Wastewater Based Epidemiology applicability in a real scenario at São José do Rio Preto- highlights in a year-long experience with SARS-CoV-2 community surveillance

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SARS-CoV-2 is the major health concern globally. Undergoing a pandemic scenario, the investigation of the effluents generated by a given population can provide important data for the correct management of the sanitary emergency. Wastewater-based epidemiology is a methodology previously used in the analysis of drugs, medications, emerging contaminants and later applied in the analysis of biomolecules, pathogens and more recently on pathogens with sustained transmission in each community. This approach follows an investigative, multidisciplinary, and complementary line to clinical molecular tests (applied in an individual context) and the interest in community data on the persistence of a disease and seeking to fill gaps have made this methodology widespread recently with the massive emergence of studies on this topic, as a result of COVID-19 pandemic. So, the aim of this study the detection and quantification of SARS-CoV-2 RNA in the wastewater during the period of one year and correlate the findings with the epidemiological data. For this, the raw sewage samples were collected in the Wastewater Treatment Plant (WWTP) of the São José do Rio Preto city (São Paulo, Brazil). From 15 Jul 2020 to 16 Jul 2021, 24-hour samples were collected three times per week from the HACH Sigma SD900 AWRS refrigerated automatic sampler. The equipment was adjusted to collect one sample for every 3000 m^3 that came to processing (36 sampling events/day normally). The raw sewage samples were decanted and filtered under vacuum using PALL CORPORATION 47 mm 1 µm glass fiber membrane. After, the samples were centrifuged at 40.000xg during 3 hours at 4°C and the pellets were resuspended in TE buffer (pH 7.6). RNA was extracted by TRIzol. Viral RNA was detected and quantified by Real time PCR. The sets of primers and probe (2019-nCoV RUO Kit) as well as the positive control (2019-nCoV_N_Positive Control, 2×10^5 genome copies (gc)/µL) were provided by IDT (Integrated DNA Technologies). Data were collected using QuantStudio 12K Flex platform and analyzed with the own equipment software. The SARS-CoV-2 RNA was detected in 109 of 150 samples- or ~72,6% of the samples. The quantification of SARS-CoV-2 RNA varied between $1{,}35x10^3$ to $2{,}75x10^6$ copies of RNA per Liter of sewage during the period evaluated. Peaks of SARS-CoV-2 positive clinical cases were observed in the epidemiological weeks 29-32 and 35 of 2020 and 10-13, 19-20, 20, 23 and 29 of 2021. At the same time sewage data reports peaks with above 100,000 copies of the SARS-CoV-2 genome copies per Liter in the epidemiological weeks 34, 35, 49, 41, 43, 44, 45, 51 and 53 of 2020 and 21-23 of 2021. These results show evidence that can guide local authorities in implementing restrictive measures and value their effectiveness in real-word scenarios at real-time.

Palavras-chave: Sewage Based Epidemiology; SARS-CoV-2; absolute viral quantification; RT-qPCR

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