A QA/QC approach for implementing SARS-CoV-2 wastewater-based epidemiology

Md Ariful Islam Juel, Visva Bharati Barua, Mariya Munir

Abstract

The world is experiencing an unprecedented situation due to the outbreak of a novel coronavirus which has already caused about a million death toll, as well as social and economic disruptions. Isolation of the COVID-19 infected patients is one of the effective ways to prevent transmission of this virus. However, the person who already is carrying SARS-CoV-2 virus may not be symptomatic and getting tested but can continuously transmit virus to others. Wastewater based epidemiology (WBE), a new line of research, is becoming popular for monitoring SARS-CoV-2 in the local community and college dorms as it can provide the data of both symptomatic and asymptomatic COVID-19 patients cumulatively and serves as an early warning tool prior to a possible outbreak in a community by correlating COVID-19 wastewater testing data and public case counts. However, a precise and accurate viral copies quantification in wastewater and accurate data reporting are pre-requisites for making that tool successful. In this paper, we aimed at the quality control on the WBE focusing on the quantification methods including sample concentration, RNA extraction, RT-qPCR methods. A known concentration of Bovine Coronavirus (BCoV), a surrogate of human coronavirus, was seeded to the wastewater prior to the sample processing and quality control of the overall process maintained by monitoring the BCoV recovery throughout the whole monitoring time. An armored Hepatitis G virus (hepG) RNA was used for the quality monitoring of different batch of extracted RNA samples while multiple dilution series of the RNA sample was used for the RT-qPCR inhibition check. Our result found an average of 9 to 15% of BCoV recovery which implies that a significant portion of viruses is lost during the sample processing. On the other hand, hepG reported an average recovery of 40 – 50% indicating a loss of about 50% viruses during the RNA extraction process alone. Virus recovery may be improved by either optimizing or selecting an effective virus concentration method and RNA extraction kit. Most of the samples showed the prevalence of SARS-CoV-2 viruses in wastewater in the range of 1000 to 100000 copies/L which was resulted due to the recent COVID-19 cause surges in the community. In our analysis, about 40% of samples initially tested negative turned into COVID-19 positive while rerunning RT-qPCR of these same samples with 50% dilution, which indicates that RT-qPCR inhibition should continuously be checked during the surveillance period for the accurate result. Data reproducibility should be checked by reprocessing randomly selected samples that were previously processed. In addition, the concentration of SARS-CoV-2 quantified from the wastewater may be corrected based on the BCoV or other surrogate virus recovery percentage before calculating the correlation between wastewater data and public health data.