

Biotin Labeling Kit

For labeling 1 x 1 mg IgG

INTRODUCTION

The Biotin Labeling kit utilizes a novel chemistry to generate highly reproducible biotinylated IgG with a simple procedure.

FEATURES

- Room temperature-stable active biotinylation reagent.
- Pre-measured active biotinylation reagent coated onto a tube surface.
- Can be used with up to 10 mg/ml (1%) BSA as a carrier protein.
- Completely scalable: conjugate anywhere from 10 µg to 1 gram IgG per reaction.
- Highly efficient Biotin incorporation.
- Purification not usually required, but desalting columns are included to remove excess biotin.

PRODUCTS and CONTENTS

Catalog Number	Bio-Link-AF	Storage Conditions
Biotin Reagent for labeling 1.0 mg IgG	1 tube	2-8°C or room temperature - in container with desiccant
Quenching Reagent B	250 µL	2-8°C
100 mg/ml BSA (with 0.05% Sodium Azide)	250 µL	2-8°C

ADDITIONAL REAGENTS REQUIRED

1X Phosphate Buffered Saline, pH 7.2 – 7.4. (1X PBS)

REAGENT STORAGE

- Store vials containing the Biotin Reagent at 2-8°C or room temperature. Keep the vials in the white plastic jar containing desiccant packets.
- Store all other reagents and components at 2-8°C.

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 biotinylated 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 to 10 mg/ml BSA and up to 0.10% sodium azide.

CONJUGATION PROCEDURE for 1.0 mg of IgG

1. Add solution containing 1.0 mg of IgG to the screw-cap tube containing the Biotin Reagent.
2. Vortex thoroughly for 15 seconds, then shake or spin solution down to the bottom of the tube.
3. Incubate the reaction at room temperature for 15 minutes. Longer incubations (up to 4 hours) will also give acceptable results.
4. Optional: remove excess biotin from the reaction by size exclusion chromatography (see recommended accessories section). Then proceed to step 8 – the quenching step is not necessary.
5. Add Quenching Reagent B to the reaction tube (this step is not necessary if excess biotin has been removed by size exclusion chromatography):
 - Add 100 μ L Quenching Reagent B to the reaction mixture
6. Vortex gently for 5 seconds, then shake down to the bottom.
7. Incubate the reaction for 5 minutes at room temperature.
8. Optional: add 100 mg/ml BSA solution to achieve the desired final concentration of BSA.
9. Optional: Add glycerol to a final concentration of 40-50%.
10. Store biotinylated IgG at 2-8°C or -20°C.

RECOMMENDED ACCESSORIES

To remove unreacted biotin from the biotinylated 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 biotin labeling or for concentrating the final biotinylated 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