

Intact and Oxidized Monoclonal Antibody (mAb) Separation on Proteomix HIC

Proteomix[®] HIC Butyl NP5, 5 μ m, 4.6 X 100 mm
(PN: 431NP5-4610)

Proteomix[®] HIC Butyl NP5, 5 μ m, 4.6 X 35 mm
(PN: 431NP5-4603)



Rituximab Oxidation Analysis on HIC Butyl

HICM1006

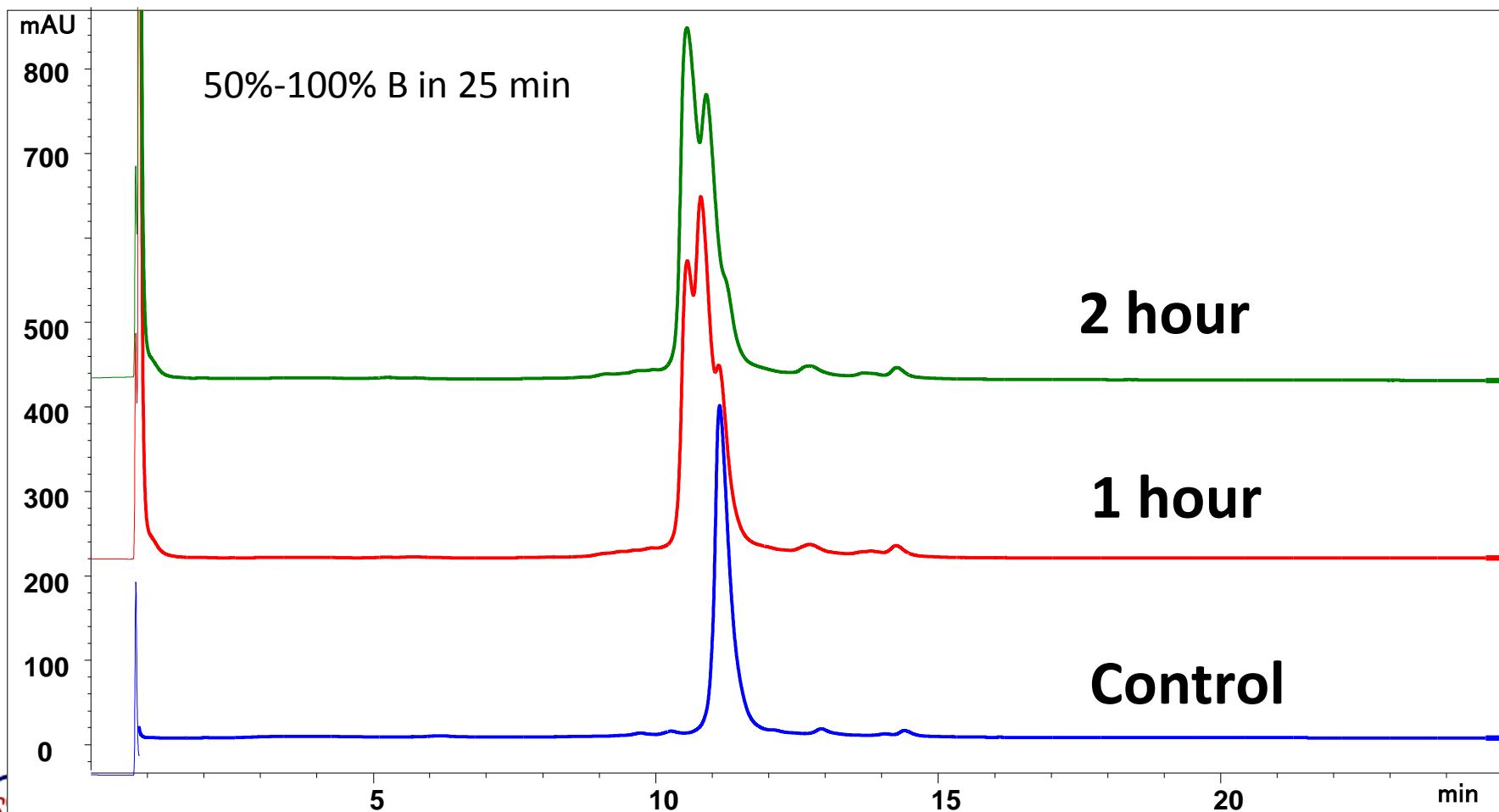
Column: Proteomix® HIC Butyl NP5, 5 µm, 4.6 x 100 mm (PN: 431NP5-4610)

Mobile phase: A: 100 mM sodium phosphate buffer, 2 M ammonium sulfate, pH 7.0, B: 100 mM sodium phosphate buffer, pH 7.0;

Flow rate: 0.8 mL/min, Pressure: 170 bar, System: UHPLC, Detector: UV 214 nm, Column temperature: 25°C,

Injection: 10 µg mAb sample, 20 µg oxidized mAb,

MAB oxidation: 10 mg/mL was diluted to 5 mg/mL with water and 70% t-BHP was added to a final 5% concentration, incubate in dark and take time point, dilute to 2.5 mg/mL in 500 mM ammonium sulfate, 25 mM phosphate buffer



Rituximab Oxidation Analysis on HIC Butyl – 1 hour HICM1006

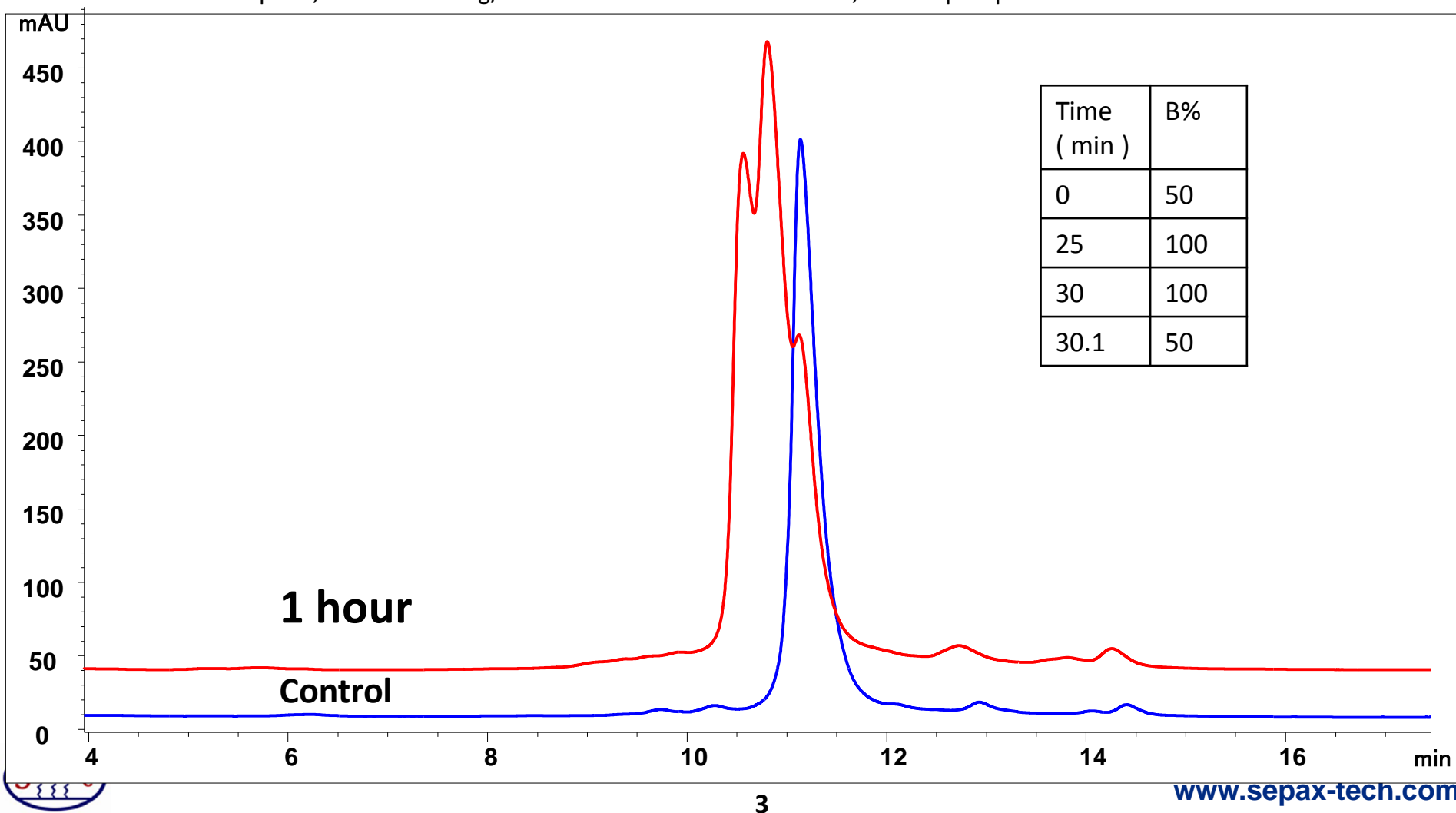
Column: Proteomix® HIC Butyl NP5, 5 µm, 4.6 x 100 mm (PN: 431NP5-4610)

Mobile phase: A: 100 mM sodium phosphate buffer, 2 M ammonium sulfate, pH 7.0, B: 100 mM sodium phosphate buffer, pH 7.0;

Flow rate: 0.8 mL/min, Pressure: 170 bar, System: UHPLC, Detector: UV 214 nm,, Column temperature: 25°C,

Injection: 10 µg mAb sample, 20 µg oxidized mAb,

MAB oxidation: 10 mg/mL was diluted to 5 mg/mL with water and 70% t-BHP was added to a final 5% concentration, incubate in dark and take time point, dilute to 2.5 mg/mL in 500 mM ammonium sulfate, 25 mM phosphate buffer



Rituximab Oxidation Analysis on HIC Butyl – 2 hour HICM1006

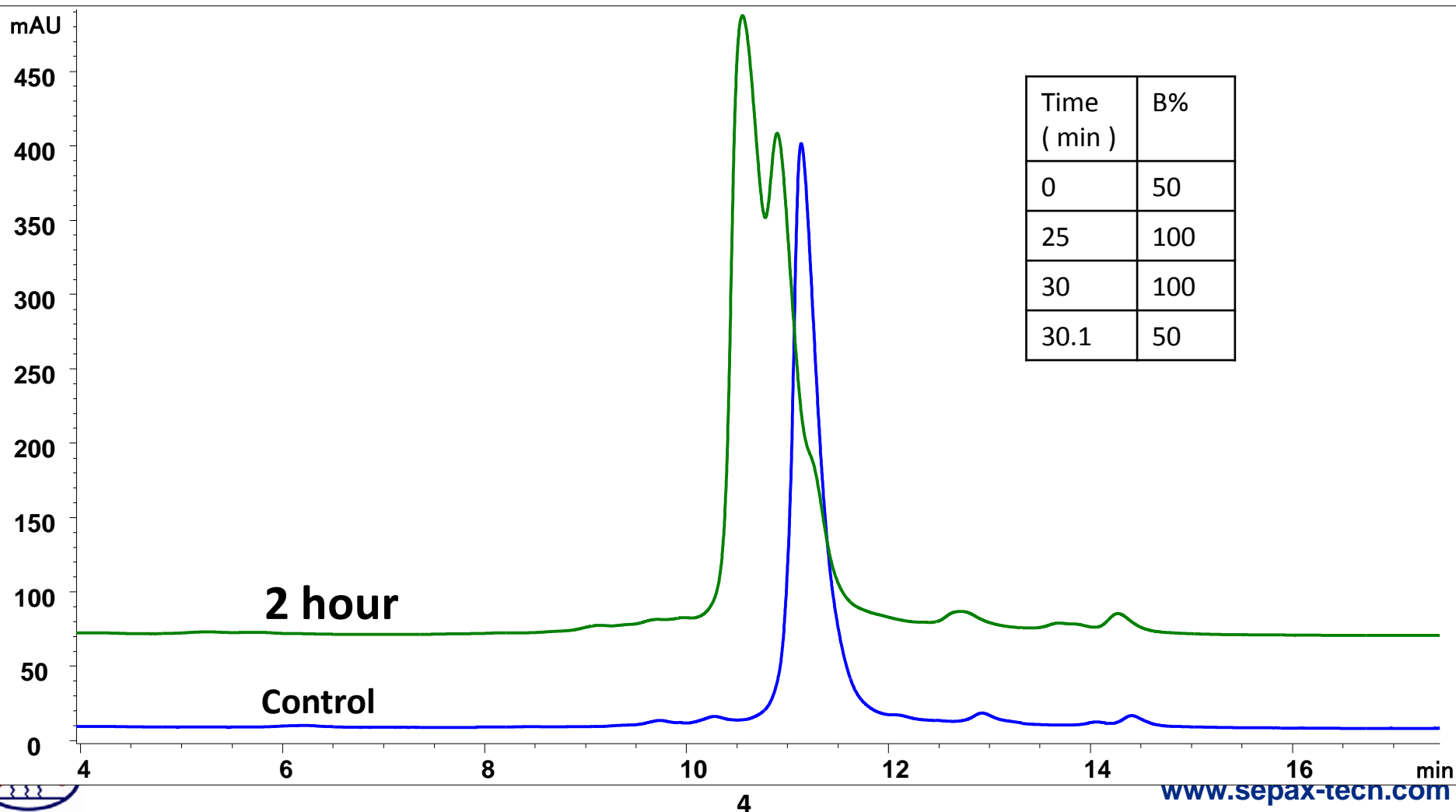
Column: Proteomix® HIC Butyl NP5, 5 µm, 4.6 x 100 mm (PN: 431NP5-4610)

Mobile phase: A: 100 mM sodium phosphate buffer, 2 M ammonium sulfate, pH 7.0, B: 100 mM sodium phosphate buffer, pH 7.0;

Flow rate: 0.8 mL/min, Pressure: 170 bar, System: UHPLC, Detector: UV 214 nm,, Column temperature: 25°C,

Injection: 10 µg mAb sample, 20 µg oxidized mAb,

MAb oxidation: 10 mg/mL was diluted to 5 mg/mL with water and 70% t-BHP was added to a final 5% concentration, incubate in dark and take time point, dilute to 2.5 mg/mL in 500 mM ammonium sulfate, 25 mM phosphate buffer



RituximAb- Proteomix HIC Butyl NP5

Column Length Impact - 4610 vs. 4603

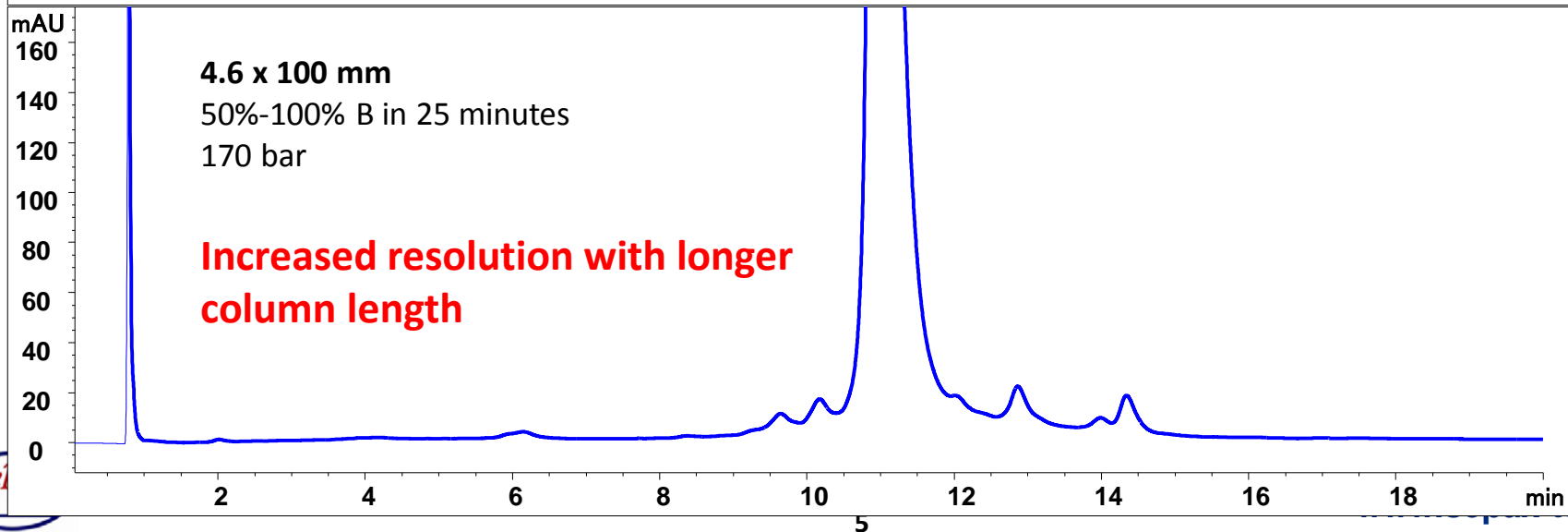
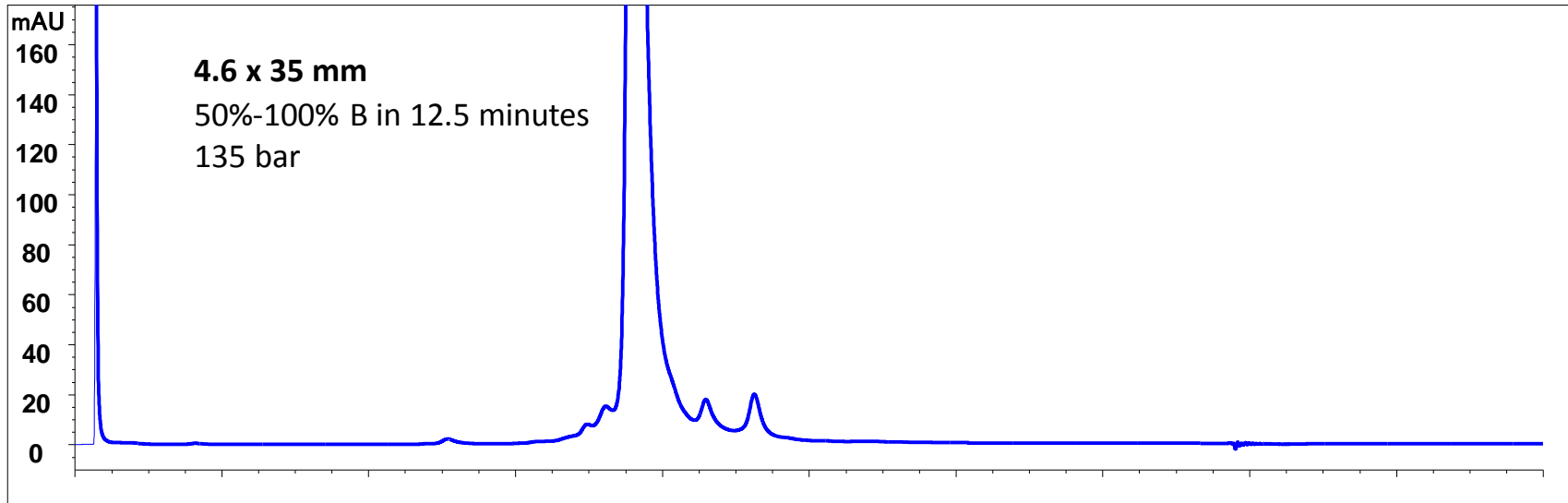
HICM1006

Column: Proteomix® HIC Butyl NP5, 5 µm, 4.6 x 35 mm, 4.6 x 100 mm (PN: 431NP5-4603, PN: 431NP5-4610)

Mobile phase: A: 100 mM sodium phosphate buffer, 2 M ammonium sulfate, pH 7.0, B: 100 mM sodium phosphate buffer, pH 7.0;

Flow rate: 0.8 mL/min, System: UHPLC, Detector: UV 214 nm, Column temperature: 25°C

Injection: 10 µg mAb for 4603, 20 µg for 4610, 2.5 mg/mL rituximab in 500 mM ammonium sulfate, 25 mM phosphate buffer



Rituximab Separation Sepax vs. Vendor T

(same condition as previous slide)

