FUNCTIONAL ANALYSIS OF THE TRANSCRIPTION FACTOR XLNR INVOLVED IN THE REGULATION OF TRANSCRIPTION OF CELLULASES- AND HEMICELLULASES-ENCODING GENES IN ASPERGILLUS NIGER

Gustavo Henrique Goldman
Ribeirão Preto School of Pharmaceutical Sciences / University of São Paulo (USP)
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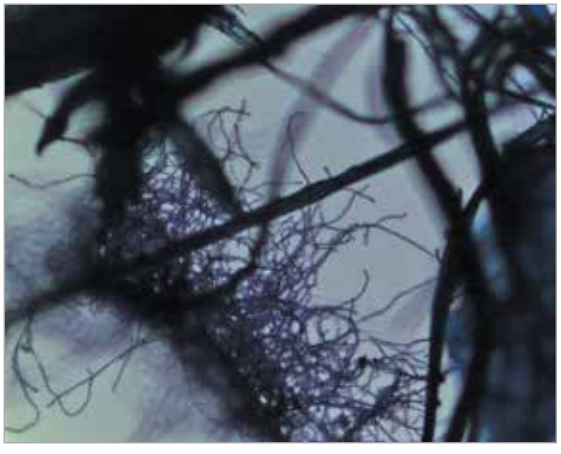

Growth of the Aspergillus Niger *strain* N_4O_2 *át 30C in steam-exploded sugarcane bagasse*

The high cost of hydrolyzing biomass polysaccharides to fermentable sugars remains a major obstacle to be overcome before cellulosic ethanol can effectively be commercialized. The costs of cellulases and hemicellulases contribute substantially to the price of bioethanol. New studies to understand and improve cellulase efficiency and productivity are of paramount importance. Filamentous fungi like *Aspergillus niger* and *Trichoderma reesei* are impressive producers of hydrolytic enzymes already applied in a series of industrial processes, e.g. food, feed, pulp, paper, and textile industries. The *A. niger* xlnR transcription factor is a master regulator that activates enzymes of the xylanolytic system, a number of endocellulases and two cellobiohydrolases. The study of transcriptional regulators involved in the activation of genes that encode enzymes responsible for the degradation of cellulose and hemicellulose could provide several advantages for further genetical improvement of biomass-degrading microorganisms. A potential strategy to modify expression patterns of cellulase and/or hemicellulase could follow either a constitutive or induced expression of modified versions of the regulatory proteins. The design of constitutively activated or even structuraly modified transcription factors may lead to strains allowing inducer substance-independent enzyme production. Our project aims to provide basic information about the fine regulation at transcriptional level of *A. niger* genes that encode hydrolytic enzymes under the control of the transcription factor XlnR. Furthermore, we also plan to identify protein partners of XlnR that could be involved either in its down-regulation, for example via carbon repression, or additional proteins that help in the assembly of the transcription machinery aiming the establishment of the gene expression regulated by XlnR.

SUMMARY OF RESULTS TO DATE AND PERSPECTIVES

We are currently evaluating mRNA accumulation by real-time RT-PCR for several genes encoding cellulases and hemicellulases, as well as establishing assays for enzymatic activity of cellulases and hemicellulases. We are deleting the gene encoding the catabolite repressor creA and overexpressing the gene encoding the transcriptional activator, xlnR. We are using sugarcane bagasse pretreated by steam explosion to evaluate the better enzymatic performance of these genetically modified strains.

Meanwhile, we are constructing strains with S-tag epitopes fused to these proteins aiming to identify protein partners involved in their post-translational regulation.

MAIN PUBLICATIONS

von Zeska Kress MR, Harting R, Bayram Ö, Christmann M, Irmer H, Valerius O, Schinke J, Goldman GH, Braus GH. 2012. The COP9 signalosome counteracts the accumulation of cullin SCF ubiquitin E3 RING ligases during fungal development. *Mol Microbiol*. **83(6)**: 1162-77. DOI: 10.1111/j.1365-2958.2012.07999.x. Epub 2012 Feb 22. PubMed PMID: 22329854.

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Gustavo Henrique Goldman

Faculdade de Ciências Farmacêuticas de Ribeirão Preto / Universidade de São Paulo (USP)
Departamento de Ciências Farmacêuticas
Av. do Café, s/n
CEP 14040-903 – Ribeirão Preto, SP – Brasil

+55-11-3062-4280
ggoldman@usp.br