Antibody Purification and On-cartridge Deglycosylation/Ides Proteolysis

Extracellular Domain

Protein L

Experimental

Evaluation of Deglycosylation/Ides Proteolysis

Comparison of deconvoluted spectra of antibody purified Trastuzumab without and with Ides Protein F on the extracellular domain. Complete deconvolution of (A) ECD captured mAb and (B) protein L captured mAb. Comparison of deconvoluted spectra of antibody purified Trastuzumab without and with Ides on-cartridge proteolysis show complete deconvolution of (C) ECD captured mAb and (D) protein L captured mAb. The mass traces indicate elution and similar traces indicate flowthrough.

Conclusions

Agilent provides a rapid and versatile antibody characterization solution that includes automated affinity purification and on-cartridge enzymatic reaction with the AssayMAP Bravo. Subsequently, spectra were acquired with the 1298 Infinity UPLC coupled to a 0.5 TDF 6500, and were deconvoluted with the MassHunter BioConfirm software. This protein characterization solution offers:

- Decreases variability, probability of human error while simultaneously increasing scalability with minimal additional labor
- Purifies mAbs from spent CHO cell medium with high yield and purity
- Generates high resolution spectra
- Provides easy antibody intact mass characterization.

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