P99

Identification of important cellular targets for antimicrobial photodynamic therapy in yeast cell through FT-IR spectroscopy

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Antimicrobial resistance is now recognized as one the most serious global threats to human in the 21 st century. The persistent problem of antibiotic resistance has created a strong demand for new methods for therapy. Antimicrobial photodynamic therapy (aPDT) has been reported to be effective in eradicating wide variety of pathogens, including antibiotic-resistant microorganisms. Different microbial cells present many variances in different levels as cellular morphology, structure, metabolism and environment adaptive alterations. In face of these alterations it would be interesting to identify important cellular targets for aPDT.

The aim of this study was identify components in yeast cells that are potential targets for fungal photoinactivation using Fourier transform infrared (FT-IR) spectroscopy.

Candida albicans ATCC 90028 in stationary growth phase (48-h) were submitted to aPDT mediated by methylene blue (50 μM) and exposed to a 660 nm-LED (P= 360 mW). Pre-irradiation time was 10 min and exposure times were 12, 15 and 18 min. FT-IR was employed to evaluate the photodynamic effect in cells following 15 min of irradiation.

Control groups, i.e., untreated cells without irradiation and MB, and cells treated only with light or MB did not show any reduction. Following aPDT, the reduction in cell viability was fluence-dependent ($p<0.05$). Similar FT-IR spectra were observed for untreated and treated cells grown for 48-h. However, following aPDT, more intense absorptions bands were detected in all spectral regions that characterize the major cellular constituents (1200-900 cm^{-1}; 1500-1200 cm^{-1}; 1800-1500 cm^{-1} and 3000-2800 cm^{-1}). FT-IR spectroscopy showed that cellular targets in *C. albicans* in stationary growth phase were preferentially the polysaccharides of the extracellular matrix (EPS) and the cell wall, as well as the lipids of the cell membrane.

We conclude that EPS presence plays an important role for aPDT. In addition, for a good clinical outcome aPDT should be performed aiming two different targets: the EPS surrounding cells and the cell itself.

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