LT BIOTECH UAB
Mokslininkų g. 6A,
LT-08412 Vilnius,
Lithuania
www.ltbiotech.lt

Lepta DEAE

1. Basic product information

Lepta DEAE is a weak anion-exchange chromatography resin that takes advantage of differences in the nature and magnitude of charges of different molecules under specific conditions to separate them. This chromatographic resin can be used successfully for the separation and purification of various biological molecules such as recombinant proteins, antibodies, nucleic acids, viruses and virus-like particles, and polysaccharides.
Compared with traditional weak anion-exchange chromatography resin, Lepta DEAE shows better performance:
(1) The improved Lepta base frame is more rigid, so it can achieve a higher process flow rate at lower back pressure and improve process efficiency.
(2) There is an increase in the dynamic binding capacity of the chromatography resin by upgrading the ligand coupling method.

2. Chromatography resin parameters

Resin type	Weak anion exchange
Functional group	Diethylaminoethyl
Matrix	Highly cross-linked agarose
Median particle size	90 μm
Total ionic capacity	0.29–0.35 mmol Cl^-/ml
Dynamic binding capacity	>90 mg ovalbumin/ml*
Recommended flow rate	90~500 cm/h
Maximum flow rate	700 cm/h
Maximum working pressure	5 bar
Working temperature	4–30°C

** Measuring conditions of dynamic binding capacity: packing height, 10 cm; test flow rate, 600 cm/h; test buffer: 0.05M Tris-HCl solution, pH 8.0; test sample: 4 mg/ml ovalbumin, when ovalbumin breakthrough is 10% of starting concentration.*

3. Chemical resistance

pH stability*	2–14
Chemical stability	All commonly used aqueous buffers, 30% isopropanol**, 75% ethanol**, 1M NaOH, 1M acetic acid, 6M guanidine hydrochloride, 8M urea
Avoid	Oxidising agents, anionic detergents

** 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 2–14 for 7 days.*
*** v/v, volume ratio*

LT BIOTECH UAB
Mokslininkų g. 6A,
LT-08412 Vilnius,
Lithuania
www.ltbiotech.lt

4. Method of use

4.1 Chromatographic conditions

(1) Buffer selection: a buffer salt whose buffer group does not interact with the chromatographic medium should be selected. If the binding and elution mode is used, the equilibration buffer should be a buffer with low salt (less than 5 mS/cm) and high pH (usually 1 pH unit higher than the isoelectric point of the target molecule) to facilitate the binding of the target molecule. Consider the stability of the sample in the buffer; the elution buffer is usually a buffer with a high concentration of salt (such as 1M NaCl) added to the equilibration buffer. If the flow-through mode is used, the equilibration buffer should adopt conditions that are conducive to the binding of impurities. After the target molecule has completely flowed through, it should be washed directly buffer with with high-concentration salt.
(2) Flow rate: generally choose a linear flow rate of 90~500 cm/h according to the column bed height .
(3) 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 volumes (CV).
Note: The equilibration buffer usually uses a low-concentration salt solution (conductivity less than 5 mS/cm), and its pH value is usually higher than the isoelectric point of the target molecule.
(2) Sample loading*: determine the loading volume and loading amount of the sample on Lepta DEAE according to the binding capacity measured in the small test.
(3) Washing*: use equilibration buffer or other suitable buffer to wash the chromatography column until the UV stabilises and returns to the baseline.
(4) Elution*: elution is achieved by increasing the concentration of salt ions. The concentration of salt ions in the elution buffer can be gradually increased through a linear gradient or a step gradient to elute molecules with different binding strengths. pH gradient elution or mixed elution can also be used.
(5) Regeneration: rinse the column with a high-salt buffer (such as 2M NaCl).
(6) Re-equilibration: re-equilibrate the column with equilibration buffer.
* If the flow-through mode is used, the ‘sample loading’ step should be set to collect; the ‘washing’ step should stop the collection after all the target molecules have passed through; the ‘elution’ step should directly use high-salt buffer – impurities just can be washed away.

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.

LT BIOTECH UAB
Mokslininkų g. 6A,
LT-08412 Vilnius,
Lithuania
www.ltbiotech.lt

• Removal of strongly hydrophobic proteins and precipitated proteins: first wash with 5 CV of 1M NaOH solution, then wash the lye with 5–10 CV of ultra pure or pure water.
• Removal of lipoproteins and lipids: first wash with 5 CV of 70% ethanol or 30% isopropanol, then rinse with 5–10 CV of ultra pure or pure water.
Note: 70% 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 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 DEAE

Product Cat. No	Package
604-00025	25 ml
604-00100	100 ml
604-00500	500 ml
604-01000	1 L
604-05000	5 L
604-10000	10 L
604-20000	20 L