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MESGEN[™]
INNOVATION BIOTECHNOLOGY

Description

MesGen Ni-IDA Resin is an 4% cross-linked Resin medium covalently coupled to a chelating agent that binds Ni²⁺ by three coordination sites for high-affinity purification of polyhistidine-tagged recombinant proteins. Ni-Charged Resin has low Ni²⁺ leakage, high protein-binding capacity and stability, and is compatible with a wide range of additives used in protein purification. This makes Ni-IDA Resin the excellent choice for high performance purification of polyhistidine-tagged proteins.

Binding Capacity

≥40 mg of purified 6 x His-tag protein, MW=27k Da

Additional Materials Required

Note: The buffers listed below are recommended. To decrease nonspecific binding and increase yield, adjustments to the imidazole concentration might be required for specific proteins.

For native conditions prepare the following buffers:

- Equilibration Buffer: 20mM sodium phosphate, 300mM sodium chloride (PBS) with 10mM imidazole; pH 7.4
- Wash Buffer: PBS with 25mM imidazole; pH 7.4
- Elution Buffer: PBS with 250mM imidazole; pH 7.4

For denaturing conditions prepare the following buffers:

- Equilibration Buffer: PBS with 6M guanidine•HCl and 10mM imidazole; pH 7.4
- Wash Buffer: PBS with 6M guanidine•HCl and 25mM imidazole; pH 7.4
- Elution Buffer: PBS with 6M guanidine•HCl and 250mM imidazole; pH 7.4

For resin regeneration prepare the following buffer:

- MES Buffer: 20mM 2-(N-morpholine)-ethanesulfonic acid, 0.1M sodium chloride; pH 5.0

Procedure for Purification of His-tagged Proteins using a Gravity-flow Column

Perform the procedure at room temperature or at 4°C.

1. Pack column with an appropriate amount of Ni-IDA resin. Allow storage buffer to drain from resin by gravity flow.
2. Prepare sample by mixing protein extract with an equal volume of Equilibration Buffer.
3. Equilibrate column with two resin-bed volumes of Equilibration Buffer. Allow buffer to drain from the column.
4. Add the prepared protein extract to the resin.
5. Wash resin with two resin-bed volumes of Wash Buffer and collect the flow-through.
6. Elute His-tagged proteins from the resin with two resin-bed volumes of Elution Buffer. Repeat this step twice, collecting each fraction in a separate tube.
7. Monitor protein elution by measuring the absorbance of the fractions at 280nm or by Protein Assay Dye Reagent (Product No. MG1030). The eluted protein can be directly analyzed by SDS-PAGE.

Procedure for Ni-IDA Resin Regeneration

The Ni-IDA resin may be used at least three times without affecting protein yield or purity. Between each use, perform the procedure as described below to remove residual imidazole and any nonspecifically adsorbed protein.

1. Wash resin with 10 resin-bed volumes of MES Buffer.
2. Wash resin with 10 resin-bed volumes of ultrapure water.
3. Store resin as a 50% slurry in 20% ethanol.

Storage condition

2-8°C

Shelf life

18 months when stored unopened

For Research Use Only. Not for use in diagnostic procedures.