

Cy5-Antibody Conjugation Kit

Cat No: BIOK1964

Description

Immunofluorescence technique is to label a known antibody or antigen molecule with fluorescein. When it reacts with the corresponding antigen or antibody, there is a certain amount of fluorescein on the formed complex. Below you can see the fluorescent antigen-antibody binding site and detect the antigen or antibody. The main features of this technology are: strong specificity, high sensitivity and fast speed.

Fluorescein-labeled antibody is abbreviated as fluorescent antibody (FA), which is currently widely used in immunopathology, cytochemistry, flow cytometry, virology and autoantibody clinical immunodiagnosis of specific, sensitive, qualitative and localized immune Chemical reagents. Antibodies used for labeling require high specificity and high affinity. The antiserum used should not contain antibodies against normal tissues in the specimen. Generally, IgG and IgM need to be purified and extracted before labeling.

The fluorescein in this kit uses high-quality imported Cy5. The cyanine dye Cy5, SE is one of the few near-infrared fluorescent labeling dyes. It produces a low red fluorescence background, high fluorescence intensity, and good fluorescence stability. Cy5, SE are often used in biomolecule labeling, fluorescence imaging and other fluorescent biological analysis, such as labeling peptides, proteins and nucleic acids. Cy5, SE fluorescent labeling dye combined with protein can significantly enhance the fluorescence intensity.



PRODUCT INFORMATION

Cy5	50ul*
Buffer	50ul
纯化柱	2 支
收集管	2 支

注意*: Cy5 荧光素 1-2 周可以存放 2-8℃,长期-20℃可以保存半年,其他组份存放 2-8℃。



NOTES

1. Store the kit at $2-8^{\circ}$ C, do not freeze.

2. 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.

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

4. DMSO is slightly toxic, permeable to human skin and irritating to the eyes. Avoid contact with skin, eyes and mucous membranes when using it.

5. 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.

Steps

1. Antibody preparation

1.1. 建议抗体浓度在 1-2mg/ml 之间。

1.2. 抗体中不要含有 BSA 或其它蛋白质成分。

1.3. 抗体缓冲液中不要含有氨基的盐(如: Tris, NaN3等), pH 在 6.5-8.5 为 宜。

2. Antibody labeling

2.1. 抗体中加入适量调整缓冲液(每 100ul 抗体中加入 10ul 调整缓冲液);

2.2. 将荧光染料 Cy5 加入抗体中(按照每 100ug 抗体中加入 1.0-2ul Cy5),用移 液枪反复吹打或 vortex 混匀,缓慢加入。

2.3. 将抗体-Cy5 混合物置水平摇床或旋转混匀仪,在摇动状态下室温避光(反应管可包裹锡箔纸)反应 1h。

注: 如果需要较高 F/P 值,可适当延长抗体和 Cy5 偶联时间。

3. Free Cy5 removal

3.1. 取出纯化柱, 装入分子筛填料, 将纯化柱 3000rpm 离心 2min。

3.2. 暂时保留收集管中的缓冲液。

3.3. 将纯化柱移至新的收集管上,吸取抗体-Cy5 偶联物置纯化柱中填料的表面。 如果样品体积小于 50ul,应先用 3.2 保留的缓冲液将液体量补足 50ul。上样体 积最大不要超过 100ul。

3.4. 待样品渗入填料后,3000rpm 离心 2min。为保证去除效果,此操作可重复 1-2 次。

3.5. 通常将抗体-Cy5标记物 4℃避光保存待用,长期请在-20℃避光保存。偶联 抗体的最佳储存条件必须根据实验、抗体稳定性和最佳储存条件确定

3.6. 拆开的试剂盒荧光素在-20℃可避光保存1年。



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