

Enhancing wastewater-based epidemiology of pathogenic *Campylobacter*

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SUMMARY

The global pandemic of COVID-19 has raised great concerns of public health. Fast and efficient surveillance methodology is urgently required to strengthen the ability of infectious disease monitoring. Wastewater-based epidemiology (WBE) has shown great potential and made great contributions in disease monitoring. Unlike clinical testing, WBE can reveal disease information on asymptomatic and presymptomatic individuals who are shedding pathogens in the community, thus making it a powerful tool for early warning and timely intervention of infectious disease. Pathogenic *Campylobacter* is one of four major causes of diarrhoeal diseases and is considered as the most common bacterial cause of human gastroenteritis worldwide. However, the existing surveillance of *Campylobacter* infection still relies on clinical report, which is hindsight, and the lack of research on *Campylobacter* in wastewater has limited the application of WBE in its timely monitoring.

To strengthen the *Campylobacter* surveillance systems through WBE study, we will focus on three main objectives. First, to develop a fast and accurate quantification methodology based on multiplex qPCR for *Campylobacter* in wastewater. Two specific species, *Campylobacter jejuni* (*C. jejuni*) and *Campylobacter coli* (*C. coli*), are selected as targets because they are most frequently related to illness, accounting for 80 to 85% and 10 to 15% infections, respectively. *Campylobacter sputorum* (*C. sputorum*) will be used as an internal sample processing control to correct the underestimation of target species induced by low DNA recovery. In addition, *C. sputorum* assay can also be used

as an internal amplification control to exclude the presence of PCR inhibitors.

Secondly, laboratory-scale sewer reactors including rising main (RM) and gravity sewer (GS) reactors will be employed to evaluate the in-sewer persistence of three *Campylobacter* species by using the established multiplex qPCR and each simplex qPCR assay. The decay rates of each species and different qPCR detection methods are compared to provide further instruction for accurate estimation of *Campylobacter* prevalence in wastewater.

And lastly, impactor factors such as temperature, pH and nutrient of wastewater, and the existence of biofilms will be investigated to delineate the decay of *Campylobacter* in wastewater.

In conclusion, this study will develop and standardize a fast and accurate quantification methodology for *Campylobacter* analysis in wastewater. The evaluation results of the in-sewer persistence and of the environmental impact factors can largely support the back-estimation, and correct the underestimation of *Campylobacter* prevalence in a community, thus can highly strengthen the existing WBE surveillance system for pathogenic *Campylobacter* monitoring.

FINDINGS TO DATE

Multiplex qPCR method for quantifying three *Campylobacter* spp. has been established. The performance of the internal sample processing control for wastewater sample is being evaluated.