Emerging Infectious Diseases: Challenges and Surveillance through Wastewater Monitoring

Razzolini, E.*, Oliveira, J.*, Etchepare, R.*, Possetti , G.** and Vicente, V.*

* Federal University of Paraná
** Sanepar – Paraná Sanitation Company

Highlights:

Emerging Infectious Diseases: Origins, rapid spread, and public health challenges.

Wastewater Surveillance: Methodology for monitoring diseases, including sample collection and genetic sequencing.

Keywords: Emerging Pathogens; Wastewater Surveillance, Environmental Monitoring

INTRODUCTION

Emerging infectious diseases are illnesses caused by previously unknown or uncommon pathogens that arise and spread rapidly, posing a significant challenge to public health. These diseases can originate from various sources, such as viruses, bacteria, fungi, or parasites, and can affect both humans and animals (Morens et al., 2004). Emerging diseases can have substantial impacts on public health and the economy, leading to outbreaks and even pandemics. Notable examples of emerging infectious diseases include severe acute respiratory syndrome (SARS), influenza caused by the H1N1 virus, arboviruses like Zika fever, and more recently, COVID-19 ("COrona VIrus Disease 2019") caused by the SARS-CoV-2 coronavirus (Morens et al., 2004; Morse et al., 2012).

On the global stage, the emergence of infectious diseases has become increasingly frequent. The rapid dissemination of these agents is due to a combination of factors, including environmental changes like deforestation and urbanization, increased interaction between humans and animals, international travel, and intensive agricultural practices (Woolhouse and Gowtage-Sequeria, 2005). Studies have shown that viruses can be excreted in the feces of infected individuals, even in those who are asymptomatic (Wu et al., 2020). Since viral particles can be excreted in feces, quantifying the viral load in domestic sewage has proven to be a promising tool for monitoring the dynamics of emerging infectious diseases, as seen during the SARS-CoV-2 pandemic (Ahmed et al., 2020a; Medema et al., 2020).

This approach has been applied not only in urban areas but also in rural regions and countries across different parts of the world. Quantifying the viral load in domestic sewage can provide important information about the presence and magnitude of infection affecting a population in a specific region, even before cases are clinically diagnosed. This was especially relevant during the COVID-19 pandemic because many cases were asymptomatic or exhibited mild symptoms, making it difficult to fully identify the extent of the virus's spread. This approach is known as wastewater-based epidemiological surveillance, providing an estimate of the collective viral load of a given population.

METHODOLOGY

The methodology implemented was based on the weekly collection of raw sewage samples at the wastewater treatment plants (WWTPs) in the city of Curitiba/Paraná (Figure 1), from March 2021 to April 2023, using automatic samplers for 4 hours in the morning. This was followed by the extraction of the total genetic material from the samples, quantification of the viral load by qPCR, and sequencing of the circulating strains through next-generation sequencing (NGS). Additionally, samples from patients who tested positive for SARS-CoV-2 were occasionally sequenced.
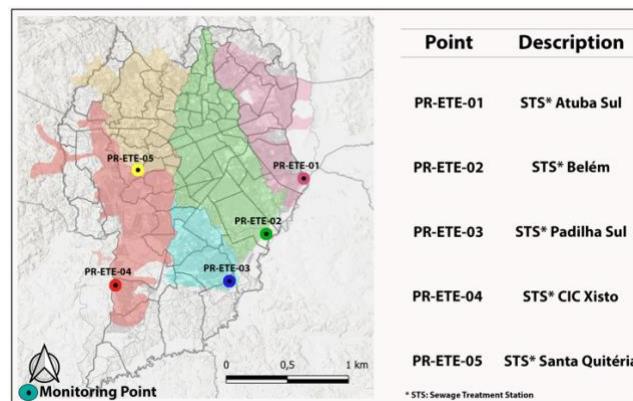

Figure 1: Points of the sewage treatment system in the city of Curitiba where epidemiological monitoring was carried out.

RESULTS AND CONCLUSIONS

The COVID-19 pandemic, even four years after the first reported case in Wuhan, China, still represents a public health situation that demands attention. Despite advances in vaccination and the application of booster doses, new strains with the capacity to cause outbreaks, even with low mortality, are still being reported, especially those related to the XBB subvariant of the Omicron strain.

The city of Curitiba reported an average of 12,726 new cases per month during the pandemic (March 2020 to May 2024). Sequencing of the genetic material present in wastewater identified that the Wild type (A), Alpha, and Omicron strains were the most frequently observed, consistently appearing every month. In June 2021, strains A, Alpha, BA.2*Omicron (BA.2.X), Beta, Epsilon, Gamma, Iota, Lambda, Mu, and Omicron were detected (Figure 2). The absence of sequencing and/or the presence of few identified strains can be attributed to the degradation of genetic material, presence of contaminants, seasonal and geographical variation, as well as the possible occurrence of natural inhibitors present in domestic wastewater samples (Schrader et al., 2012; Saito and Doi, 2021).
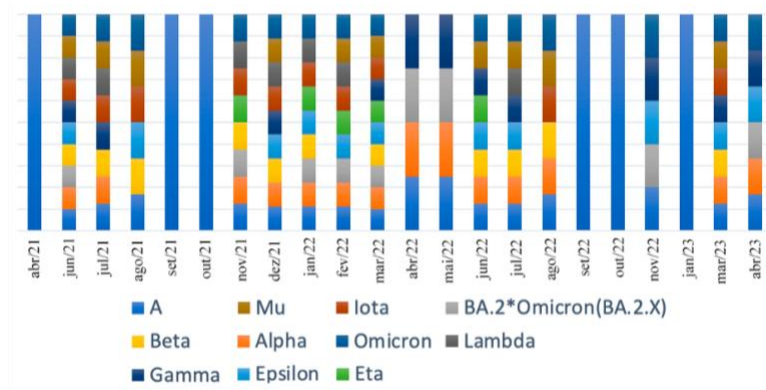
Figure 2: Distribution of SARS-CoV-2 lineages identified during the study.

In addition to the presence of SARS-CoV-2, it was also possible to identify the circulation of other pathogens in the population of Curitiba, including Poliovirus, Monkeypox, Measles, and Enterovirus D68 (Table 1). Furthermore, with the aid of sequencing samples from patients, it was possible to identify the circulation of new strains responsible for peaks in cases in the population (i.e., Eris strain).

In conclusion, continuous monitoring and genetic sequencing are essential to identify new viral strains and understand their dissemination dynamics. This allows for a more effective response to changes in the epidemiological landscape and the protection of public health.

Pathogen	Cq
Measles	21.2028
Enterovirus D68	21.8475
Poliomyelitis	27.5086
MPox	35.1360

Table 1: Cq Values for detection of different pathogens in Real-Time qPCR samples.

ACKNOWLEDGMENTS

REFERENCES

Ahmed, W., Harwood, V. J., Gyawali, P., Sidhu, J. P. S., & Toze, S. (2015). Comparison of concentration methods for quantitative detection of sewage-associated viral markers in environmental waters. Applied and Environmental Microbiology, 81(6), 2042–2049.

Medema, G., Heijnen, L., Elsinga, G., Italiaander, R., & Brouwer, A. (2020). Presence of SARS-Coronavírus-2 RNA in Sewage and Correlation with Reported COVID-19 Prevalence in the Early Stage of the Epidemic in The Netherlands. Environmental Science & Technology Letters, 7(7), 511–516.

Morens, D. M., Folkers, G. K., & Fauci, A. S. (2004). The challenge of emerging and re-emerging infectious diseases. Nature, 430(6996), 242-249.

Morse, S. S., Mazet, J. A., Woolhouse, M., Parrish, C. R., Carroll, D., Karesh, W. B., ... & Daszak, P. (2012). Prediction and prevention of the next pandemic zoonosis. The Lancet, 380(9857), 1956-1965.

Saito, T., & Doi, H. (2021). A model and simulation of the influence of temperature and amplicon length on environmental DNA degradation rates: a meta-analysis approach. Frontiers in Ecology and Evolution, 9, 623831.

Schrader, C., Schielke, A., Ellerbroek, L., & Johne, R. (2012). PCR inhibitors–occurrence, properties and removal. Journal of applied microbiology, 113(5), 1014-1026.

Woolhouse, M. E., & Gowtage-Sequeria, S. (2005). Host range and emerging and reemerging pathogens. Emerging Infectious Diseases, 11(12), 1842-184.

Wu, Y., Guo, C., Tang, L., Hong, Z., Zhou, J., Dong, X., ... & Xu, K. (2020). Prolonged presence of SARS-CoV-2 viral RNA in faecal samples. The Lancet Gastroenterology & Hepatology, 5(5), 434-435.