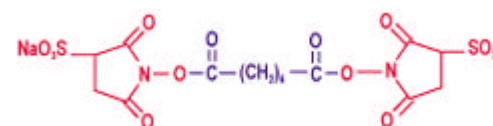


## DSS, BS3(Sulfo-DSS) Homobifunctional cross-linkers

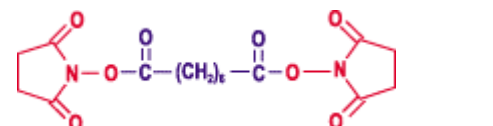
### Product Description

**Catalog number:** UP54940A, 100 mg                      UP54940B, 50 mg  
**Name:** **BS3**, Sulfo-DSS  
**Formula :** Bis-(Sulfo-succinimidyl)-Suberate  
 CAS: 82436-77-9 , C<sub>16</sub>H<sub>19</sub>N<sub>2</sub>O<sub>11</sub>Na, **M.W.= 572.4**

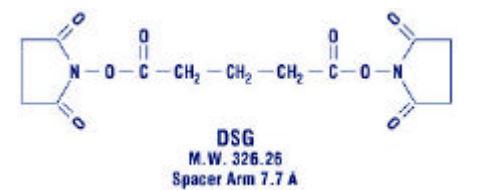


**Storage :** -20°C (possible at 4°C) , protect from moisture and light. (M)

**Catalog number:** UP28065A, 1 g                      UP28065B, 5x1 g  
**Name:** **DSS**  
**Formula :** DiSuccinimidyl Suberate  
 CAS: 68528-80-3 , C<sub>16</sub>H<sub>20</sub>N<sub>2</sub>O<sub>8</sub>, **M.W.= 368.4**



**Catalog number:** UP29859A, 50 mg                      Inquire  
**Name:** **DSG**  
**Formula :** DiSuccinimidyl Glutarate  
 C<sub>14</sub>H<sub>16</sub>N<sub>2</sub>O<sub>8</sub>, **M.W.= 326.26**



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

### General Considerations

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. Considering the final result, an important other thing is the nature and length of the spacer. **DSS** and **BS3** react toward amines, through the succinimide group.

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

- Obtention of immunogens carrier-hapten
- Obtention of di-specific affine probes
- Obtention of biologically active conjugates
- Obtention of oligomeric conjugates : poly- peptides
- Studies of di- or oligo-meric proteins for structural investigations
- Coating of polystyrene surfaces for immunoassay
- Grafting peptides onto gels for chromatography separations
- Grafting haptens onto cells and particules (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 aminoacid) and in a lower proportion on  $NH_2$  located in terminal peptidic chains. The reaction competes with hydrolysis, that increases with pH, and with the high dilutions of the molecule that should be biotinylated.
- The sulfonyl moiety ( $NaSO_3$ ) introduces a hydrophilic group, that allows the product not to cross biological membranes. An other interest of the sulfonyl group is to permit the solubilisation of the product directly in aqueous buffers, up to 10 mM, avoiding the use of organic solvents like DMSO or DMF, which are possibly nocive to cells or applications.
- The spacer arm of DSS and BS3 are 8 atoms long, for a length of 11.4 Angstroms

## Use

### Protocole : Irreversible immobilisation of antibodies onto an affinity column

This standard protocole describes the coupling attachment of IgG onto a proteinA agarose gel. It should be optimized to each application (volume, type of Ig and gel...). For applications from 1 to 10 mg of IgG, 1 ml of gel is usually needed (pipette a 2ml of 50% suspension). Incubations and washes are performed in batch, with 5 ml buffer per ml of gel.

- 1- Regenerate if needed the agarose protein A gel. Wash well with PBS (150mM NaCl, 20mM phosphate, pH7.4)
- 2- Incubate the antibody in appropriate quantity onto Protein A, during 30 min at room temperature under constant and gentle agitation  
Uptima recommends a IgG amount equal to the IgG binding capacity of the gel, but lower quantity may be applied depending on the antibody availability and further application requirements
- 3- Wash well with PBS. Absorbance at 280nm should be less than 0.05.  
Rem: the unbound antibody can be recovered in washes and quantified by a protein assay (BC Assay #UP40840A) to determine the loading capacity of the gel, by difference with initial added IgG.
- 4- Prepare a DSS solution at 20 mM in anhydrous DMSO. This solution should be used immediately. The BS3 is dissolved at 20 mM in aqueous buffer.
- 5- Add very slowly 175 $\mu$ l of DSS solution or BS3 solution per ml of gel in 2ml of PBS while stirring. This is advantageous done in 2 or 3 steps separated by 5 min intervals. Incubate for 1H under constant agitation at room temperature.
- 6- Wash the gel with PBS (at least 3 washes of 5 ml per ml of gel during 5minutes)
- 7- Incubate the gel with a saturating agent, 5 ml/ml of gel, during 1H at room temperature. Usually, BSA (#UP900100), normal serum, or SeaBlock (#UP40301) are used, diluted at 5% in PBS.

## Other Information

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

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

213 av.J.F.Kennedy, 03103 Montluçon, fax :04 70 03 82 60, hotline Interbiotech : 04 70 03 76 06

### Related products :

MaxiBind™ : activated proteins for immunization and screening conjugates

CelluSep : dialysis membranes

### Literature:

Loster K, et al, Chemical cross-linking leads to two high molecular mass aggregates of rat alpha 1 beta 1 integrin differing in their conformation but not in their composition, FEBS Lett., 1995, **373**, 234-238; DSS

Jordan J.A., et al, Targeting of mouse erythrocytes by band 3 crosslinkers; Biochim.Biophys.Acta, 1291, 27-34 (1996)

Staros J.V.; N-hydroxysuccinimide active esters: bis(N-hydroxysuccinimide) esters of two dicarboxylic acids are hydrophilic, membrane-impermeant, protein crosslinkers; Biochem. 1982, **21**, 3950-3955; DSS

Waugh, S.M., DiBella, E.E. and Pilch, P.F. Isolation of a proteolytically derived domain of the insulin receptor containing the major site of cross-linking/binding. Biochemistry 1989, **28**, 3448-3455; EGS

rev : B12E

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