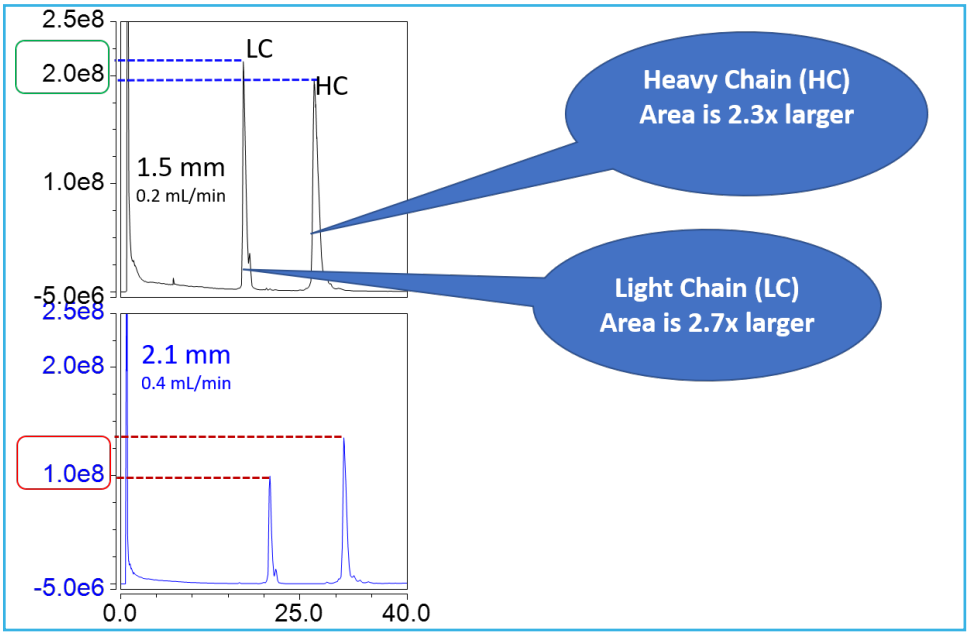
HALO®

BIOPHARMACEUTICALS

Demonstration of Increased Sensitivity with Reduced Trastuzumab Using a 1.5 mm ID Column

286-BIO



PEAK IDENTITIES

LC = Light chain
HC = Heavy chain

TEST CONDITIONS:

Column: HALO 1000 Å Diphenyl, 2.7 µm, 1.5 x 150 mm
Part Number: 9212X-702
Column: HALO 1000 Å Diphenyl, 2.7 µm, 2.1 x 150 mm
Mobile Phase A: Water/0.1% DFA
Mobile Phase B: 50% Acetonitrile/50% n-propanol/0.1% DFA
Gradient:

Time (min)	%B
0.0	27
40.0	36
40.1	27
45.0	27

Flow Rate: 0.2 mL/min for 1.5 mm ID
0.4 mL/min for 2.1 mm ID
Back Pressure: 252 bar (1.5 mm)
272 bar (2.1 mm)
Temperature: 60 °C
Detection: ESI +
Injection Volume: 3 µL of 1.0 mg/mL reduced and alkylated trastuzumab
Sample Solvent: Water/0.1% TFA
LC System: Shimadzu Nexera X2
MS System: ThermoFisher Q Exactive

MS CONDITIONS:

Spray Voltage (kV): 3.8
Capillary temperature: 320 °C
Sheath gas: 35
Aux gas: 10
RF lens: 50

A separation of intact Trastuzumab was performed on a HALO 1000 Å Diphenyl column. The switch from a 2.1 mm ID column to a 1.5 mm ID allows for an increase in sensitivity and reduces overall solvent consumption. In this example both peak intensity and area are increased. This sensitivity was acheived by optimizing the post-column tubing. The 1.5 mm ID column is ideal for achieving more performance from a UHPLC system saving on the investment of a specialized low flow HPLC.

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