

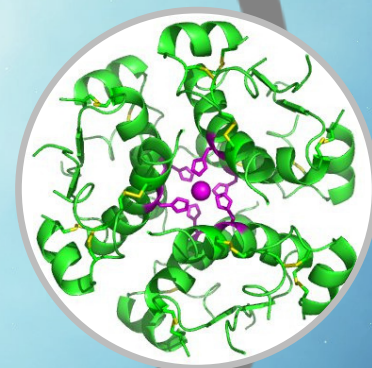
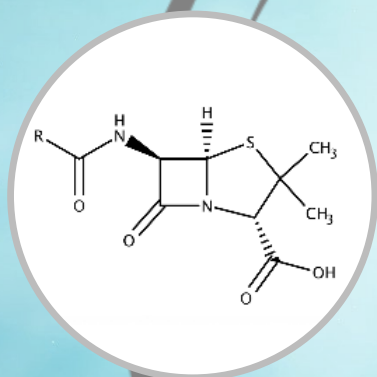


Sepax Technologies

PolyRP™

REVERSED PHASE CHROMATOGRAPHY

POLYMERIC PROCESS MEDIA FOR
BIOMOLECULE SEPARATIONS



Our Specialty

Sepax Technologies, a Delaware US-based company, provides cutting edge products and services for liquid chromatography (LC). Sepax specializes in the development and manufacture of LC analytical, preparative and process separation & purification columns, bulk resins and systems in a wide range of modalities, such as SEC, IEX, HIC, Affinity, and RP.

Sepax also provides LC services, including analytical testing, method optimization, purification, custom resin development, and ligand immobilization. Certified to the ISO 9001-2015 standards, Sepax focuses on customer & market needs, and is continuing to expand its presence and supply chain around the globe in three business platforms: Analytical Chromatography, Industrial Purification and Medical Diagnostics.

Our Commitment

At Sepax, we create value through serving customers' needs and solving their chromatographic separation and purification challenges. Through innovative technologies and solution-based approaches, Sepax delivers products and services that build lasting relationships with customers, achieving a strong leadership role in the industry. At Sepax, we firmly believe that there is nothing too complicated or challenging for us to consider.

Our Strategy

Whether you are conducting analytical research, in need of customized resins, or scale-up purification, Sepax Services offers unmatched technical capabilities and expertise. Working in tandem with our technical team and our customers, Sepax offers highly individualized services to meet your specific requirements, achieving project goals in an efficient and cost-effective manner.



PolyRP™

Introduction

PolyRP bulk media are highly crosslinked spherical resins made of styrene and divinylbenzene. These highly rigid resins are narrowly dispersed particles with particle size selection of 10, 15, and 30 μm and pore size selection of 100, 300, 500 and 1000 \AA . These resins have abundant phenyl surface functional groups that enable hydrophobic interaction which is useful in reversed phase separation.

PolyRP bulk media are highly stable over a variety of operational conditions. They are stable to resist high temperatures up to 200°C (operating temperature up to 80°C). They are compatible with many commonly used organic solvents and aqueous buffers. PolyRP bulk media have a long life time. With a well-controlled polymer resin manufacturing process, PolyRP bulk media are very reproducible from batch to batch. Compared to silica based reversed phases, PolyRP bulk media are more stable at extreme pH (1- 14) with a similar separation efficiency and unique selectivity. **Figure 1** shows 10 μm PolyRP resins with pore sizes of 100, 300, and 1000 \AA .

SEM Analysis on Particle Morphology

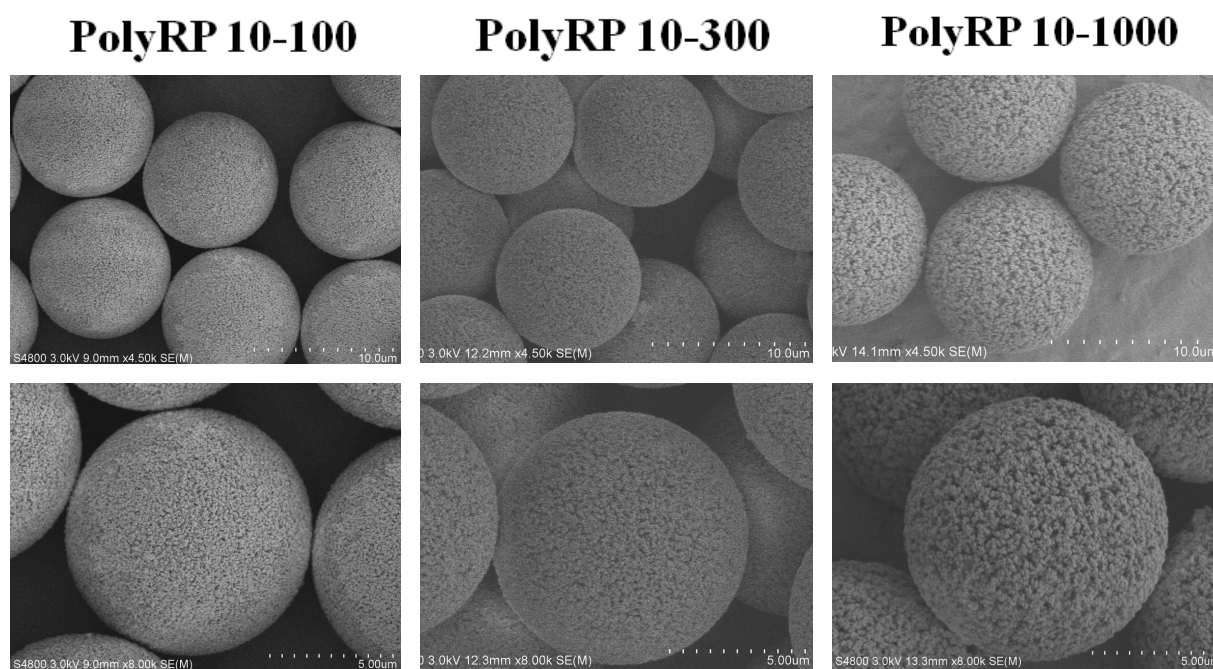


Figure 1. Rigid, spherical, monodispersed, porous microspheres. Precise control on particle morphology: bead size, pore size, surface area, pore volume.

Features

- PolyRP resins are narrowly dispersed particles
- Well controlled particle size 10, 15, 30 μm
- Well controlled pore size at 100, 300, 500 and 1000 \AA
- Strong mechanical strength
- Wide pH operation range 1-14
- High capacity and loading
- High separation, high resolution and efficiency

Resin Technical Specifications

Resin	PolyRP 10	PolyRP 15	PolyRP 30
Matrix	Porous polystyrene/divinylbenzene microspheres		
Particle Size (μm)	10.0 \pm 1.0	15.0 \pm 1.5	30.0 \pm 3.0
Particle Size Distribution (D_{90}/D_{10})	≤ 1.3		
Average Pore Size (\AA)	100, 300, 500, 1000		
Bulk Density (g/mL)	0.26 – 0.32	0.20 – 0.32	0.20 – 0.32
Specific Surface Area (m^2/g)	200 - 1000	200 - 1000	200 - 1000
Specific Pore Volume (mL/g)	0.9 – 1.4	0.9 – 2.4	0.9 – 2.4
Expansion in Methanol (vol%)	< 5	< 5	< 5
Max Pressure	15 MPa (150 bar)	10 MPa (100 bar)	10 MPa (100 bar)
Operation temperature	$\leq 80\text{ }^\circ\text{C}$		
pH range	1 – 13 for use; 1 – 14 for CIP		
Compatible Solvents	Compatible with many commonly used organic solvents and aqueous solution such as a mixture of water and acetonitrile, acetone, methanol, ethanol, n-propanol, THF; 1 M HCl, 1 M NaOH, 1 M HCl/ 90% methanol, 90% HAc, 0.45 M NaOH/40% isopropanol, 6 M guanidine		
CIP and Regeneration	Following solvents can be used alone or in combination: 0.5-1 M NaOH, 1 M HCl, 90% acetonitrile or isopropyl alcohol, 90% acetic acid, 3% TFA		
Autoclavable	20 min at 121 $^\circ\text{C}$		
Storage	2-30 $^\circ\text{C}$, 20% ethanol		

Evaluation of DAC Column

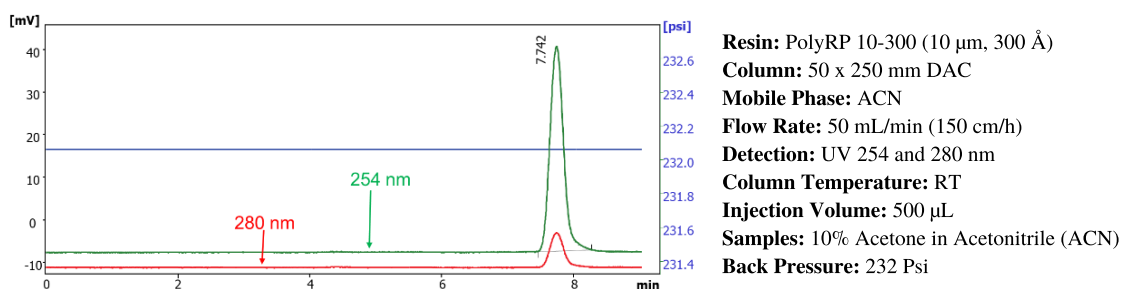


Figure 2. PolyRP 10-300 efficiency, $N=27,028/\text{m}$ and peak symmetry of 1.14.

Mobile Phase Impact on Back Pressure

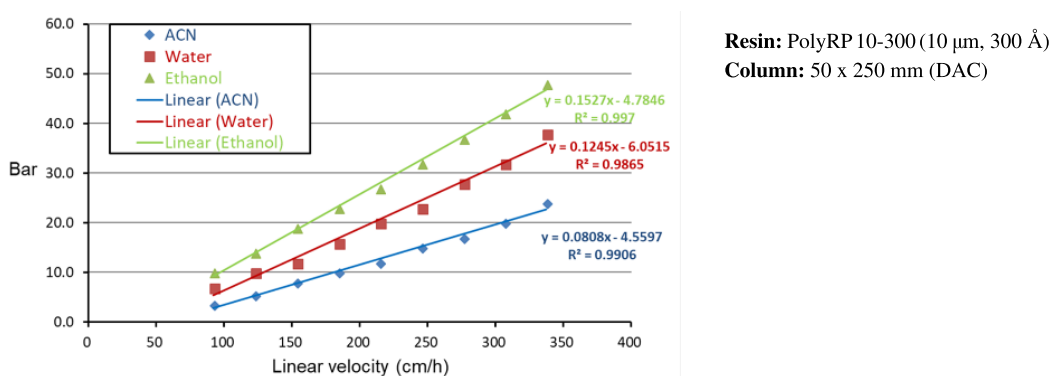


Figure 3. Net back pressure vs. flow-rate characteristics of PolyRP10-300. Typical flow rate range is 100-600 cm/h.

Insulin Analog Separation Using PolyRP 10-300 Scale up

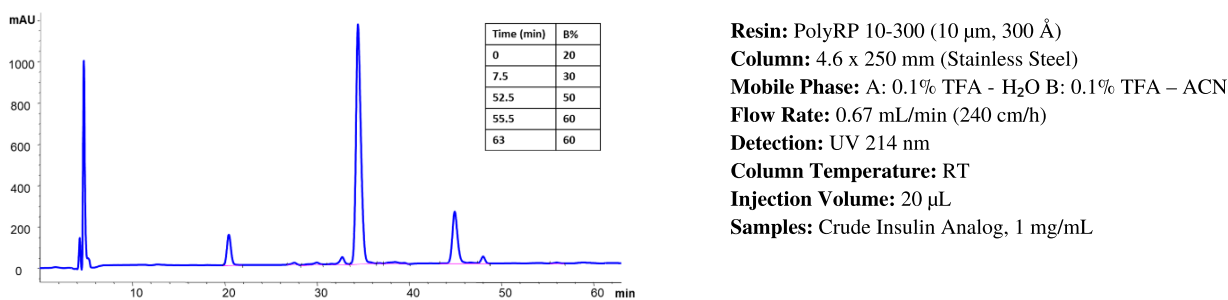


Figure 4. PolyRP 10-300 bulk media separated crude Insulin analog based on hydrophobic interaction.

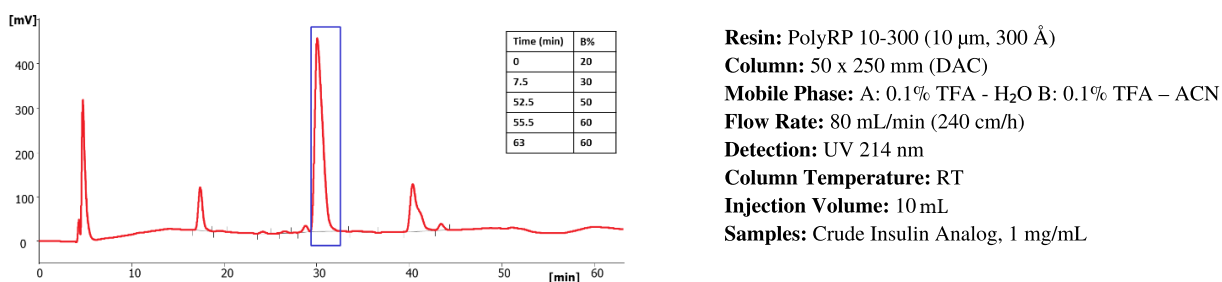


Figure 5. Scale-up is performed successfully from 4.6 x 250 mm column to 50 x 250 mm DAC column.

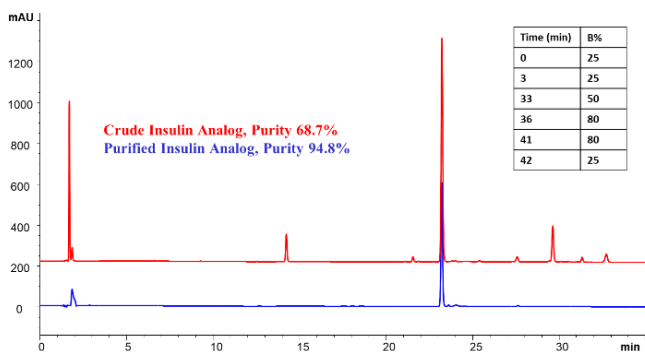


Figure 6. Purity analysis of crude and purified insulin analog by HPLC.

Resin: GP-C18 (3 μm , 120 \AA)
Column: 4.6 x 150 mm (Stainless Steel)
Mobile Phase: A: 0.02 M Na_2SO_4 -Triethanolamine (1%), pH2.3 B: ACN
Flow Rate: 1.0 mL/min (360 cm/h)
Detection: UV 214 nm
Column Temperature: 40°C
Injection Volume: 5 μL
Pressure: 97-172 bar

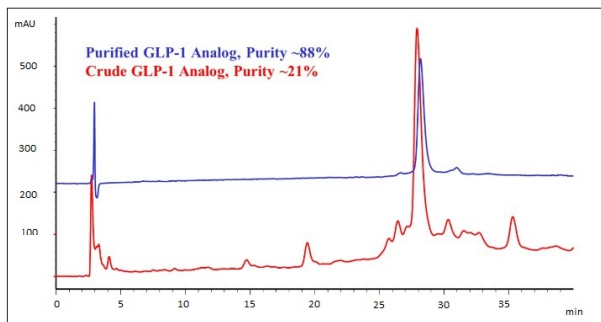


Figure 7. PolyRP separated crude GLP-1 analog based on hydrophobic interaction.

Resin: PolyRP 10-300 (10 μm , 300 \AA)
Column: 4.6 x 250 mm (Stainless Steel)
Mobile Phase: A: 0.1% TFA - H_2O , B: 0.1% TFA - ACN
Flow Rate: 1.0 mL/min (360 cm/h)
Detector: UV 214 nm
Column Temperature: RT
Injection Volume: 100 μL
Gradient: 0-40 min, 30-60% B

Pentapeptide Separation Using PolyRP 10-300

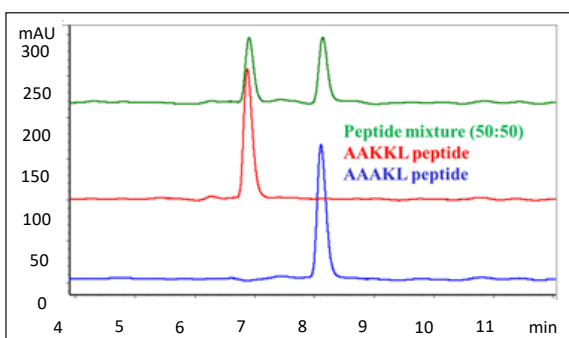
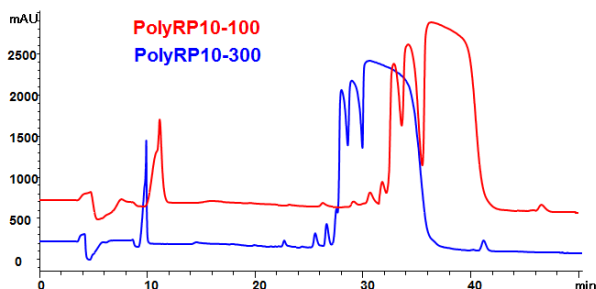


Figure 8. PolyRP 10-300 separated two pentapeptides which are different by only one amino acid. More polar AAKKL pentapeptide eluted first. Alanine (A), Lysine (K), Leucine (L).

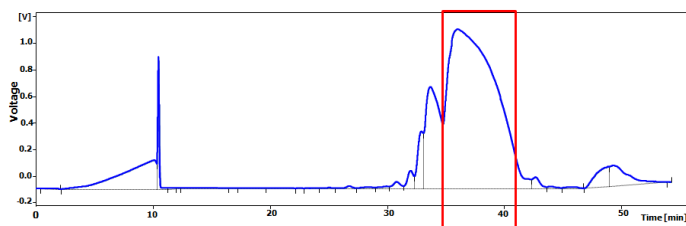
Resin: PolyRP 10-300 (10 μm , 300 \AA)
Column: 4.6 x 250 mm (Stainless Steel)
Mobile Phase: A: 0.1% TFA - H_2O , B: 0.1% TFA - ACN
Flow Rate: 1.0 mL/min (360 cm/h)
Detector: UV 214 nm
Column Temperature: 25°C
Injection Amount: 4 μg
Gradient: 0-20 min, 5-50% B

Polypeptide Separation Using PolyRP 10-100



Resin: PolyRP 10-100 & 10-300 (10 μm , 100 \AA & 300 \AA)
Column: 10 x 250 mm (Stainless Steel)
Mobile Phase: A: 0.1% TFA - H_2O , B: 0.1% TFA - ACN
Detector: UV 210 nm
Column Temperature: RT
Flow Rate: 3.0 mL/min (230 cm/h)
Sample: Polypeptide ~3000 Da crude purity 72%

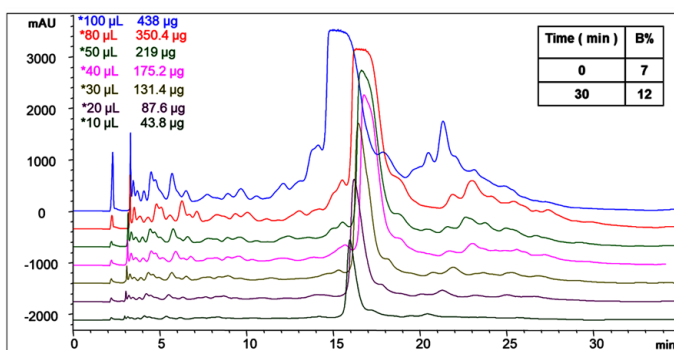
Figure 9. Due to a pore size impact on separation resolution, PolyRP 10-100 performed better than PolyRP10-300 for a customer's polypeptide. Additionally, the customer's demanding purification targets (>95% purity and >90% recovery yield) were met.



Resin: PolyRP 10-100 (10 µm, 100 Å)
Column: 50 x 250 mm (DAC)
Mobile phase: A: 0.1% TFA - H₂O B: 0.1% TFA – ACN
Detector: UV 210 nm
Flow Rate: 40 mL/min (120 cm/h)
Sample: Polypeptide ~3000 Da crude purity 72%

Figure 10. PolyRP10-100 process chromatography was successfully scaled up to 50 DAC and to 300 DAC (at customer site and met customer’s purification targets). The customer’s overall purification cost was greatly reduced due to high separation resolution and high loading capacity of PolyRP10-100.

ssDNA Primer Separation Using PolyRP 10-300



Resin: PolyRP 10-300 (10 µm, 300 Å)
Column: 4.6 x 150 mm (Stainless Steel)
Mobile Phase: A: 100 mM TEAA (pH 7.0) B: ACN
Flow Rate: 0.8 mL/min (290 cm/h)
Detector: UV 260 nm
Column Temperature: 30°C
Sample: 32 nucleotides 4.38 mg/mL in water
Pressure: 38 bar

Figure 11. PolyRP 10-300 separated 32 nucleotides ssDNA primer from smaller ssDNA primers and protein impurity. After separation the purity increased from ~72% to 99%, and recovery yield was 91%.

Ordering Information

Resin	Particle Size	Pore Size	Part Number
PolyRP 10-100	10 µm	100 Å	260110101
PolyRP 10-300	10 µm	300 Å	260110301
PolyRP 10-300 HC	10 µm	300 Å	260510301
PolyRP 10-500	10 µm	500 Å	260110501
PolyRP 10-1000	10 µm	1000 Å	260110951
PolyRP 15-100	15 µm	100 Å	260115101
PolyRP 15-300	15 µm	300 Å	260115301
PolyRP 15-500	15 µm	500 Å	260115501
PolyRP 15-1000	15 µm	1000 Å	260115951
PolyRP 15-1000 HC	15 µm	1000 Å	260515951
PolyRP 20-1000	20 µm	1000 Å	260120951
PolyRP 30-100	30 µm	100 Å	260130101
PolyRP 30-300	30 µm	300 Å	260130301
PolyRP 30-300 HC	30 µm	300 Å	260530301
PolyRP 30-500	30 µm	500 Å	260130501
PolyRP 30-1000	30 µm	1000 Å	260130951

*HC -- High loading capacity resin

Standard packing size:
 1L, 5L, 10L, 25L, 50L, 100L
 Additional pack sizes are available

Additional particle and pore sizes are available. Pre-packed stainless-steel columns for sample preparation and separation process development/ scale-up are available.

Please contact your regional sales agent for more information.

Better Surface Chemistry For Better Separation



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