Title:

Detection of SARS-CoV-2 RNA from two wastewater treatment plants of Nepal

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Abstract:

There have been several studies conducted throughout the world presenting the applicability of wastewater-based epidemiology (WBE) as an alternative diagnostic tool for the mass detection of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) from environmental samples including wastewater. This study reported the presence and reduction of SARS-CoV-2 at two wastewater treatment plants (WWTPs) in Nepal between July 2020 and February 2021. All together eighty-four grab water samples were collected from two WWTPs [n = 40; WWTP A (oxidation ditch) and n = 28; WWTP B (non-aerated lagoons)]. The positive ratios of SARS-CoV-2 RNA were 53% (21/40) and 61% (17/28) at WWTP A and B, respectively, using at least one of the four quantitative PCR assays tested (CDC-N1, CDC-N2, NIID_2019-nCOV_N, and N_Sarbeco). The highest incidence of SARS-CoV-2 RNA was obtained from the influent of both WWTPs. The highest concentration was observed for an influent sample of WWTP A (5.5 ± 1.0 $\log_{10}$ copies/L) using the N_Sarbeco assay. $\log_{10}$ reduction values (LRVs) of SARS-CoV-2 RNA at WWTP B (0.93 ± 1.45 $\log_{10}$) were significantly higher than those of the crAssphage (-0.40 ± 1.08 $\log_{10}$) ($p < 0.05$), proposing that SARS-CoV-2 was resistant to wastewater treatment. One influent sample of WWTP A tested positive for N501Y variant using the variant-specific qPCR, highlighting a need for further typing of water samples of other variants. Detection of SARS-CoV-2 was observed before, after, and during the increased number of clinical cases, suggesting that WBE has an excellent opportunity to emphasize disease estimation in developing countries. Moreover, due to a strong correlation with that of SARS-CoV-2 and abundance in wastewater, crAssphage can be propose the indictor of SARS-CoV-2.
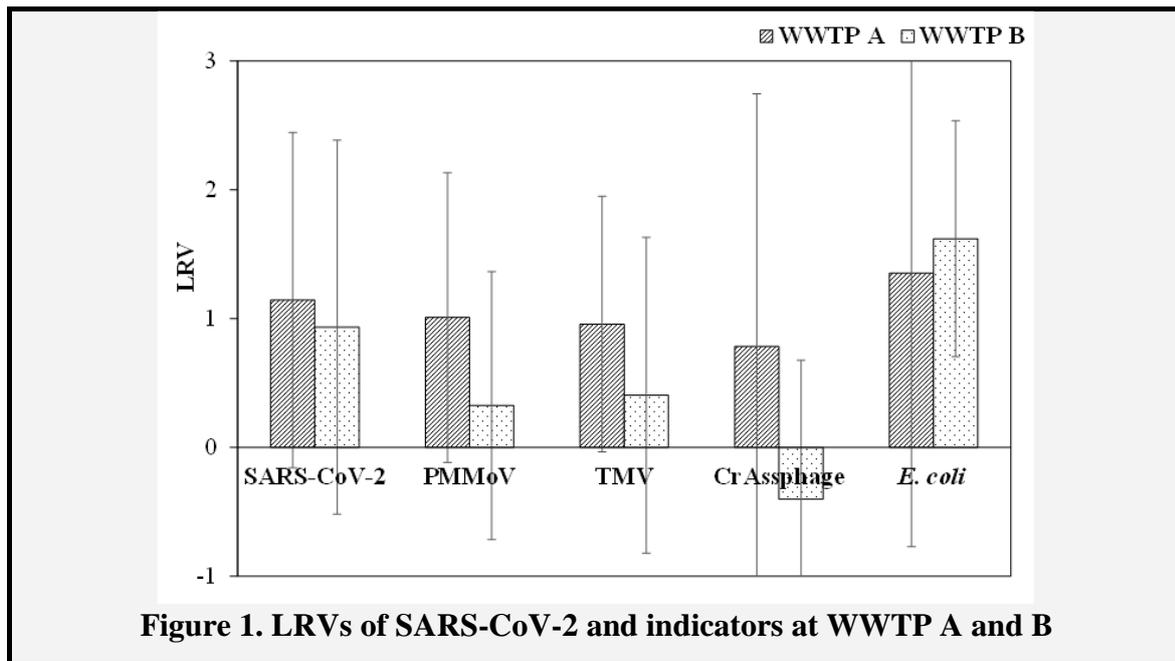
Figure 1. LRVs of SARS-CoV-2 and indicators at WWTP A and B