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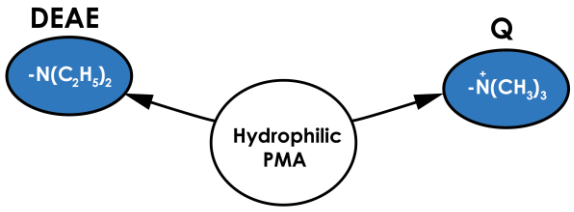
Monomix Alkaline-Resistant Anion Exchange Resins User Manual

1. Product Description

Monomix alkaline-resistant anion exchange resins (AEX) are are based on hydrophilic treated polymethacrylate. The resins have particle size of 60 μm, and pore size of 1000 Å. The resins have good physical and chemical stability. The resins are surface treated through Sepax's proprietary technology and have tentacle structure that are composed of long linear polymer chains carrying functional groups. The polymer chains are covalently bonded to the matrix, and this configuration provides excellent hydrophilicity and a high surface area that allows biomolecules to bind accessible functional groups without much steric hindrance and largely avoid non-specific binding to biological samples. Ion-exchange functional groups are covlently bonded on the surface of hydrophilic substrates to obain strong anion exchange resin (Monomix Mab60-Q) and weak anion exchange resin (Monomix HC-60 DEAE Excel) and form an ion exchange layer of high functional group density and homegeneousness. The resin chemistries are optimized to improve alinlie resistanct against the early generation of Monomix AEX resins. Monomix alkaline-resistant AEX have found wide applications in the separation and purification of biological samples such as antibodies, vaccines, insulins, proteins, nucleic acids, and heparins.

Resin Chemistry Scheme

Figure 1. Monomix Mab60-Q, Monomix HC60-DEAE Excel resin chemistry scheme



Characteristics

- High binding capacity and excellent biocompatibility
- Capable of withstanding high pressure and high flow rate due to rigid base bead matrix
- High separation resolution, efficiency and recovery
- High lot-to-lot consistency
- Easy to scale up
- Highly hydrophilic surface with minimal non-specific binding
- Small volume change under normal packing conditions
- Supply capacity: >100L per order

Technical Specifications

Table 1: Monomix alkaline-resistant AEX: Monomix Mab60-Q, Monomix HC60-DEAE Excel

Resin Type	Monomix Mab60-Q	Monomix HC60-DEAE Excel
Matrix	Hydrophilic Polymethacrylate	
Functional Group	$-N^{+}(CH_3)_3$	$-N(CH_2CH_3)_2$
Particle Size (μm)	60	
Pore Size (Å)	100	
Dynamic Binding Capacity *	≥ 80 mg BSA /mL resin	
Maximum Linear Flow rate	1000 cm/h	
Operating Temperature	≤ 40 ℃	
pH Stability	2-12	
Maximun Operating Pressure	≤ 1 MPa (10 bar)	
Compatible Mobile Phase	Compatible with conventional buffer salt systems (Tris, phosphate, acetate buffer, etc.) and conventional organic phase/aqueous systems (acetonitrile, ethanol, etc.)	
Long-term Storage	50% (v/v), store in 20% ethanol, 2% benzyl alcohol or 10 mM NaOH	
Regeneration	1-2 M NaCl	
CIP	0.5 M HCl or 0.5-1.0 M NaOH. For impurities bound with strong hydrophobicity, use Tween or Triton X-100 at a concentration of 0.1-1%	

**Dynamic Binding Capacity measurement conditions:*
Monomix Mab60-Q: linear flow rate 180 cm/h, 2.0 mg/mL of BSA in 50 mM Tris buffer (pH = 8.5)
Monomix HC60-DEAE Excel: linear flow rate 180 cm/h, 2.0 mg/mL of BSA in 50 mM Tris buffer (pH = 8.0)

2. Instructions for Use

2.1 Safety Precautions

For information on the safe use of this product, please refer to the Safety Data Sheet (SDS).

2.2 Exchange packing buffer

Monomix CEX cationic resin is stored in 20% ethanol at a resin volume ratio of ~50%. Before packing, 20% ethanol storage solution needs to be replaced with 1.0 M NaCl packing buffer. Resins are allowed to settle overnight, decant supernatant. Make a resin slurry using 1-2 CV of 1.0 M NaCl packing buffer, stir genetlly, and let the resin slurry settle for about 2 hours, remove top layer liquid which may contain fines. Repeat this step 2 more times. Make a final resin packing slurry using 1.0 M NaCl and target resin solid 50-60% V:V.

2.3 Column Packing

2.3.1 Calcuate resins amount (V)
$V_{slurry}=V/\ 50\% =S\times H\times F/50\%$
V: targeted column volume
S: cross-section area of the column
F: Compression factor (for resin stored in 20% ethanol, the compression factor is about 1.15)
H: targeted packing height

2.3.2 As described in Exchange packing buffer, after re-homogenization, close bottom valve to allow packing media to settle naturally. After resin bed top surface settle down to more than 5cm, install the column head where air bubbles are removed. Tighten the seal ring, open the bottom valve and turn on the low-pressure packing station. Use 1 M NaCl packing buffer and a linear flow rate of 50cm/h to accelerate the resin bed sedimentation.

2.3.3 After the resin bed settles and stabilizes, turn off the packing station. Then loosen the seal ring, lower the column head to about 2 cm above resin bed surface and then tighten the seal ring. Turn on the packing station, incrase liner flow rate stepweisely 50cm/h, 100cm/h, 150cm/h, 200 cm/h, 250cm/h, 300 cm/h …to compress the resin bed gradually. The flow rate of each stage is maintained for 3 min. Untile the column back pressure reaches 3 bar and then maintain the linear flow rate for 15 min. Mark the height of the column bed at this time. Turn off the packing station, after the column pressure drop to zero close the bottom valve. Rotate the 4-way valve on the column head to the drain line location, thencompress the resin bead at a ratio of 1:1.02-1.05 (based on the resin bed height marked before). Lower the column head to the target height, and the column packing is finished.

2.4 Column Efficiency Evaluation

Sample	1 M NaCl
Sample Volume	2.0% CV
Mobile Phase	0.1 M NaCl
Flow Rate	60 cm/h
Detector	Conductivity
Eligibility Criteria	Tailing factor：0.8-1.5； Monomix 60 mm Plate Count: ≥2000 /m

2.5 Solutions to Non-ideal Column Efficiency

- Reduce the slurry concentration
- Increase packing flow rate
- Extend packing time

If peak fronting occurs, solutions are the opposite of the above

3. Use of Chromatographic Column

3.1 Select and optimize a buffer system according to specific characteristics (isoelectric point, ion-exchange group, etc.) of targeted molecules to be separated/purified or analyzed;

3.2 Equilibrate the column with about 5 CV of buffer until conductivity and pH of effluent are constant, consistent with those of the fresh equlibration buffer;

3.3 **Sample Preparation:** Solid samples can be dissolved in a buffer solution; low concentration samples can be dialyzed with a buffer to increase concentration; high concentration samples can be diluted with a buffer. Samples with insoluble impurities should be filtered first to avoid clogging the column and to prolong column life;

3.4 **Loading**: A sample loading volume should be determined according to resin loading capacity and purity of targeted molecule in crude sample; after loading is completed, continue to pump buffer until a stable baseline is obtained;

3.5 **Elution:** According to the characteristics of targeted molecule, choose a method (increasing the salt concentration or changing pH) to elute targeted molecule bound to the resin bed;

3.6 **Regeneration**: After each use of purificatio / separation, rinse the column with 1-2M NaCl solution to remove impurities adsorbed on the resin bed;

3.7 **Cleaning (CIP)**: If impurities cannot be removed through the regeneration step, causing column

clogging, increased back pressure, or decreased flow rate, the performance of the column can be restored by forward or reverse online cleaning. In general, online cleaning can result in increased back pressure of the column, so it is recommended to use linear flow rates within 0.5 times of normal operation conditions. Specific CIP methods are determined based on the characteristics of the impurities, some examples below:,

a) For impurities such as precipitated or denatured substances, wash with 5 CV of 0.5-1.0 M NaOH; if the result is not satisfactory, use 0.5-1.0 M NaOH plus 1M NaCl, and then use at least 5 CV of 0.22um filtered buffer (pH6-8) to wash the column;

b) For impurities bound with strong hydrophobicity, wash the column with 2 CV of non-ionic detergent (such as Tween or Triton X-100 at a concentration of 0.1-1%), and then immediately use at least 5 CV of filtered buffer (pH6-8) to wash the column; it can also be washed with 3-4 CV of 70% ethanol or 30% isopropanol, and then at least 5 CV of steriled and filtered buffer (pH6-8) to wash.

4. Product Strorage Conditions

When Monomix alkaline-resistant AEX are not immediately used, they can be stored at 4-35 °C in a sealted container after washing them with 20% ethanol, 2% benzyl alcohol or 10mM NaOH aqueous solution for 3-5CV. Monomix alkaline-resistant AEX can be stored in 10mM NaOH for more than 6 months. But it is recommended not store the resins in alkaline solutions for long-term in order to prolong resin life and to avoid resin performance decline.

5. Product Ordering Information

Product Name	Functional Group	Particle Size/Pore Size	Order Number
Monomix Mab60-Q	Strong Cation Exchange	60 µm, 1000 Å	285060950
Monomix HC60-DEAE Excel	Weak Cation Exchange	60 µm, 1000 Å	285160950

**Resin package sizes are available in 5L, 10 L, 50 L, and prepacked column sizes are available in 1, 4.2, 5 mL.*
For resin products or prepacked columns not listed above, please contact us.