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Monitoring of SARS-CoV-2 in Porto Alegre's Sewage Sludge

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Monitoring SARS-CoV-2 virus in the population is of great importance to assist governmental decisionmaking and to contain and mitigate damage caused by the COVID-19 epidemic. However, the scarcity of rapid tests and inputs for molecular diagnostic tests make mass testing of the population unfeasible. A viable alternative is the detection and quantification of the virus in sewage networks. SARS-CoV-2 is released into the feces of infected patients, including asymptomatic patients, and have already been detected in sewage in several studies. It is known that coronaviruses have a greater affinity for solids compared to other nonenveloped viruses. Therefore, extracting the virus directly from the solid and concentrated part of the sewage, the sludge, can be a more effective alternative for environmental monitoring of SARS-CoV-2 than the monitoring of wastewater. To assist in the environmental monitoring of SARS-CoV-2, our work aims to analyze the viral load in sludge samples from the Serraria sewage treatment station, considered the most representative of the city of Porto Alegre. Sludge samples have been collected weekly; the RNA have been extracted using the RNeasey PowerSoil Total RNA kit (Qiagen), and the presence of the virus have been quantified by RT-qPCR, using the US CDC's primers N1 for the virus nucleocapsid, while the human RNase P gene will be tested as a control. The methodology is currently being tested; the weekly sampling and extractions since December 1, 2020 have been done successfully and the viral RNA in the tested samples have varied from $2,15x10^6$ to $0,358x10^6$ copies L^{-1} . The quantifications have shown that the number of virus copies in the sludge can be up to 20 times higher than found in wastewater and have a strong correlation coefficient (R²=0,877) with the week's moving-average of new Covid cases from the city; however, we are still working on the human RNase P control to validate the data. In parallel, reactions for viral quantification have also been made using reverse transcriptase and Tag enzymes produced at the Centro de Biotecnologia from our University (UFRGS). If these latter enzymes prove feasible, environmental monitoring of SARS-Cov-2 will become more economically accessible and can thus be widely implemented. Even after mass vaccination and control of the pandemic, implementation of this surveillance could be used to monitor the rise of new SARS-Cov-2 variants and foresee a possible new outbreak.

Palavras-chave: Sars-Cov-2; Sludge, Wastewater, Surveillance, Sewage.