

Lepta PlasmidCap HR

1. Basic product information

Lepta PlasmidCap HR is a thiophilic affinity chromatography resin. The principle of thiophile affinity chromatography is to use the interaction between electron donors and acceptors to separate and purify target molecules. This interaction is strengthened in a high-salt environment and weakened in a low-salt environment. Lepta PlasmidCap HR selectively binds closed-circle supercoiled plasmid DNA but not open-circle plasmid DNA. Therefore, it can be used to prepare high-quality plasmid DNA. Lepta PlasmidCap HR couples the sulfurcontaining compound 2-mercaptopyridine to a new high-rigidity base frame, which provides better pressure-flow rate performance for large-scale separation and purification of plasmid DNA, increases the flexibility of process design and improves productivity. At the same time, finer particle size provides better resolution.

2. Chromatography resin parameters

Resin type	DNA affinity chromatography
Matrix	High degree cross-linking agarose
Functional group	2-mercaptopyridine
Median particle size	40
Ligand density	27–50 µmol/ml
Dynamic binding capacity	>3.0 mg plasmid/ml
Recommended flow rate	90~500 cm/h
Maximum flow rate	600 cm/h
Maximum working pressure	5 bar
Working temperature	4–30°C

** Measurement conditions of dynamic binding capacity: packing height, 10 cm; flow rate, 200 cm/h; test buffer, 2 M ammonium sulfate, 10 mM EDTA, 0.1M Tris, pH 7.5. No breakthrough observed during 3 minutes of retention time.*

3. Chemical resistance

pH stability*	2–14
Chemical stability	All commonly used aqueous buffers, 30% isopropanol**, 75% ethanol**, 1M acetic acid, 0.1M NaOH

* The physical and chemical properties and functions of the chromatographic resin have no obvious changes after being placed in an environment of 40°C and pH 2–14 for 7 days.

** v/v, volume ratio

4. Method of use

4.1 Chromatographic conditions

1. **Buffer selection:** Equilibration/binding/washing buffer: 2.0M (NH₄)₂SO₄, 10 mM EDTA, 0.1 M Tris, pH 7.5.

Elution buffer: 1.7M (NH₄)₂SO₄, 10 mM EDTA, 0.3M NaCl, 0.1 M Tris, pH 7.5.

1. **Flow rate:** generally, choose a linear flow rate of 90–500 cm/h according to the bed height of the column.
2. **Sample pretreatment:** to prevent the sample from clogging the column, the sample needs to be filtered with a 0.45 µm microporous membrane before loading. It is recommended that the pH and conductivity of the sample is adjusted to be consistent with the equilibration buffer (dilution, ultrafiltration can be used and desalting to adjust the pH and conductivity of the sample).

4.2 Chromatography steps

1. **Equilibration:** use equilibration buffer to fully equilibrate the chromatography column until the pH and conductivity are stable and basically consistent with the equilibration buffer. This step usually requires 3–5 column bed volumes (CV).
2. **Sample loading:** according to the binding capacity measured in the small test, determine the sample loading volume and loading amount of the sample on Lepta PlasmidCap HR.

3. **Impurity washing:** use equilibration buffer or other suitable buffer to wash the chromatography column until the UV stabilises and returns to the baseline.
4. **Elution:** use elution buffer; this step usually requires 5–10 CV of buffer.
5. **Regeneration:** rinse the column with a high-salt buffer (such as 2M NaCl).
6. **Re-equilibration:** re-equilibrate the column with equilibration buffer.

5. Cleaning and regeneration

Contaminants (such as lipids, endotoxins and proteins) accumulate on the column as the number of uses of the chromatography resin increases. Regular cleaning-in-place (CIP) is essential to keep the column in a stable working condition. 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 denatured or precipitated proteins:** first wash with 5 CV of 0.1M NaOH solution, and then use 5– 10 CV of ultrapure or pure water.
- **Removal of lipoproteins and lipids:** first wash with 5 CV of 30% isopropanol, then rinse with 5–10 CV of ultrapure or pure water.

Note: 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 NaOH solution is used to treat the chromatography 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: Lepta PlasmidCap HR

Product Code in Catalogue	Product Volume
681-00025	25 ml
681-00100	100 ml
681-00500	500 ml
681-01000	1 L
681-05000	5 L
681-10000	10 L
681-20000	20 L

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