Sulfhydryl-Terminated Magnetic Beads

A sulfhydryl is a functional group that contains a sulfur atom linked to a hydrogen atom. In chemistry, the sulfhydryl group, often known as a thiol, is denoted by the suffix "-thiol" and the prefix "mercapto-" or "sulfanyl." Thiols have a strong preference for soft metals.

BcMag™ Sulfhydryl-Terminated Magnetic Beads are uniform, silica-based superparamagnetic beads grafted with a high density of sulfhydryl (SH) functional groups on the surface (Fig.1). The Sulfhydryl-Magnetic Beads are specially designed for conjugation of protein/peptides modified by SMCC (Sulfosuccinimidyl 4-[*N*-maleimidomethyl]cyclohexane-1-carboxylate). SMCC contains an amine-reactive *N*-hydroxysuccinimide (NHS ester) and a sulfhydryl-reactive maleimide group. NHS esters react with primary amines at pH 7-9 to form stable amide bonds. Maleimides react with sulfhydryl groups at pH 6.5-7.5 to form stable thioether bonds. The procedure is quick and straightforward for sample preparation without laborious repeat pipetting and centrifuging. BcMag™ Sulfhydryl-Terminated Magnetic Beads are ideal for conjugating large proteins. BcMag™ Long-Arm Sulfhydryl-Terminated Magnetic Beads are recommended for conjugating small peptides.

Fig.1 Structure of Sulfhydryl-Terminated Magnetic Beads

Workflow

BcMag™ Sulfhydryl-Terminated Magnetic Beads work perfectly as solid resin for various affinity purifications to refine molecules, cells, and parts of cells into purified fractions. After conjugation with ligands, add the beads to a solution containing the target molecules, then mix, incubate, wash and elute the target molecules (Fig.2)

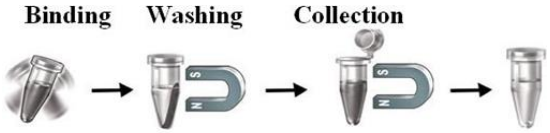

Fig.2

Specification		
Composition	Magnetic beads are grafted with a Sulfhydryl group on the surface.	
Number of Beads	~ 1.68 x 10^9 beads/mg (1µm beads) ~ 5x 10^7 beads /mg (5µm beads)	
Magnetization	~40-45 EMU/g	
Type of Magnetization	Superparamagnetic	
Effective Density	2.5 g/ml	
Formulation	Lyophilized Powder	
Ligand Density	1µm Magnetic Beads	~250 µmole / g of Beads
	5µm Magnetic Beads	~200 µmole / g of Beads
	1µm Long-Arm Magnetic Beads	~220 µmole / g of Beads
	5µm Long-Arm Magnetic Beads	~185 µmole / g of Beads
Storage	Upon receipt, store at 4°C	

Protocol

Materials Required:

- Conjugation buffer: PBS, PH 7.2

Phone: +1-858-909-0079 **Bioclone Inc**. Fax: +1-858-909-0057

- SMCC (Sulfosuccinimidyl 4-[*N*-maleimidomethyl]cyclohexane-1-carboxylate (Pierce, cat# 22360 22122) or Sulfo-SMCC (Pierce, cat# 22122)
- Magnetic rack (for manual operation): Based on sample volume, the user can choose one of the following magnetic Racks: BcMag rack-2 for holding two individual 1.5 ml centrifuge tubes (Cat. # MS-01); BcMag rack-6 for holding six individual 1.5 ml centrifuge tubes (Cat. # MS-02); BcMag rack-24 for holding twenty-four individual 1.5-2.0 ml centrifuge tubes (Cat. # MS-03); BcMag rack-50 for holding one 50 ml centrifuge tube, one 15 ml centrifuge tube, and four individual 1.5 ml centrifuge tubes (Cat. # MS-04); BcMag™ rack-96 for holding a 96 ELISA plate or PCR plate (Cat. # MS-05).

Protein /Peptide Preparation:

1. Dialyse the protein/Peptides against 50 volumes of PBS, pH 7.2, 5mM EDTA.
2. Add the appropriate amount of SMCC to the protein solution and mix very well.
   Notes: Prepare SMCC solution (3.7mg/ml in DMF. If the SMCC does not completely dissolve, place the tube in a 50°C water bath for several minutes.) Add 100 ul of SMCC solution to 1 ml protein solution (10mg/ml). or 50 ul of SMCC solution to 1 ml protein solution (<1mg/ml).
3. Incubate at + 4°C for 2 hours or room temperature for 30 minutes.
4. Remove free SMCC and unmodified proteins on a Sephadex G15 column. Elute with PBS buffer. The elution of the protein-SMCC can be monitored at 206 nm with a spectrophotometer.

Magnetic Beads Preparation:

Note: Weight, suspend the magnetic beads with PBS (Concentration: 30mg/ml), disperse the beads by vigorous vortexing, and store at 4˚C. Shake the bottle to resuspend the Magnetic Beads before use *completely*.

1. Shake the bottle to resuspend the BcMag™ Sulfhydryl-Terminated- Magnetic Beads thoroughly.
2. Transfer 100µl-300 µl of the Beads (30 mg/ml) to a tube. Place the tube on the magnetic rack for 1-3 minutes. Remove the supernatant while the tube remains on the rack.
3. Remove the tube and resuspend the beads thoroughly with 200µl PBS buffer. Leave the tube at room temperature for 2-3 minutes. Place the tube on the magnetic rack for 1-3 minutes. Remove the supernatant while the tube remains on the rack.
4. Repeat step 3 two times.
5. Incubate the Magnetic Beads with 1 ml of DTT (dithiothreitol 3 mg/ml) for 15 minutes at room temperature and wash the Beads three times with PBS buffer.
6. Mix the reduced beads with the protein-SMCC conjugate, and incubate for 12 hours at +4°C.
7. Wash the beads and resuspend them in the desired buffer.

Related Products	
Amine-Terminated Magnetic Beads	Iodoacetyl-Activated Magnetic Beads
DADPA-Activated Magnetic Beads	Peptide conjugation buffer Kit-I
Carboxyl-Terminated Magnetic Beads	Peptide conjugation buffer Kit-II
Epoxy-Activated Magnetic Beads	DVS-Activated Magnetic Beads
Hydrazide-Terminated Magnetic Beads	NHS-Activated Magnetic Beads
Glycoprotein and Antibody Conjugation Kit-I	Hydroxyl-Terminated Magnetic Beads
Glycoprotein and Antibody Conjugation Kit-II	Sulfhydryl-Terminated Magnetic Beads
Aldehyde-Activated Magnetic Beads	Tosyl-Activated Magnetic Beads
Silica-Modified Magnetic Beads	CDI-Activated Magnetic Beads
Alkyne-Activated Magnetic Beads	Thiol-Activated Magnetic Beads
Azide-Activated Magnetic Beads	Cleavable NHS-Activated Magnetic Beads
Cleavable Amine-Terminated Magnetic Beads	Cleavable Azide-Activated Magnetic Beads
Cleavable Carboxyl-Terminated Magnetic Beads	Cleavable Alkyne-Activated Magnetic Beads
Cleavable Epoxy-Activated Magnetic Beads	Cleavable Iodoacetyl-Activated Magnetic Beads
Cleavable Hydrazide-Terminated Magnetic Beads	Cleavable Tosyl-Activated-Magnetic Beads
Cleavable Aldehyde-Activated Magnetic Beads	Streptavidin Magnetic Beads
Boronate Affinity Magnetic Beads	Cleavable Streptavidin Magnetic Beads
Monomer Avidin Magnetic Beads	