

HIGH THROUGHPUT SCREENING FOR PURIFICATION OF PEGYLATED PROTEINS USING MIXED MODE RESINS.

Postdoctoral researcher: Karla Mayolo-Deloisa
e-mail: K.P.MayoloDeloisa@tudelft.nl
Supervisor: Dr. Ir. Marcel Ottens
Institute: Delft University of Technology
Department of Biotechnology
Bioprocess Engineering Group
Project term: November 2015 – October 2016
Financed by: CONACyT, Tecnologico de Monterrey, TU Delft



DESCRIPTION

PEG–protein conjugates, or PEGylated proteins, are an important class of modern therapeutic drugs. However, PEGylated proteins must be characterized and purified before use in order to meet the stringent regulatory requirements that demand proven clinical efficacy and safety. The process of PEGylation involves formation of a stable covalent bond between activated PEG (polyethylene-glycol) and the polypeptide drug of interest. PEGylation changes the physical and chemical properties of the biomedical molecule, such as its conformation, electrostatic binding, and hydrophobicity; resulting in an improvement in the pharmacokinetic behavior of the drug [1].

Conjugation may result in the generation of a family of species characterized by a distribution in number and position of attached PEG groups [2]. As PEGylated variants are not equal in their effectiveness, it is desirable to resolve heterogeneous conjugate mixtures into effective subpopulations using adequate separation strategies.

Due the above, in this project the use of a High Throughput Screening for separation of PEGylated proteins using mixed mode resins is proposed [3]. The main contribution of this work is the study of the interaction between mixed mode resins and PEGylated proteins. Since until now these resins have not been exploited for the separation of such proteins. To know the effects of several factors as: molecular weight of PEG, type and concentration of salt and type of resin upon the adsorption isotherms will permit to understand the mechanisms that acting in the partition. In addition to establish a new process of purification of PEGylated conjugates.

REFERENCES

1. Cisneros-Ruiz M, Mayolo-Deloisa K, Rito-Palomares M and Przybycien TM (2014). Separation of PEGylated variants of Ribonuclease A and apo- α -Lactalbumin via Reversed Phase Chromatography. *Journal of Chromatography A* 1360:209-216.
2. Mayolo-Deloisa K, Lienqueo ME, Andrews B, Rito-Palomares M and Asenjo JA (2012). Hydrophobic Interaction Chromatography for Purification of monoPEGylated RNase A. *Journal of Chromatography A* 1242:11-16.
3. Nfor B, Noverraz M, Chilamkurthi S, Verhaert P, van der Wielen L, Ottens M. High-throughput isotherm determination and thermodynamic modeling of protein adsorption on mixed mode adsorbents. *Journal of Chromatography A* 1217:6829-6850.