TITLE: MONITORING OF SARS-COV-2 IN PORTO ALEGRE'S SEWAGE SLUDGE

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ABSTRACT:
Monitoring SARS-CoV-2 virus in the population is of great importance to assist governmental decision-making and to contain and mitigate damage caused by the COVID-19 epidemic. However, the scarcity of rapid tests and inputs for molecular diagnostic tests makes 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 has been detected in the sewers since the start of the pandemic.
It is known that coronaviruses have a greater affinity for solids compared to other non-enveloped 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 are being collected weekly; the RNA is being extracted using the RNeasey PowerSoil Total RNA kit (Qiagen), and the presence of the virus is being quantified by RT-qPCR, using the US CDC's primers N1 for the virus nucleocapsid.
The weekly sampling and extractions since December 1, 2020 have been done successfully and although the quantifications have been showing that the number of virus copies in the sludge are higher than the ones found in the wastewater. Although they have not presented a strong correlation with epidemiological data from the city, we are still working on modifications on sample collection and use of different controls to normalize the data.
Reactions for viral quantification are also being made using reverse transcriptase and Taq enzymes being produced at 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.
In parallel the first attempts to identify SARS-CoV-2 variants in sludge samples through PCR have been successful, the aim is to still validate the data through sequencing of the samples that can also provide more information. The identification of variants in sludge samples may be of great importance for monitoring of the pandemic and possible future outbreaks of new variants.

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