

# **Combination Biotinylated Molecular Operation Process**

# 1. Preparation

- 1.1 Buffer: the following is the commonly used buffer composition, users can adjust the salt concentration of buffer and pH according to the need.
- 1.2 Buffer I (Suitable for binding biotinylated nucleic acids): 10 mM Tris-HCl (pH 7.5), 1 mM EDTA, 1 M NaCl,  $0.01\%^{\circ}0.1\%$  Tween-20.
- 1.3 Buffer II (Suitable for binding biotinylated antibodies/proteins): PBS, pH 7.4, contain 0.05% Tween-20, add 0.01%~0.1% BSA according to the need.
- 1.4 Magnetic separator.
- 1.5 Vortex generator.
- 1.6 Rotating mixer.
- 1.7 Pipette and suction head.
- 1.8 Suitable centrifugal tubes.

### 2. The combination of biological nucleic acid

2.1 Put the magnetic bead bottle on the vortex oscillator for 20 s, and the oscillating magnetic beads are suspended. With a pipette to remove 100  $\mu$ L beads to the new centrifuge tube. Put the centrifuge tube on a magnetic separator and placed for 1 min (hereinafter referred to as magnetic separation). Use a pipette to suck out the supernatant and remove the centrifuge tube from the magnetic separator.

Note: according to the number of biotinylated molecules and the amount of magnetic beads in the product information table, user can calculate the amount of magnetic beads to be used. It is suggested that the amount of biotinylated molecules is 1~2 times of magnetic beads, so that the magnetic beads are saturated.

2.2 Add 1 mL Buffer I to the centrifuge tube, cover the centrifuge tube cover, fully shake the suspended magnetic beads. Then magnetic separation, and remove supernatant.

Note: when step 2.1 takes the volume of magnetic beads larger than 1 mL, add Buffer I with the same size as the magnetic beads.

2.3 Repeat step 2.2 once.

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- 2.4 Adding 500  $\mu$ L diluted with Buffer I biotinylated nucleic acid (the magnetic beads concentration of 2 mg/mL), fully oscillating and resuspend magnetic. Put the centrifuge tube on a rotating mixer and rotated at room temperature for 30 min.
- 2.5 Magnetic separation, transfer the supernatant to a new centrifuge tube.
- 2.6 Washing magnetic beads three times according to "step 2.2" method.
- 2.7 According to the requirements of subsequent experiments, add with appropriate low salt buffer to resuspend magnetic beads. At this point, the biotin nucleic acid step is completed. Magnetic beads can be used for subsequent operations.
- 2.8 Users can determine the concentration of nucleic acid before and after reaction, then calculate amount of the nucleic acid binding to the beads, ((before the reaction concentration after the reaction concentration) \* the reaction solution volume).

#### 3. Combination of biotinylated antibody/protein manipulation process

3.1 Put the magnetic bead bottle on the vortex oscillator for 20 s, and oscillating and suspended magnetic beads. Use a pipette to remove 100  $\mu$ L magnetic beads into a new centrifuge tube. Magnetic separation, use a pipette to suck out the supernatant then remove the centrifuge tube from the magnetic separator.

Note: according to the number of biotinylated molecules and the amount of magnetic beads in the product information table, user can calculate the amount of magnetic beads to be used. It is suggested that the amount of biotinylated molecules is 1~2 times of magnetic beads, so that the magnetic beads are saturated.

3.2 Add 1 mL Buffer II to the centrifuge tube, cover the centrifuge tube cover, fully shake the suspended magnetic beads. Magnetic separation, then remove supernatant.

Note: when step 3.1 takes the volume of magnetic beads larger than 1 mL, add Buffer II with the same size as the magnetic beads.

- 3.3 Repeat step 3.2 twice, washing three times in total.
- 3.4 Adding 1 mL diluted with Buffer II biotinylated antibody/protein (the magnetic beads concentration of 1 mg/mL), fully oscillating and resuspend magnetic. Put the centrifuge tube on a rotating mixer and rotated at room temperature for 30 min.
- 3.5 Magnetic separation, transfer the supernatant to a new centrifuge tube.
- 3.6 Washing magnetic beads five times according to "step3.2" method.

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3.7 According to the requirements of subsequent experiments, add with Buffer II or other appropriate buffer to resuspend magnetic beads. At this point, the biotinylated antibody/protein step is completed. Magnetic beads can be used for subsequent operations.

# **Product Application Scope**

Legend	Application direction	Sketch
(	Immunoassay,	Streptavidin can specifically bind
	separation of protein,	biotinylated antibody or antigen, as
	cell sorting, etc.	immune detection, ELISA solid-phase
		reaction carrier, or used for sorting cells.
10000E	Isolated nucleic acid,	Streptavidin can specifically combine
Carried States	Preparation of Nucleic	biological nucleic acid probe in the
	acid probes.	hybridization experiments that widely
		used in DNA, RNA.
Va. BAARDAS	DNA-Study on protein	Streptavidin specifically targets with
	interaction protein.	biotinylated DNA or RNA fragments can
		be used to study the interaction
		between proteins and nucleic acids.
→ ↑		man bearing have
SA Nanotin Antibody	Antigen Complementary I nucleic acid chain	Nucleic acid DNA binding Labelled probe protein antibody

Note: the application directions listed above have many forms of implementation, not limited to illustrations.

#### Note

- 1. Avoid freezing magnetic beads and other operations.
- 2. In order to reduce the loss of magnetic beads, the time of magnetic separation should be no less than 1 min.
- 3. The magnetic beads should be fully shake and suspended evenly before the magnetic beads are removed from the magnetic storage tube. Bubbles should be avoided during operation.

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- 4. It is recommended to use a good pipette suction head and a reaction tube to avoid losses due to adhesion of magnetic beads and solution.
- 5. The size of biotinylated molecules affects the magnetic bead loading. Users need to determine the load of magnetic beads to specific biotinylated molecules according to the experiment.
- 6. The amount of biotinylated molecules should be 1~2 times of the magnetic beads, in order to saturate the magnetic beads.
- 7. This product is for research use only.

# **Ordering Information**

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