

Evaluation of the Effect of Photodynamic Inactivation on Persistent Bacteria

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Bacterial persistence, a phenomenon in which a subpopulation of cells enters a state of metabolic dormancy and temporarily becomes tolerant to antibiotic action, enables persistent bacteria to survive under adverse conditions, such as antibiotic treatment. The project's main objective is to standardize Photodynamic Inactivation (PDT) protocols using curcumin (as the photosensitizer) and light (at 450 nm) to evaluate phenotypic changes contributing to the reduction of persistent bacterial formation. Given that the selective pressure exerted by subtherapeutic doses of antibiotics can favor the survival of cells that naturally enter a state of dormancy, thereby increasing their proportion in the population, a secondary objective of the project is to assess the influence of PDT on a population of persistent bacteria in the context of subtherapeutic antibiotic therapy dosages. To this end, we aim to evaluate the effects of PDT on both acquired and intrinsic persistence profiles of two bacterial strains of the same species, *Staphylococcus aureus*, including a clinical isolate of MRSA (methicillin-resistant *Staphylococcus aureus*). The evaluation of the phenotypic characteristics of persistent bacteria will be conducted through the analysis of microbial survival capacity under adverse conditions, including the presence of subtherapeutic antibiotic dosages, using: (1) antibiotic susceptibility tests to evaluate the Minimum Inhibitory Concentration (MIC); (2) survival curve assays; (3) growth rate analyses to determine the impact of persistence in the latency phase; and (4) scanning electron microscopy (SEM) to assess morphological changes associated with persistence, such as cell size, shape, and the presence of specific structures. Therefore, understanding the influence of PDT on persistent bacteria could provide valuable insights for developing more effective therapies against infectious diseases where antibiotics fail.

Key words: Photodynamic inactivation, persistence, *Staphylococcus aureus*.