## Nosocomial contamination and bacterial resistance in intensive care units after disinfection: an emerging challenge under biofilm and planktonic growth condition

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## Abstract

Disinfectants are widely used in healthcare facilities to prevent the occurrence of hospitalassociated infections (HAIs), especially in intensive care units (ICUs). Microorganisms adhere to different surfaces and become physically structured in biofilms by secreting extracellular polymeric substances (EPSs) which act as an external shield for the microorganisms, reducing the penetration and diffusion of biocidal substances. This study analyzed the presence of phenotypically resistant Gram-negative rods in ICUs and investigated whether biofilms are responsible for the persistence of multidrug-resistant bacteria in these specialist hospital wards. Multidrug-resistant Gram-negative rods were isolated after ICU disinfection and evaluated for susceptibility to three disinfectants [benzalkonium chloride and biguanide (BCB), sodium hypochlorite (NaClO), and hydrogen peroxide (H<sub>2</sub>O<sub>2</sub>)] under two different conditions: when they are in a planktonic state (unicellular-life phase) and after induction to form biofilms (multicellular-life phase). We compared the efficacy of these chemical disinfectants in removing monospecies biofilms by quantifying the mature biofilm biomass using the crystal violet technique, and thorough evaluation using scanning electron microscopy (SEM). All disinfectants tested showed bactericidal activity against bacteria growing in suspension. After inducing the bacteria to form a biofilm, we observed that BCB was unable to reduce biofilm biomass, while  $H_2O_2$  was only weakly effective against A. baumannii, A. calcoaceticus complexes ACB (p<0.05), and E. coli ATCC 25922 (p<0.05). SEM images corroborated crystal violet data regarding the pronounced NaClO activity observed against all bacteria (p<0.0001) and allowed a better evaluation of the action of BCB, demonstrating variation in sensitivity to this disinfectant for each species evaluated. Multidrug-resistant bacteria isolated from ICU inanimate surfaces did not show the same sensitivity to the assessed disinfectants when they were in a biofilm as when they were in a single cell state, and it appeared that biofilms are able to reduce the activity of disinfectants and contribute to the spread of bacteria involved in the occurrence of HAIs.

Keywords: Healthcare-associated infections<sub>1</sub>. Biofilms<sub>2</sub>. Disinfection<sub>3</sub>. Antimicrobial Resistance<sub>4</sub>. Intensive care units<sub>5</sub>.

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