

(FOR RESEARCH USE ONLY. DO NOT USE IT IN CLINICAL DIAGNOSIS!)

# Allophycocyanin(APC) Antibody Quick Labeling Kit

Catalog No.: RE80006

Size: 0.5mg

If you have any questions or need further help during experiment, please don't hesitate to contact us through the following methods:

Tel: 0086-27-87002838

Website: www.enkilife.com

**Shelf life:** Please refer to the label on the outer package.

**Techsupport:** In order to provide you with better service, please inform us the lot number on the label of the outer package.

#### **Product Introduction**

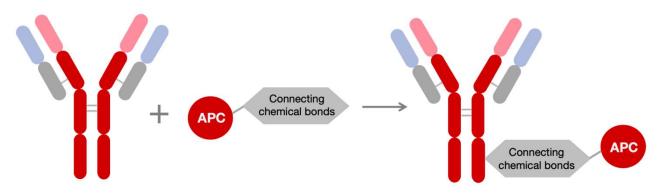
Allophycocyanin (APC) is a high-brightness phycobiliprotein purified from Spirulina sp., a blue-green alga, with a molecular weight of 104kDa. It is a far-red fluorescent protein with a high quantum yield. It emits light when excited by laser at 594 nm and 633 nm, with the maximum absorption peak at 650 nm and the maximum fluorescence emission peak at 660 nm. APC is 100 times stronger than ordinary chemiluminescence. In ordinary applications, it can at least increase sensitivity by 5-10 times. It is mainly used in flow cytometry, microarray analysis, ELISAs, etc.

### **Product Features**

- •The kit has complete components and is easy to operate. High-quality APC-conjugated antibodies can be obtained by following the operating steps.
- •The APC provided in this kit is a high-purity, high-activity fluorescent protein with higher fluorescence efficiency.
- •The kit uses directional coupling technology, and neither the fluorescent protein nor the antibody will self-couple, ensuring the specificity and uniformity of the conjugate.
- •The cross-linker used in the kit has an extended arm, ensuring that the luminous efficiency is not affected by protein steric hindrance.
- -Due to the protein skeleton characteristics of APC, it has stronger anti-quenching capabilities.

## **Labeling Principle**

This kit uses the free amino groups on the antibody and APC to covalently couple the antibody molecules with APC using directional docking coupling technology.



## Components

Components	Size
Activated APC Protein	0.75mg, can be used to label 0.5mg of antibody
Labeling Buffer	30ml
Antibody Modification Reagent	30ul * 1 vial (store at -20°C)
30KMWCO Ultrafiltration Tube	2 pieces
Blocking Reagent	50ug * 1 vial, mix with 5ul DMSO before use
DMSO	200ul*1vial
Instruction Manual	1 piece

# Storage

The antibody modification reagents in the kit can be stored at -20°C, and the remaining components can be stored at 2-8°C for 6 months.

# **Operation process**

- 1. Antibody modification
- 1.1. Take the antibody to be labeled (purity>90%), adjust the concentration to about 5mg/ml, add 2µl antibody modification reagent solution per mg of antibody, mix gently, and react at room temperature in the dark for 90 minutes.
- 1.2. After the reaction is completed, transfer it to a 30KMWCO ultrafiltration tube and ultrafilter it with labeling buffer 3-5 times (12000rpm 5min each time). The volume of the liquid in the tube core each time should not exceed 1/4 of the original volume. After the last ultrafiltration is completed, the liquid in the tube core is the modified antibody.

- 2. Activated APC protein and antibody coupling
- 2.1. Redissolve the activated APC protein with labeling buffer and adjust the concentration to 5mg/ml (0.75mg activated APC protein is re-dissolved in 150ul labeling buffer).
- 2.2. Mix the modified antibody and activated APC protein in a mass ratio of 1:1.5 (1.5mg activated APC protein for every mg modified antibody) and react at room temperature in the dark for 2 hours.
- 2.3. Add 5ul DMSO to the blocking reagent tube to prepare a 10mg/ml blocking reagent solution; add 5ul of each 0.5mg antibody to the reaction product of step "2.2" to block the unreacted active groups.
- 2.4. Divide the labeled antibodies, add appropriate protective agents, and store at -20  $^{\circ}$ C for future use.

#### **Notes**

- •The antibody modification reagent and blocking reagent in this kit are highly active reagents, please store at -20°C; the rest of the components should be stored at 2-8°C, DO NOT freeze.
- •The components of the kit may be turned upside down during transportation, which may cause the liquid or dry powder reagent to stick to the tube wall or bottle cap. Please centrifuge before use to allow the liquid or dry powder reagent attached to the tube wall or bottle cap to settle to the bottom of the tube.
- •The activated APC protein in this kit should be stored at low temperature away from light. Once reconstituted, please use it all at once; try to keep it away from light during the labeling process.
- •The blocking agent needs to be prepared before use, and the dry powder cannot be stored for a long time after dissolution.
- •When using this kit to label antibodies, the specificity of the antibodies should be high, with a purity of not less than 90%. It is best to use monoclonal antibodies, and the solution environment should not contain free amino groups, preferably PBS solution; NaN3 and BSA must be removed from the antibody before labeling. Operations such as dialysis, concentration, and concentration determination of antibodies can cause loss of antibody quantity, so it is necessary to consider the optimal amount of antibody when preparing antibodies before labeling.
- •Since the groups carried by the modified antibodies are easily reoxidized, the modified antibodies need to be coupled with activated APC as soon as possible.
- •The ultrafiltration tubes in this kit are specially treated to effectively avoid fluorescence quenching and protein adsorption loss, and ultrafiltration tubes on the market may affect the labeling efficiency.
- •Some reagents in this kit are highly active and have certain hazards to the skin and body, Please wear gloves throughout the operation; DMSO is slightly toxic, permeable to human skin, and irritating to the eyes. Avoid contact with skin, eyes and mucous membranes when using.