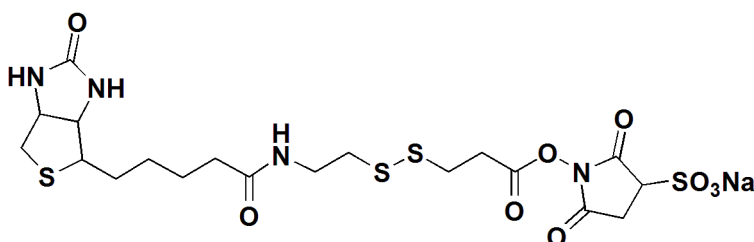


Sulfo-NHS-SS-Biotin Protocol and Product Information Sheet

| | |
|----------------------|---|
| Product Category: | Biotinylation Reagents |
| Catalog Number(s): | b2104-100mg , b2104-1gm , b2104-custom |
| Product Name: | Sulfo-NHS-SS-Biotin |
| Alternative Name(s): | Sulfosuccinimidyl-2-(biotinamido)-ethyl-1,3'-dithiopropionate; Biotin disulfide N-hydroxysulfosuccinimide ester |
| CAS Number: | 325143-98-4 |
| Chemical Formula: | C ₁₉ H ₂₇ N ₄ NaO ₉ S ₄ |
| Molecular Weight: | 606.69 |
| Spacer Length: | 24.3 Å |
| Storage: | Upon receipt store at 4°C (shipped at ambient temperature). |



General Sulfo-NHS-SS-Biotin Protein Biotinylation Protocol

1. Allow vial of Sulfo-NHS-SS-Biotin to equilibrate to ambient temperature before opening.
2. Dissolve protein at a concentration of 1-10 mg/mL in 100 mM sodium phosphate, 150 mM NaCl, pH 7.2-7.5 or other suitable amine-free buffer.
3. Immediately before use, create a 6 mg/mL Sulfo-NHS-SS-Biotin stock solution (~10mM) in water or buffer (same as step 2). Anhydrous DMF ([cr8106-25ml](#)) or DMSO ([cr8105-25ml](#)) can also be used to make a stock solution ahead of time.
4. Add sufficient Sulfo-NHS-SS-Biotin stock solution to the protein solution to obtain 10-20 fold molar excess of biotinylation reagent over protein.
Note: Alternatively, an amount of Sulfo-NHS-SS-biotin can be added to the protein solution required to give 10-20 fold molar excess. Dilute protein solutions (i.e. 1-2 mg/mL) may require increased molar excess of Sulfo-NHS-SS-Biotin (i.e. ≥ 20 fold) to yield similar biotinylation of a more concentrated protein solution.
5. Allow biotinylation reaction to proceed for 30-60 minutes at room temperature or ≥ 2 hours at 4°C.
6. Desalt biotinylated protein through dialysis or gel filtration with a resin, such as Sephadex® G-25 ([q4109](#)) or equivalent.
7. To reverse the biotin tag, through reduction of the disulfide linkage, incubate biotinylated sample with 50mM DTT ([cr8101-5x10mg](#)) for 90-120 minutes at room temperature or 1 hour at 45°C.

References:

Hermanson, G.T. 1996. Bioconjugate Techniques. Academic Press, San Diego, CA, USA.