Kia Ni-NTA Resin

KIAGEN
Science Base Company

Cat. No. KP101-01

Storage: at 2-8°C (20% ethanol) for two years

Description

Ni-NTA Resin is used for one-step purification of His tagged proteins. The His-tagged proteins bind to Ni^{2+} cations, which are immobilized on the Ni-NTA resin by 4 metal-chelating sites. After unbound proteins are washed away, the target proteins are recovered by gradient elution. It is suitable for both native and denatured protein purification.

Resin Specifications

Resin	6% agarose
Ligand	NTA
Shape	sphere
Pore size	45-165 μm
Binding capacity	10-20 mg of proteins/ml
Recommended flow rate	<300 cm/h
Highest resistance of atmospheric pressure	0.3 Mpa
pH stability	3-13

Procedures

1. Prepare Ni-NTA purification column
   (1) Resuspend the Ni-NTA resin in its bottle by inverting.
   (2) Transfer the resin into a purification column. Allow the resin to settle.
   (3) Equilibrate the column with 5-10 bed volume of equilibration buffer.
2. Prepare samples
   To avoid blocking column, samples should be centrifuged and filtrated before loading.
3. Load samples and wash
   Load samples and wash with 5-10 bed volume of equilibration buffer and collect the f1owthrough in one tube.
4. Elute
   Elute target protein with different concentration imidazole or different pH equilibration buffer.

5. Resin regeneration

(1) 2 bed volume of 6M GuHCl, 0.2 M acetic acid
(2) 5 bed volume of deionized water
(3) 3 bed volume of 2% SDS
(4) 1 bed volume of 25% ethanol
(5) 1 bed volume of 50% ethanol
(6) 1 bed volume of 75% ethanol
(7) 5 bed volume of 100% ethanol
(8) 1 bed volume of 75% ethanol
(9) 1 bed volume of 50% ethanol
(10) 1 bed volume of25% ethanol
(11) 1 bed volume of deionized water
(12) 5 bed volume of 100 mM EDTA, pH 8.0
(13) 10 bed volume of deionized water
(14) 5 bed volume of 100 mM NiSO4

Notes

Samples should be centrifuged and filtrated with 0.45 μm filter before loading .

- Equilibration Butler for soluble protein
  300 mM NaCl, 50 mM sodium phosphate butler, 10 mM imidazole, 10 mM Tris-Cl pH 8.0
- Equilibration Butler for inclusion body
  6 M GuHCl, 100 mM sodium phosphate butler;
  or 8 M urea, 100 mM sodium phosphate butler, 10 mM Tris-Cl pH 8.0

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