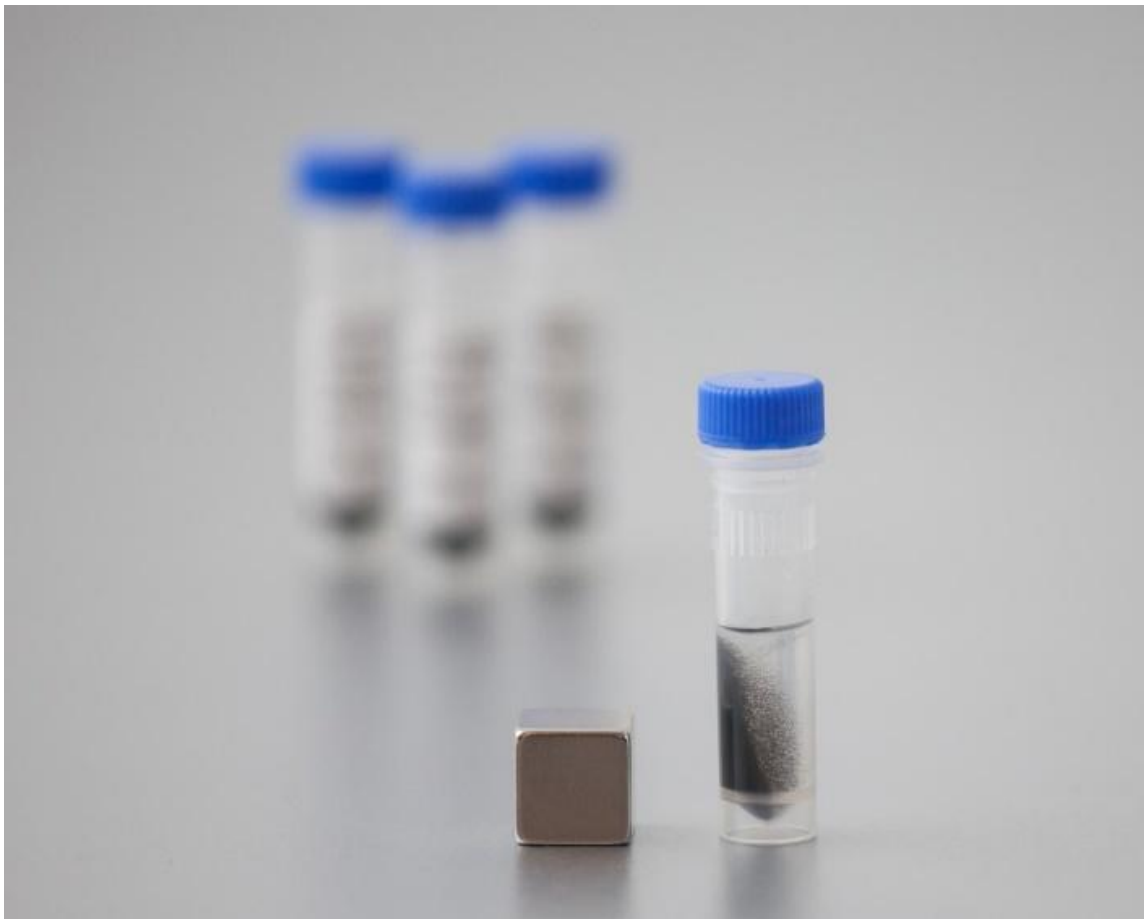
INSTRUCTION

MAGicBeads Ni-IMAC



- Super-paramagnetic porous beads
- Exceptional magnetization and zero magnetic remanence
- High binding capacity
- Minimal non-specific interaction

Please read through this manual carefully before using MAGic™Beads Ni-IMAC.

Intended use

This product is intended for purification of biomolecules carrying a polyhistidine tag.

For research use only.

Bead suspension

The product is a 10% bead suspension; however, it may appear to contain a larger bead volume when bead–bead interactions occur, altering bead configurations. Once the beads are properly resuspended, these interactions are disrupted, revealing the true bead volume. This phenomenon is batch-specific but does not affect bead performance.

General information

MAGicBeads Ni-IMAC provides a simple and effective system for small to medium scale isolation of biomolecules carrying a polyhistidine tag using magnetic separation techniques. The quantity of high-capacity beads can easily be scaled up or down to match the target concentration and sample volume. The beads are suitable for separations using appropriate magnetic separators, such as the MAGicAccio line.

MAGicBeads Ni-IMAC consists of super-paramagnetic agarose beads covalently coupled with an NTA ligand and loaded with nickel ions to obtain a matrix with affinity for histidine residues. This product is intended for purification of biomolecules carrying a polyhistidine tag.

The MAGicBeads Ni-IMAC magnetic particles are easily attracted to external magnets, allowing efficient separation. The agarose matrix enables minimal nonspecific binding of proteins due to its hydrophilic nature. The black beads are easily observed by the naked eye, making them easy to follow and collect, and they do not aggregate, which facilitates the resuspension.

Product data

Table 1. Characteristics for MAGicBeads Ni-IMAC

Product	MAGicBeads Ni-IMAC, 10% bead suspension
Target molecule	Histidine-tagged proteins
Matrix	Super-paramagnetic porous agarose
Ligand	Nitrilotriacetic acid loaded with nickel ion (Ni-NTA)
Average particle size (DV50)[1]	90 µm
Particle size distribution	50–150 µm
Binding capacity[2]	20-30 mg 30 kDa his_6-protein/ mL settled beads
Storage	+2 to +8°C in 20% ethanol. Do not freeze

1 The median particle size of the cumulative volume distribution
2 The binding capacity will depend on the nature of the his-tag and the protein

Materials supplied

- MAGicBeads Ni-IMAC supplied as a 10% bead suspension in 20% ethanol. 10 mL 10% bead suspension contains 1 mL beads.

Additional materials needed

- **Equilibration/Washing buffer** – For washing of beads, use PBS (137 mM NaCl, 2.7 mM KCl, 10 mM phosphate) + 0.5M NaCl pH 7.5
- **Elution buffer** – PBS + 0.5M NaCl + 0,5M Imidazole pH 7.5
- **Storage buffer** – Store beads in 20% ethanol.
- **Mixer** – Mix samples during incubations using an end-over-end mixer, a benchtop shaker, or a rocking table. Manual inversion of the vial can also be applied.
- **Magnetic separator** – MAGicAccio LAB (Product No. 2000, 2100) are suitable for separations in 0.5–5 mL volumes. For separation of volumes larger than 5 mL, use MAGicAccio PILOT 50 (Product No. 2200) for volumes up to 50 mL, MAGicAccio PILOT 500 (Product No. 2300) for volumes up to 500 mL, or MAGicAccio PILOT 2000 (Product No. 2400) for volumes up to 2000 mL.
- Additional vials/tubes, pipettes and pipette tips.

Handling instructions

Dispensing the bead suspension

- The bead suspension should be well suspended before dispensing. Mix thoroughly by manual inversion or by vortexing, between each pipetting from the vial.

Magnetic bead separation

- MAGicAccio LAB can be used to collect the beads from liquid volumes up to 5 mL. For volumes from 5 mL up to 50 mL it is recommended to use the MAGicAccio PILOT 50 separator. Use the MAGicAccio PILOT 500 separator for volumes up to 500 mL or the MAGicAccio PILOT 2000 for volume up to 2000 mL. Refer to the manual of the separators for detailed instructions.
- Use the magnetic separator to attract the magnetic agarose beads to the wall of the test tube or bottle before each liquid removal step.
- Remove liquid carefully, trying not to disturb the magnetic beads. To avoid sample loss, make sure that no beads are removed.
- Move the tube away from the magnetic field, add new liquid and resuspend the beads by mixing.

Incubation

- Incubations should be performed with continuous mixing, using either an end-over-end apparatus, a bench-top shaker, or a rocking table. Short incubations, e.g., for elution, can be performed by using manual mixing/inversion of the test tube or bottle.
- Binding and elution can be performed at room temperature, as well as in a cold room.

Product operation

Intended use

- This product is intended for purification of biomolecules carrying a polyhistidine tag.

Bead input

- The amount of beads and the binding time strongly depends on the target concentration in the sample.

Binding

- The MAGicBeads Ni-IMAC bind polyhistidine tags, in the range of pH 7.5.
- Purification can be performed directly in cell broth or cell lysis, without diluting the sample. However, the pH must be within the given range.

Washing

- In most applications, it is sufficient to wash the beads PBS + 0.5M NaCl pH 7.5.

Elution

- The recommended elution buffer is PBS + 0.5M NaCl + 0,5M Imidazole pH 7.5.

Regeneration of magnetic beads

- Ni-IMAC beads should be regenerated/recharged on a regular basis every 5^{th} to 10^{th} run
- Regeneration/recharging
    - 3 washes with dH_2O
    - 3 washes with 50mM sodium acetate pH 6.0
    - 1 wash dH_2O
    - 2.5% Ni^{2+} solution
    - 2h incubation
    - 4 washes dH_2O
    - 1 wash with wash buffer, let incubate for 10 min
    - 1 wash dH_2O
    - 6 washes with Tris-HCl pH 7.5
    - 1 wash dH_2O
    - Resuspend in storing buffer

Storage

- The MAGic™ Beads Ni-IMAC should be stored as a 10% bead suspension at +2 to +8°C in 20% ethanol.

Practical notes

- Beads caught in the lid or on the walls of the reaction vial can be recovered by washing with solution using a pipette or removed with a quick spin in a microcentrifuge.
- If low amount of target is recovered, increase the amount of magnetic beads and/or increase the time of incubation.
- It is recommended to optimize the coupling time of target to beads depending on sample source and target concentration.
- When reusing beads, it is recommended to use the beads for purification of the same target to avoid any cross-contamination between purification runs.
- If there is a need to elute the bound target in a smaller volume than is possible with the magnetic separators, the magnetic beads can be transferred and eluted using a basic gravity flow column setup.

Disclaimer

The product is not fully tested. For research use only.

Ordering information

Products	Quantity	Product No.
MAGicBeads Ni-IMAC	1 mL beads	1100
MAGicBeads Ni-IMAC	10 mL beads	1101
MAGicBeads Ni-IMAC	50 mL beads	1105
MAGicBeads Ni-IMAC	100 mL beads	1110
MAGicBeads Ni-IMAC	250 mL beads	1125
MAGicBeads Ni-IMAC	500 mL beads	1150

Related products	Product No.
MAGicBeads ACT	13XX
MAGicAccio LAB rack	2000
MAGicAccio LAB cube	2100
MAGicAccio PILOT 50	2200
MAGicAccio PILOT 500	2300
MAGicAccio PILOT 2000	2400
MAGicAccio PROCESS	3100

MAGIC BIOPROCESSING
Virdings Allé 28
SE-754 50 Uppsala, Sweden

Email: info@magicbioprocessing.com
Web: www.magicbioprocessing.com