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# Ni Smart Magarose Beads

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## 1. Product Description

**Ni Smart Magarose Beads** is a new kind of IMAC packing, chelating nickel ions ($Ni^{2+}$)solider. **Ni Smart Magarose Beads** can be used to purify 6xHis-tagged proteins expressed in series of expression vectors, such as *E.coli*., yeast, insect cells and mammalian cells. This form is very stable octahedral structure of nickel ions in the center, which can protect the nickel ions from attack of the competitive small molecule. The structure of Ni-SMART is compatible with a certain concentration of reducing agents, denaturing agents, detergents and other additives.

Table 1. Characteristics of **Ni Smart Magarose Beads**

| Item | Description |
|---|---|
| Matrix Spherical | Magnetic agarose |
| Static Binding Capacity | ＞10mg 6×His-tagged protein/ml medium |
| Particle size（μm） | 30-100um |
| Beads concentration | 20%(V/V) slurry |
| Storage Solution | 1×PBS containing 20% ethanol |
| Storage Temperature | 2-8℃ |

Table 2. Chemical compatibilities for **Ni Smart Magarose Beads**

| Reagent | Stability |
|---|---|
| 0.01M NaOH | 1week |
| 10mM EDTA,1M NaOH, 5mM DTT, 5mM TCEP, 20mM β-mercaptoethanol, 6MGua-HCl | 24hour |
| 500mM imidazole，100mM EDTA | 2hour |
| 30% isopropanol | 20min |

## 2. Purification Procedure

### 2.1 Buffer Preparation

The basic principle of the following recommended buffer and other buffer is low concentration of imidazole in Lysis and wash buffer and high in elution buffer. Water and chemicals used for buffer preparation should be of high purity. It is recommended to filter the buffers by passing them through a 0.22μm or 0.45 μm filter before use.

**Lysis Buffer**:50 mM $NaH_2PO_4$, 0.5M NaCl, pH7.4

**Wash Buffer**:50 mM $NaH_2PO_4$, 0.5M NaCl, 0-5mM imidazole，pH7.4

**Elution Buffer**:50 mM $NaH_2PO_4$, 0.5M NaCl, 250mM imidazole，pH7.4

### 2.2 Sample Preparation

1) Centrifuge the homogenized lysate at 7,000rpm(7,500×g) for 20min at 4℃ to clarify sample. Save supernatant.
2) The sample contains a tolerable range of reagents and does not require dialysis or concentration. It can be sampled directly.

### 2.3 Preparation of the Magnetic Beads

The protocol uses 100 μl **Ni SMART Magarose Beads**, but this may be scaled up or down as required.

1) Completely resuspend the beads by shaking or vortexing the vial.
2) Transfer 500 μl **Ni Smart Magarose Beads** (20% v/v) into a clean tube.
3) Place the tube on a magnetic separation rack to collect the beads. Remove and discard the supernatant.
4) Add 1 ml Lysis Buffer to the tube and invert the tube several times to mix. Use the magnetic separation rack to collect the beads and discard the supernatant. Repeat this step twice.

### 2.4 Protein Purification

1) Transfer the 6X His-tagged Protein lysate to **Ni Smart Magarose Beads.** Incubate the tube at room temperature with mixing (on a shaker or rotator) for 10 -30 minutes. Place the tube on a magnetic separation rack to collect the beads. Remove and discard the supernatant. If necessary, keep the supernatant for analysis.
2) Add 1ml Wash Buffer to the tube and mix well, use the magnetic separation rack to collect the beads and discard the supernatant. Repeat the wash step three more times.
3) Add 200-300μl Elution Buffer, mix well and incubation for 5-10min. Use the magnetic separation rack to collect the beads and transfer the supernatant into a new tube. Repeat this step one more time.

### 2.5 Analysis

Identify the fractions containing the His-tagged protein. Use UV absorbance, SDS-PAGE, or western blot.

## 3. Additional Information

1) Please read the product instruction carefully before using the product.
2) In the process of magnetic beads preservation, operations such as freezing, drying and high-speed centrifugation should be avoided, otherwise the structure of magnetic beads will be damaged and the binding capacity of proteins will be seriously affected.
3) Before using the magnetic beads, please oscillate gently and fully to keep the beads in a uniform suspension state.
4) The baeds can be reused to purify the same protein; when different proteins are purified, it is recommended to use new magnetic beads to avoid cross-contamination.

## 4. Related Products

| Product | Cat. No. | Size |
|---|---|---|
| Ni IDA Magarose Beads | SM001001 | 1 ml |
| | SM001005 | 5 ml |
| | SM001025 | 25 ml |
| | SM001100 | 100 ml |
| | SM00101L | 1 L |
| Ni NTA Magarose Beads | SM008001 | 1 ml |
| | SM008005 | 5 ml |
| | SM008025 | 25 ml |
| | SM008100 | 100 ml |
| | SM00801L | 1 L |
| Ni Smart Magarose Beads | SM025001 | 1 ml |
| | SM025005 | 5 ml |
| | SM025025 | 25 ml |
| | SM025100 | 100 ml |
| | SM0501L | 1 L |