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Technical Information

ICG NHS ester labeling kit

Overview

ICG NHS ester Labeling Kit is designed for effective antibody or protein labeling. ICG (Indocyanin green) dye is clinically approved near infrared (NIR) fluorescent dye. NIR fluorescence allows to observe the deep image from the surface of skin and being utilized in a wide range of research fields. The maxima of Ex/Em values are at 785/811nm. ICG might be excited using 750-800 nm laser line or LED and displays excellent optical property. NHS esters readily react with primary amine on antibody or protein by forming stable amide bond and, ipso facto, it is most popular and effective functional group incorporated into fluorescent reagent for protein or antibody labeling. ICG NHS ester labeling kit contains all required contents for antibody and protein labeling. We offer ICG NHS ester labeling kit as a convenient method which enable the labeling process to be easy.

Table 1. Components of ICG NHS ester Labeling Kit

Part No.	Contents	Amount	Storage
ILSM01	Component A, ICG NHS ester	6 µL	-20 ℃
ILSM02	Component B, Reaction buffer	500 µL	2-8 ℃
ILSM03	Component C, Wash buffer	2.5 mL	2-30 ℃
ILSM04	Component D, Filtration tube	3 tubes	

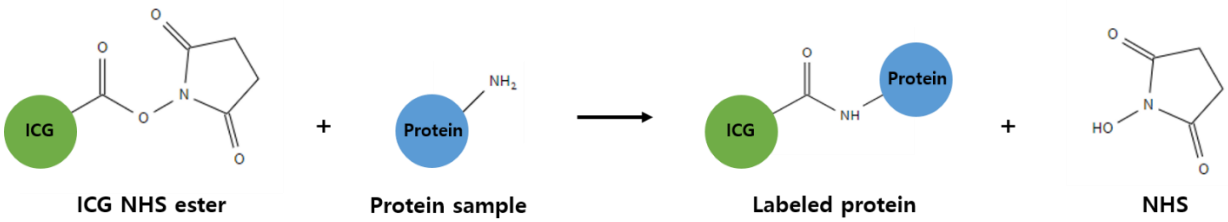


Figure 1. NHS ester reaction scheme for chemical conjugation to a primary amine.

Stable amide bond is formed and N- hydroxysuccinimide is released after the reaction.

Application

ICG NHS ester labeling kit enable detection of biomarkers. ICG-conjugated antibody can specifically visualize the target cells expressing the receptor. For example, peptides, proteins, and antibodies are cancer-specific ligands that form biological conjugation with ICG through ICG NHS ester labeling kit protocol. And ICG labeling kit can be used for labeling of FDA-approved monoclonal antibodies such as Panitubumab, Daclizumab, or Trastuzumab. The ICG-conjugated monoclonal antibodies are switch optical imaging probes

that activated only in target cells.

Before beginning of labeling Experiment

Materials and equipment required but not provided

- Micropipette
- 37 ℃ incubator
- Microcentrifuge
- Microtubes
- PBS or appropriate buffer

Experimental protocols

Labeling of protein or antibody with ICG NHS ester labeling kit

1. Add 100 µL of wash buffer and sample solution containing 50-200 µg protein.
2. Centrifuge at 8,000 rcf for 15 min.
3. Add 100 µL of reaction solution and 1.74 µL of ICG NHS ester.
4. Mix by pippeting and incubate at 37℃ for 10 min
5. Add 100 µL of wash buffer
6. Centrifuge at 8,000 rcf for 15 min.
7. Add 200 µL of wash buffer.
8. Centrifuge at 8,000 rcf for 15 min.
9. Repeat step 7 and 8. A_{800}
10. Resuspend with 200 µL of PBS or appropriate buffer.
11. Transfer the resultant to microtube and store at 4℃.

Filtered solution in the collection tube after centrifugation can be discarded.

Calculation of dye / protein ratio

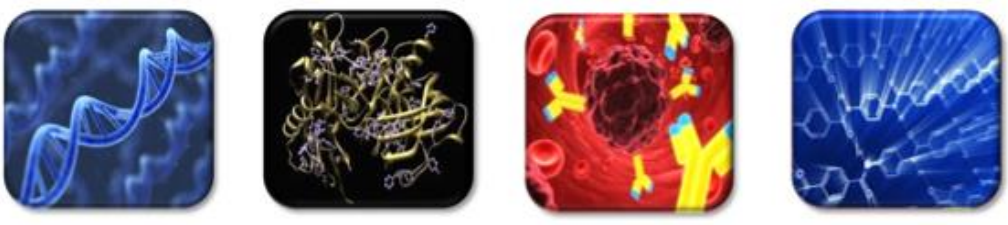
- Ratio (dye / protein) = $$\frac{A_{800}/147{,}000}{(A_{280}-A_{800}\text{x}0.075)/(\varepsilon \text{ of protein})}$$

A800: absorbance at 800 nm
A280: absorbance at 280 nm
ε: molar absorption coefficient of protein at 280 nm

- ε of IgG is 216,000
- ε of ICG in PBS is 147,000

Custom Labeling Service

Based on accumulated know-how and technologies, BioActs provide a wide range of custom services such as protein fluorescence labeling, organic synthesis, oligonucleotide synthesis upon customers' request. Our reliable technology has acknowledged by our clients from domestic and overseas universities, institutions, in vitro diagnostic and pharmaceutical companies and has enabled to steadily conduct their requirements. In addition, we can introduce fluorescent materials to many other compounds such as organic and inorganic compounds, drugs, hormones, polymer, peptides, proteins, antibodies, etc. We also can provide chemical and optical analytical data, along with cell and animal experiments.



Technical Support

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CoA (Certificate of Analysis) provides detailed quality information of each product. To see CoA, check the lot number written on each product's page at www.bioacts.com, when having trouble with check, contact to our technical support team

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