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Investigating the photophysical properties and photodynamic inactivation response of methylene blue on perfusate solutions for organ preservation

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Organ transplantation is a life-saving therapeutic option, particularly for patients suffering from conditions like end-stage organ failure or severe chronic diseases. However, the current supply of transplanted organs meets less than 10% of global demand, leading to long waiting lists. A significant factor contributing to the discard of potentially viable organs is contamination by microorganisms (1) such as bacteria, fungi, or viruses, which can cause life-threatening infections in recipients. The limited availability of suitable organs for transplantation and the emergence of drug-resistant microorganisms have driven our research group to explore decontamination methods that do not contribute to the rise of antimicrobial resistance. Methylene blue (MB) is a well-known photosensitizer that has shown promising results for microbiological control using photodynamic therapy (2). Nevertheless, insufficient antimicrobial response in *ex vivo* graft experiments was observed by the use of antimicrobial photodynamic therapy (aPDT) for decontamination of organs for transplantation. Other studies have also reported impaired aPDT response when biological fluids are present (3). The present study aimed to investigate the photodynamic response of MB in the organ preservation solutions STEEN and Custodiol HTK, based on its *in vitro* photodegradation kinetics and resulting bacterial inactivation. UV-visible absorption spectroscopy and liquid chromatography coupled with mass spectrometry (LC-MS) were used to investigate the optical properties of MB in the two perfusate solutions and the formation of photodegradation products under irradiation at 660 nm. A strain of *Staphylococcus aureus* was used to monitor the antimicrobial response of aPDT at different treatment conditions. The study provided significant insights into the photophysical behavior of MB in the presence of perfusate biomolecules and their consequences to the lack of antimicrobial effect. The results suggest that the perfusate biomolecules are responsible for quenching the photodynamic action of MB, possibly due to the interaction with the photosensitizer, trapping its molecules, and not allowing its interaction with the bacterium cells.

Palavras-chave: Spectroscopy; Photodynamic therapy; Transplantation.

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