

Protocol for Labeling with 6-TAMRA NHS Ester

This protocol describes a general method for labeling primary amine-containing biomolecules (such as proteins, peptides, or oligonucleotides) with 6-Carboxytetramethylrhodamine NHS Ester (6-TAMRA SE)

Materials Required:

- 6-TAMRA NHS Ester (dye)
- Target biomolecule (e.g., antibody, oligo, peptide)
- Dimethyl sulfoxide (DMSO), anhydrous
- Sodium bicarbonate buffer (0.1 M, pH 8.3–8.5)
- Desalting column or spin filter (for purification)
- Microcentrifuge tubes, pipettes

Step-by-Step Protocol:

1. Dissolve 6-TAMRA NHS Ester in anhydrous DMSO to a concentration of 10 mg/mL. Store protected from light.
2. Prepare your amine-containing biomolecule in 0.1 M sodium bicarbonate buffer at ~1–10 mg/mL.
3. Add 6-TAMRA NHS Ester to the biomolecule solution at a molar ratio of 5–10:1 (dye:biomolecule).
4. Incubate the reaction for 1 hour at room temperature, protected from light.
5. Purify the labeled product using a desalting column or spin column to remove free dye.
6. Store labeled conjugates at 4°C in the dark, or aliquot and freeze at –20°C.

Notes:

- Do not use buffers containing primary amines (e.g., Tris) during the reaction.
- Optimal dye-to-protein ratio may require empirical adjustment depending on the application.

Applications:

- Fluorescence-based detection
- Antibody and peptide conjugation
- Fluorescent probe synthesis