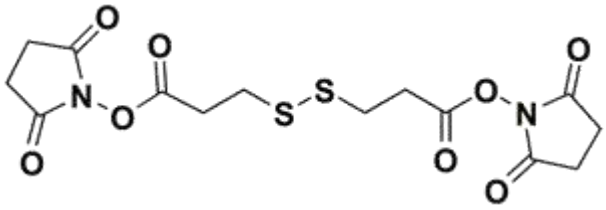
DSP Crosslinker Protocol and Product Information Sheet

Product Category:	Homobifunctional Crosslinkers
Catalog Number(s):	c1106-100mg, c1106-1gm, c1106-custom
Product Name:	DSP Crosslinker
Alternative Name(s):	DSP; Lomant's Reagent; 3,3'-Dithiodipropionic acid di(N-hydroxysuccinimide ester); Di(N-succinimidyl) 3,3′-dithiodipropionate; Dithiobis-succinimidyl propionate
CAS Number:	57757-57-0
Chemical Formula:	$C_{14}H_{16}N_2O_8S_2$
Molecular Weight:	404.42
Spacer Arm Length:	12.0 Å
Storage:	Upon receipt store at -20°C (shipped at ambient temperature). Protect from moisture (i.e. humidity); blanket under desiccated, inert gas.



General DSP Crosslinking Protocol

1. Allow vial of DSP Crosslinker to fully equilibrate to ambient temperature before opening to prevent condensation inside the vial (DSP is moisture-sensitive and will hydrolyze).
2. Prepare a 50 mM solution of DSP crosslinker, by dissolving 10mg DSP in 495 µL of dry DMSO or dry DMF solvent. (Note: The NHS esters of DSP are susceptible to hydrolysis. If aliquoted into an aqueous solution and stored in it, then the DSP will degrade.)
3. Using a 20-fold molar excess approach (20:1 Crosslinker:Protein), add crosslinker solution to the protein sample in non-amine containing buffer (i.e. 25 mM Sodium Phosphate, pH 7.4), so that the final crosslinker concentration is between 0.5 to 5 mM. Optimal pH range is from 7 to 9.
4. Allow the sample to react at room temperature for 30-45 minutes. Allow slightly longer if sample must be kept on ice (recommended 2-3 hours). This reaction rate is not highly temperature sensitive.
5. Quench any unreacted DSP with 25 mM to 200 mM Tris, pH 7.4. Allow to react for 10-15 minutes at room temperature.
6. Desalt sample to remove unreacted DSP crosslinker (i.e. gel filtration, dialysis, etc.).

Intracellular DSP Crosslinking Protocol

1. Remove media by washing cells twice with non-amine containing buffer (i.e. 25 mM Sodium Phosphate, pH 7.4).
2. The crosslinking solution as noted in steps 1 and 2 above, then add the crosslinker solution to the cells in a final concentration of ~2 mM.
3. Incubate the reaction mixture for 30-45 minutes at room temperature or for 2-3 hours if on ice.
4. Quench any unreacted DSP with 25 mM to 200 mM Tris, pH 7.4. Allow quenching reaction to proceed for 10-15 minutes at room temperature.

Reference:

Wong, S.S. (1993) CRC Chemistry of Protein Conjugation and Crosslinking. CRC Press, Boca Raton, Florida.

ProteoChem * 5570 W. 290 N. Suite 2 * Hurricane, UT 84737
Toll Free: 888.501.2436 * Phone: +1.720.239.1630 * Fax: +1.720.239.1631
Technical Support E-mail: ts@proteochem.com
www.proteochem.com