

The Improvement of Photoinactivation of *Escherichia coli* by using Silica Nanoparticles Loaded with Toluidine Blue

Larissa, S.Amaral^{1,*,#}, Elias, P.P. Neto², Ubirajara, P. Rodrigues Filho², Janice, R.Perussi^{1,2}

¹Universidade de São Paulo, Programa de Pós-Graduação Interunidades em Bioengenharia EESC/FMRP/IQSC, Av. Trabalhador São Carlense, 400 - Parque Arnold Schmidt, São Carlos - SP, 13566-590, Brazil.

²Universidade de São Paulo, Instituto de Química de São Carlos, Av. Trabalhador São Carlense, 400 - Parque Arnold Schmidt, São Carlos - SP, 13566-590, Brazil.

#Corresponding author: larissa.bioengenharia@usp.br

The use of photosensitizers adsorbed on silica nanoparticles (SiNp) instead of antibiotics as antimicrobial agents is a promising alternative for to avoid resistance of bacteria to antibiotics. This technique is based on the use of a photosensitive molecule excited by light in the presence of oxygen. SiNp can be employed in this method once they do not exhibit light absorption in the visible range ^[1]. The SiNp also present penetrability/affinity for gram-negative bacteria being nontoxic to mammalian cells ^[2]. The purpose of this study was to analyse the influence of SiNp in the photoinactivation of *E.coli* by Antimicrobial Photodynamic Therapy (a-PDI) using Toluidine Blue (TB) and white light. In the first step it was performed a 2³ factorial design in which Incubation Time (IT); Light Dose (LD) and Concentration of photosensitizer (C) were 8 and 16 min; 15 and 30 J cm⁻² and 20 and 45 μmol L⁻¹ of TB. Calculations and new experiments were performed with the obtained results in the model in order to optimize the photoinactivation. Then a quadratic model with axial points was used followed by statistic treatment leading to the optimum point of 100% inactivation with the use of SiNp: 18 μmol L⁻¹ of SiNp-TB incubated with *E. coli* by 25 min and irradiation with 48 J cm⁻². On the other hand using only TB without SiNp, the C of MB was 37 μmol L⁻¹; IT of 18 min and LD of 39 J cm⁻². We can conclude that the use of SiNp allows the use of 51% less photosensitizer, which was probably due to the hydrophilic character of the TB causing a greater affinity to the gram negative bacteria in presence of the SiNp and thus, it allows a greater toxicity when irradiated.

[1] Chatterjee, D. K; Fong, L.S et al, Nanoparticles in photodynamic therapy: An emerging paradigm. *Advanced Drug Delivery reviews*. **60**, (2008) 1627-1637.

[2] Capeletti, L.D; Oliveira, L.F; Gonçalves, K.A et al. Tailored silica-antibiotic nanoparticles: overcoming bacterial resistance with low cytotoxicity. *Langmuir, American chemical society*. **30**, (2014) 7456-7464.