

## **Molecular Biotechnology Programme**

Uppsala University School of Engineering

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Author	
Rickard Frost	
Title (Fire 15.1)	
Title (English)  Fed botch cultivation for the expression of Affibedy® melecules	
Fed-batch cultivation for the expression of Affibody® molecules	
with minimized phosphogluconoylation	
Abstract Recombinant proteins produced in <i>Escherichia coli</i> are susceptible to phosphogluconoylation, a partial post-translational modification first described by Geoghegan <i>et al.</i> in 1999. The modification causes an additional mass of 258 Da and its dephosphorylation results in an excess mass of 178 Da. Homogeneous protein products are desirable and therefore the fraction of modified proteins is unwelcome. The aim of this study was to minimize the abundance of the modification by altering the conditions during fed-batch cultivation. In order to decrease the cell content of the precursor glucose-6-phosphate the carbon source was altered from glucose to glycerol. The influence of the specific growth rate on the modification was also evaluated. Data obtained from several fed-batch and continuous cultivations show that the modified proteins are less abundant on the model protein when glycerol is used as substrate, and when the specific growth rate is high (above 0.30 h <sup>-1</sup> ).	
Keywords Phosphogluconoylation, <i>Escherichia coli</i> , fed-batch, chemostat, Affibody® molecule	
Supervisors	
Finn Dunås	
Affibody AB Scientific reviewer	
Mikael Widersten	
Department of biochemistry, Uppsala University	
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Biology Education Centre Biomedical Center Husargatan 3 Uppsala	
Box 592 S-75124 Uppsala Tel +46 (0)18 4710000 Fax +46 (0)18 555217	