INSTRUCTIONS

Carboxyl Magnetic Beads

Catalog No.: FE03001 FE03002

AvanBio Inc.
239 New Road, Suite A-107
Parsippany, NJ 07054
U.S.A.

Phone: +1 (800) 278-9865
Fax: +1 (855) 378-6845
Email: support@avanbio.com

【Introduction】

EmerTher® Carboxyl Magnetic Beads are nano-superparamagnetic beads coated with carboxylic acid functional groups, which can be activated (e.g. using EDC and NHS/sulfo-NHS) and then reacts with amines on nucleic acid, protein, antibodies or other molecules to form stable amide linkages. After that, the beads can be separated from the solution using magnet for downstream experiments, such as enzymatic reactions or immunopurification of other large molecules.

【Product Specifications】

Diameter： 500 nm

Magnetic response rate: >30 emu/g

pH stability： 4-12

Solvent： 20% ethanol

Binding capacity: 10-100 μg protein per mg carboxyl magnetic beads

【Product Content】

Catalog Number	Conc. (mg/ml)	Volume (ml)	Amount of Beads (mg)
FE03001	20	1	20
FE03002	100	1	100

【Storage】

Shipped at room temperature. Stored at 2-8°C, 2 years

【Coupling Protocol】

The following protocol provides general guidelines for coupling of proteins or antibodies to EmerTher carboxyl magnetic beads. The protocol is scalable. Optimization of the coupling conditions (protein concentration, bead concentration, coupling buffer, pH, and incubation time) for the ligand of interest is recommended. Other general activation protocols are also applicable.

A. Notes:

1. Amine-containing buffers (e.g., Tris and glycine) inhibit coupling of protein to the magnetic beads. If protein is dissolved in a buffer containing primary amine, remove the interference in the buffer using dialysis or desalting.
2. The optimal protein concentration should be optimized. Too low a protein concentration may result in bead crosslinking. For coupling of expensive antibodies that may not be available in enough quantity to reach the desired concentrations, another protein (i.e., BSA) may be added to take up remaining reactive sites.
3. Do not freeze, dry, or centrifuge at a high speed during use or storage of beads. Otherwise, this may decrease the binding capacity of the beads.

B. **Additional materials required**:

1. Activation Buffer: 50 mM MES, pH 7.0
2. Coupling Buffer: 150 mM NaCl, 0.01% Tween-20, 50 mM MES, pH 7.0
3. Sulfo-NHS and EDC
4. Blocking Buffer: 150 mM NaCl, 0.01% Tween-20, 100 mM Tris-HCl, pH 7.0
5. Protein solution: Dissolve protein in 0.5 ml of Coupling Buffer. Protein concentration is typically 1-10 mg/mL.
6. A magnetic stand and a rotator

C. Two-step EDC/sulfo-NHS activation and coupling procedure:

Step 1: Activation (using EDC/sulfo-NHS activation as an example)

1. Gently mix the magnetic beads thoroughly before use by shaking.
2. Place 1 mg of magnetic beads into a microcentrifuge tube. The amount of beads used can be scaled up or down.
3. Place the tube into a magnetic stand, collect the beads and discard the supernatant.
4. Wash the beads three times with Activation Buffer (1 ml each time) by magnetic separation, and re-suspend the beads in 1 ml of Activation Buffer.
5. Add 5 mg of EDC and 5 mg of sulfo-NHS at the same time. Mix to dissolve. To facilitate faster dissolution, EDC and NHS may be dissolved immediately before use as a concentrated stock solution in Activation Buffer and then an aliquot of this solution is added to the bead suspension to obtain the correct final concentration.
6. Incubate for 15 min at room temperature.
7. Collect the beads with a magnet. Quickly wash the beads once with Coupling Buffer (1 ml) by magnetic separation, and then re-suspend the beads in 0.5 ml of Coupling Buffer.

Step 2. Coupling with protein

1. Add 0.5 ml of the protein solution to 0.5 ml of the bead suspension. Mix thoroughly and incubate for 2-4 hrs at room temperature or overnight at 4 °C on a rotator.
2. Collect the beads with a magnet and save the supernatant for analysis if needed.
3. Add 10 ml Blocking Buffer. Mix thoroughly and incubate for 2-4 hrs at room temperature or overnight at 4 °C on a rotator.
4. Collect the beads with a magnet and discard the supernatant.
5. Wash the protein-coupled beads twice (15 min each time) with Blocking Buffer (1 ml each time) by magnetic separation and discard the supernatant.
6. Suspend the beads with a desired volume of Blocking Buffer or in a buffer compatible with the attached protein. Store at 2-8º C until ready for use.

Find out more at AvanBio

This product is for research use only and is not for use in diagnostic procedures. Please visit our website at www.avanbio.com for MSDS information and more life science research products from AvanBio. For technical support, please email us: support@avanbio.com.