5th Conference on Physiology of Yeast and Filamentous Fungi

4 – 7 June 2013
Montpellier, France

SYMPOSIUM BOOK

Organised by UMR1083 SPO,
INRA, 2 place Viala, F-34060
Montpellier Cedex 2 France
http://www5.montpellier.inra.fr/spo/

Under the auspices of the European Federation of Biotechnology

Unique trancriptional responses of *Ashbya gossypii* to endoplasmatic reticulum stress

Tatiana Q. Aguiar [1], Orquídea Ribeiro [1], Mikko Arvas [2], Marilyn G. Wiebe [2], Merja Penttilä [2], Lucília Domingues [1]

[1] Centre of Biological Engineering, Universidade do Minho, Campus de Gualtar 4710-057 Braga, Portugal
[2] VTT Technical Research Centre of Finland, P.O. Box 1000, 02044 VTT Espoo, Finland

In eukaryotic cells, proteins that enter the secretory pathway must pass through the endoplasmic reticulum (ER) to be properly folded and modified. When the demand for protein folding exceeds the ER capacity, unfolded or misfolded proteins accumulate in this compartment, leading to ER stress. To relieve stress, eukaryotic cells generally activate the unfolded protein response (UPR), which induces a comprehensive gene expression program that adjusts the protein folding capacity of the ER.
Ashbya gossypii is a filamentous hemiascomycete phylogenetically close to *Saccharomyces cerevisiae*, which has been exploited as a host for the production of heterologous proteins. To analyse the impact of recombinant protein secretion on the gene expression profiles of this biotechnologically relevant fungus we conducted genome-wide transcriptional analyses in a recombinant *A. gossypii* strain expressing the *Trichoderma reesei* endoglucanase I (EGI) using DNA microarrays. A corresponding empty vector strain was used as control. The transcriptional responses of EGI-producing cells to ER stress were also examined by exposing them to 10 mM dithiothreitol (DTT) for 30 min, 1 h and 4 h. This strong reducing agent is known to disrupt protein folding in the ER and, consequently, to induce the UPR in several organisms. Surprisingly, our data revealed that secretion of EGI did not cause prominent variations on the *A. gossypii* transcriptional profiles and that a conventional UPR was not activated in response to DTT-induced ER stress, as the expression levels of several well-known UPR target genes (*HAC1*, *BIP1*, *IRE1*, and *PDI1* homologs) remained unaffected. In addition, a consensus UPRE motif was not found in the promoter region of genes up-regulated by DTT treatment, as described in other fungi, supporting the absence of a conventional HAC1-dependent UPR. However, several genes involved in the ER-associated degradation (ERAD) were highly up-regulated after DTT treatment, namely genes involved in proteasome assembly, proteolysis and vesicle-mediated transport, suggesting that an alternative ER quality control system exists in *A. gossypii*. Unexpectedly, several genes involved in protein glycosylation were down-regulated by DTT treatment, an effect usually observed in other organisms after exposure to the N-glycosylation inhibitor tunicamycin.
This study has unveiled unique ER stress responses in *A. gossypii*, which now merit further investigation.

Acknowledgments: This work was financially supported by Fundação para a Ciência e a Tecnologia (FCT), Portugal, through Project AshByofactory (grant PTDC/EBB-EBI/101985/2008), MIT-Portugal Program (PhD grant SFRH/BD/39112/2007 to T. Q. Aguiar) and grant SFRH/BD/30229/2006 to O. Ribeiro.

Keywords: *Ashbya gossypii*; endoplasmatic reticulum stress; protein secretion; unfolded protein respons