High Performance Magnetic Nanoparticles

FG beads®

The FG (Ferrite-glycidyl methacrylate) beads developed by Tokyo Institute of Technology consist of 200 nm-diameter ferrite nanoparticles coated firmly with a polymer layer. The FG Beads are used as carriers for affinity purification of target proteins. ¹)

Features

- **Extremely low non-specific binding**
  Poly-GMA (Glycidyl methacrylate) coated magnetic nanoparticles.
- **Excellent recovery of target proteins**
  200 nm particles have a large surface area and a high dispersibility.
- **High stability in organic solvents**
  Various compounds can be immobilized on the beads.

Comparison with other magnetic beads ²)

<table>
<thead>
<tr>
<th>Linker and Function Group</th>
<th>Ligands to be fixed</th>
<th>Linker and Function Group</th>
<th>Ligands to be fixed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Plain beads</td>
<td>R-NH₂ (Amino group)</td>
<td>Streptavidin</td>
<td>Biotinylated compounds</td>
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<tr>
<td>Linker beads (epoxy beads)</td>
<td>R-SH (Thiol group)</td>
<td>NeutrAvidin™</td>
<td>Biotinylated compounds</td>
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<td>NH₂ beads</td>
<td>R-COOH (Carboxy group)</td>
<td>Protein A</td>
<td>IgG</td>
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<td>COOH beads</td>
<td>R-NH₂, R-NHR ²</td>
<td>Protein G</td>
<td>IgG</td>
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<td>NHS beads</td>
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<tr>
<td>Azide beads</td>
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Purification process

Purification of target protein of Thalidomide (elucidation of the teratogenic mechanism) 

CRBN (Cereblon) and DDB1 were isolated from human cell extract using thalidomide fixed beads. As a result, the teratogenic mechanism of thalidomide was elucidated.

Purification of novel target protein of MTX (methotrexate) 

When MTX is fixed via different site, a novel protein is purified and identified as deoxycytidine kinase (dCK). As a result, a possible mechanism of synergistic effect between MTX and ara-C on malignant lymphoma was proposed.

Purification of target protein of Capsaicin 

Prohibitin 1 and prohibitin 2 were isolated from human myeloid leukemia NB4 cell extract using capsaicin derivative (Cap-NH2) fixed beads. As a result, the apoptosis induction mechanism of capsaicin was elucidated.

Elucidation of the mechanism of enteropathogenic E. coli infection 

EspB is a protein of enteropathogenic E. coli (EPEC) essential for infection in humans. Myosin was isolated from human cell extract using EspB fixed beads. As a result, the mechanism of EPEC infection was elucidated.
Protein A / Protein G beads

- **High recovery**  IgG binding capacity – more than twice the amount of a competitor.
- **High purity**  Extremely low non-specific adsorption.
- **Quick processing**  30 minutes for IgG binding

**Applications**
- IgG purification
- Immunoprecipitation (IP)
- Chromatin Immunoprecipitation (ChIP)
- Protein separation

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IgG Purification

Sample:
1. HeLa Extract + IgG
2. FG beads (Protein A)
3. Competitor (Protein A)
4. FG beads (Protein G)
5. Competitor (Protein G)
6. Input IgG

Detection: Silver Staining

We checked the performance of FG beads (Protein A and Protein G) in an immunoprecipitation experiment. By using FG beads, antigen HSA was immunoprecipitated with high recovery and extremely low non-specific adsorption.

1. Add 10 μg of IgG into 200 μg of HeLa cell extracts (200 μl).
2. Add 300 μg of each beads to the HeLa cell extracts.
3. React for 10 min at 4°C and separate beads from the HeLa cell extract.
4. Elute bound IgG by adding Glycine-HCl.

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Immunoprecipitation

Sample:
1. HeLa Extract + HSA
2. FG beads (Protein A)
3. FG beads (Protein G)
4. Input IgG

Detection: Silver Staining

We checked the performance of FG beads (Protein A and Protein G) in an immunoprecipitation experiment. By using FG beads, antigen HSA was immunoprecipitated with high recovery and extremely low non-specific adsorption.

1. Immobilize anti-Human Serum Albumin antibody on FG beads.
2. Add 400 ng of HSA into 200 μg of HeLa cell extracts (200 μl).
3. Add 0.1 mg of each beads to HeLa cell extracts.
4. React for 120 min at 4°C and separate beads from the HeLa cell extract.
5. Elute bound IgG and HSA by adding Glycine-HCl.

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Sample:
1. Input
2. FG beads (Streptavidin)
3. Competitor (Streptavidin)
4. Competitor (Streptavidin)

Detection: Silver Staining

We compared the performance of biotinylated drug MTX (Methotrexate) immobilized FG beads with the beads of a competitor in a target protein purification experiment. By using FG beads, MTX target protein DHFR was purified with extremely low non-specific adsorption.

1. Immobilize biotinylated MTX on Streptavidin beads.
2. Add 0.5 mg of each beads into 600 μg of HeLa cell extracts (200 μl).
3. React for 120 min at 4°C and separate beads from the HeLa cell extract.
4. Elute bound DHFR by adding elution buffer.

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Streptavidin / NeutrAvidin beads

- **High recovery**  Biotin binding capacity – more than twice the amount of a competitor.
- **High purity**  Extremely low non-specific adsorption.

**Applications**
- Immunoprecipitation (IP)
- Chromatin Immunoprecipitation (ChIP)
- Cell separation
- Affinity purification of drug target proteins

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Immunoprecipitation

Sample:
1. HeLa Extract + HSA
2. FG beads (Streptavidin)
3. Competitor (Streptavidin)
4. Competitor (Streptavidin)

Detection: Silver Staining

We compared the performance of biotinylated drug MTX (Methotrexate) immobilized FG beads with the beads of a competitor in a target protein purification experiment. By using FG beads, MTX target protein DHFR was purified with extremely low non-specific adsorption.

1. Immobilize biotinylated MTX on Streptavidin beads.
2. Add 0.5 mg of each beads into 600 μg of HeLa cell extracts (200 μl).
3. React for 120 min at 4°C and separate beads from the HeLa cell extract.
4. Elute bound DHFR by adding elution buffer.
Magnetic Stand

- **Quick cooling down**
The magnetic stand made of metal can quickly cool down samples on ice. You can experiment without protein denaturation.

- **High speed separation**
The magnetic stand separates magnetic nanoparticles in shorter time than competitors because shape and placement of magnets are well designed.

**Reference**
1) S. Sakamoto et al., Chem. Rec. 9 (2009) 66
2) K. Nishio et al., Colloids Surfaces. B. 64 (2008) 162
3) T Ito et al., Science 327 (2010) 1345
6) Y. Iizumi et al., Cell Host & Microbe. 2 (2007) 383

**Ordering Information**

<table>
<thead>
<tr>
<th>Product Name</th>
<th>Storage</th>
<th>Product No.</th>
<th>PKG Size</th>
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(Storage) RT: Room Temperature  R: Refrigerator  F: Freezer

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