et al., 2014). Various parameters have been developed to control the biological quality, among which the total cell count is one of the most widely used parameters (Carter et al., 2000; Chen et al., 2007; LeChevallier et al., 1991). Cultivation-based heterotrophic plate count (HPC) method has a long history to be recommended in most guidelines, but it detects only a small fraction of the total microbes (Allen et al., 2004; Bartram et al., 2003). Flow cytometry (FCM) coupled with nucleic acid targeting stains has been recently developed and proven to be an sensitive tool for measuring the total cell concentration in water. The two methods can be integrated to elucidate the dynamic of cultivable and uncultivable microbial biomass in DWSs (Hammes et al., 2008).

In terms of community composition, 16S rRNA gene based sequencing revealed that DWSs harbored diverse microbes, including some opportunistic pathogens and disinfectant resistant bacteria in the distribution networks (Pinto et al., 2012; Berry et al., 2006; Holinger et al., 2014; Hwang et al., 2012; Lautenschlager et al., 2014; Lin et al., 2014; Mi et al., 2015). The microorganisms that survived after water treatment processes are considered to be an important source for the potential pathogens at tap faucets. Moreover, the seeded bacteria in some bio-enhanced filters that used to remove specialized pollutants posed a potential risk of leaking from filter biofilms (Zhang et al., 2013).

Some studies concurred that filtration and disinfection had more significant effects on the microbial community compared with other processes, such as coagulation and sedimentation, and that the biofiltration process may determine the characteristics of the downstream microbiome (Holinger et al., 2014; Kwon et al., 2011; Pinto et al., 2012; Wang et al., 2013). Meanwhile, another study showed no major changes occurred after sand filtration (Eichler et al., 2006). The discrepancies found among studies may result from different factors such as study areas, treatment process chains, time scale of sampling and sequencing methods. The temporal fluctuation is a critical consideration for rigorous statistical tests and high validity of information on the processes. However, longitudinal surveys on the dynamics of microbial communities through treatment processes were still limited.

In the present study, the temporal and spatial changes in microbial communities of a full-scale drinking water treatment plant (DWTP) in Beijing, China, were investigated over 9 months. HPC, FCM and 454 pyrosequencing were used to measure the microbial biomass and bacterial compositions of

both bulk water and granular activated carbon (GAC) biofilms. An in-depth characterization of the microbial dynamic patterns in a DWTP was conducted, the results of this study may help to extend our knowledge about the microbial quality of water in DWTPs. The primary objective of this work was to determine (1) how the treatment processes and the temporal variation contribute to the microbial biomass and the bacterial community structure, (2) the influence of GAC biofilm on the water microbiology and (3) the distinct and persistent bacteria that present throughout the treatment processes.

1. Materials and methods

1.1. Drinking water treatment processes and sampling schedule

The DWTP monitored in this study produces 60% of the drinking water of Beijing, China. During the sampling period, its water source consists of two reservoirs (Miyun reservoir in Beijing and Huangbizhuang reservoir in Hebei) as well as groundwater from Huairou aquifer. The average volume mix ratio of these water sources is 4:1:2. The treatment processes include pre-chlorination, coagulation, clarification and coal-sand dual media filtration as the conventional treatment and GAC filtration as the advanced treatment (Fig. 1). The GAC tanks are backwashed every 6 days. Free chlorine is added to the GAC effluent at a concentration of 1.2–1.8 mg/L for 5 hr. 0.2 mg/L ammonia is added post clear well to produce a chloramine residual of 0.7–0.8 mg/L in the distribution system.

Samples were collected in 6 months over a period of 9 months in 2012 (May, August, October, November and December) and 2013 (January). The pre-chlorinated raw water (RW), the coal-sand filter effluent (SE), the GAC tank effluent (CE) and the finished water (FW) were water samples and the GAC particles (CB) were biofilm samples. The GAC particles were taken from the top of the filter tank. Samples were collected in sterile bottles, which were taken to the laboratory within 4 hr. Water quality parameters were listed in Appendix A Table S1.

1.2. Heterotrophic plate count

1-mL aliquots of ten-fold serial dilutions of each water sample were mixed with 20 mL R2A agar (Difco, BD, USA) and incubated at 20°C for 7 days. All HPC determinations were performed in triplicate.

1.3. Total cell concentrations measured by flow cytometry

Total cell concentrations were determined according to the method introduced by Hammes et al. (2008) with a Cell Lab Quanta SC flow cytometer (Beckman Coulter, Inc., Brea, CA, USA). The total cell concentration of all the samples should be maintained between 3×10^3 and 2×10^6 cells/mL.

1.4. DNA extraction

10–40 L of bulk water were filtered through a 0.22- μ m pore size membrane (47 mm diameter, Millipore, USA) with a 90-mm Filter Holder (Millipore, USA). For GAC biofilm samples, about

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