NeoTrap Co-NTA FPLC Columns

Cat# NB-19-0077-1mL Size: 1 ml

Cat# NB-19-0077-5mL Size: 5 ml

Introduction

NeoTrap Co-NTA FPLC Columns are designed for simple, one-step and rapid purification histidine-tagged recombinant proteins by immobilized metal ion affinity chromatography (IMAC).

Compatible with all common liquid chromatography instruments (including ÄKTA™ FPLC's), peristaltic pumps and syringes.

Specifications

PRODUCT	NeoTrap Co-NTA FPLC Columns	
Cat. No.	NB-19-0077-1mL	NB-19-0077-5mL
Column volume	1 ml	5 ml
Resin	Cross-linked 6% agarose	
Bead size	50-150 µm	
Ligand	Nitrilotriacetic acid (NTA)	
Binding capacity*	40 mg/ml resin	
pH stability	2-14 (<2 h); 3-12 (up to one week)	
Chemical stability	Stable in: 0.1M HCl and 1% SDS - 30 mins; 0.5 M NaOH and 30% acetic acid - overnight; 0.01M HCl, 0.1M NaOH and 0.2M acetic acid - one week.	

*Depends on the type of proteins and binding conditions

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Recommended Protocol for Purification:

Buffers needed:

Binding Buffer:	Sodium phosphate 20 mM, NaCl 500 mM, Imidazole 20 mM, pH 7.4. The presence of low concentration of imidazole is recommended. The exact concentration is protein and metal ion dependent with a guided range of 10 – 50 mM (e.g., 20 mM)
Washing Buffer:	Can be the same as the binding buffer or may contain additional reagents (e.g., detergents, alcohol and increased imidazole concentration etc.) in order to remove as much weakly bound impurities as possible.
Elution Buffer:	Sodium phosphate 20 mM, NaCl 500 mM, Imidazole 250-500 mM, pH 7.4

Buffers should be sterilized using a filter of 0.22 µm.

INSTRUCTIONS:

1. Column preparation

Connect the NeoTrap column to the pump by removing the end of the column and the top stop plug (save it for storage). Avoid introducing air in the column.

2. Column equilibration

Equilibrate the column with 5 - 10 column volumes of binding buffer until the signal reaches the baseline or becomes stable.

3. Sample application

It is recommended to dilute the sample containing the His-tagged protein 1:1 with Binding Buffer to avoid ionic and pH changes.

All samples should be filtered through a 0.22 µm filter in order to remove particles before applying it into the column.

4. Column washing

Wash with the binding buffer or washing buffer until the O.D. 280 nm reaches the baseline level again, normally 10-15 column volumes.

5. Purified protein elution

Elute the His-tagged protein with 5-10 column volumes of elution buffer and collect the fractions on ice.

For the correct storage of the protein, it is recommended to remove the imidazole by dialysis or ultrafiltration.

6. Storage of the column

Put the top and bottom stop plugs in the column and keep at 4-30°C in 20% ethanol. **Do not freeze.**

For reference only

For Research Use Only. Not for Diagnostic or Therapeutic Use.

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