

G25+ AgaDex Size Exclusion Resin

Product information

Product Description

G25+ AgaDex Size Exclusion Resin is a gel filtration resin with a highly cross-linked agarose and dextran matrix, designed for desalting, buffer exchange and removal of small molecules (≤ 10 kDa) from proteins, polysaccharides and nucleic acids. It can also be used to purify peptides, oligonucleotides, antibiotics, synthetic drugs and natural products.

Product Features

- Fast desalting and buffer exchange of biomolecules
- High flow velocity (up to 300 cm/h) and pressure tolerance (≤ 0.5 MPa)
- Withstands stringent cleaning: 1M NaOH, 6M guanidine HCl, 8M urea

Storage

Store at 4-30 °C. Valid for 12 months.

Safety Information

Always wear appropriate lab PPE: coat, gloves and goggles. Please consult the Material Safety Data Sheet (MSDS) for more information.

Note: This product is intended for research use only.

Technical Parameter

Resin	Highly cross-linked agarose and dextran
Particle Size Range	45-165 μm
Average Particle Size	$\sim 90 \pm 10 \mu\text{m}$
Separation Range	≤ 10 kDa (globular molecules)
pH Stability	3-12 (long term) 1-14 (short term)
Chemical stability	All common buffers: 8M urea, 6M guanidine hydrochloride, 70% ethanol, 1M sodium hydroxide
Operating Pressure	≤ 0.5 MPa
Recommended Flow	150 cm/h
Maximum Flow	300 cm/h
Storage Solution	20% ethanol or 2% benzyl alcohol, 0.2M sodium acetate
Storage Temperature	4-30 °C

Product Description	Catalog No.
G25+ AgaDex Size Exclusion Resin (25ml)	HNVO120350-25
G25+ AgaDex Size Exclusion Resin (100ml)	HNVO120350-100
G25+ AgaDex Size Exclusion Resin (500ml)	HNVO120350-500
G25+ AgaDex Size Exclusion Resin (1000ml)	HNVO120350-1000

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Protocol

A. Column Packing

Note: Allow the resin to reach room temperature before packing. Degas all buffers and water before use.

1. *Resin quantity:* Settled resin volume needed = column cross-sectional area (cm²) × bed height (cm) × compression factor (settled bed height / packed bed height). The compression factor of G25+ AgaDex is ≈ 1.15.
2. *Slurry volume:* Slurry volume needed = settled resin volume / slurry concentration. G25+ AgaDex is supplied as an 80% slurry. If the slurry concentration is unknown, pour the slurry into a measuring cylinder, let it settle overnight (12-20 h) and measure the settled resin volume.
3. *Resin washing:* Attach a glass filter funnel to a filtering flask, resuspend the resin, transfer the required volume of slurry to the funnel and wash 3 times with approximately 3 mL purified water per mL of resin. Gently resuspend the resin after each addition. Do not use a magnetic stirrer, as it breaks the resin particles.
4. *Slurry preparation:* Transfer the washed resin to a beaker with purified water and prepare a 50-75% slurry.
5. *Column preparation:* Add purified water to the clean column to remove air bubbles from the bottom filter. Close the stop plug when approximately 1 cm of water remains, and make sure the column is vertical.
6. *Pouring:* Pour the slurry into the column in one continuous motion (use a packing reservoir if needed), avoiding air bubbles.
7. *Connecting:* Fill the packing reservoir with purified water and insert the column adaptor. Fill the connecting tubing from the pump with packing solution, connect the pump to the top of the column and remove the bottom stop plug.
8. *Bed consolidation:* For a 60 cm bed height, set the flow velocity to 30 cm/h. Open the bottom plug, start the pump and run until the bed has consolidated. Mark the bed height.
9. *Compression:* Remove the packing reservoir (if used) and attach the top adaptor, making sure no air is trapped. Lower the adaptor to 0.5 cm above the bed and continue compressing at 240 cm/h until the bed has consolidated. Mark the bed height.
10. *Final adjustment:* Stop the flow, close the bottom stop plug, disconnect the pump and slightly loosen the O-ring. Press the adaptor to about 0.3 cm below the marked position, tighten the O-ring and close the stop plug. The column is ready for testing.

B. Column Efficiency Test

Test the column after packing and regularly during its working life, especially if separation performance declines. Column efficiency is expressed as the height equivalent to a theoretical plate (HETP) and the asymmetry factor (As).

1. *Test sample:* Apply 1.0% CV of test sample and elute at 30 cm/h using one of the methods below:
Acetone method: sample 1.0% (v/v) acetone in water; eluent purified water; detection UV 280 nm.
NaCl method: sample 0.8 M NaCl in water; eluent 0.4 M NaCl in water; detection conductivity.
2. *Calculation:* $N = 5.54 \times (VR/Wh)^2$; $HETP = L/N$; $N/m = N \times 1000/L$; $As = b/a$
L = bed height (cm); N = number of theoretical plates; VR = volume eluted from sample application to peak maximum; Wh = peak width at half peak height (VR and Wh in the same units); a, b = ascending and descending parts of the peak width at 10% of peak height.
3. *Acceptance criteria:* At least 3000 plates/m and an asymmetry factor of 0.7-1.5. Use the same test conditions when comparing columns.

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C. Chromatography

1. *Sample preparation:* Filter the sample through a 0.45 µm membrane before loading.
2. *Loading volume:* Load 1-5% of the column volume (CV) when the target and impurities are close in size, and 10-25% of CV when they differ greatly (e.g. desalting or buffer exchange).
3. *Buffer selection:* Aqueous buffers are generally used. Buffer pH and conductivity have little effect on separation; choose a buffer in which the sample is stable.
4. *Flow velocity:* Run at the recommended flow velocity (150 cm/h; maximum 300 cm/h). The taller the column, the slower the flow velocity should be.

D. Cleaning-in-Place (CIP)

Clean the column when efficiency decreases, backpressure increases, separation performance declines or the resin becomes discoloured.

1. Rinse 2 CV with purified water.
2. Rinse 1 CV with 1 M NaCl.
3. Rinse 1 CV with 0.2 M NaOH.
4. Rinse 4 CV with purified water.

E. Sanitization

Storage solutions (20% ethanol or 2% benzyl alcohol containing 0.2 M sodium acetate) cannot be sterilized. For microbial control, treat the resin with 0.5-1 M NaOH at the recommended flow velocity before and during use.

F. Storage

Store in 20% ethanol or 2% benzyl alcohol containing 0.2 M sodium acetate at 4-30 °C in a sealed container. Replace the storage solution every 3 months to prevent evaporation and microbial growth.

G. Disposal

G25+ AgaDex is not readily biodegradable. Incineration is recommended.

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Troubleshooting

Problem description	Possible reasons	Solutions
Poor resolution between the target peak and the impurity peak	The injection volume is too large	Decrease the injection volume to 0.5% CV
	The sample has high viscosity	Dilute the sample accordingly
	The flow rate is too high	Decrease the flow rate
	The chromatographic column is too high	Select a longer or thinner chromatographic column
	Poor column resin	Re-pack the column or use pre-packed column
	The sample is not filtered	Filter the sample with 0.22 µm filter or 0.45 µm filter membrane
	Uneven temperature during use	Maintain a constant temperature
No expected elution peak	There is an ion interaction between the protein and the resin	Maintain the ionic strength of the buffer at 0.05 - 0.15 NaCl
	There is a hydrophobic interaction between the protein and resin	The hydrophobic interaction can be minimized by lowering the ionic strength or by increasing the pH and adding mild detergent.
	Proteins and lipids precipitate in the column	Wash the column or replace with a new column
	Microbial growth in the resin	Replace with new column and store in 20% ethanol
Elution peak appears earlier	There are gaps in the packed column	Re-pack the column
	The target substance aggregates and precipitates under the eluent condition	Check the stability of the target substance under eluent conditions (salt concentration, pH, etc.)
Elution peak is delayed	There is an ion interaction or hydrophobic interaction between protein and resin	Maintain the ionic strength of the buffer at 0.05 - 0.15 NaCl
	Dirty resin, filter membrane, or the top of the column bed	Clean and re-equilibrate the column
	Microbial growth in the resin	Replace with new column and store in 20% ethanol
Air bubbles are introduced in the column bed	There is a temperature difference during use, or there is residue in the tube lines	Re-pack the column
Rapid Increase in chromatographic peak	Overly tight packing of the resin	Repack the column
Slow or tailing chromatographic peak	Loose packing of the resin	Repack the column
Slow flow rate	Aggregation of the proteins or lipids	Clean the resin or membrane promptly
	Protein precipitation in the resin	Adjust composition of the equilibrium and elution buffers to maintain stability of the target substance and the binding efficiency of the resin
	Microbial growth in the column	After using the resin, store it correctly and promptly to prevent microbial growth
Pressure increased	The sample is turbid	Prepare fresh sample before use
	The tube lines and sieve plate are blocked	Clean the tube lines, sieve plate and resin, and re-pack the column.