# **Biotin Conjugation Kit**

#### Cat No: BIOK1960

### Description

Biotin-Avidin-System (BAS) is a very widely used biological reaction amplification system. The interaction between biotin and avidin is currently known to be the strongest non-covalent interaction. The high-affinity and firm binding between biotin and avidin and the multi-stage amplification effect make BAS immunolabeling and related tracers The analysis is more sensitive. Biotin-labeled antibodies are currently widely used immunochemical reagents that combine specificity, sensitivity, qualitative and localization in immunopathology, cytochemistry, pathology and clinical immunity. Under alkaline conditions, the activated biotin binds to the free amino groups of the protein to produce labeled biotinylated protein.

The fluorescein in this kit uses high-quality imported biotin (Biotin). The principle of biotin-labeled antibody is to use -NHS group on biotin and free -NH2 on the antibody to chemically form Biotin-antibody conjugate. There are 86 lysine residues in IgG molecule, and generally can bind 15-20 at most, and IgG molecule can bind 2-8 molecules of Biotin.

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# **PRODUCT INFORMATION**

Biotin	1 vial (Protect from light)
Buffer	50ul
DMSO	150ul
Purification column	2 vial
Collection tube	2 vial



## Steps

- 1. Antibody preparation
- 1.1. The recommended antibody concentration is between 1-2mg/ml.
- 1.2. Do not contain BSA or other protein components in the antibody.
- 1.3. The antibody buffer should not contain amino salts (such as Tris, NaN3,
- etc.), and the pH should be 6.5-8.5.
- 2. Antibody labeling
- 2.1. Take out the Biotin and centrifuge for a few seconds, and shake the 北京百欧泰生物科技有限公司 Tel: +86 136 8125 6816 Email: biotyscience@163.com Address: 北京市房山区良乡凯旋大街建设路 18 号-D3032

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Biotin powder in the tube to the bottom of the tube;

2.2. Add 50ul DMSO to the tube and pipette repeatedly or vortex until the Biotin is completely dissolved;

2.3. Add an appropriate amount of adjustment buffer to the antibody (add10ul adjustment buffer to every 100ul antibody);

2.4. Add the dissolved Biotin to the antibody (add 4.0ul Biotin to every 100ug of antibody), pipette repeatedly or vortex to mix.

2.5. Place the antibody-Biotin mixture on a horizontal shaker or a rotating mixer, and react for 1 hour at room temperature while shaking (the reaction tube can be wrapped in tin foil).

Note: If a higher Biotin coupling ratio is required, the coupling time of antibody and Biotin can be extended appropriately.

3. Free Biotin removal

3.1. Take out the purification column and centrifuge it at 3000 rpm for 2 minutes.

3.2. Temporarily keep the buffer in the collection tube.

3.3. Move the purification column to a new collection tube, suck the antibody-Biotin conjugate and place it on the surface of the packing in the purification column. If the sample volume is less than 50ul, you should first make up the liquid volume to 50ul with the buffer reserved in 3.2. The maximum sample volume should not exceed 100ul.

3.4. After the sample infiltrates the packing, centrifuge at 3000rpm for 2min.

3.5. Store the antibody-Biotin marker at 4 ° C until use. The final product can

be stored at 4 ° C for 1 year.

NOTES

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1. Store the kit at 2-8°C, do not freeze.

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The components of the kit may be upside down during transportation, causing liquid or dry powder reagents to stick to the tube wall or bottle cap.
Please centrifuge before use to make the liquid or dry powder reagent attached to the tube wall or bottle cap settle to the bottom of the tube.
Biotin needs to be prepared for immediate use, and the dissolved Biotin cannot be stored for a long time.

4. The buffer in the purification column contains the toxic component sodium azide (NaN3). Avoid contact with skin, eyes and mucous membranes when using it.

5. DMSO is slightly toxic, permeable to human skin and irritating to the eyes.
Avoid contact with skin, eyes and mucous membranes when using it.
6. The dialysis, concentration and concentration determination of the antibody before labeling will all cause the loss of the amount of antibody.
Therefore, when preparing the antibody before labeling, consider the most appropriate amount of antibody according to the specific situation.

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