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Yeast Copper Proteins and Reactive Oxygen Species in Effecting Lifespan

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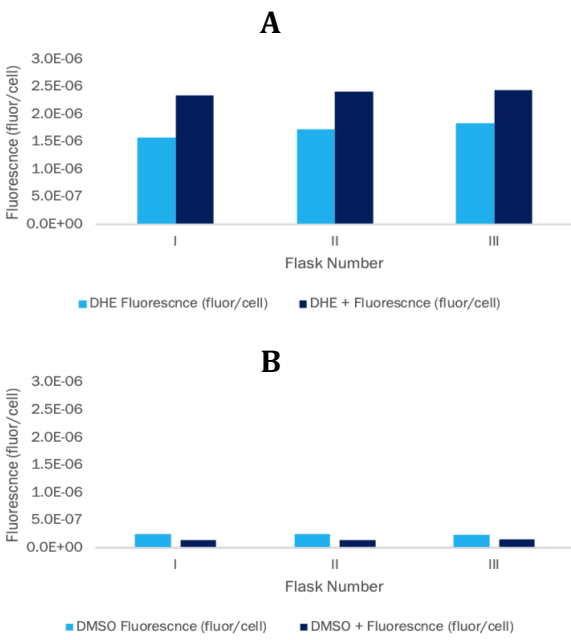


Figure 1. WT yeast cultures (50 mL in YPD) were cultured for one to three days (I-III) at 30°C, either in the presence (+) or absence of the ETC inhibitor Antimycin A. Cellular ROS levels were determined by staining cells with the fluorescent dye dihydroethidium (DHE, 2.5 μ M). Cells were washed with PBS and then treated with either DHE (panel A), DMSO (negative control, panel B), or no treatment. The treated samples were incubated for 30 minutes at 30°C, and then assayed via plate reader (Molecular Devices, SPECTRAmax, Gemini XPS) for DHE fluorescence with an excitation of 460 nm and emission of 580 nm. The no treatment sample was used to determine total number of cells in the samples using an OD600 reading. Results are presented as fluorescence normalized to the total number of cells. The presence of Antimycin A in the culture media clearly shows an increase in the detectable ROS produced.