

Magnosphere™ MS160/Tosyl

PRODUCT DESCRIPTION

Magnosphere™ MS160/Tosyl beads are magnetic microparticles designed for covalent immobilization of ligands containing -NH₂ groups for bioseparation. Their surfaces are covered with a JSR Life Sciences proprietary hydrophilic polymer, on which Tosyl group is incorporated as an active group. The Tosyl group makes it possible to immobilize ligands such as antibody without using coupling reagents. As the hydrophobic Tosyl group is eliminated through a coupling reaction, surface of the beads became hydrophilic after the reaction.

This chemistry enables ligands to keep their function high and achieve a high yield and low non-specific binding bioseparation.

Uniform particle size and superparamagnetic property of the **Magnosphere™ MS160/Tosyl** beads help good magnetic separation and re-suspension response. These characters of the beads are ideal for variety of applications such as enzyme immunoassay, immunoprecipitation-western blot.

FEATURES

- Uniform particle size
- Superparamagnetic
- Rapid magnetic responsiveness
- Low non-specific binding of proteins

EXAMPLE APPLICATIONS

Immunoassay, Immunoprecipitation-Western blot, nucleic acid hybridization

SPECIFICATIONS

| | |
|-----------------------------|------------------------------------|
| Package volume | 4 mL |
| Solid content in slurry | 1 % (10 mg/mL) |
| Dispersion media | 0.01%ProClin950 / H ₂ O |
| Bead diameter | 1.5 μm |
| Bead magnetite content | 28 % approx. |
| Surface tosyl group density | 150 nmol/mg bead approx. |
| Shelf life | Labeled on the bottle |

STORAGE

Stored at 2-8 °C. Do not freeze the vial. Vortex the vial or pipette gently up and down to obtain a homogeneous dispersion before use.

Stability of the beads after immobilization of ligand molecules depends on the condition of the preparation.

RECOMMENDED PROTOCOLS

An example of chemical coupling protocol of antibody onto the **Magnosphere™ MS160/Tosyl** beads is shown below. The optimum condition may depend on the toughness of the antibody used.

Reagent and equipment requirement

| | |
|-----------------------------|---|
| Binding Buffer: | 0.1 M Borate buffer pH 9.5 |
| Washing Buffer: | TBS-T (25 mM Tris-HCl, pH 7.2, 0.15 M NaCl, 0.05 % Tween20) |
| Catalytic reagent solution: | 3M Ammonium sulfate / 0.1M Borate buffer, pH9.5 |

| | |
|-------------------|---|
| Blocking reagent: | 10 % BSA / H ₂ O |
| Equipment: | Magnetic separator. Vortex tube mixer. Tube rotator with temperature control (37 °C) |

[Protocol I] Chemical antibody coupling onto the beads

1. Suspend the **Magnosphere™ MS160/Tosyl** beads well using Vortex mixer and put 1 mL of the suspension (i.e., 10 mg beads) into a microtube.
2. Place the tube on a magnetic separator for 1 minute and remove the supernatant carefully.
3. Add 900 μL (= A μL) of Binding Buffer and suspend the beads by vortexing.
4. Add 200 μg (= B μL) of antibody (200 μL, if antibody was diluted to 1 mg/mL) and suspend the beads by vortexing.
5. Add (A+B)/2 μL of Catalytic reagent solution and suspend the beads by vortexing.
6. Keep rotate the beads for 18 hours at 37 °C.
7. Add 10 μL of Blocking reagent and continue to react for 6 hours at 37 °C.
! Blocking is important to eliminate the unreact Tosyl groups
8. After the reaction, remove the supernatant as in step 2.
9. Add 500 μL of Washing Buffer and suspend the beads by vortexing.
10. Remove the supernatant as in step 2.
11. Repeat steps 9 & 10 for a total of 3 times.
12. Suspend the beads with a desired buffer suitable for downstream applications and store at 2-8 °C until needed.

APPENDIX [Appendix I] Time course of antibody coupling reaction

Antibody, which added to the **Magnosphere™ MS160/Tosyl** beads, is physically adsorbed on the beads at first. Then, the antibody is coupled covalently in progression (Fig.1). The beads became hydrophilic through the progress of coupling since the tosyl is a leaving group.

Physically adsorbed antibody sometime causes loss of long-term stability due to the detachment of the antibody. Increase in reaction time, temperature and addition of salt (such as (NH₄)₂SO₄ or Na₂SO₄) reduce the adsorbed antibody. It is also effective to add excess amount of blocking reagent, such as BSA, at the latter period of reaction to get off the antibody from the beads.

Physically adsorbed antibody can be washed out through a washing with ionic surfactant, such as 0.5% SDS, but washing with ionic surfactant may cause loss of antibody affinity.

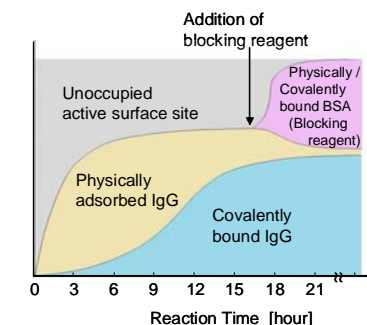


Fig.1 Time course of bound IgG on the beads

IMPORTANT NOTICE

- This product is for research use only and not intended for therapeutic or *in vivo* diagnostic use.
- The specificity of this product may change without notice.
- JSR Life Sciences Corporation does not guarantee that this product will be continuously available.
- JSR Life Sciences Corporation makes no warranties as to this product including, but not limited to, implied warranties of merchantability or fitness for a particular purpose.

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