

TITLE: IMPACTS OF THE OVEREXPRESSION OF AZF1 TRANSCRIPTION FACTOR IN CELLULASE PRODUCTION BY *Trichoderma reesei*.

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ABSTRACT:

The fungus *Trichoderma reesei* is a significant producer of enzymes that degrade lignocellulosic biomass for producing value-added bioproducts. *T. reesei*'s cellulolytic system is controlled by several transcription factors that efficiently regulate the production of these enzymes. Among them is the positive regulator Azf1. This work aimed to contribute to the understanding of the role of Azf1 in the regulation of cellulase gene expression in the fungus *T. reesei* using the overexpression approach. For this, a vector was constructed with *azf1* gene under the control of the constitutive promoter *Pcdna1*, which overexpressed Azf1 in inducing and repressive conditions of cellulase production, with a fold increase of at least 10-fold in the gene expression. Although more abundant in transcriptional and protein levels, overexpression of Azf1 did not activate the transcription of cellulases in glucose. While in cellulose, in the early stages of cultivation, the expression of the protein *Swollenin* and β -glucosidases *cel1a*, *cel1b*, and *cel3b* increased. This increase in the production of these β -glucosidases, in turn, caused a rise in the hydrolysis rate of cellobiose, which activated the carbon catabolite repression (CCR) and caused repression of the cellulase genes and the transcription factor Xyr1 in the later stages of cultivation. In addition, the overexpression of Azf1 caused an increase in the activity of cellulases by *T. reesei* in cultivation on cellulose and sugarcane bagasse. Therefore, this work presented new roles for Azf1 in regulating the cellulase expression in *T. reesei*, contributing to the development of strains with greater production of cellulases.

Keywords: 1. *Trichoderma reesei*. 2. Cellulases. 3. Transcription factor. 4. Azf1. 5. Overexpression.

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