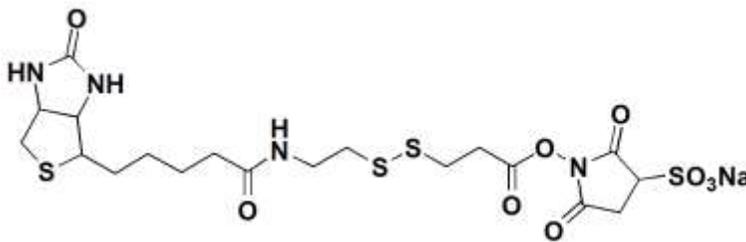


Sulfo-NHS-SS-Biotin Protocol and Product Information Sheet

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 -20°C (shipped at ambient temperature). Protect from moisture (i.e. humidity); blanket under desiccated, inert gas.



General Sulfo-NHS-SS-Biotin Protein Biotinylation Protocol

1. Allow vial of Sulfo-NHS-SS-Biotin to fully equilibrate to ambient temperature before opening to prevent condensation inside the vial (Sulfo-NHS-SS-Biotin is moisture-sensitive).
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 ([g4109](#)) 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.

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