Zenix™ SEC-80

Having trouble applying size exclusion chromatography to peptides under 10,000 Da? What if there was a size exclusion column specifically designed for small protein and peptide separations?

Sepax Zenix™ SEC-80 provides a valuable solution

Highlighted FACTS:

***3um and 80Å**:* high efficiency 3um, 80Å pore for high resolution separation of peptides with molecular weights in the range of 1kD to 6kD.

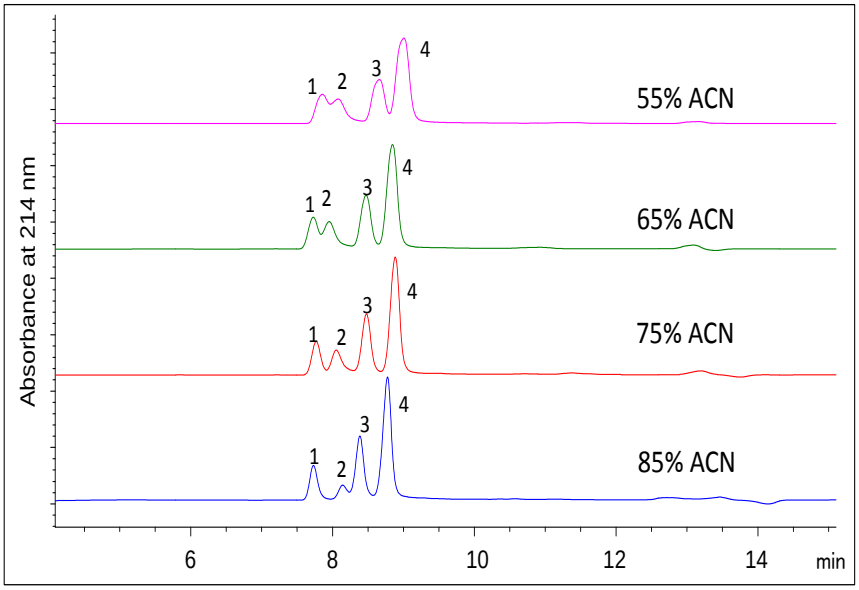
***Mobile Phase Compatibility**:* compatible with different mobile phases, including TFA/acetonitrile and methanol additives for peptide separation.

***Peptides**:* insulin from porcine pancreas, glucagon, Angiotensin I and Bradykinin are separated on Zenix™ SEC-80 with good resolution.

***Complex Peptide Mixtures**:* E. coli tryptic digests are fractionated successfully on Zenix™ SEC-80.

Effect of Acetonitrile Mobile Phase Concentration

Column: Zenix™ SEC-80 (3 µm, 7.8x300 mm)
Mobile phase: 0.1% TFA with the indicated percentage of acetonitrile
Flow rate: 0.8 mL/min; Injection volume: 5 µL
Detection: UV214 nm; Temperature: Ambient (~25 ℃)
Sample:
1. Insulin (0.5mg/mL)
2. Glucagon (0.5mg/mL)
3. Angiotensin I (0.5mg/mL)
4. Bradykinin (0.5mg/mL)



Separation parameters for peptide mixture using 75% acetonitrile with 0.1% TFA in water

Peak	Protein	MW (Da)	Retention time (min)	Resolution	Plate counts
1	Insulin (porcine)	5778	7.75		16711
2	Glucagon	3483	8.03	1.07	12132
3	Angiotensin I	1297	8.46	1.58	19741
4	Bradykinin	1060	8.86	1.65	21060

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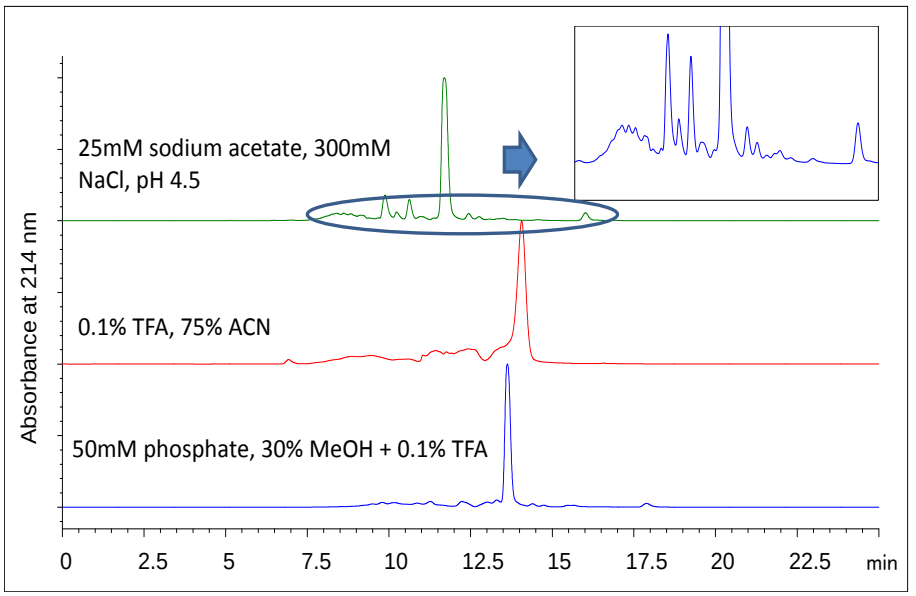
What is Zenix™ SEC-80

Sepax Zenix™ SEC-80 (Size Exclusion): Ultra-high efficiency and resolution SEC column. Zenix™-80 SEC 3um is specifically designed for small protein and peptide separations. Delivering unrivaled resolving power and reproducibility. Made of uniform, hydrophilic, and neutral nanometer thick proprietary surface coating chemically bonded on silica, offers long column lifespan and negligible non-specific interactions.

Technical Specifications:

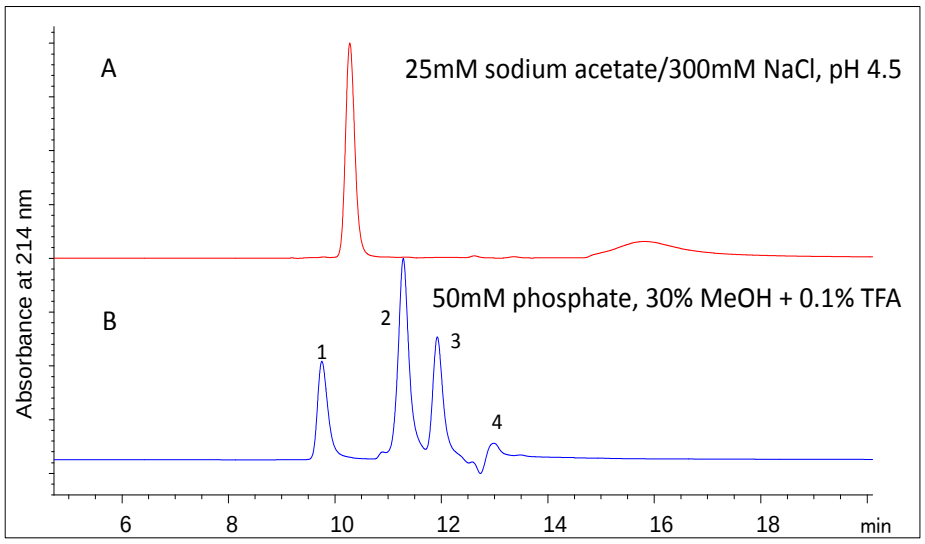
Phase	Zenix™ SEC-80
Material	Neutral, hydrophilic film bonded silica
Particle size (μm)	3
Pore size (Å)	~ 80
Protein MW range (native)	*Up to 50,000*
pH stability	2 – 8.5 (pH 8.5-9.5 can be tolerated temporarily)
Backpressure (psi) for 7.8x300 mm (1.0 mL/min)	~ 1,500
Maximum backpressure	~ 4,500
Salt concentration range	20 mM - 2.0 M
Maximum temperature	~ 80 °C
Mobile phase compatibility	Aqueous and organic



Separation of E. coli digests on Zenix™ SEC-80 (7.8x300mm)

For complex E. coli tryptic digest, a mobile phase of 25 mM sodium acetate/300 mM NaCl gave the best separation. The bottom chromatogram was run with 50mM phosphate/30%MeOH/0.1% TFA. The middle one was run with 0.1% TFA/75% ACN. The top chromatogram represents the best degree of peptide separation and was run with 25mM sodium acetate/300mM NaCl, pH 4.5.

Effect of Salt and Organic Additives on Peptide Separation



Peptide separation on Zenix™ SEC-80 (7.8 x 300mm).

Panel A was with mobile phase 25mM sodium acetate and 300mM NaCl, pH 4.5. Panel B was with mobile phase 50mM phosphate, 30% MeOH and 0.1% TFA. Peak elution order: 1. Insulin, 2. Bradykinin, 3. Angiotensin I, 4. Glucagon. The flow rate was 0.8 ml/min.

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