Multi-dimensional chromatographic approaches to characterize protein biopharmaceuticals

Davy Guillarme*[1], Valentina D'Atri[1], Alexandre Goyon[1], Alain Beck[2], Sarah Cianferani[3], Sabine Heinisch[4], Dwight Stoll[5], Jean-Luc Veuthey[1], Szabolcs Fekete[1]

[1] Institute of Pharmaceutical Sciences of Western Switzerland, University of Geneva, CMU-Rue Michel Servet 1, 1211 Geneva 4, Switzerland

[2] Center of Immunology Pierre Fabre, 5 Avenue Napoléon III, BP 60497, 74160 Saint-Julien-en-Genevois, France

[3] Laboratoire de Spectrométrie de Masse BioOrganique, Université de Strasbourg, CNRS UMR7178, IPHC, 67000 Strasbourg, France

[4] Université de Lyon, Institut des Sciences Analytiques, CNRS UMR5280, Université de Lyon, ENS, 69100 Villeurbanne, France

[5] Gustavus Adolphus College, Department of Chemistry, St. Peter, MN, USA

Gold statement
- Learn about protein biopharmaceuticals characterization
- Understand the challenges and benefits of multidimensional liquid chromatography
- Understand which chromatographic modes can be used in both dimensions of 2D-LC-MS

Introduction
The characterization of therapeutic monoclonal antibodies (mAbs) and antibody-drug conjugates (ADCs), is a tremendous challenge to state-of-the-art analytical technologies. Indeed, subtle changes in these large (> 150 kDa) molecules can have profound effects on efficacy and pharmacokinetic properties, thus it is important to have the ability to rapidly and accurately assess changes in the distribution of different isoforms (e.g., glycosylation, oxidation, deamidation, lysine truncation…) of such biomolecules.

Body
Today, the most widely used analytical approaches for therapeutic protein characterization are liquid chromatography (LC) and mass spectrometry (MS), probably due to the remarkable developments of these strategies in the past few years, enabling a new level of performance. However, some chromatographic methods require tedious and time-consuming procedures, especially when the separation cannot be directly coupled with MS detection (e.g., because of the presence of non-volatile salts required for some separation modes) [1].
The aim of this presentation will be to review the possibilities and trends of multidimensional LC and LC-MS, which provides rich opportunities to increase both the efficiency of the characterization process, and the value of the information gained from these analyses. During this presentation, various combinations of chromatographic dimensions will be explored for the deep characterization of biopharmaceuticals at the intact protein, middle-up and bottom-up levels of analysis [2].

Conclusion
Multidimensional LC combined with mass spectrometry appears as a powerful tool to make non-denaturing modes of chromatography compatible with MS, and also to automate the peptide mapping and bottom-up approach.

References
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