FITC IgG Labeling Kit

For labeling 1 x 10 mg IgG

REAGENT STORAGE

Kit Component	Storage Temp	Storage Notes
FITC Labeling Reagent	-20ºC	Keep the vial in the desiccated container as supplied in the kit
Quenching Reagent B	-20°C or 2-8ºC	Does not need to be kept desiccated.
100 mg/ml BSA (with 0.05% Sodium Azide)	-20°C or 2-8ºC	Does not need to be kept desiccated.

INTRODUCTION

The FITC-IgG Labeling Kit utilizes a novel chemistry to generate highly reproducible fluorescein-labeled IgG with a simple procedure.

FEATURES

- Pre-measured active labeling reagent.
- Can be used with up to 10 mg/ml (1%) BSA as a carrier protein.
- Completely scalable: conjugate anywhere from 0.1 to 1 gram IgG per reaction.
- Highly efficient FITC incorporation.
- Optimized FITC:IgG ratio by labeling time and temperature.
- Purification not usually required.

PRODUCTS and CONTENTS

Catalog Number	**CNJ93104-010**
Components	**Storage Conditions**
FITC Labeling Reagent for 10 mg IgG	1 X 500 µL
Quenching Reagent B	1000 µL
100 mg/ml BSA (with 0.05% Sodium Azide)	1000 µL

ADDITIONAL REAGENTS REQUIRED

None

SHELF LIFE

The performance of the product is guaranteed for a minimum of 12 months when stored as directed.

IgG Amount and Concentration and Buffers

The IgG to be labeled should be at a concentration 0.5 - 2.0 mg/ml in 1X PBS, pH 7.2 – 7.5. The IgG solution may contain up 10 mg/ml (1%) BSA.

CONJUGATION PROCEDURE for 10 mg of IgG

Note: Fluorescein is light sensitive. Perform reactions in the dark and keep light exposure to a minimum.

1. Let kit come to room temperature for at least 1 hour. Kit can be removed from -20ºC up to 24 hours before use.
2. Spin the screw-cap tube containing FITC Labeling Reagent for 1 minute at high speed (> 14,000 x g).
3. Add 10 mg IgG solution to the tube containing the FITC Labeling Reagent.
4. Vortex thoroughly for 10 seconds, then shake solution down to the bottom.
5. Incubate the labeling reaction at 37ºC for 60 minutes in a heat block.

   Note: Room temperature incubations will be successful. 15 minute – 4 hour incubation at 37°C will be successful.
6. Remove excess FITC from the reaction by size exclusion chromatography (see Recommended Accessories section for spin desalting columns). Then proceed to step 8 – quenching is not necessary.
7. Add Quenching Reagent to the tube (not necessary if excess FITC has been removed by size exclusion chromatography):
   - For 10 mg IgG, add 1000 µL per reaction.
8. Store FITC-labeled IgG in the dark at 2-8ºC.

   Optional: Add 100 mg/ml BSA solution to achieve the desired final concentration of BSA.

   Optional: Add glycerol to a final concentration of 40-50%.

RECOMMENDED ACCESSORIES

To remove excess FITC from the labeled IgG - Order from ThermoFisher:

Sample Size	Description	Cat #
2 – 12 µL	Zeba Spin Desalting Columns, Micro (75µL), 7K MWCO	89877, 89878
30 - 130 µL	Zeba Spin Desalting Columns, 0.5mL, 7K MWCO	89882, 89883
200 – 700 µL	Zeba Spin Desalting Columns, 2mL, 7K MWCO	89889, 89890
500 – 2000 µL	Zeba Spin Desalting Columns, 5mL, 7K MWCO	89891, 89892
700 – 4000 µL	Zeba Spin Desalting Columns, 10mL, 7K MWCO	89893, 89894

For concentrating IgG before labeling or for concentrating the final FITC-labeled IgG – Order from MilliporeSigma:

Sample Size	Description	Cat #
Up to 500 µL	Amicon Ultra-0.5 Centrifugal Filter Unit with Ultracel-50 membrane	Z740176
Up to 2 mL	Amicon Ultra-2 Centrifugal Filter Unit with Ultracel-50 membrane	UFC205024
Up to 4 mL	Amicon Ultra-4 Centrifugal Filter Unit with Ultracel-50 membrane	UFC805008
Up to 15 mL	Amicon Ultra-15 Centrifugal Filter Unit with Ultracel-50 membrane	Z648000