TITLE: EVALUATION OF BIOFILM FORMATION IN STRAINS *MYCOBACTERIUM ABSCESSUS* IN THE PRESENCE OF SUBINHIBITORY CONCENTRATIONS OF GLUTARALDEHYDE

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ABSTRACT:

In 2008, the National Health Surveillance Agency (ANVISA) reported that from 2003 to 2008, more than 2,000 cases of hospital infections caused by Rapid Growth Mycobacteria had been reported in the country, mainly related to videolaparoscopic procedures. Some of these infections had as a common factor the predominance of a particular clone of Mycobacterium abscessus subsp. bolletii (Mab) and mainly affected the skin and the subcutaneous cellular tissue of the patients. After the occurrence of these outbreaks, several measures were taken by Anvisa, such as the suspension of chemical sterilization by immersion of surgical instruments and health products, by the use of any liquid sterilizing agent. The investigations conducted by Anvisa concluded that the outbreaks were due to failures in reprocessing of critical medical equipment, which were sterilized mainly by the use of 2% glutaraldehyde (GA). These decisions were based on empirical observations, based exclusively on the observation of what was happening in hospital practice. To date, the relationship between Mab and its high tolerance to GA has not been elucidated. The objective of this study was to evaluate in parallel the strains of M. abscessus subsp. bolletii CBRVS 00594 and Mycobacterium abscessus subsp. abscessus ATCC 19977, in order to contribute to the elucidation of mechanisms that confer resistance to GA. For this, the biofilm (BF) formation in different concentrations of GA was verified (0.5%; 1.0%; 1.5%). The BFs were developed in polycarbonate and stainless steel discs for UFC analysis and cell culture plates for analysis of laser confocal scanning microscopy (MVCL). The results of the comparative analysis of BF formation by Mab demonstrated that the microorganism was able to form BF on both disc types, even at high concentrations of GA. There was a reduction in the formation of BF when exposed to concentrations of 1.0% and 1.5% of GA, without being completely destroyed. M. abscessus subsp. abscessus was also able to form BF on both disc types, but BF was totally destroyed at concentrations of 1.0% and 1.5% GA. The two strains analyzed by MVCL developed BF after 7 and 14 days of incubation. It was also observed the presence of viable cells in the BF, even after 14 days of growth, in the presence of high concentrations of GA. These results confirmed the ability of the *M. abscessus* species to survive and develop in glutaraldehyde and may be related to the occurrence of the outbreaks.

Keywords: *Mycobacterium abscessus* subsp. *bolletii*; *Mycobacterium abscessus* subsp. *abscessus*; Glutaraldehyde; Biofilm

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