ABSTRACT

This doctoral dissertation is focused on multidrug resistant microorganisms that belong to the ESKAPE group. ESKAPE is an acronym for a group of microorganisms (***E****nterococcus* spp.*,* ***S****taphylococcus aureus,* ***K****lebsiella pneumoniae,* ***A****cinetobacter baumannii,* ***P****seudomonas aeruginosa and* ***E****nterobacter cloacae*) that have an increased risk due to acquired mechanisms and can easily escape from the biocidal activity of antibiotics. According to the data presented in the Antimicrobial Resistance (AMR) report, the number of deaths from drug-resistant microorganisms could exceed the number of deaths from cancer by 2050. In addition, the number of deaths from AMR in 2050 could also be as high as 10 million per year if no action is taken to stop the progressing problem of antibiotic resistance. In face of the progressing crisis, all activities that help minimize the use of antibiotics or eliminate microorganisms with a different mode of action have become attractive tools to fight AMR.

This work focuses on the use of photodynamic inactivation to sensitize clinical isolates from the ESKAPE group to the action of antibiotics. Photodynamic inactivation (aPDI) is based on the use of visible light in the range of 380 nm - 740 nm, oxygen and photosensitizing compounds (both exogenous or endogenous) in the case of antimicrobial blue light inactivation (aBL). As a result of the excitation of these compounds, reactive oxygen species (ROS) are created, which in turn cause various structures in bacterial cells to become damaged and ultimately perish. Numerous scientific studies have demonstrated the effectiveness of utilizing aPDI/aBL photoinactivation and antibiotics to eradicate multidrug-resistant microorganisms. However, many of these experiments were performed in the wrong way, i.e., using inappropriate synergy testing methods, which has evidenced that combining these two monotherapies is efficient.

The research presented in this doctoral dissertation was carried out based on my own protocol for testing the interaction between photoinactivation and antibiotics, the use of recommended methods and implementation of antibiotics, which originated from various classes and categories and exhibited various mechanisms of activity. A properly implemented synergy testing protocol provides a full picture of the possibilities of aPDI/aBL photoinactivation as a tool to sensitize ESKAPE microorganisms with different resistance profiles. Moreover, another aim of this study was to explain the mechanisms behind the synergistic effect between

aBL/aPDI and antibiotics and to determine the measurable impact of photoinactivation on the tested microorganisms. An additional objective of this dissertation was to verify the photo- and cytotoxicity of the potential use of visible light (blue) against eukaryotic and prokaryotic cells. The last element of the research was verifying the potential of combined therapy (photoinactivation and antibiotics) using an *in vivo* mouse model infected with *Staphylococcus aureus* and *Pseudomonas aeruginosa*.

The results of my research that are presented in this doctoral dissertation have been published in 5 scientific publications, which are attached. The whole work is a set of thematically coherent results.