**TITLE**: EVALUATION OF A NEW TRILOCUS SEQUENCE-BASED MULTIPLEX-PCR TO DETECT MAJOR ACINETOBACTER BAUMANNII CLONAL COMPLEXES CIRCULATING IN BRAZIL

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## ABSTRACT

Acinetobacter baumannii is an opportunistic and emergent nosocomial pathogen of increasing interest due to its widespread resistance to antimicrobials. Dissemination of carbapenem-resistant Acinetobacter baumannii (CRAB) is mainly due to the spread of clonal lineages, particularly those included into the clonal complexes (CC) CC1, CC2, CC15, CC25, and CC79. Multilocus sequence typing (MLST) is the current standard for investigating the population structure of bacterial species but as alternative to MLST, PCR based methods can be used for genotyping of A. baumannii and might be more appropriate for strain phylogeny in large-scale epidemiology We evaluated the usefulness of a trilocus sequence-based typing (3LST), a recently modified trilocus sequence-based typing (m3LST) in comparison with the standard MSLT of 7 housekeeping genes as per the Institute Pasteur Scheme. A collection of 78 CRAB isolated from 67 different Brazilian health institutions was submitted to the three methodologies, and concordance rates were calculated. The collection studied included mainly isolates belonging to endemic Brazilian Clonal Complexes (CC1, CC15, CC25 and CC79, n=71, 92.3%) but also singletons sequence types (ST) with low prevalence in our country (ST107, ST113, ST188, ST317, ST584, ST733, n=6; 7.7%). Traditional 3LST was able to correctly assign only 24 strains (concordance of 39.7%), all of them belonging either to CC1 or to non-identified groups. On the other hand, m3LST correctly assigned all the isolates into the main CC responsible for the CRAB dissemination in Brazil. All the singletons ST were not misidentified as prevalent lineages, in both methods. m3LST is a powerful tool to investigate molecular epidemiology of A. baumannii representative of prevalent Brazilian clonal complexes 1, 15, 25 and 79. This technique correctly identified the CC in a large collection of well-characterized CRAB from diversified institutions, including representative strains of ST in low prevalence in the country.

**Keywords:** molecular epidemiology; multiplex-PCR; MLST; Acinetobacter baumannii; 3LST

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