# PHENOTYPICAL AND MOLECULAR CHARACTERIZATION OF Stenotrophomonas maltophilia STRAINS ISOLATED FROM PATIENTS DURING COVID-19 PANDEMIC FIOCRUZ SANTOS, M.C.S.<sup>1</sup>; MIRANDA, C.A.C.<sup>2</sup>,\*; PASCHOAL, R. P.<sup>3</sup>; VILLAS BÔAS, M.H.S.<sup>4</sup>; BRANDÃO, M.L.L.<sup>1</sup>

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### INTRODUCTION

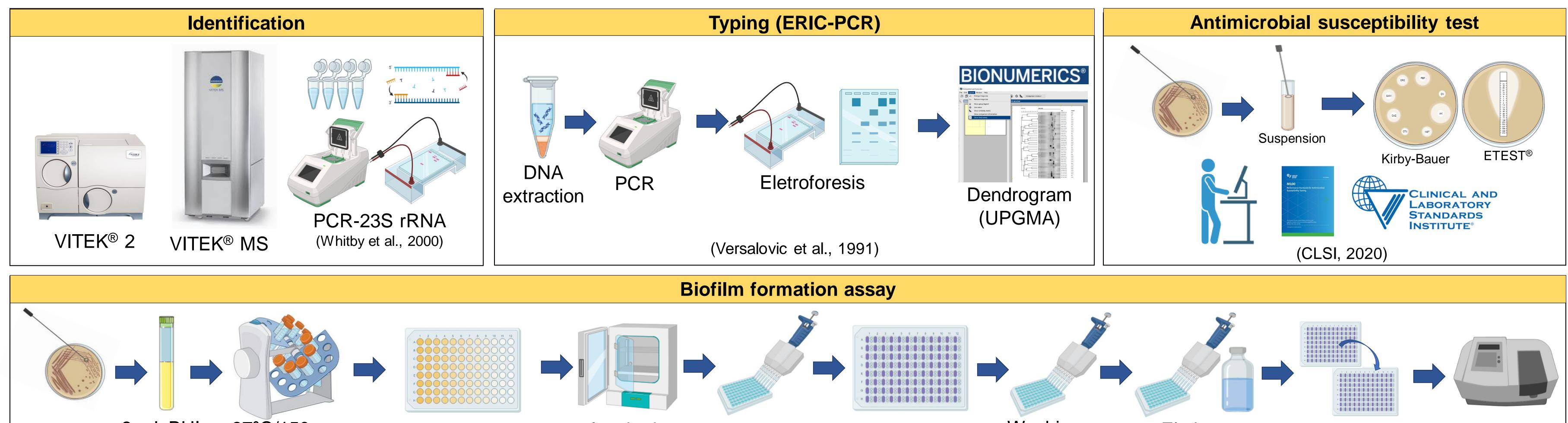
Antimicrobial resistance threatens the effective prevention and treatment of an ever-increasing range of infections caused by bacteria (WHO, 2020). During COVID-19 pandemic, the incidence of bacterial infection in hospitalized COVID-19 patients was high, especially those caused by multidrug-resistant Gram-negative bacteria as *Stenotrophomonas maltophilia* (CHONG et al., 2021). *S. maltophilia* is an emerging multidrug-resistant global opportunistic pathogen, being more commonly associated with respiratory infections in humans. The aim of this study was to characterize *S. maltophilia* strains isolated from hospitalized patients during COVID-19 pandemic.

# METHODOLOGY



February-September/2021 Rio de Janeiro/RJ S. maltophilia (n=9)

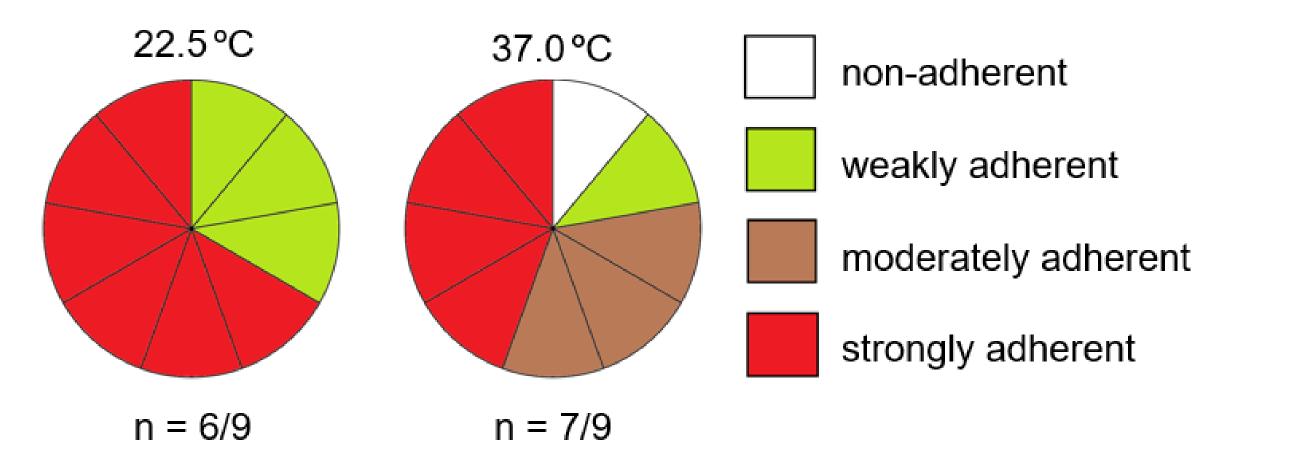
- nine patients (8 males and 1 female) with ages varying from 64 to 86 years;
- two with COVID-19 co-infection, five only with S. maltophilia;
- Clinical specimens included tracheal swab (n=4), blood (n=2), sputum (n=1), lung mass (n=1), and soft tissue fragment (n=1).



3 mL BHI 37°C/150 rpm Washing 96-well polystyrene Incubation Washing Violet crystal Elution Transfer 150 µL to a Absorbance  $H_2O(5X)$ 22.5 and 37°C Ethanol 96% PBS (5X) 0.41%/ 45 min 600 nm plates new microplate (Umeda et al., 2017)

#### **RESULTS AND CONCLUSIONS**

All strains were identified as *S. maltophilia* by VITEK<sup>®</sup>2 and MALDI-TOF (Fig 1) and were positive using 23S PCR. Nine distinct band profiles were obtained by ERIC-PCR (Fig. 1). All strains were susceptible to cefiderocol, minocycline, and resistant to ceftazidime. Five (55.5%) were resistant to ticarcillin-clavulanate and four (44.5%) susceptible, increased exposure. Three (33.3%) were resistant to chloramphenicol and six (66.7%) intermediate. One (11.1%) strain was resistant to trimethoprim-sulfamethoxazole and one (11.1%) to levofloxacin. Six (66.7%) strains were classified as strongly adherent and three were weakly adherent at 22.5°C. Four (44.5%) strains were classified as strongly, three (33.3%) as moderately, one weakly and one non-adherent at 37.0°C (Fig 2).



| -20 | 60 40 | 80 | 100 | Key          | Pacient | Туре     | Gender | Age | Origem               | Date     | MIN | LVX | SXT | Bionumber | MALDI TOF/MS (%)      | Biofilm (22.5°C) B | ofilm (37.0ºC) |
|-----|-------|----|-----|--------------|---------|----------|--------|-----|----------------------|----------|-----|-----|-----|-----------|-----------------------|--------------------|----------------|
|     |       |    | -   | <br>SM008/22 | G       | N-Covid. | М      | 74  | soft tissue fragment | Feb/2021 | S   | I   | S   | I         | S. maltophilia (96.5) | 3                  | 2              |
| _   |       |    | -   | SM009/22     | G       | N-Covid. | Μ      | 74  | blood                | Feb/2021 | S   | S   | S   | V         | S. maltophilia (92.1) | 3                  | 2              |
|     |       |    | -   | SM003/21     | С       | Covid-19 | F      | 68  | tracheal swab        | Sep/2021 | S   | S   | S   | II        | S. maltophilia (96.1) | 3                  | 3              |
|     |       |    | -   | SM004/22     | D       | Covid-20 | М      | 72  | tracheal swab        | Feb/2021 | S   | S   | S   | III       | S. maltophilia (92.1) | 3                  | 3              |
|     |       |    | -1  | SM005/22     | Е       | N-Covid. | Μ      | 77  | tracheal swab        | Feb/2021 | S   | S   | R   | II        | S. maltophilia (99.9) | 1                  | 0              |
|     |       |    | - [ | SM001/21     | А       | N-Covid. | Μ      | 64  | tracheal swab        | Jun/2021 | S   | S   | S   | I         | S. maltophilia (80.1) | 3                  | 3              |
|     |       |    | -1  | SM010/22     | G       | N-Covid. | М      | 74  | blood                | Mar/2021 | S   | S   | S   | V         | S. maltophilia (80.1) | 1                  | 2              |
|     |       |    | -   | SM006/22     | F       | N-Covid. | Μ      | 83  | lung mass            | Feb/2021 | S   | S   | S   | IV        | S. maltophilia (93.5) | 1                  | 1              |
|     |       |    | -   | SM002/21     | В       | N-Covid. | М      | 86  | sputum               | Jun/2021 | S   | S   | S   | II        | S. maltophilia (96.1) | 3                  | 3              |

**Figure 1**. Cluster analysis of *S. maltophilia* strains (n=9) patterns resolved by ERIC-PCR. The dendrogram was evaluated using Dice coefficient and Unweighted Pair-Group Method with Arithmetic mean (UPMGA) with BioNumerics software. Legend: Male (M), Female (F), Susceptible (S), Intermediate (I), Resistant (R), Levofloxacin (LVX), Minocycline (MIN), Sulfamethoxazole-trimethoprim (SXT), Matrix-assisted laser desorption/ionization time-of-flight (MALDI TOF/MS).

The Antimicrobial susceptibility test was similar between strains, neither with they were from patients co-infected with COVID-19 or not. All strains presented different ERIC-PCR profiles, indicating that they were not clonal, and possible have different origins. Considering the necessity to use broadspectrum antibiotics in cases of bacterial infections, especially in cases of co-infection with COVID-19, the resistant found to four classes of

Figure 2. Biofilm formation of *S. maltophilia* strains (n=9) in 96-well polystyrene plates in at 22.5 and 37.0°C.

antimicrobial agents ( $\beta$ -lactam, cephems, folate pathway antagonists, and fluoroquinolones) is worrisome. In conclusion, the continuing monitoring of the antimicrobial susceptibility profile and clonality of *S. maltophilia* is important to understand the epidemiology of this bacteria.

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