

DST, Sulfo-DST

Homobifunctional ox-cleavable crosslinkers

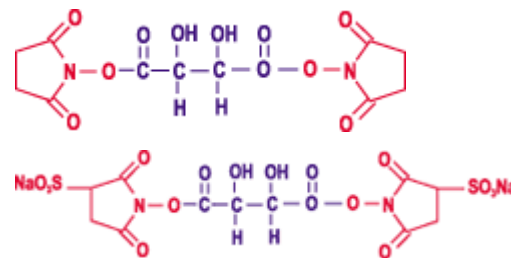
Description

Oxidizer cleavable homobifunctional amine reactive crosslinker

Catalog number: UP28068A, 1g
Name: DST
Formula : DiSuccinimidyl Tartarate
M.W.= 344.24, CAS: 70539-42-3

Catalog number: UP24864A, 100mg UP24864B, 50mg
Name: Sulfo-DST
Formula : DiSulfoSuccinimidyl Tartarate
M.W.= 548.32

Storage : +4°C (possible at -20°C), protect from moisture and light. (L)



Introduction

Cross-linkers are chemical reagents used to conjugate molecules together by a covalent bond. Several atoms separate the 2 molecules, forming the 'spacer arm'. The conjugate associates the characteristics and biological activities of each component.

Cross-linkers have become important tools for the preparation of conjugates used in a lot of immunotechnologies, and for protein studies (structure, interactions, activity, degradation...). **Homobifunctional** cross-linkers present 2 identical reactivities. The choice of the reactivities is determinant to the design of the right conjugate. EGS crosslinker reacts toward amines, through the succinimide group, and contains a **oxidizer cleavable** linkage.

Uptima offers a high quality DST and its sulfonated form to answer the needs of coupling proteins and peptides for biological studies and immunoassays like (other cross-linkers are available): (see literature below)

- Obtention of conjugates for structural studies (receptors, ligand interactions...)
- Reticulation of proteins (complexes) in solution or immobilized (receptors)
- Obtention of oligomeric conjugates : poly- peptides
- Immobilization on polystyrene or glass surfaces for immunoassays and biosensors
- Grafting peptides onto gels for chromatography separations
- Grafting haptens onto cells and particles (beads) for diagnostics...

Scientific and technical Information

- The chemical group **N-hydroxysuccinimide (NHS)** reacts in aqueous phase on primary ($-NH_2$) and secondary amines ($=NH$) (in fact on its deprotonated form), optimally at neutral pH or higher : amines present in proteins (Lys amino acid) and in a lower proportion on NH_2 located in terminal peptidic chains. The reaction occurs in few minutes in organic media at room temperature, and also in aqueous buffers but in competition with hydrolysis, that increases with pH, and with the high dilutions of the molecule that should be coupled. The reaction with amines occurs typically at pH 6.5-8.5 in 1 hour. n-Hydroxysuccinimide is released and should usually be removed before use of the conjugate.
- The **sulfonyl moiety ($NaSO_3$)** introduces a negatively charged hydrophilic group, that allows the product not to cross biological membranes. It reacts outside cells, when the crosslinker reacts also in the membrane and in the cells. An other interest of the sulfonyl group is to permit the solubilisation of the product directly in aqueous buffers, avoiding the use of organic solvents like DMSO or DMF, which are possibly noxious to cells or applications.
- The **spacer arm** of DST measures 6.3 Angstroms length. It is stable, allowing to prepare and use the conjugate. It can however be cleaved further by mild oxidizer agents, like periodate. DST has also been used to modify protein before electrophoresis analysis, and conjugates can then be cleaved by soaking the gel in 0.012M sodium periodate, 0.1% SDS, 0.02M sodium phosphate pH 7.0 for 2 hours at room temperature. (Smith 1978)
- Examples of protocols are given in the literature. As guidelines, here are some information for reversible couplings:

Contact your local distributor

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FT-28068A

Coupling of proteins in solution: the molecule(s) to be coupled is (are) prepared in PBS (20mM phosphate, 150mM NaCl, pH7.5). Other suitable buffers include HEPES, carbonate and borate (but not Tris) provided the pH is kept between 7 and 9. Crosslinker is added at 5 to 40 molar excess over the protein. Incubation may last 30min to 1H incubation at room temperature (or 1-2H at +4°C if thermolabile proteins). The crosslinking leads to conjugates (dimers...) and to reticulated forms. If different molecules are mixed, homo and hetero conjugates are obtained.

Immobilization of proteins: a soluble protein is conjugated to an aminated (or protein-coated) support (microplate, gel...). Crosslinker is used at 1-5mM with the protein of interest at 1-5mM.

Cell crosslinking: the cells at 1-10% suspension, mixed with the protein to be coupled at 2-10nM, are incubated with 0.5-4mM of crosslinker. +4°C is recommended for many cells, and agitation should be mild but continuous.

Immobilization of proteins: the protein is incubated with the crosslinker on the desired protein that is coated on polystyrene or other support. The concentration of protein and crosslinkers should be determined depending on protein nature and coating density.

If a precipitate is observed, protein and crosslinker concentrations should be decreased, or DMSO added up to 20% final concentration in the reaction mixture.

A stop reaction may be useful, for example with 20mM Lysine or with a Tris buffer during 15-30min.

A separation technique is usually necessary to isolate conjugates (gel filtration, dialysis, cell washing...)

Other information regarding NHS reactivity are available ([NT-NHS](#): buffers, conditions of use...).

Literature: DST

Bragg P.D. and Hou C.; A crosslinking study of the Ca²⁺ Mg²⁺-activated adenosine triphosphate of Escherichia coli; Eur. J. Biochem. 1980, 106-495-503

Farries, T.C. and Atkinson, J.P. (1989). Biosynthesis of properdin. J. Immunol. 142, 842-847.

Park, L.S., Friend, D., Gillis, S. and Urdal, D.L. (1986). Characterization of the cell surface receptor for a multi-lineage colony-stimulating factor (CSF-2).

Smith R.J., apaldi R.A., Muchmore D. and Dahlquist F.; crosslinking of ubiquinone cytochrome c reductase (complex III) with periodate-cleavable bifunctional reagents; Biochemistry 1978, 17, 3719-37

Other information

For use *in vitro* only, not for diagnostic.

For any information, please contact Uptima, or your local distributor.

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Related products :

See [BioSciences Innovations catalogue](#) and [e-search tool](#).

-Desalting tools ([CelluSep](#) dialysis, desalting columns)

-Other cleavable homobifunctional amine reactive crosslinkers:

- DSP [UP18971](#) (thiol cleavable), EGS [UP28067](#) (mild alkaline cleavable),...

-Homobifunctional amine reactive crosslinkers (non cleavable): [BS3](#) and [NHS-PEO-NHS](#)

-Heterobifunctional crosslinkers:

- MAL-PEOx-NHS #[AL6580](#) (hydrophilic spacer)
- SMPB #[UP28072A](#) & Sulfo-SMPB #[UP52757A](#) • SMCC-hydrazide #[BI1281](#)

-Other conjugation technologies:

- Hydralink Conjugation kit #[BL1501](#) and [crosslinkers](#) (SANH #[BL9270](#), MHPH #[BL9401](#))

-Useful modifiers:

- SATA #[84235A](#), Iminothiolane #[42425A](#)

Catalog size quantities and prices may be found at <http://www.interchim.com>.

Please inquire for higher quantities (availability, shipment conditions).

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