LT BIOTECH UAB
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Helios 50-HQ

1. Basic product information

Helios 50-HQ is a strong anion-exchange resin based on polymers. Compared with traditional agarose chromatography resin, it can be operated under high pressure and high flow rate. It has low backpressure during use, which is convenient for linear amplification and good stability. At the same time, it has high salt tolerance, high resolution, and high and stable protein binding capacity, which can reduce dilution in the purification process, reduce the operation steps of dialysis and ultrafiltration, and improve the process flexibility. It is convenient for the connection of upstream and downstream purification processes, and is widely used in the separation and purification of monoclonal antibodies, recombinant proteins, vaccines, blood products, viruses, DNA and polypeptide products.

2. Chromatography resin parameters

Resin type	Strong anion exchange
Matrix	Polystyrene-divinylbenzene
Median particle size	50 μm
Total ionic capacity	0.13–0.17 mmol/ml
Dynamic binding capacity	≥70 BSA mg/ml
Protein separation (conductivity mS/cm)	Ovalbumin: 20.4–21.7 Soybean trypsin inhibitor: 28-30
Pressure-resistant flow rate	300 cm/h

3. Chemical resistance

pH Stability	2~12 (working range), 1~14 (CIP)
Chemical stability	All commonly used ion exchange buffers

** The physical and chemical properties and functions of the chromatographic resin have no obvious change after being placed in an environment of 40°C and pH 2–12 for 7 days.*

4. Method of use

4.1 Column packing
The slurry concentration is equal to the volume of the resting gel divided by the total volume after homogenisation. The best packing effect can be obtained by using 0.5M NaCl with a slurry concentration of 60–70%. Methods are:
1) The column bed volume (CV) of the chromatographic column is $V=Ac\times L$, where $Ac=\pi\times r^2$. (Ac: cross-sectional area of the chromatographic column; L: length of the chromatographic column; r: radius of the chromatographic column.)
2) Agitate the resin to form a homogenious slurry, and measure the required mass or volume. It should be about 1.2 CV to prevent shrinkage.
3) Replace 20% ethanol with 0.5M NaCl solution and equilibrate overnight.
4) Before loading the column, use 0.5M NaCl solution to adjust the concentration of the slurry to 65–70%; pour the homogenate into the chromatography column all at once and mark the height after settling, to balance.

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5) Install the distributor and adjust the height so the compression coefficient is 1.05~1.10; then start the infusion pump, and use 1.5~2 working flow rate to stabilise the column bed.
6) Determination of column efficiency and symmetry according to SOP must meet predetermined standards.
4.2 Evaluation of column efficiency
After packing, the chromatographic column is washed with 3–5 CV of ultrapure or pure water. Balance at a flow rate of 100 cm/h and conduct a column efficiency test.
Column efficiency test method for ion exchange chromatography column:
Sample: 2M NaCl solution
Loading volume: 1~5 % CV
Eluent: 0.5M NaCl solution
Linear velocity: 100 cm/h
Detection: conductivity detector
4.3 Rinsing
Packed columns should be rinsed with at least 5 CV of ultrapure or pure water.
4.4 Equilibration
Equilibrate the column with an appropriate 5–10 CV buffer until the conductance and pH of the effluent remain unchanged (consistent with the equilibration solution). For example, equilibration buffer could be 20 mM PBS, pH 7.0. Screening and optimisation should be carried out according to the stability and isoelectric point of the target protein, and the type of ion-exchange resin.
4.5 Sample loading
Solid samples can be prepared by dissolving in equilibration buffer. Low-concentration sample solutions can be dialysed with equilibration buffer; high-concentration sample solutions can be diluted with equilibration buffer. To avoid column clogging, samples should be centrifuged or membrane filtered (preferably 0.45 or 0.22 μm). The amount of feed is calculated according to the loading capacity of the resin and the content of the target protein in the feed solution. Before loading the sample, ensure that the sample buffer is as consistent as possible with the equilibration buffer.
4.6 Elution
After loading the sample, continue to rinse with the equilibration buffer until the baseline is stable. According to the situation, the method of increasing the salt concentration or changing the pH of the mobile phase can be used to elute the samples adsorbed on the chromatographic resin in sequence.

5. Cleaning and regeneration

Contaminants (e.g. lipids, endotoxins and proteins) accumulate on the column as the number of uses of the chromatography resin increases. Determine the frequency of CIP according to the degree of contamination of the chromatography resin (if the contamination is considerable, CIP is recommended after each use to ensure repeatability of results and to prolong the working life of the chromatography resin). For different types of impurities and contaminants, the recommended cleaning conditions are as follows:
• Removal of strongly binding proteins: wash with 5 CV of 2M NaCl solution, or use a high salt buffer not lower than pH 2, such as 1M NaAc solution.
• Removal of strongly hydrophobic proteins and precipitated proteins: first wash with 0.2–0.5M NaOH solution (contact time 1-2 hours), then wash 5-10 CV of equilibration solution and 5 CV of ultra pure or pure water.
• Removal of lipoproteins and lipids: first wash with 5 CV of 50% ethanol or 30% isopropanol (contact time 0.5-1 hour), then rinse with 5–10 CV of ultra pure or pure water. It can also be cleaned with alkaline or acidic solution containing non-ionic surfactant, such as 0.1~0.5% Triton X-100 + 0.1M acetic acid for 1–2 hours, and rinsed with 50% ethanol above 5 CV to remove the detergent. Rinse with 5 CV of ultrapure or pure water as above (when using high-concentration organic solvents, the method of gradually increasing the concentration of organic solvents should be adopted to avoid air bubbles).

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Note: 50% ethanol or 30% isopropanol should be degassed before use; the flow rate should be 30–60 cm/h during CIP. Reverse cleaning should be used when the clogging is severe.

To reduce the microbial load, it is recommended that 0.5~1M NaOH solution is used to treat the chromatographic resin. Treatment time is 15~30 minutes.

6. Storage

Keep the unopened chromatography resin in the original container and store at 4~30°C in a well-ventilated, dry and clean place. Do not freeze. Wash the used column with 2–3 CV of 20% ethanol solution and store at 2~8°C.

7. Destruction and recycling

Since chromatography resin is difficult to degrade in nature, it is recommended that the waste chromatography resin is incinerated to protect the environment. For chromatography resin that has been in contact with biologically active samples such as viruses and blood, follow the local biosafety requirements before destroying or disposing of it.

8. Packing method

Detailed information on resin packaging is available on request. Please contact your local distributor.

9. Ordering information

Product name: Helios 50-HQ

Product Cat. No	Package
206-00025	25 ml
206-00100	100 ml
206-00500	500 ml
206-01000	1 L
206-05000	5 L
206-10000	10 L