Product Data Sheet

Product Name	ToLuminate CD-HA Photogel

Catalog Number PG96-1

Description

The ToLuminate CD-HA 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 hyaluronic acid crosslinker CD-HyLink. When the two reagents are combined and illuminated in the presence of LAP (Lithium phenyl-2,4,6-trimethylbenzoylphosphinate) thiol groups on CD-HyLink 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. The hydrogel allows cell spreading and migration if cell adhesion peptides are present in the gel.

CD-HyLink 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 SH-reactive groups on N-Dextran 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.

Note: Dextran hydrogels crosslinked with CD-HyLink cannot be dissolved by the addition of dextranase.

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-HyLink, freeze-dried	4x 200 µL	10 mmol/L* thiol groups	Lyophilisate and after reconstitution: -70°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 reconstutution of lyophilisate.

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Reconstitution

CD-HyLink:

1. Briefly centrifuge vial containing CD-HyLink lyophilisate to make sure that the entire material is at the bottom of the reaction tube.
2. Add 188 µl *3-D Life* Water for a concentration of 10 mmol/L thiol groups.
3. Close tube and briefly vortex.
4. Incubate for 60 min at room temperature.
5. Briefly vortex and centrifuge again.
6. CD-HyLink 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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