MagListo™ His-tagged Protein Purification Kit

[Cat. No.] K-7200

Introduction

MagListo™ His-tagged Protein Purification Kit allows easy purification of His-tagged proteins through magnetic separation by Ni-NTA Magnetic Nanobeads and buffers. Ni-NTA Magnetic Nanobeads are silica beads with an average of 400 nm and a range of 200-800 nm diameter. Nickel-chelating nitrilotriacetic acid (Ni-NTA) groups coated on the surface of the magnetic nanobeads can interact with His-tagged proteins. After washing out unbound proteins, the target proteins can be purified through an elution process.

Components

Components	Amount
Ni-NTA Magnetic Nanobeads (10%)	1 ml x 5 ea
Binding/Washing buffer	100 ml
Elution buffer	15 ml
Nd magnet	3 ea

*** Note:** For research use only. Not for use in diagnostic or therapeutic procedures.

Specifications

Ni-NTA Magnetic Nanobeads	
Composition	Silica based magnetic nanobeads
Binding capacity	3 mg of protein/ml of beads
Size	Average 400 nm
Concentration	100 mg/ml

Buffer Composition

Binding/Washing buffer	50 mM sodium phosphate, 500 mM NaCl, 10 mM imidazole, pH 8.0
Elution buffer	50 mM sodium phosphate, 500 mM NaCl, 500 mM imidazole, pH 8.0

Storage Buffer

Ni-NTA Magnetic Nanobeads are supplied as a 10% (v/v) suspension in 20% ethanol.

Storage

- Ni-NTA Magnetic Nanobeads and Buffers should be stored at 2-8°C.

Precautions

- Determine whether to purify proteins under native conditions or denaturing conditions depending on the protein folding state.
- An exact protocol may need to be optimized by the user.

Online Resources

Korean

English

Visit our **product page** for additional information and protocols.

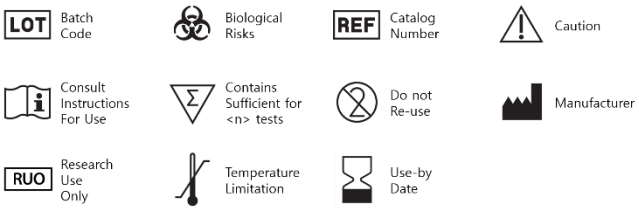
Ordering Information

Description	Cat. No.
MagListo™ His-tagged Protein Purification Kit	K-7200

Notice

BIONEER corporation reserves the right to make corrections, modifications, improvements and other changes to its products, services, specifications or product descriptions at any time without notice.

Explanation of Symbols



Experimental Procedures

Steps		Procedure Details
	Before you begin	
	 **Preparation of *E. coli* lysate**	1. Prepare 3 ml of pre-induced bacterial culture with IPTG. 2. Harvest the cells by centrifugation for 1 min at 12,000 rpm at 4°C and discard the supernatant. 3. Resuspend the cells in 500 µl of Binding/washing buffer. 4. Sonicate the cells using a sonicator equipped with a micro-tip on ice. 5. Centrifuge the *E. coli* lysate for 5-10 min at 12,000 rpm at 4°C to pellet the cell debris and save the supernatant. 6. Take 10 µl of the supernatant for SDS-PAGE analysis. *** Note:** Supernatant from this step is the **Cleared lysate sample** from *E. coli* for checking the total proteins.
1	 **Equilibrating Ni-NTA magnetic nanobeads**	1. Transfer 1 ml of Ni-NTA magnetic nanobeads to a 1.5 ml tube and place the tube on a Neodymium (Nd) magnet for 1 min. 2. Remove the supernatant. 3. Equilibrate by adding 1 ml of Binding/washing buffer to the bead slurry and mix briefly. 4. Place the tube on the Nd magnet for 1 min and remove the supernatant. 5. Repeat step 3 and 4 one more.
2	 **Protein binding**	6. Load up to 500 µl of cleared lysate onto the pre-equilibrated magnetic nanobeads. 7. Mix by inverting 5-10 times. 8. Place the tube on the Nd magnet for 1 min and remove the supernatant. *** Note:** Supernatant from this step is the **Unbound sample** for checking the binding efficiency.
3	 **Washing magnetic nanobeads**	9. Add 1 ml of Binding/washing buffer and wash the magnetic nanobeads by gently pipetting. 10. Place the tube on the Nd magnet for 1 min and remove the supernatant. *** Note:** Supernatant from this step is the **Washing sample** for checking the washing conditions. 11. Repeat step 9 and 10 three times. *** Note:** After the final wash, the remaining Binding/washing buffer should be removed completely.
4	 **Eluting target proteins**	12. Add 250-300 µl of Elution buffer to elute target proteins from magnetic nanobeads and gently mix. *** Note**: If more concentrated protein solutions are required, elute in lesser volumes. 13. Incubate for 1 min at room temperature. 14. Place the tube on the Nd magnet for 1 min and collect the supernatant. *** Note:** Supernatant from this step is the **Elution sample** for checking the final target proteins. 15. Repeat the Elution step once more. 16. Identify the purification steps by analyzing each sample with SDS-PAGE.
	 **Option**	• If you want to purify proteins with a centrifuge instead of a Nd magnet, Centrifuge for 30 sec at 12,000 rpm in step 1, 4, 8, 10, 14. • You can also use BIONEER's *MagListo*™-2 Magnetic Separation Rack **(Cat. No. TM-1010)** instead of a Nd magnet. Refer to the Manual of this product for additional information.