

Product Data Sheet

Product Name ToLuminate CD Photogel

Catalog Number PG91-1

Description

The ToLuminate CD Photogel kit provides reagents for setting up cell-compatible hydrogels formed by illumination with blue light (365-405 nm) on the basis of the thiol-ene chemistry. Its major components are norbornene-modified dextran (N-Dextran) and the thiol-modified crosslinker CD-Link. When the two reagents are combined and illuminated in the presence of LAP (Lithium phenyl-2,4,6-trimethylbenzoylphosphinate) thiol groups on CD-Link are activated and form stable thioether bonds with norbornene groups on N-Dextran. The components are mixed at physiological pH (pH 7.2) for optimal cell compatibility. There is ample time to place the pre-gel solution in any culture dish or container, since gelation starts only after illumination.

CD-Link contains a matrix metalloproteinase (MMP)-cleavable peptide sequence (Pro-Leu-Gly-Leu-Trp-Ala), which is cleaved by a broad range of MMPs including MMP1, MMP2, MMP3, MMP7, MMP9 and MMP13 [1, 2, 3]. It allows cells to locally degrade the polymer network, if they produce the indicated MMPs.

Cell adhesion peptides (e.g. *3-D Life* RGD Peptide, Cat. No. 09-P-001) can be covalently attached to a portion of the norbornene groups on N-Dextran with the same chemical reaction as CD-Link to provide a cell-adhesive matrix.






3-D Life AgaFloat (Cat. No. A10-3) can be added to the pre-gel solution to avoid sedimentation of cells before illumination.

Dextran hydrogels crosslinked with CD-Link can be dissolved by the addition of dextranase (*3-D Life* Dextranase, Cat. No. D10-1), which allows the recovery of chemically fixed or live cells for post-culture analyses (e.g. RT-PCR) or for further cultivation.

For more information and instructions, please consult the General Protocol GP-5 "Preparation of ToLuminate Photogels".

Quantity Up to 4 mL hydrogel.

Components

Material	Quantity	Concentration of reactive groups	Storage
 N-Dextran	600 µl	30 mmol/L	-20°C to -80°C
 CD-Link, lyophilized	3x 200 µl*	20 mmol/L* thiol groups	Lyophilisate and after reconstitution: -70°C to -80°C
 10x CB (pH 7.2) Phenol Red-free	600 µl	n/a	Short term (≤2 months): 4°C Long term: -20°C to -80°C
 LAP, 10 mM	600 µl	10 mmol/L	-20°C to -80°C
 Water	2x 1500 µl	n.a	Room temperature or lower

All materials are filter-sterilized.

*Volume/concentration after reconstitution of lyophilisate.

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Reconstitution

CD-Link:

1. Briefly centrifuge vial containing CD-Link lyophilisate to make sure that the entire material is at the bottom of the reaction tube.
2. Add 188 μ l 3-D Life Water per tube for a concentration of 20 mmol/L thiol groups.
3. Close tube and briefly vortex.
4. Incubate for 5 min.
5. Briefly vortex and centrifuge again.
6. CD-Link is now ready for use.

References

[1] Knight, C. G. et al. FEBS 296:263-66 (1992) [2] Patterson, J. and Hubbell, J. A. Biomaterials 31, 7836-7845 (2010) [3] Deng, S. et al. J. Biol. Chem. 275, No. 40, 31422–31427 (2000)

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