

Cyclodextrins as modulators for separation of charged variants of mAbs by capillary zone electrophoresis

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Abstract

Charge heterogeneity is a critical quality attribute for mAb-based therapeutics. Its characterization involves the identification of charge variant species in the final formulation. Capillary zone electrophoresis (CZE) is a valuable tool to assess charge heterogeneity, but typically offers incomplete separation of the different charged isoforms. Cyclodextrin-mediated chiral CE is a well-established electromigration method. In this study, we propose the use of cyclodextrins as modulators for CZE based separation of mAb charge variants. Uncoated fused silica capillaries have been used for examining charge heterogeneity in biosimilars of Rituximab and Trastuzumab in presence of different cyclodextrins. Both the negatively charged CM β CD and uncharged HP β CD have been explored. The effect of cyclodextrin concentration has been found to impact the extent of modulation of charge variants resolution. Addition of 10 mM CM β CD not only improved resolution but also resulted in separation of two additional basic and one additional acidic species for Rituximab. Similarly, besides improvement in the resolution of separation, one additional basic variant was observed with 2.5 mM CM β CD with another sample, Trastuzumab. The study demonstrates that the effect of CM β CD is mAb specific. No improvement in resolution was, however, observed with HP β CD, suggesting the selective nature of cyclodextrins for mAb variants.