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# Wastewater Surveillance of *Candida auris* in Belo Horizonte: a tool under development

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**Highlights:**

- *Candida auris* was detected in the sewage of Belo Horizonte, despite no reported clinical cases.
- The method of concentration used significantly impacts the detection of the pathogen.
- Insufficient data is available to determine trends in the activity or transmission of *C. auris*.

**Keywords:** *Candida auris*, methodology; wastewater surveillance.

## INTRODUCTION

The fungus *Candida auris* is a dangerous pathogen that can cause severe systemic infections, especially in long-term healthcare settings. It was first identified in 2009 and has since been reported in over 40 countries (AHMAD; ALFOUZAN, 2021; VILA *et al.*, 2020). The World Health Organization (WHO) and the U.S. Center for Disease Control and Prevention (CDC) have classified it as a significant public health concern and an urgent threat, respectively.

In Brazil, the first case of *Candida auris* was detected in 2020, and it has been under surveillance by the Health System since 2017 (ANVISA, 2020). In the United States, the first reported case was in 2013 (VALLABHANENI *et al.*, 2016), and the number of cases has been increasing significantly. Like other types of Candida, this pathogen has been found in various parts of the human body, including the skin, wounds, blood, sputum, urine, faeces, vaginal secretions, and rectal swab samples(AHMAD; ALFOUZAN, 2021; FORSBERG *et al.*, 2019; PACILLI *et al.*, 2020; WELSH *et al.*, 2017) The fungus has been found in various parts of the human body. Infected individuals may carry it without showing symptoms for over 24 months after exposure (AHMAD; ALFOUZAN, 2021).

Despite the methodological applicability, in practical terms, monitoring indicators without linking them to potential treatment or prevention tools and techniques would serve more as a data collection task than as a solution capable of generating significant impact in protecting the population. While the collection of information is crucial and underscores the relevance of this work, associating surveillance with rapid detection methodologies and treatment intervention systems that actively mitigate the presence of *Candida auris* would be highly valuable.

This study explores the potential use of sewage-based epidemiological surveillance to generate more accurate and comprehensive estimates of *Candida auris* incidence and prevalence in Belo Horizonte.

## METHODOLOGY

Raw and treated sewage samples are gathered weekly from two Wastewater Treatment Plants (WWTP) in Belo Horizonte, catering to 3.5 million people. Composite samples are collected over a 24-hour period at 4°C and then dispatched to the laboratory. The concentration methods were assessed using membrane filtration (30 ml filtered into a 0.45µm sterile membrane), centrifugation (12,000rpm for 20 min at 4°C), and culturing (HiCrome Candida, incubated at 37°C for 48 hours). The DNA extraction was performed using FastKitDNA (MPBiologicals), and the identification process was carried out in accordance with the methodology described in a previous study (BARBER *et al.*, 2023).

## RESULTS AND CONCLUSIONS

Previous studies at the sewershed scale have shown that detecting *Candida auris* in wastewater indicates ongoing outbreaks in the contributing population (BABLER *et al.*, 2023; BARBER *et al.*, 2023). To our knowledge, this study represents the first Brazilian wastewater surveillance effort to monitor *C.auris* in sewage. The initial findings suggest that the marker gene *Candida auris* has been detected in the wastewater of Belo Horizonte despite no reported clinical cases of this particular strain, suggesting a significant gap in the clinical database. This fact highlights the potential of using wastewater surveillance tools to monitor pathogens that pose a public health risk. Overall, *C. auris* was detected in 25% of the raw sewage samples from both WWTPs. However, it is essential to note that the concentration method can affect this frequency. The percentage of positive samples during the period was 27.6% (8 out of 29) for culturing, 29.4% (10 out of 34) for membranes, and 11.8% (2 out of 17) for pellets (Figure 1).

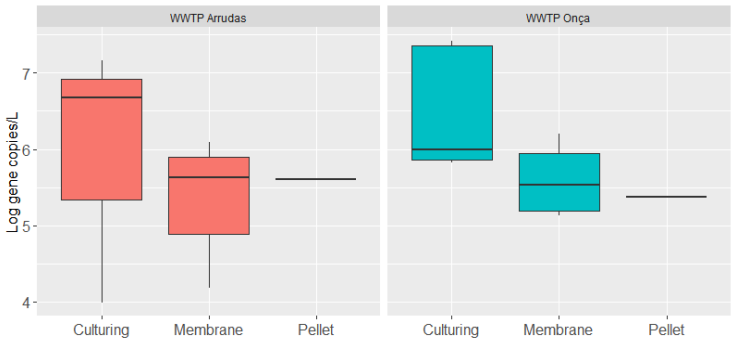


**Figure 1** Concentrations of *C.auris* in $Log_{10}$ copies genes/L in the two sewage treatment plants and in different concentration methods.

The culturing concentrations ranged from 4.9 to 7.4 log10 gc/L, membrane 4.2 to 6.2 log10 gc/L, and pellet 5.3 to 5.6 log10 gc/L. These results align with a previous study(BARBER *et al.*, 2023) that evaluated *C. auris* nucleic acids in sewage pellets and found a range from 4.0 to 7.42 log10 gc/L. This

initial result emphasizes two critical points for epidemiological surveillance. Firstly, directly identifying the *C. auris* gene using qPCR seems less sensitive than the cultivation method. Secondly, there is a high likelihood for *C. auris* to be viable in domestic sewage, even though it has not yet been isolated in the study.

The detection of *C. auris* nucleic acids in wastewater highlights the urgent need for increased clinical testing and reporting capacity. It also demonstrates the potential of wastewater testing to act as a sentinel surveillance tool for emerging pathogens. However, despite the methodological applicability of such monitoring, relying solely on the collection of data without integrating it with treatment or prevention strategies would limit its practical impact on public health. While the information gathered is crucial, its true value lies in pairing it with rapid detection methodologies and intervention systems that can actively mitigate the pathogen's presence. Therefore, the combination of wastewater surveillance with robust treatment and prevention measures is essential to ensure meaningful protection of the population and to enhance the relevance of this work in public health efforts.

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