A chemogenomic screen reveals *YPL257W* as a pleiotropic determinant of polyphenol and heavy metal resistance in *Saccharomyces cerevisiae*

Ioana Nicolau*, Lavinia L. Ruta, Ileana C. Farcasanu

Department of Organic Chemistry, Biochemistry and Catalysis, Faculty of Chemistry, University of Bucharest, Sos. Panduri 90-92, 050663 Bucharest, Romania

*e-mail: ioana.nicolau@chimie.unibuc.ro

Plant polyphenols are often used as food supplements due to their antioxidant, cardio-vascular disease prevention, hepatoprotective, anti-inflammatory, or anticancer activities. In spite of the general belief that high intake of natural polyphenols is benefic for human health, there are cases when these antioxidants turn villain, eliciting contradictory responses [1-3]. To unravel new cell targets of polyphenol action we used the model eukaryotic microorganism *Saccharomyces cerevisiae*. Starting from the observation that chlorogenic acid - one of the most abundant polyphenol compounds in the human diet - is toxic to *S. cerevisiae* cells, we screened a haploid yeast deletion library [4] for increased tolerance to chlorogenic acid, with the aim to identify components involved in polyphenol uptake. One hit indicated the uncharacterized ORF *YPL257w* which encodes a putative transmembrane protein of 193 amino acids. We found that the knock-out strain *ypl257wΔ* was tolerant to chlorogenic aid, but also to exogenous oxidative stress or heavy metal stress imposed by surplus Cu(II) or Mn(II). Accumulation of chlorogenic as well as of Cu(II) or Mn(II) was significantly reduced in *ypl257wΔ* cells, a plausible explanation for the tolerant phenotypes observed. Quantitative RT-PCR indicated that *YPL257w* expression in yeast wild type cells was not significantly influenced by exposure to chlorogenic acid, but was stimulated by Cu(II) or Mn(II). Expressing *YPL257w-GFP* from a centromeric plasmid led to plasma membrane localization of the tagged protein. Overexpression of *YPL257w-GFP* from an in inducible promoter indicated initial localization at the plasma membrane followed by a shift to vacuole membrane, once the GFP-tagged product accumulated. As the vacuolar membrane localization of the YPL257w-GFP was accompanied by a massive Cu(II) accumulation in the vacuole, the possibility to utilize YPL257w-overexpressing yeast cells as heavy metal hyperaccumulators is discussed. In summary, our screen uncovered a possible role of YPL257w in non-specific transport across plasma and vacuolar membranes.

Keywords: *Saccharomyces cerevisiae*, chemo-genomic screen, YPL257w, chlorogenic acid, copper, manganese

Reference

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