

RESEARCH PROJECT

 Institute
Life Technologies

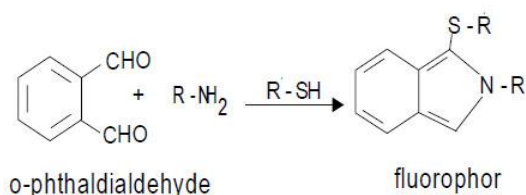
Development of a simple analytical test method for the quantification of L-Glu or GlutaMAX™ during bioprocesses

Partenaire(s) HES-SO Valais

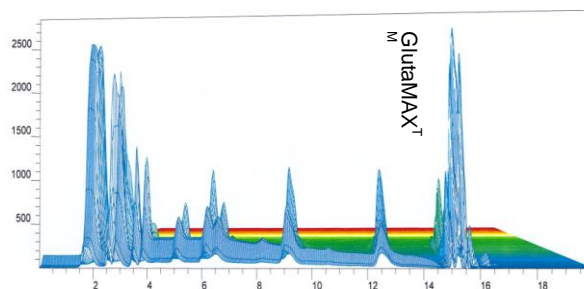
Collaborateur(s) A.Fornage, M. Mayor, M. Pusnik & F. Kalman

Description The objective of this work was to develop a time & cost-efficient analytical test method for the quantification of L-Glutamine or GlutaMAX™ (Ala-Glu) in different mammalian cells cultures. Monitoring L-glutamine and GlutaMAX™ levels during a bioprocess is crucial in order to control cell metabolism and to optimize the respective cultivation processes. Due to the absence of chromophores the sensitive detection of L-Glu and GlutaMAX™ in complex matrices like fermentation juices is quite challenging. Existing methods are based on enzymatic reactions, kits or automated special devices which are expensive and / or time consuming.

Using a common HPLC system with UV-detection a robust and selective isocratic RP-HPLC separation system was developed for each compound. A simple, but efficient sample preparation strategy was established where L-Glutamine or GlutaMAX™ containing samples are derivatised with OPA directly and in an automated manner in the HPLC auto sampler for 1 min prior to separation. The pre-column reaction parameters were optimized in order to assure complete and quantitative reaction. The UV-detection allows determination of the metabolites in the μM to mM range, usually sufficient to control bioprocesses. However even more sensitive fluorescence detection (fmol range) can be performed if needed. The accuracy of both methods is better than $\pm 10\%$, the precision is better than 5% for the content of the two compounds in fermentation juices. Matrix effects were not observed testing many different media & fermentation juices.



Reaction scheme of the derivatization of the primary amine of L-Glu and GlutaMAX™ with OPA



Example chromatogram of the media SFM4CHO (HyClone SH30548) using GlutaMAX™ as substrate at different wavelengths

URL <http://itv.hevs.ch>

Contact Dr. Franka Kalman
franka.kalman@hevs.ch
T +41 27 606 86 56