

Nickel Magnetic Particles

Product No. 544070 (2 mL), 544071 (5 mL)

1. General Description

The Abraxis' superparamagnetic nanoparticles contain nickel-charged compounds, and are utilized in the magnetic separation and isolation of polyhistidine-tagged proteins from protein extraction samples. The particles have a large surface area and four metal-binding sites with high capture efficiencies.

2. Safety Instructions

Reagent contains 20% ethanol. Take precaution in coming in close contact with high heat or fire.

3. Storage and Stability

The Nickel Magnetic Particles should be stored in the refrigerator (4-8°C). The reagent must be allowed to reach room temperature (20-25°C) before use and may be used until the expiration date on the box. Do not freeze, dry, or centrifuge the particles as they may result in loss of binding activity and aggregation.

4. Test Principle

The nickel magnetic particles are incubated with diluted extracted protein solution and then separated by magnets. After the unbound particulates are washed from the particles, the bound his-tagged proteins are eluted from the particles using the elution buffer. The particles are then magnetically separated from the eluted solution, which is removed manually.

5. Warning and Precautions

-Do not freeze the reagent.

-Prior to use, ensure that the product has not expired by verifying that the date of use is prior to the expiration date on the label.

-Ensure that reagent bottle caps are tight after each use to prevent drying of reagents.

-Avoid close contact with high heat or fire.

-Mistakes in handling the test can also cause errors. Possible sources for such errors can be:

Inadequate storage conditions of the test kit (or reagents), incorrect pipetting sequence or inaccurate volumes of the reagents, too long or too short incubation times, and/or short magnetic separation times.

6. Characteristics

Particle mean diameter: ~0.5 µm

Particle concentration: 12.5 mg/mL

Binding capacity: ≥40 µg pure his-tagged protein/mg of particles

7. Protein Isolation

A. Materials Provided

1. Nickel magnetic particles, 12.5 mg/mL

B. Additional Materials (not provided with the kit)

1. Binding/Wash Buffer: PBS (0.1M Sodium Phosphate, 0.3M Sodium Chloride), 0.05% Tween 20 detergent, pH 8.0
2. Elution Buffer: PBS, 0.05% Tween 20, 0.5M Imidazole pH 8.0
3. Neutralization Buffer: 1M Tris pH 8.0, 1 mL
4. Micro-pipettes with disposable plastic tips (10-200 and 200-1000 μ L)
5. 1.5 mL or 2.0 mL Eppendorf or microcentrifuge vials
6. Timer
7. Rotator
8. Distilled or deionized water
9. Vortex mixer
10. Solo or Multi-6 Microcentrifuge Separator (PN 472270; PN 472260)

C. Procedures

1. Add 100 μ L (0.5 mg) of particles to 1 mL of binding buffer in each tube to wash particles.
2. Magnetically separate using a magnetic separator for 2 minutes or when the supernatant is clear.
3. Remove the supernatant and wash once more by adding 1 mL of binding buffer.
4. Repeat step 2 and remove the supernatant.
5. Resuspend particles by adding 450 μ L of binding buffer.
6. Add 50 μ L of extracted protein supernatant to the particles.
Note: Sample volume can be modified according to user preference. If the sample volume is < 500 μ L, dilute it to a final volume of 500 μ L with Binding/Wash Buffer.
7. Gently mix using vortex or rotator for 30 minutes.
8. Magnetically separate using a magnetic separator for 2 minutes or when the supernatant is clear.
9. Remove supernatant and wash with 0.5 mL Binding/Wash buffer to remove unbound proteins.
10. Repeat steps 8 and 9 once more. Remove supernatant.
11. Add 25 μ L of elution buffer to particles and mix well.
12. Incubate at room temperature for 10 minutes with occasional gentle mixing or vortex.
13. Separate for 2 minutes and remove the eluent to a new tube containing 15 μ L of neutralization buffer.
14. Repeat the elution steps 11, 12, and 13 once more. Combine the two eluates, if desired.

Ordering Information

Description	Size (mL)	Part Number
Nickel Magnetic Particles	2, 5	544070/71
Multi-6 Microcentrifuge Separator		472260
Solo Tube Separator		472270
15/50 mL Tube Separator		472250
Multi Purpose Magnetic Separator (15/50/microcentrifuge)		472280

General Limited Warranty: Abraxis LLC warrants the products manufactured by the Company, against defects and workmanship when used in accordance with the applicable instructions for a period not to extend beyond the product's printed expiration date. **Abraxis LLC makes no other warranty, expressed or implied. There is no warranty of merchantability or fitness for a particular purpose.**

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