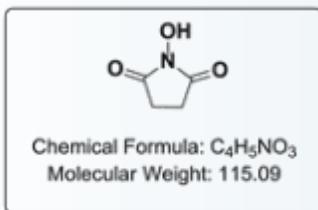


N-Hydroxysuccinimide (NHS)

Product Number: 13504-25, 13504-custom



Product Background

Alternative Names: N-hydroxysuccinimide; NHS; hydroxysuccinimide; 1-Hydroxy-2,5-pyrrolidinedione; HOSu;

N-Hydroxysuccinimide (NHS) is a catalyst used in carboxylate and amine coupling reactions involving EDC-HCl (EDAC-HCl) or other carbodiimide. NHS is involved in stabilizing the protein-carbodiimide intermediate, which allows for a more efficient coupling to occur.

This compound is also available in the sulfonated form, which key benefit is increased solubility of the intermediate. For more information, see Sulfo-NHS on www.covachem.com.

General Protein Coupling Protocol with EDC and NHS

- To a 2 mL sample of protein #1 (~10 mg/mL, MW=60,000 Da) dissolved in reaction buffer (such as Phosphate buffered saline, pH 7.2), add 0.77 mg of EDC-HCl (4.0 μmol).
- To this solution, add 1.15 mg of N-Hydroxysuccinimide (10.0 μmol), and mix well.
- Allow this activating reaction to proceed at room temperature for 10 to 15 minutes.
- (Optional Steps) Inactivate residual EDC with excess 2-mercaptoethanol (2.5 μL). Separate the small molecule by-products (EDC, NHS, and 2-mercaptoethanol) via gel filtration recovering the activated protein #1.
- Prior to proceeding with the amine coupling portion of this reaction, ensure that the buffer pH is pH 7 to 8. Adjust the pH to within this range by adding concentrated phosphate buffer or sodium carbonate. Note: avoid any compounds containing amines, such as Tris.
- Add Protein #2 (directly as a solid or as a 1-10 mg/mL solution in PBS or other suitable buffer) to the activated protein #1 solution. Mix. Allow to react for 2 to 2.5 hours at room temperature.
- (Optional) Quench the reaction with hydroxylamine, glycine or other suitable amine at a final concentration of 10-20 mM.
- (Optional) Remove unreacted small molecules via gel filtration with Sephadex G-25.

Notes: A good starting point for this coupling reaction is where Protein:EDC:NHS mole ratios are roughly 1:10:25. This protocol reflects these ratios, although these are routinely modified by the end user. Adjustments may need to be made on an individual basis.

*suitable reaction buffers include: PBS, (100 mM Sodium Phosphate, 150 mM NaCl, pH 7.2) or MES

Product Information Bulletin

Product Description

Package Size and Product Numbers

25 gram.....13504-25

Product Name:

N-Hydroxysuccinimide (NHS)

CAS Number:.....[6066-82-6](#)

Product Specifications

- Appearance: White Solid
- Infrared Spectroscopy: Passes
- Gas Chromatography ≥ 97.5 %

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