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Genome-wide analysis of protein production physiology in the filamentous fungus Trichoderma reesei

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Background

Trichoderma reesei is a filamentous fungus used widely as a protein production host by the enzyme industry. It is an excellent protein secretor; the highest native enzyme production levels in the streamlined industrial process are above 100 g/l [1]. T. reesei is also used as a host for recombinant protein production. The unfolded protein response (UPR) pathway has been characterized from this fungus in our group and we have discovered a novel secretion stress response, repression under secretion stress (RESS) affecting the genes encoding secreted proteins. The genome of T. reesei was sequenced recently by the Joint Genome Institute.

Results

The methods for genome-wide analysis of *T. reesei* physiology have been set up in our laboratory. For transcriptomics we use the method TRAC and commercial genome-wide oligonucleotide arrays. As a rapid and semiautomated method TRAC is very useful in focused transcript analysis of multiple samples e.g. in assessment of the quality of chemostat cultures, in choosing the right samples for genome-wide analysis and in transient experiments with many sample time points. In proteomics we use conventional 2D-electrophoresis techniques. The developed methodology has been used to address the physiology of *T. reesei* that is relevant for industrial production of recombinant and native proteins. A strain expressing the *Melanocarpus albomyces* laccase, a protein not inducing the unfolded protein response, was investi-

gated by transcriptomics and proteomics. Surprisingly few physiological changes were found, and most of them were detected only at the proteome level. We have also addressed the effects of certain key bioprocess parameters on productivity and cell physiology with our experimental platform, including $\rm O_2$ deficiency, growth rate and cell density. The results of these studies will be discussed.

Conclusion

An experimental platform for genome-wide physiological studies in *Trichoderma reesei* has been founded and used for elucidation of the effects of recombinant protein production and certain key bioprocess parameters. This work has started to accumulate useful data that can contribute to increasing the understanding on protein production in eukaryotic microbes.

References

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