Title: EVALUATION OF THE HYDROGEN PEROXIDE EFFECT IN GENE EXPRESSION OF Gluconacetobacter diazotrophicus PAL5 strain

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Abstract:

Nitrogen is a nutrient absorbed in larger quantities by plants and, despite of being abundant in the atmosphere, it is a non-assimilable form by plants. Among the alternatives of input of N to ecosystems, biological nitrogen fixation (BNF) is considered as the most viable, since it reduces the costs and damage to the environment, the dependence on nitrogen fertilizer and contributes to the soil fertility. This process is performed by the nitrogenase enzyme that requires large amounts of ATP provided by the aerobic respiration. During this process, reactive oxygen species (ROS) may be generated, which can damage the cell and inhibit the nitrogenase activity. To circumvent this effect, the bacteria must resist efficiently or repair the damage caused by the reactive oxygen species. *Gluconacetobacter diazotrophicus* is an endophytic diazotrophic bacterium that, when subjected to oxidative stress, expresses proteins that directly eliminate oxidants. In this study, the effect of oxidative stress induced by H₂O₂ in gene expression of *G. diazotrophicus* PAL5 strain using quantitative qPCR will be assessed. The wild type strain was grown in triplicate in biological LGI-P liquid medium, followed by cells exposure or not to 100 µM of H₂O₂ for sixty minutes. After this time, cells were harvested for total RNA extraction and subsequent analysis by RT-qPCR (quantitative real time RT-PCR). The results demonstrated that the relative expression of katA and katC genes were similar at both conditions (with and without stress). In contrast, the relative expression of the sodA gene was higher in the presence of H₂O₂. We believe that another enzyme may be initiating cellular detoxification process, such as the Alkyl hydroperoxide reductase enzyme, which in *Escherichia coli* is the primary scavenger of H₂O₂. Additional studies are required in order to assess more accurately the contribution of other antioxidant enzymes in the detoxification induced by H₂O₂.

Key words: Oxidative Stress, RT-qPCR, Hydrogen Peroxide

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