

# **Bioligand Rapid Conjugation Kit**

### Cat No: AKIT-M-20

### Description

The surface of the magnetic bead in the bioligand rapid conjugation kit is modified by special groups, which can form stable chemical bonds with proteins and other molecules with primary amine groups for affinity and purification of antibodies, antigens and other biomolecules. Compared with traditional carboxyl and amino magnetic beads, the biological ligand containing primary amino group is simply dissolved in the Coupling buffer or PBS provided with the kit, and the biological ligand can be covalentially coupled to the magnetic bead by mixing the biological ligand with the magnetic bead for 1~2 h at room temperature. It has the advantages of simple operation, mild coupling conditions and fast and efficient coupling of biological ligands. The magnetic bead coupling process must be carried out in a buffer solvent without any amino group. In manual operation, the magnetic separation frame is used to separate the magnetic bead from the solvent. It can also be operated with automated equipment, which is suitable for the screening of multiple samples.

For custom sizes, formulations or bulk quantities please contact our customer service department. Website: <u>www.abvigen.com</u> Phone: +1 929-202-3014 Email: <u>info@abvigenus.com</u>

#### Characteristics

#### 1. Kit Component

Biological ligands rapidly coupled magnetic beads	1 mL
Washing Buffer	5 mL
Coupling Buffer	5 mL
Blocking Buffer	5 mL
Storage Buffer	10 mL
Magnetic rack (double row 16 holes)	1



#### 2. Basic information about magnetic beads

Particle size	200 nm
Binding ability	$\geq$ 100 µg Rabbit IgG/mg magnetic bead
Concentration	10 mg/mL
Preservative solution	ddH <sub>2</sub> O
Storage condition	Stable storage at 2 ~ 8°C for one year

Note: The binding ability is related to the characteristics of the biological ligand itself, and the value here is only for reference.

### 3. Binding ability with different molecular weight proteins

Protein	Molecular weight (kDa)	Magnetic bead bonding ability ( $\mu$ g/mg of bead)
lgG	150	100
Streptavidin	53	50
Protein A/G	50	45
Protein G	29	60

Note: The amount of binding is affected by the number of active primary and secondary amine groups in the protein.

# **Operation Process**

# Coupling method of magnetic beads and biological ligands

The following procedure is described by taking a magnetic bead sample of 500  $\mu$ L and using a 2 mL EP tube. Users can adjust according to their own needs:

# 1. Preparation of ligand solution:

Appropriate amount of ligand to be coupled is dissolved by Coupling Buffer and prepared into a ligand solution with a concentration of  $0.5 \sim 5.0$  mg/mL. Ligands that have been stored in buffers need to be completely removed from the original buffers containing primary amine groups by means of dialysis or desalting. Then Coupling Buffer can be used to prepare ligand solutions with a concentration of  $0.5 \sim 5.0$  mg/mL, and the prepared ligand solutions can be stored at 4°C for further use.

**Note:** (1) In order to achieve better performance, when the ligand concentration is  $\geq$  2 mg/mL, the coupling efficiency will be higher, but it should be comprehensively considered according to the cost and use requirements;



(2) The ligand solution cannot contain components with primary amino groups, such as Tris, glycine, gelatin, BSA, etc.;

**2. Magnetic bead cleaning:** After mixing the magnetic bead, take 500  $\mu$ L magnetic bead into 2 mL EP tube, magnetic separation to remove the supernatant, wash it twice with 1 mL Washing Buffer, and magnetic separation to remove the supernatant;

**3. Biological ligand coupling:** Add 500  $\mu$ L ligand solution to the EP tube equipped with magnetic beads (appropriate dosage and concentration need to be optimized according to the specific experiment), gently mix, coupling at 25°C for 1 ~ 2 h, or place the coupling at 4°C overnight, keep the magnetic beads suspended during the coupling (can be inverted mixing by vertical mixer);

**4. Magnetic bead sealing:** Put the EP tube on the magnetic rack to magnetically separate and remove the superliquid, add 1mL Blocking Buffer to re-suspend the magnetic bead, close it at 25°C for 2 h, or place it at 4°C for overnight, keep the magnetic bead suspended during the closure (can be inverted mixing by vertical mixer);

**5. Preservation:** The EP tube was placed on the magnetic rack for magnetic separation to remove the supernatant, washed with 1 mL Storage Buffer for 3 times, and then re-suspended in 500 µL Storage Buffer. The concentration of magnetic beads after coupling ligand was finally 10 mg/mL, and finally stored at 4°C. The amount of preservation solution can be determined according to the need to adjust the concentration of the coupled ligand magnetic beads, and 0.05% sodium azide or 0.1% proclin-300 can be added to the preservation solution to inhibit bacterial growth if necessary.

#### Notes

1. Magnetic beads are stored in  $ddH_2O$ , freezing, drying and centrifugation will cause the agglomeration of magnetic beads, which is not easy to be re-suspended and dispersed, and affect the chemical activity of functional groups on the surface of magnetic beads.

2. Before using this product, be sure to fully oscillate or ultrasonic to keep the magnetic beads in uniform suspension.

3. The buffer containing substances with primary amine will inhibit the protein coupling to the surface of the magnetic bead, and the removal of primary amine substances can be used by dialysis and desalination.

4. In order to ensure the best experimental results, please select the appropriate ligand for covalent coupling reaction.

5. This product is for scientific research only.

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**Ordering Information** 

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