INSTRUCTIONS

Sulfo-NHS Acetate

26777

1503.2

Number	Description
26777	**Sulfo-NHS Acetate,** 100mg Molecular Weight: 259.17

Storage: Upon receipt store at -20°C. Product shipped at ambient temperature.

Introduction

Thermo Scientific™ Sulfo-NHS Acetate (Figure 1) is a protein modification reagent for blocking primary amines. The NHS-ester group of this reagent reacts with primary amines at pH 7.0-9.0, allowing modifications to occur in many standard non-amine-containing buffers. Once reacted, the amine is irreversibly capped with an acyl group. For reversible amine blocking, Citraconic Anhydride (Product No. 20907) is used.

Sulfo-NHS Acetate is typically used to prevent polymerization when performing protein cross-linking reactions and when conjugating peptides to carrier proteins for immunogen production. Blocking amines on the peptide allows directed conjugation of carboxylic acids on the peptide to primary amines on the protein using EDC (Product No. 29980).

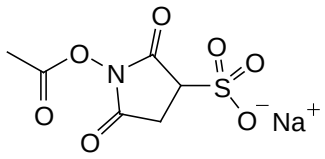

Figure 1. Chemical structure of Sulfo-NHS Acetate.

Important Product Information

- Reconstitute Sulfo-NHS Acetate immediately before use. Do not prepare stock solutions for storage because the NHS-ester moiety readily hydrolyzes and becomes non-reactive. Discard any unused reconstituted reagent.
- Sulfo-NHS Acetate is moisture-sensitive. To avoid moisture condensation onto the product and subsequent hydrolysis of the reagent, the vial must be equilibrated to room temperature before opening.

Procedure for Blocking Amines on Proteins or Peptides

A. Additional Materials Required

- Reaction Buffer: 0.1M sodium carbonate ($NaHCO_3$) buffer, pH 8.5 or other amine-free buffer such as phosphate buffered saline (PBS; e.g., 100mM sodium phosphate, 0.15M NaCl; pH 7.2; Product No. 28372) or HEPES, pH 7.5-8
- Quenching Buffer (optional): 1M Tris•HCl, pH 7.5 (1M glycine or lysine may also be used)
- Desalting Column (optional): Dextran Desalting Column, 5K MWCO (Product No. 43230) or Dialysis Unit (e.g., Thermo Scientific™ Slide-A-Lyzer™ Dialysis Cassette 0.1-0.5mL, 10K MWCO; Product No. 66385). For peptides larger than 2kDa, use Polyacrylamide Desalting Columns, 1.8K MWCO (Product No. 43426).

B. Procedure

1. Dissolve or buffer-exchange the protein or peptide sample into the Reaction Buffer.

   Note: If the peptide is insoluble in the Reaction Buffer, dissolve peptide with DMSO or DMF and then dilute with an equal volume of Reaction Buffer.

2. Dissolve Sulfo-NHS Acetate in ultrapure water to 10mM (2.6mg/mL). Prepare a 25-fold molar excess of Sulfo-NHS Acetate to amino groups in the sample and add it to the reaction. Alternatively, if the number of amines in the sample is not known, adding an equal mass amount of the Sulfo-NHS Acetate to the sample will provide sufficient molar excess.
3. Mix reaction and incubate for 1 hour at room temperature.

4. If desired, add Quenching Buffer to quench any NHS esters that did not react or remove the non-reacted reagent by dialysis or desalting.

Troubleshooting

Problem	Possible Cause	Solution
Amines not blocked	Reagent hydrolyzed	Completely equilibrate reagent vial to room temperature before opening to prevent condensation in the vial
		Dissolve reagent immediately before use – do not make a stock solution for storage
	Improper buffer was used	Use only non-amine containing buffers at pH 7-9 – at higher pH, hydrolysis is a competing reaction that reduces efficiency, and at lower pH, the reaction is extremely slow

Additional Information Available on Our Website

- Tech Tip #18: Block amino groups to prevent polymer formation in peptide-carrier protein conjugations
- Tech Tip #3: Determine reactivity of NHS ester biotinylation and crosslinking reagents
- Tech Tip #43: Protein stability and storage
- Tech Tip #20: Dialysis: an overview

Related Thermo Scientific Products

28372 **BupH™ Phosphate Buffered Saline Packs,** 40 packs, each pack results in 500 ml

28390 **BupH MES Buffer Saline Packs**, 10 packs, each yields 500mL of 0.1M 2-[morpholino]-ethanesulfonic acid, 0.9% NaCl, pH 4.7 when dissolved in 500mL deionized water (5L total)

77149 **EDC (carbodiimide)**, 10mg

22980 **EDC (carbodiimide)**, 5g

23031 **Ethylenediamine Dihydrochloride**, 10g

77171 **Imject Bovine Serum Albumin (in MES buffer)**, 2mg

77110 **Imject Bovine Serum Albumin (in PBS)**, 5 × 20mg

Product Reference

Whaseon Lee-Kwon, *et al.* (2003). Lysophophatidic acid stimulates brush border Na+/H+ exchanger 3 (NHE3) activity by increasing its exocytosis by an NHE3 kinase A regulatory protein dominated mechanism. *J Biol Chem* **278(19):**16494-501.