Conjugation Protocol for Absolute Mag™ Streptavidin Magnetic Particles, 1 μm

Cat. No: WHM-X053

Introduction

Absolute Mag™ streptavidin magnetic particles offer easy affinity isolation or handling of bionylated nucleic acids, antibodies, or other biotinylated ligands and targets without columns or centrifugation. Our streptavidin conjugated magnetic beads are uniform and superparamagnetic beads with a monolayer of recombinant streptavidin covalently coupled to the surface. The magnetic beads are re-blocked with biocompatible polymers with significantly low non-specific binding. The high affinity interaction between streptavidin and biotin ($Kd=10^{-15}$) is used in a vast number of applications.

Features

- Narrow size distribution, CV≤5%
- Significant low non-specific binding to proteins and nucleic acid
- Easily adapted to automated processes
- No iron exposure to ensure lowest autosignal, particularly with respect to chemiluminescence
- Compatible with PCR reaction

Binding Capacity

1 mg of 1 μm Absolute Mag™ streptavidin magnetic particles typically binds:

- ~3,500 pmoles free biotin
- ~560 pmol biotinylated peptides
- ~25 μg biotinylated IgG
- ~25 μg ds-DNA
- ~700 pmol ss-oligonucleotides

Beads treatment for RNA Manipulation

Reagents and Materials Required

- Absolute Mag™ Streptavidin Magnetic Particles, 1 μm (WHM-X053), resuspend the magnetic beads before use.
- Washing Buffer 1: DEPC-treated 0.1 M NaOH
- Washing Buffer 2: DEPC-treated 0.1 NaCl
- Biotinylated RNA
- Magnetic Separator

1 μm Absolute Mag™ streptavidin magnetic particles are not provided in RNase-free solutions, please follow the following steps for RNA applications.

45-1 Ramsey Road Shirley, NY 11967, USA
Tel: 1-631-624-4882 Fax: 1-631-938-8221
E-mail: info@creative-diagnostics.com
www.cd-bioparticles.com

Procedures

1. Transfer 1 mL Absolute Mag™ Streptavidin Magnetic Particles into a 1.5 mL centrifuge tube.
2. Place this tube in a magnetic separator and remove the supernatant after the supernatant is clear.
3. Add 1 mL Wash Buffer 1 to the magnetic beads pellet and resuspend the magnetic beads by vortexing for 15 seconds.
4. Wash the magnetic beads twice with Washing Buffer 1 and resuspend the magnetic beads in 1 mL Wash Buffer 2.
5. Wash the magnetic beads once with Washing Buffer 2 and resuspend the magnetic beads in 1 mL Wash Buffer 2.

The magnetic beads now ready to bind biotinylated RNA.

Immobilization Protocol for biotinylated antibodies or proteins

Reagents and Materials Required:

• Absolute Mag™ Streptavidin Magnetic Particles, 1 μm (WHM-X053), resuspend the magnetic beads before use.
• Washing & Binding Buffer: PBS buffer 0.01% BSA, pH 7.4 or PBS buffer 0.01% tween 20 depending on preference or concern about any downstream applications.
• Biotinylated Protein
• Biotinylated RNA
• Magnetic Separator

Procedures

1. Wash the magnetic beads with Washing & Binding Buffer three times by a magnetic separator.
2. Incubated the magnetic beads with biotinylated protein in Washing & Binding Buffer for 30 mins at room temperature using gentle rotation.
3. Separate the magnetic beads with a magnetic separator after the supernatant is clear.
4. Wash the magnetic beads 5 times with Washing & Binding Buffer.
5. Resuspend the magnetic beads with the desired concentration for downstream applications.

Immobilization Protocol for biotinylated Nucleic Acid

Reagents and Materials Required

• Absolute Mag™ Streptavidin Magnetic Particles, 1 μm (WHM-X053), resuspend the magnetic beads before use.
• Washing & Binding Buffer: 10 mM Tris-HCl, 1 mM EDTA, 2 M NaCl, pH 7.4
• Biotinylated nucleic acids
• Magnetic Separator

Procedures

1. Wash 0.1 mL magnetic beads with Washing & Binding Buffer three times by a magnetic separator.

45-1 Ramsey Road Shirley, NY 11967, USA
Tel: 1-631-624-4882 Fax: 1-631-938-8221
E-mail: info@creative-diagnostics.com
www.cd-bioparticles.com

2. Resuspend the washed beads in 0.2 mL Washing & Binding Buffer to prepare a magnetic beads suspension with concentration of 5 mg/mL.
3. Add 0.2 mL biotinylated DNA/RNA in distilled water.
4. Incubate the magnetic beads with biotinylated nucleic acids for 15 mins at room temperature with gentle rotation.
5. Separate the magnetic beads with a magnet until the supernatant is clear.
6. Wash the magnetic beads 3 times with Washing & Binding Buffer.
7. Resuspend the magnetic beads with the desired concentration for downstream applications.

Releasing immobilized biotinylated molecules

The biotin-streptavidin bond can be broken by harsh conditions. 5 min incubation at 65 °C or 2 min at 90 °C in 10 mM EDTA pH 8.2 with 95% formamide will typically dissociate > 96% of immobilized biotinylated DNA. Alternatively, boil the sample for 5 min in 0.1% SDS for protein dissociation. Please note that proteins will be denatured by such treatment and the streptavidin conjugated cannot be re-used.

45-1 Ramsey Road Shirley, NY 11967, USA
Tel: 1-631-624-4882 Fax: 1-631-938-8221
E-mail: info@creative-diagnostics.com
www.cd-bioparticles.com