

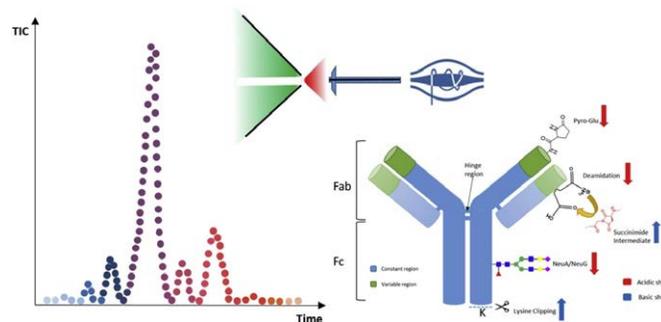
ZipChip

In-depth analysis of monoclonal antibodies using microfluidic capillary electrophoresis and native mass spectrometry

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ABSTRACT: Charge variant profiling of therapeutic proteins is required by the International Council for Harmonisation guidelines and is traditionally performed by capillary electrophoresis or ion exchange chromatography. Recently, improvements in the hyphenation of capillary electrophoresis with mass spectrometry and the introduction of mass spectrometry compatible background electrolytes has allowed the implementation of native mass spectrometric determination of the charge variant profile obtained from the electrophoretic separation. The low flow operation of the microfluidic electrophoretic platform significantly boosts mass spectrometric sensitivity and increases the dynamic range, even when using sample amounts as low as 1 ng in capillary. In the current study, rituximab, trastuzumab and bevacizumab drug products were analysed using the ZipChip microfluidic CE-ESI-MS platform that facilitated confident identification of proteoforms with an average mass accuracy of < 15 ppm. Up to 52 proteoforms were identified for trastuzumab drug product, while rituximab sample revealed the presence of fragments and sialylated N-glycans. Overall, the CE-ESI-MS platform proved to be a fast and robust tool for therapeutic protein charge variant profiling and facilitated efficient coupling with native mass spectrometry for the generation of highly informative characterisation data.



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