Up-take and localization of photosensitizers in fungi by confocal fluorescence microscopy

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Photodynamic antimicrobial chemotherapy (PACT) is a strategy to kill microorganisms. It is based on the use of intracellular photosensitive drugs, which upon irradiation by specific light sources react with oxygen to create reactive oxygen species (ROS). One of the most promising PACT applications is related with killing fungi in resistant infections.¹ However, there is relatively few information concerning the up-take and localization of photosensitizers in the different fungi life-forms. Therefore, we performed this study aiming to understand the uptake and localization of two photosensitizers, i.e., Methylene Blue (MB) and Hypericum extract (HYP) in Trichophyton rubrum (Tr). Both MB and HYP guickly enter hyphae. Both photosensitizers also enter conidia but with much lower efficiency. Co-localization experiments with rodamine-123 indicate that both MB and HYP are partially localized in mitochondria of hyphae. The constituents of HYP, which are hypericin and chlorophyll, have different localization, hypericin localizing preferentially in mitochondria. The fast penetration of the photosensitizers in hyphae may be related with the high level success in the treatment. The slow penetration in conidia and its low metabolic activity, may explain the 15% recurrence that is observed in the clinical cases.¹ We are searching strategies to facilitate photosensitizer uptake by conidia, which may decrease the level of recurrence.

1. Tardivo et al Photodyagnosis and Photodynamic Therapy 2005, 2/3, 175-191.

PDT, methylene blue, fungi, cytolocalization