

Assessing glycolytic flux alterations resulting from genetic perturbations in *E. coli* using a biosensor - DTU Orbit (09/11/2017)

Assessing glycolytic flux alterations resulting from genetic perturbations in *E. coli* using a biosensor

We describe the development of an optimized glycolytic flux biosensor and its application in detecting altered flux in a production strain and in a mutant library. The glycolytic flux biosensor is based on the Cra-regulated *ppsA* promoter of *E. coli* controlling fluorescent protein synthesis. We validated the glycolytic flux dependency of the biosensor in a range of different carbon sources in six different *E. coli* strains and during mevalonate production. Furthermore, we studied the flux-altering effects of genome-wide single gene knock-outs in *E. coli* in a multiplex FlowSeq experiment. From a library consisting of 2126 knock-out mutants, we identified 3 mutants with high-flux and 95 mutants with low-flux phenotypes that did not have severe growth defects. This approach can improve our understanding of glycolytic flux regulation improving metabolic models and engineering efforts.

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Authors: Lehning, C. E. (Intern), Siedler, S. (Intern), Ellabaan, M. M. H. (Intern), Sommer, M. O. A. (Intern)

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