

## **SCREENING AND IDENTIFICATION OF MULTIRESTANT BACTERIA IN PATIENTS, HEALTH CARE WORKERS AND HOSPITAL ENVIRONMENT: COMPARATIVE STUDY USING MOLECULAR BIOLOGY TECHNIQUES.**

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Healthcare-associated infections (HAI), especially those caused by multidrug-resistant bacteria (MDR), are a public health problem that needs efficient surveillance programs. HAI can seriously affect patient health, increasing costs and duration of hospitalization. Here, a pilot study for screening and identification of MDR Gram-negative bacteria (GNB) was carried out at the Professor Polydoro Ernani de São Thiago University Hospital (Florianópolis, SC, Brazil). Samples from patients, healthcare workers and high-touch surfaces were collected from two Surgical Inpatient Units (SU-I and SU-II) and the Surgical Center (SC) using sterile swabs. A swab containing Amies medium (MC samples) was seed on MacConkey agar following identification and antimicrobial susceptibility test (AST) performed using Vitek2 kits (BioMérieux). A second dry-swab (MB samples) was collected on the same sites for molecular analysis via Illumina sequencing of the 16S rDNA and PCR detection of  $\beta$ -lactamases genes OXA-23, OXA-51, KPC, CTX-M-1 and NDM-1. The results were visualized using Epiome software (Neopropecta S/A). A total of 136 samples (68 MB and 68 MC) were collected. HAI-related GNB were isolated in 17.6% of MC samples, while sequencing was able to detect high concentrations of these 16S rDNA bacterial sequences in 20.6% of MB samples. The most important hot-spots were: snack room of health care workers, rectal swabs from patients, soap dispensers and faucets in patients bathrooms, bed rails and health care workers hands. Comparison of microbiological and molecular techniques has shown that 71.4% of high concentration samples by sequencing (BM) had positive GNB culture (MC); however sequencing showed the highest bacterial diversity. Vitek2 and 16S rDNA sequencing had identical results for 76.2% of the identified species. The presence of resistance-related genes was mainly identified in samples collected from the snack room of health care workers, nurse station, bandage and medicine rooms workstations, in addition of samples from patients. Samples with sensitive AST profile were also negative for the resistance-related genes; however, 3 MB samples were positive for KPC or OXA-51 genes and presented negative culture for the corresponding MC samples. Finally, the use of molecular biology techniques revealed to be effective for the detection of microbial diversity as well as antibiotics resistance, indicating to be a powerful tool to increase the efficiency in identifying MDR bacteria.

**Key words:** Healthcare-associated infections (HAI), Multidrug-Resistant Bacteria (MDR), Resistance-associated genes, Next Generation Sequencing (NGS).

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