Contribution ID: **80** Type: **Poster**

Quantification of monoclonal antibodies by middle-up capillary electrophoresis-mass spectrometry

Monoclonal antibodies (mAbs) are biopharmaceuticals widely used in treatment of various diseases. Due to their intrinsic complexity and post-translational modifications, therapeutic mAbs are highly heterogeneous molecules requiring extensive quality control. Quantitative analysis typically relies on liquid chromatography coupled to tandem mass spectrometry (LC-MS/MS) and the gold-standard bottom-up (BU) proteomic approach. This study presents an alternative middle-up (MU) approach combined with capillary zone electrophoresis–mass spectrometry (CZE–MS). CZE has previously demonstrated suitability for mAb analysis and is recognised as a quality control technique in the pharmaceutical industry. Here, we focus on the quantitative MU CZE–MS analysis of the immunoglobulin G (IgG) mAb infliximab (IFX) in the pharmaceutical product Remicade and two other biopharmaceuticals Avastin and MabThera.

IFX calibration standards were prepared at six concentrations, and mAb standard solution of adalimumab (ADA) (Merck, Darmstadt, Germany) as internal standard was added to each sample. Next, samples were reduced with TCEP reducing agent at 37 °C for 1 hour, then analysed using an Agilent 7100 CE system coupled to an Agilent 6410 triple quadrupole mass spectrometer (both Agilent, Santa Clara, USA). Pharmaceutical formulations of Remicade, Avastin, and MabThera were diluted to 20 µg/mL and analysed using the same MU CZE–MS workflow.

The developed method was validated in accordance with the ICH Q2(R1) guidelines for analytical method validation. It demonstrated good accuracy and precision for IFX quantification in Remicade, and was further applied to quantify bevacizumab and rituximab in their respective drug formulations. The results indicate that the optimised MU CZE–MS method is suitable for the quantitative analysis of various IgG therapeutic mAbs.

User consent

yes

Author: HAVLIKOVA, Jana (Comenius University Bratislava)

Co-authors: MARAKOVA, Katarina (Comenius University Bratislava); MIKUS, Peter (Comenius University Bratislava)

Presenter: HAVLIKOVA, Jana (Comenius University Bratislava)

Session Classification: Poster Session 2