

## How does heat-shock affect the influence of titanium dioxide nanoparticles in growth and antioxidant power of *Saccharomyces cerevisiae* BY4741 ?

J Capela-Pires<sup>1,2</sup>, R Ferreira<sup>1,2</sup>, I Alves-Pereira<sup>1,2</sup>

<sup>1</sup>ICAAM – Instituto de Ciências Agrárias e Ambientais Mediterrânicas, Universidade de Évora, Évora, Portugal

<sup>2</sup>Departamento de Química, Escola de Ciências e Tecnologia, Universidade de Évora, Évora, Portugal

Nanomaterials include all substances that contain nanoscale structures sized between 1 and 100 nm. At this size, the characteristics of materials change: their strength, conductivity, and reactivity, which differ substantially from macro- or micron- sized materials, shifting the rules of physics and chemistry to the sidelines. Although, the geological origin and the ubiquitous occurrence of nanoparticles in the earth crust can lead to suppose a good phylogenetic adaptation of living beings to such substances, the unique characteristics of nanoparticles (NPs) bring a new dimension to environmental effects testing. The industrial development coupled with vast new applications of nanomaterials, have contributed to raise their environmental levels, reason because, concern over the environmental pressure of the nanoparticles in certain regions of the world as well as its effects on the biosphere has grown in recent years, since its reactivity with biomolecules mainly depends on the surface area/molecular size ratio and physicochemical factors such as pH and temperature.

Thus, the main objective of this study was to evaluate how heat-shock affects cell survival and antioxidant response of *S. cerevisiae* BY4741, a Eurocast strain, exposed to titanium dioxide nanoparticles (TiO<sub>2</sub>-NPs). Cells in exponential phase were inoculated in liquid YEPD medium 2 % (w/v) glucose at 28 °C are exposed at 0.1 or 1.0 µg/mL NP-TiO<sub>2</sub> prepared by sonication, during 200 min at 40 °C. Samples from each treatment were used to obtain the post-12000 g fractions, which were used for protein content, DPPH, glutathione antioxidant capacity and, ALP, catalase and LOX activities determinations.

The results show that the presence of TiO<sub>2</sub>-NPs in the culture medium induced cell death, response evidenced by a decrease of proliferative capacity detected by the alkaline phosphatase (ALP, EC 3.1.3.1) activity, loss of redox buffer capacity mediated by glutathione, evidenced by a decrease of GSH+GSSG contents and GSH/GSSG ratio. On the other hand, cell death also appears depend on the loss of ability to scavenge free radicals, estimated by DPPH method. We also observed an increase of lipoxygenase (LOX, EC 1.13.11.12) activity, a marker of lipid peroxidation, which may be related with a loss of antioxidant power mediated by peroxisomal catalase (CAT A, EC 1.11.1.6), probably due a slowdown of β-oxidation. Finally it was observed an increase of the antioxidant cytoplasmic catalase (CAT T, EC 1.11.1.6) in cells exposed to concentrations of 0.1 mg/mL, but a significantly decrease of this enzyme activity in cells exposed to 1 mg/mL TiO<sub>2</sub>-NPs. This apparently bimodal response indicates a loss of proliferative capacity by an active process when the level exposure was 0.1 mg/mL. However, for 1 mg/mL TiO<sub>2</sub>-NPs level, appears to occur a transition for necrosis.

**Keywords:** yeast; alkaline phosphatase; glutathione; lipoxygenase; catalase

### References

- [1] Breaudiere J, Spillman T (1984) Bergmeyer methods of enzymatic analysis, vol. II, 3rd ed., Verlag Chemie, Florida.
- [2] Hissin A, Hilf P (1976) Anal. Biochem. 74, 214–226.
- [3] Brand-William W, Cuvelier M, Berset C (1995) Use of a free radical method to evaluate antioxidant activity. Academic Press. 28, 25-30.
- [4] Carru C, Zinellu A, Sotgia S, Marongiu G, Farina M, Usai M, Pes G, Tadolini B, Deiana L (2003) J. Chrom. A. 1017, 233–238.
- [5] Gata J, Pinto M, Macias P (1996) J. Agric. Food Chem. 44, 2573–2577.
- [6] Beers R, Sizer I (1952) J. Biol. Chem. 195, 133-140.