Nickel NTA Chelating Superflow 4

Note: Chelating agents such as EDTA or citrate buffers can strip the Nickel ions from the resin. Avoid using these except for elution. The resin optimally binds at neutral to slightly basic pH (7-8).

Before you start:

1. Measure the required amount of suction-dried resin.
2. Wash the resin in a Buchner funnel or empty column with 5 bed volumes of DI water.
3. Remove the excess water by gentle suction and transfer the resin into a beaker or flask.
4. Determine Imidazole concentration for binding buffer, lower concentration gives higher yield but lower purity.

Buffers:

1. Storage Buffer: 20% Ethanol
2. Binding Buffer: 20 mM Sodium Phosphate buffer with 0.5 M NaCl, 20-40 mM Imidazole, pH 7-8
3. Alternative Binding Buffer: Tris-HCl (Tris may reduce binding affinity)
4. Elution Buffer: 20 mM Sodium Phosphate, 0.5 M NaCl, 500 mM (dependent on target protein) Imidazole
5. Alternative Elution Buffer: 10 mM EDTA

Protocol:

1. Wash the resin with at least 5 bed volumes of DI Water.
2. Equilibrate the washed resin in a column with 5-10 column volumes of binding buffer at 50-150 cm/h.
3. Load desired histidine-tagged protein sample onto the column and resin.
4. Pour binding buffer through the column and collect the flow-through. Measure absorbance of flow-through.
5. Elute with elution buffer using a stepwise or linear gradient and measure absorbance of eluent liquid.
6. Wash resin with 10-20 bed volumes of DI water to remove addition/unchelated metal ions.
7. Suction dry resin to remove residual water.
8. Place Nickel NTA Chelating Superflow 4 in 0.5 bed volumes of 20% ethanol.
9. Store resin at 2-8 °C.

Storage:

Nickel NTA Chelating Superflow 4 (cat#937410SF4) is supplied in 20% ethanol. Store at 2-8 °C. Do not freeze. Do not expose the resin to acidic or basic conditions (unless it is prescribed in the instructions).

For technical service email info@sterogene.com or call (760) 929-0455