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Antimicrobial Photodynamic Therapy with Polystyrene Nanoparticles, Encapsulated Porphyrin-Derivative and Iodine Generation against *Candida Albicans*

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Extended Abstract

Candida albicans is the most important pathogenic fungus to global health, responsible for a large part of invasive candidiasis, mainly in hospital environments [1]. *Candida albicans* has a form of resistance and virulence difficult to eradicate: the biofilm - a complex microbial community, organized in an extracellular polymer matrix, composed by lipids, polysaccharides and nucleic acids [2]. New therapies modalities are necessary to combat countless resistant forms of microorganisms, including biofilms; and Antimicrobial Photodynamic Therapy (aPDT) associated with different nanoparticles shown a promising alternative. The aPDT is a combination of light and photosensitizer (PS) to create reactive oxygen species (ROS), damaging important cellular structures for microorganisms, leading to their death. Nanoparticles with encapsulated PS can enhance the effect of aPDT via increasing ROS production [3, 4]. Numerous nanoparticles can be used to encapsulate a PS, e.g. hydrophobic porphyrin photosensitizers, to prevent their aggregation in aqueous solutions. Sulfonated polystyrene nanoparticles (NPs) are biocompatible, permeable to oxygen and transparent to visible light, allowing encapsulation of a porphyrin PS. Light-induced antifungal activity of such NPs with encapsulated 5,10,15,20-tetraphenylporphyrin (TPP) was demonstrated on *Candida* species, including *C. albicans*. In addition, potassium iodide (KI) was used to enhance the effect of aPDT with TPP-NPs, resulting in a dual effect caused by photogeneration of $O_2(^1\Delta_g)$ and oxidation of I^- by $O_2(^1\Delta_g)$ to I_2/I_3^-, serving as an additional antimicrobial component [5]. The aim of this study is to demonstrate the effect of aPDT with TPP-NPS in the presence or absence of KI against the growth and formation of *C. albicans* biofilm. *Candida albicans* (ATCC #1023) were sown in Sabouraud-Dextrose agar for 48h at 37ºC and suspended in saline solution at cell density of 10^6 or 10^7 cells. mL^{-1} for growth or biofilm experiments, respectively. Stable aqueous dispersion of TPP-NPs, with no tendency to aggregate were synthetized as previously described [6]. TPP-NPs dispersion consisted from spherical nanoparticles with an average diameter of ~10-20nm and high zeta-potential (-76 mV). For aPDT, TPP-NPs were irradiated with a blue LED lamp (415 nm) in absence or presence of KI (10 mM). A microplate reader was used to quantify absorbance of *C. albicans* growth and biofilm formation through XTT assay. In microbiological tests, TPP-NPs eradicated *C. albicans* growth in ~99% ($1x10^{10}$ TPP-NPs.mL^{-1}) in presence of KI. In absence of KI, TPP-NPs reduced *C. albicans* growth in ~25% ($1x10^{13}$ TPP-NPs.mL^{-1}) and, to produce the same effect observed in absence of KI, it was necessary a TPP-NPs' concentration 10^4 times lower (3.10^9 TPP-NPs.mL^{-1}). Moreover, NPs-TPP ($1x10^{13}$ TPP-NPs.mL^{-1}) reduces ~20, 38 and 50% the metabolic activity of biofilm formation, immediately, 20 and 50 minutes after irradiation, respectively. Combination with KI, increased the inhibition to ~95%. Results demonstrated high inhibition of *C. albicans* growth and biofilm formation in cells treated with TPP-NPs, mainly in KI presence. Thus, aPDT with TPP-NPs in combination with KI could be an efficient alternative antifungal therapy.

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