

## NHS-LC-LC-Biotin Kit

### Product Description:

NHS-LC-LC-Biotin is a double-long-chain, NHS (N-hydroxysuccinimide) ester-activated, non-sulfonated biotinylation reagent for labeling primary amines ( $-NH_2$ ), such as protein lysine, that enables simple and efficient biotinylation of antibodies, proteins, and any other primary amine-containing biomolecules in solution for detection or purification using streptavidin probes or resins.

The NHS-LC-LC-Biotin Kit is a biotin labeling kit developed based on NHS-LC-LC-Biotin, which provides a complete set of reagents for biotin labeling. The biotin contained in the kit has been activated and ready for immediate use, with up to 1 mg of antibody labeled at a time. The kit also includes buffer, ultrafiltration tube, protective solution, antibodies do not need dialysis, easy to operate, and the complete process can be completed in only 90 minutes.

This kit can be used for biotin labeling of antibodies or proteins, and the labeled antibodies are tested for most ELISA applications and some IHC applications. Compared with other biotin labeling kits, the long-arm biotin kit labeled antibody has a higher signal-to-noise ratio in ELISA applications.

### Composition and storage conditions

Components	K4401-2 T	K4401-5 T
NHS-LC-LC-Biotin	2X 0.1 mg	5X 0.1 mg
Labeling Buffer	15 mL	30 mL
DMF	200 $\mu$ L	200 $\mu$ L
Ultrafiltration tube	4X 0.5 mL	10X 0.5 mL
Antibody Stabilization Solution	1 mL	2 mL
Store the kit unopened at 2-8°C for 1 year, and the dissolved biotin can be stored at 2-8°C for 1 week.		

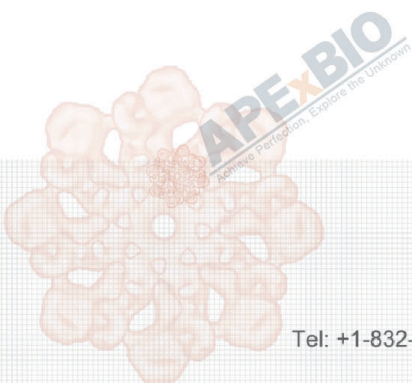
### Experimental manipulation

- Each vial (0.1 mg) of biotin lyophilized powder (NHS-LC-LC-Biotin) was dissolved in 10  $\mu$ L of DMF to obtain a concentration of 10 mg/mL biotin.

2. Add the antibody to be labeled to an Ultrafiltration tube and add an appropriate volume of Labeling Buffer to a total volume of 0.5 mL and centrifuge at 12,000×g for 10 minutes. *[Note 1] The maximum volume of the Ultrafiltration tube is 0.5 mL; [Note 2] When the concentration of the antibody to be labeled is low, the ultrafiltration can be centrifuged once, and then an appropriate amount of Labeling Buffer can be added to the Ultrafiltration tube to make the final concentration of the antibody to be labeled 2 mg/mL.*
3. Add an appropriate amount of dissolved biotin (the recommended mass ratio of antibody to biotin is 1:10, such as 0.5 mL of 2 mg/mL antibody (MW 150,000), add 10 mg/mL of biotin 10 uL), gently pipette and mix well, and incubate in a 37°C incubator for 30-60 minutes in the dark
4. Optionally, if you want to increase the efficiency of labeling, you can mix well with a gun every 10 minutes during the previous incubation.
5. After the end of incubation, centrifuge at 12,000×g for 10 min.
6. Washing: Add an appropriate amount of Labeling Buffer to the above Ultrafiltration tubes to make a final volume of 0.5 mL, mix by gentle pipetting, and centrifuge at 12,000×g for 10 minutes. Repeat this one more time.
7. Recovery: After centrifugation, add 0.2 mL of Labeling Buffer to the above Ultrafiltration tube, gently pipette, invert the filter cartridge and place it in another clean centrifuge tube, and centrifuge at 6,000×g for 10 minutes.
8. Volumetric: Collect the solution in the centrifuge tube, i.e., the biotinylated antibody. According to the need, add Labeling Buffer, adjust the concentration, and then add the same volume of Antibody Stabilization Solution and store at -20°C.

## Precautions

1. The kit can also label other antigens, HRP, and peptides containing amino groups, and the specific labeling ratio depends on the number of amino groups in the substance to be labeled. If a protein is labeled, it is recommended to adjust the protein concentration to 2 mg/mL and add it at a 1:20 ratio of protein: biotin.
2. DMF needs to be sealed and stored dry.
3. The Ultrafiltration tube provided with this kit is 10 KD, and the molecular weight of the labeled antigen or peptide is 20 KD and above.
4. If the protein is labeled, it is recommended to adjust the protein concentration to 2 mg/mL, at which time the molecular ratio of protein: biotin is 1: 20.
5. Before labeling, calculate the amount of biotin according to the amount of antibody to be labeled.
6. This product is for scientific research purposes only.



## **APExBIO Technology**

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