Monitoring of transcriptome and proteome profiles to investigate the cellular response of E. coli towards recombinant protein expression under defined chemostat conditions.

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Details

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Abstract

The use of strong promoter systems for recombinant protein production generates high product yields, but also overburdens the host cell metabolism and compromises production. Escherichia coli has highly developed regulatory pathways that are immediately responsive to adverse conditions. To gain insight into stress response mechanisms and to detect marker genes and proteins for stress specific monitoring time course analysis of controlled chemostat cultivations was performed using E. coli total microarray and difference gel electrophoresis (Ettan DIGE). In order to detect differences and consistencies of stress response as well as the impact of the inducer isopropyl-beta-d-thiogalactopyranosid on cells, expression of two recombinant proteins (hSOD and GFPmut3.1) was investigated. Genes involved in aerobic metabolism under control of

the ArcB/ArcA two component system were found to be down-regulated, and the interplay of the psp operon, ArcA system and guanosine tetraphosphate is suggested to be involved in stress regulatory mechanisms. A distinct impact of the two recombinant proteins was observed, particularly on levels of known stress regulatory genes and proteins, as well as on the response associated with ArcA and psp. Altogether, 62 genes as well as seven proteins showed consistent expression levels due to recombinant gene expression, and are therefore suggested to be appropriate monitoring targets.

Involved units

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