SCIENCE FOR ENVIRONMENT POLICY

Waste-water monitoring could help predict and prevent COVID-19 transmission

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Contact:
Dr Warish Ahmed,
Warish.Ahmed@csiro.au

People suffering from COVID-19 — whether symptomatic or not — shed the SARS-CoV-2 virus into their faeces. Consequently, techniques to monitor untreated wastewater for the presence of SARS-CoV-2 genetic material are being trialled worldwide to note occurrence across communities. To find out more, Australian scientists implemented a waste-water-based COVID-19 detection technique in the city of Brisbane, Australia, in 2020, to help predict and prevent the spread of the disease.

The COVID-19 pandemic is a public health emergency, requiring effective monitoring of infection levels and pathways in communities to mitigate the spread of the COVID-causing SARS-CoV-2 virus. In February 2020[1], the virus was found in wastewater collected at Amsterdam's Schipol Airport in The Netherlands. This study was the first to identify SARS-CoV-2 viral ribonucleic acid (RNA) in faeces, but the findings were not correlated to known positive COVID-19 cases in the community. The researchers suggested that waste-water-based disease monitoring could aid effective surveillance of the disease (as previously demonstrated for other disease-causing viruses such as poliovirus, norovirus and hepatitis A virus) — and needed to enable early warning of disease outbreaks.

Numerous studies worldwide have since detected SARS-CoV-2 RNA in untreated wastewater, sometimes before the first reported clinical diagnoses in the region[2]. To strengthen the EU's preparedness for public health emergencies such as the pandemic, the European Commission recently adopted a recommendation[3] to promote wastewater monitoring activities in Member States and establish systematic surveillance of SARS-CoV-2 and its variants in EU wastewaters, highlighting the importance of waste-water monitoring as a complementary approach to COVID-19 testing.

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This study builds on a preceding proof-of-concept study[4] conducted on wastewater in Brisbane in 2020, and provides insight into the development of methodological and molecular techniques to reliably detect and quantify SARS-CoV-2 RNA in waste-water samples from a large community.

The researchers collected 63 samples of untreated sewage from three waste-water processing locations for the Brisbane population between 24 February and 1 May 2020. They extracted SARS-CoV-2 from these samples and counted the number of RNA viral copies. An estimate of infection was calculated based on the number of RNA viral copies in the wastewater. The researchers used three assays (types of test), two targeting the N gene of the viral RNA (CDC N1 and N2), and one targeting the E gene (Sarbeco_E). COVID-19 caseload data was obtained from Queensland Health, split by hospital and health service.

SARS-CoV-2 RNA was detected in archived waste-water samples from a south Brisbane waste-water treatment plant in late February 2020, up to three weeks before the first cases were reported in that catchment. A total of 21 samples were positive for SARS-CoV-2, ranging from 135 to 11 992 gene copies (GC) per 100 milliliters (ml) of wastewater. Of the 21 positive results, 19 were achieved using the CDC N1 assay and two using the E_Sarbeco assay, with no samples testing positive using the CDC N2 assay. The N1 assay, therefore, demonstrated the greatest sensitivity for SARS-CoV-2 detection.

Positive samples were detected from two of the three waste-water treatment plants, with samples generally testing positive during the period with highest caseload data. The peak in daily cases occurred in late March; however, active case numbers were only available from 9 April, after the peak in daily cases, leaving only three days with data overlap with the quantified SARS-CoV-2 detections in wastewater. For those days the ratios of gene copies per 100 mL (GC/100 mL) to number of active cases were 1.7, 1.8, and 2.9. Overall, the decline in prevalence of RNA in wastewater matched the tapering off of the first epidemic wave in Brisbane, but numbers of RNA viral copies showed no correlation with daily case numbers. The researchers suggest this could be due to the low total number of COVID cases in Brisbane compared with other locations, and that the general variation in SARS-CoV-2 prevalence between waste-water catchments could be due to large differences in catchment size, pipe networks, water characteristics and hydraulic retention times.

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The detection of SARS-CoV-2 RNA in wastewater — found alongside limited reported clinical cases — demonstrates the potential of waste-water surveillance as an early warning system for disease outbreaks. Waste-water monitoring could assist COVID-19 management and health messaging efforts, say the researchers, for example by identifying hotspots for pop-up testing clinics to minimise further waves of infection.

1. Lodder, W. and de Roda Husman, A., (2020) SARS-CoV-2 in wastewater: potential health risk, but also data source. *The Lancet Gastroenterology & Hepatology*, 5(6): 533-534.

2. RNA was detected in waste-water samples in Milan and Turin in December 2019, for example, but the first documented case in Italy was reported in February 2020, indicating that citizens were shedding SARS-CoV-2 RNA in faeces before cases were reported by public health units: La Rosa, G., Iaconelli, M., Mancini, P., Bonanno, G., Ferraro, G.B., Veneri, C., Bonadonna, L.,Lucentini, L., and Suffredini, E., (2020) First detection of SARS-CoV-2 in untreated wastewaters in Italy. *Science of The Total Environment*, 736: 139652.

3. EC Recommendation of 17.3.2021 on a common approach to establish a systematic surveillance of SARS-CoV-2 and its variants in wastewaters in the EU.

4. Ahmed, W., Angel, N., Edson, J., Bibby, K., Bivins, A., O'Brien, J., Choi, P., Kitajima, M., Simpson, S., Li, J., Tscharke, B., Verhagen, R., Smith, W., Zaugg, J., Dierens, L., Hugenholtz, P., Thomas, K. and Mueller, J., (2020) First confirmed detection of SARS-CoV-2 in untreated wastewater in Australia: A proof of concept for the wastewater surveillance of COVID-19 in the community. *Science of The Total Environment*, 728: 138764.