

2.1 mm ID Size Exclusion chromatography for Protein Separation

Native volatile buffer
200 mM ammonium acetate, pH 7.0
Vs.
Native salt buffer
150 mM sodium phosphate, pH 7.0

Native volatile buffer can be applied to native protein SEC-LC-MS analysis without denaturing the protein.

Reference: Anal. Chem. 2012, 84, 2843–2849,
Native Intact Mass Determination of Antibodies Conjugated with Monomethyl
Auristatin E and F at Interchain Cysteine Residues



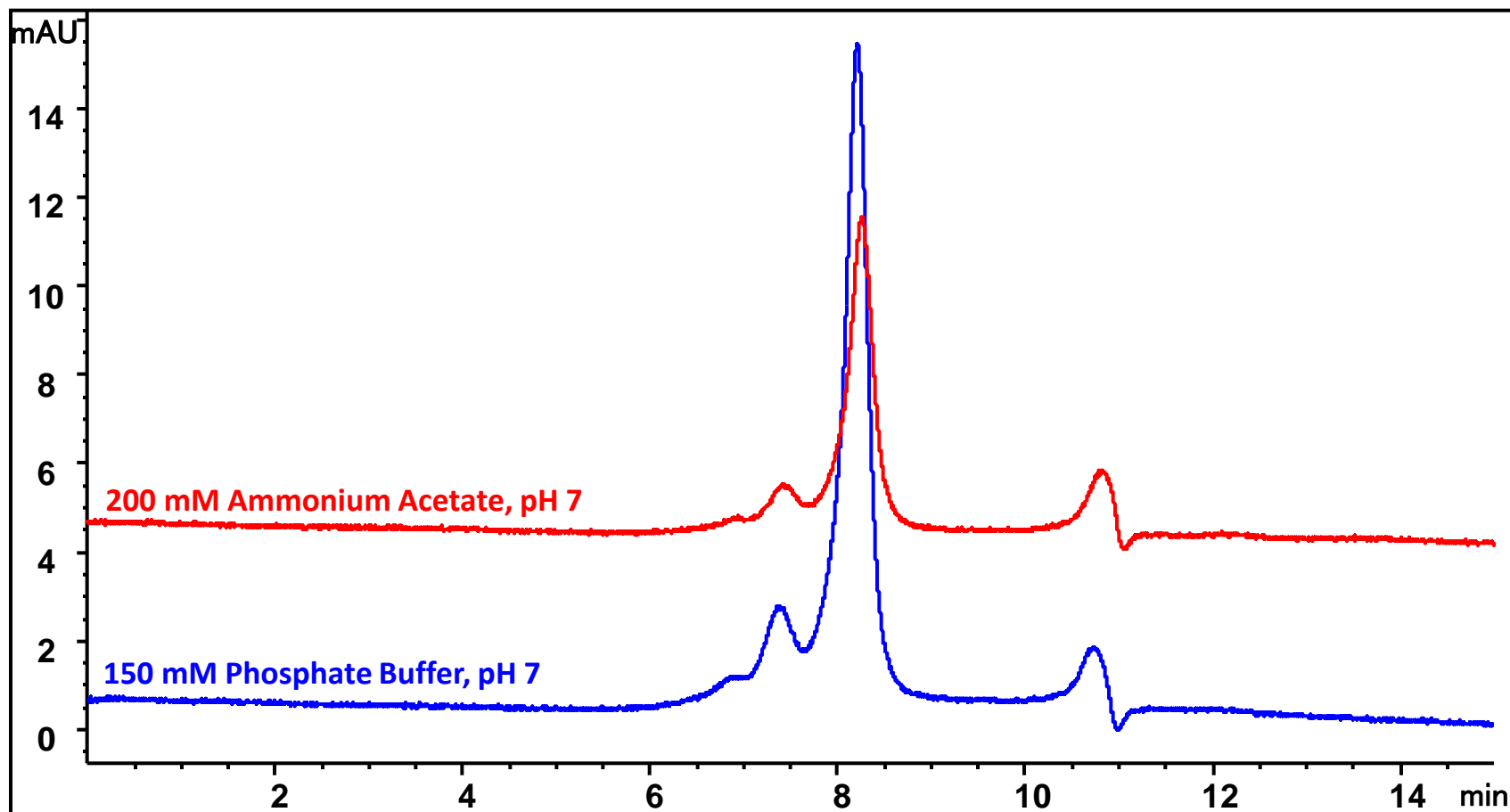
BSA SEC Analysis

Column: Zenix-C SEC-300 (3 μm , 300 \AA , 2.1 x 300 mm)

Mobile phase: See chromatogram

Flow rate: 0.08 mL/min; Detector: UV 280 nm; Column temperature: 25 $^{\circ}\text{C}$

Injection volume: 1 μL , Sample: BSA 1 mg/mL



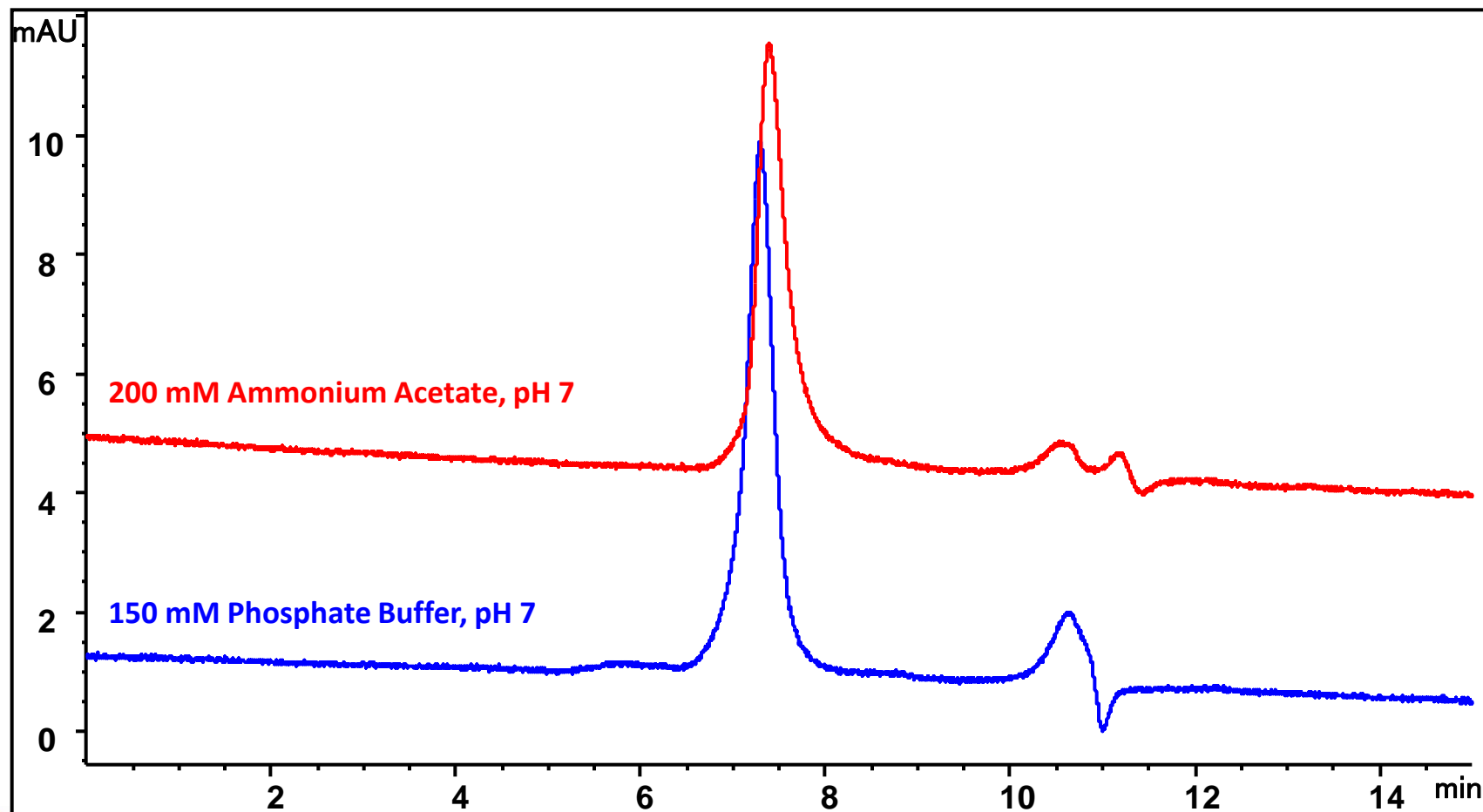
Monoclonal antibody analysis

Column: Zenix-C SEC-300 (3 μm , 300 \AA , 2.1 x 300 mm)

Mobile phase: See chromatogram

Flow rate: 0.08 mL/min; Detector: UV 280 nm; Column temperature: 25 $^{\circ}\text{C}$

Injection volume: 1 μL , Sample: mAb 1 mg/mL



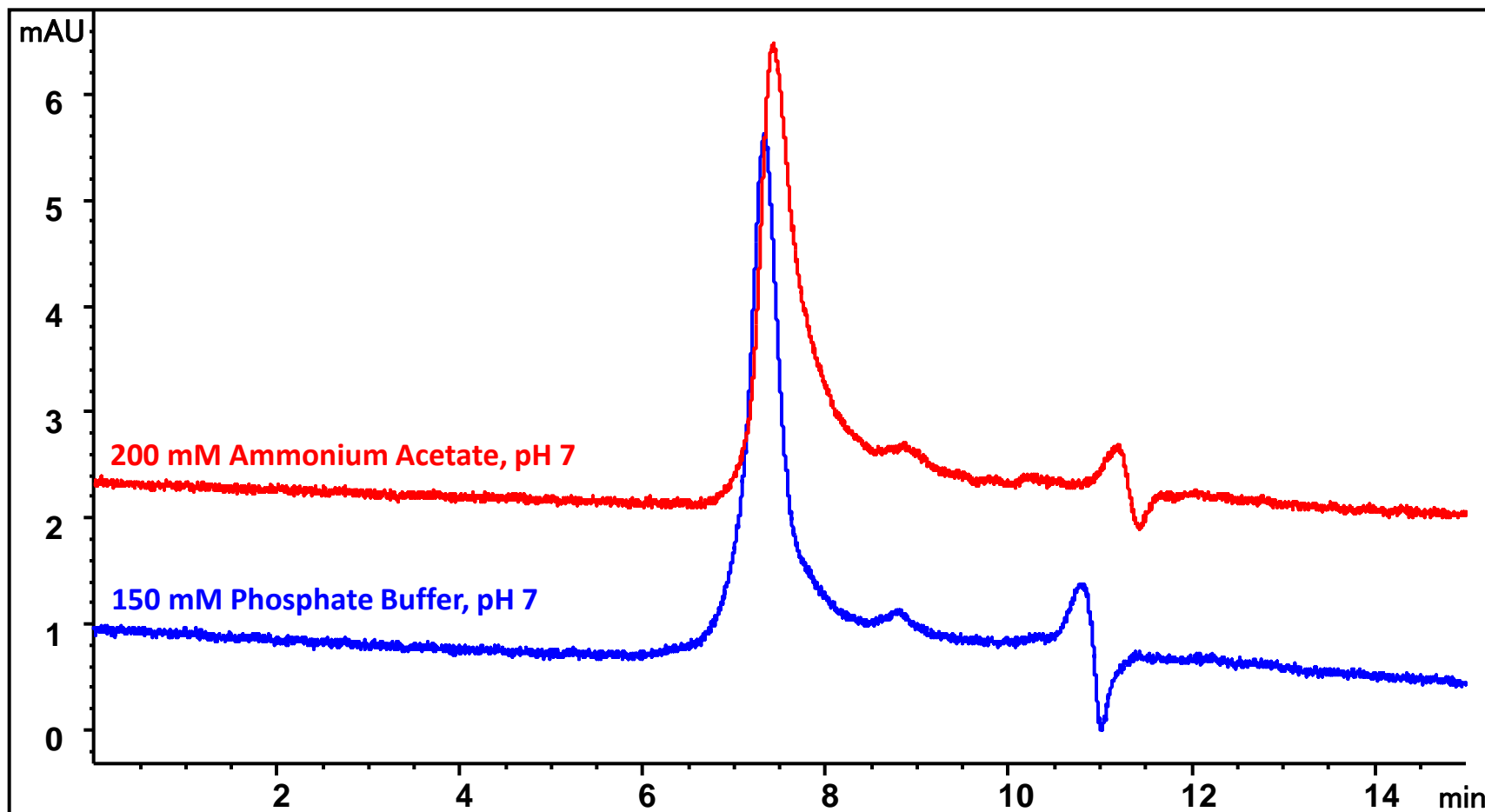
Antibody drug conjugate #1-cysteine conjugate with cleavable linker

Column: Zenix-C SEC-300 (3 μm , 300 \AA , 2.1 x 300 mm)

Mobile phase: See chromatogram

Flow rate: 0.08 mL/min; Detector: UV 280 nm; Column temperature: 25 $^{\circ}\text{C}$

Injection volume: 1 μL , Sample: ADC #1 1 mg/mL



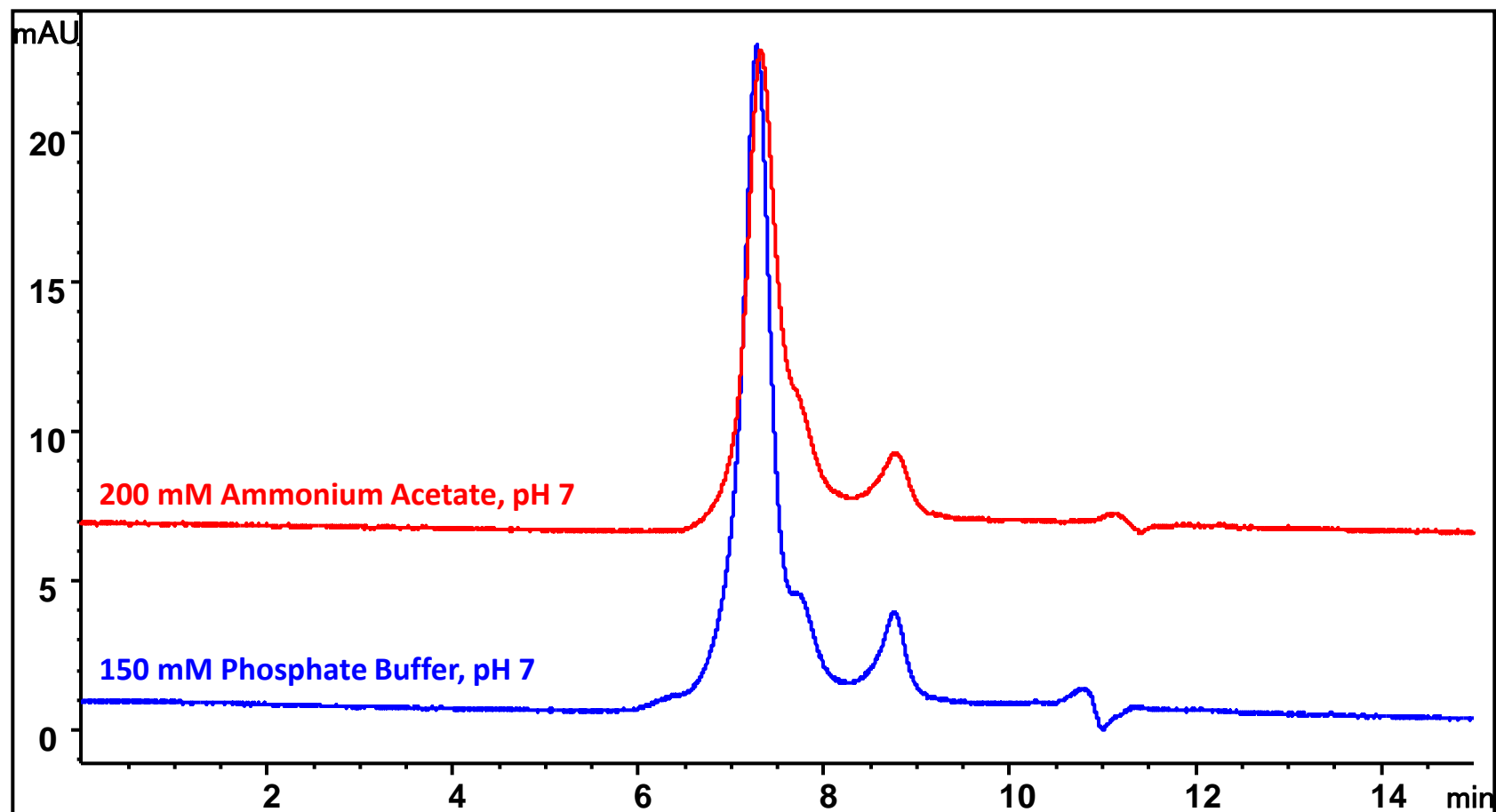
Antibody drug conjugate #2-cysteine conjugate with non-cleavable linker

Column: Zenix-C SEC-300 (3 μm , 300 \AA , 2.1 x 300 mm)

Mobile phase: See chromatogram

Flow rate: 0.08 mL/min; Detector: UV 280 nm; Column temperature: 25 $^{\circ}\text{C}$

Injection volume: 1 μL , Sample: ADC #2 1mg/mL



Antibody drug conjugate #3 - Lysine conjugate

Column: Zenix-C SEC-300 (3 μm , 300 \AA , 2.1 x 300 mm)

Mobile phase: See chromatogram

Flow rate: 0.08 mL/min; Detector: UV 280 nm; Column temperature: 25 $^{\circ}\text{C}$

Injection volume: 1 μL , Sample: ADC #3 1mg/mL

