

Biocytin C2 maleimide

Catalog number: 3085

Unit size: 5 mg

Component	Storage	Amount (Cat No. 3085)
Biocytin C2 maleimide	Freeze (< -15 °C)	1 vial (5 mg)

OVERVIEW

Biocytin maleimide readily reacts with thiol moieties of biopolymers to form a relatively stable thioether conjugate. This biocytin maleimide requires mild conjugation conditions. For example, a pH of 5.5-8.5 is usually optimal for modifying cysteine residues, and air exposure of the reaction solution should be minimized whenever possible to avoid the air oxidation of thiol substrates. Most conjugations are done at room temperature. However, either elevated or reduced temperature may be required for a particular labeling reaction. Reactions with this biotinylation reagent should be performed in buffers free of extraneous thiols (such as 6-mercaptoethanol, dithiothreitol, and mercaptoethylamine). To be biotinylated by thiol-reactive reagents, proteins or peptides must have a free thiol group (SH) available.

SAMPLE EXPERIMENTAL PROTOCOL

Labeling Proteins with Biocytin C2 Malemide

- Dissolve your thiol-containing protein at concentration of 1 10 mg/mL (3 - 10 mg is the optimal labeling concentration) using PBS buffer (20 mM, pH 7.2).
- 2. Dissolve the Biocytin C2 Maleimide in DMSO at a concentration of 5 10 mg/mL.
- Mix the Biocytin C2 Maleimide and protein solution at 2:1 molar ratio of biocytin/protein, and shake the reaction mixture at room temperature for 2 - 4 hours.
- 4. Filter the reaction mixture through a protein spin column for 100 µg to 1 mg protein labeling reaction. If the reaction scale is larger than 1 mg, purify the conjugate using gel filtration on a properly sized Sephadex G-25 column.
- Collect the desired fractions for your immediate use or freeze dry them for future use.

Labeling Small Molecules with Biocytin C2 Malemide

- Dissolve Biocytin C2 Maleimide (10 15 mg/mL) and your thiolcontaining molecule in DMSO at 1:1.2 molar ratio of biocytin/thiol-containing molecule.
- 2. Stir the reaction mixture at room temperature for 2 4 hours.
- 3. Purify the conjugate using HPLC (ammonium acetate/water and acetonitrile, pH 7.0).
- 4. Collect and pool the desired fractions.
- 5. Combine and freeze-dry the pooled fractions.

EXAMPLE DATA ANALYSIS AND FIGURES

Figure 1. Chemical structure for Biocytin C2 maleimide

DISCLAIMER

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