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# Phenyldiazirine oxLink™

Lyophilized powder, 250 μg, ≥95% pure by HPLC Product Number: CM81402-250UG and CM81402-

4x250UG

CAS Registration Number: N/A

MW: 452.39

# F<sub>3</sub>C H AqT linker O NH<sub>2</sub>

## **Product Description**

CellMosaic's Phenyldiazirine oxLink™ is a proprietary photo-crosslinking reagent developed by CellMosaic for studying biomolecular interactions. This reagent combines a highly efficient, carbenegenerating phenyldiazirine group with convenient oxime-based reversible chemical crosslinking and the super-hydrophilic AqT® linker. These structural features make this reagent highly advantageous compared to traditional photo-crosslinking reagents.

- 1) Photo-crosslinking group: oxLink™ uses a trifluoromethyl phenyldiazirine group, a highly efficient carbene-generating photo-crosslinking group. Trifluoromethyl phenyldiazirine photolyzes around 360 nm, at which photodamage to biomolecules is minimized. The generated carbene inserts C−H bonds into the neighboring biomolecular partner within picoseconds. Because the electron-withdrawing trifluoromethyl group confers stability on the intermediate diazo-isomer, no side products are detected under normal labeling conditions.
- 2) Reversable chemical crosslinking group: The oxLink™ reagent contains an aminooxy group for chemical crosslinking with biomolecules containing ketone or aldehyde functionalities. Aminooxy-ketone ligation is orthogonal to peptide chemistry, enable highly selective, solid phase-based separation of the aminooxy-labeled crosslinked products. This selective purification method allows characterization of protein complexes in complex matrices such as plasma, cellular membranes, and cell lysates, where free thiols may otherwise interfere with labeling and purification. **Figure 1** illustrates a workflow showing how reversible oxLink™ can be used to crosslink interacting biomolecule partners and identify the crosslinking site or detect the interaction partner.
- 3) Hydrophilic AqT® linkers: AqT® linkers are novel proprietary biomaterials invented at CellMosaic. Because the trifluoromethyl phenyldiazirine group is highly hydrophobic, biomolecules labeled with it using a traditional ethylene and ethylene glycol-type linkers tend to aggregate and destabilize the labeled protein. The AqT® linker greatly enhances the hydrophilicity and water solubility of oxLink™ (>27 mg/mL). It also improves biocompatibility and reduces non-specific hydrophobic interactions with other biomolecules, allowing high loading of phenyldiazirine groups.

## **Application of the Product**

- Labeling biomolecules containing aldehyde or ketone groups
- Studying dynamic biomolecule interactions via photo-crosslinking (e.g., biological complexes and networks, protein—protein interactions, protein—DNA interactions, and small ligand—protein interactions)

#### **Key Features of the Product**

• Highly water soluble (>27 mg/mL) and biocompatible

- Enable high loading with minimized aggregation
- Contains an orthogonal peptide crosslinking group capable of forming a reversible linkage
- Includes an efficient carbene-generating photo-crosslinking group

# Storage/Stability

- Stable at room temperature when kept in the dark.
- Recommended storage below -20°C, where the product remains stable for several years without signs
  of decomposition

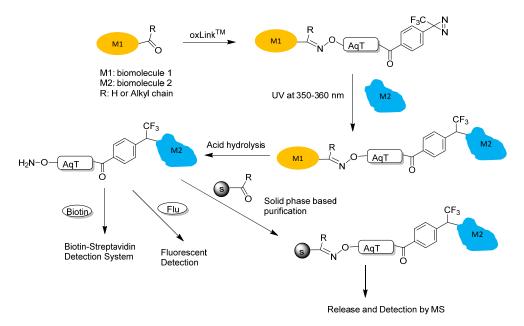
#### Usage

Supplied as a lyophilized solid. Ready to use after dissolving water or suitable labeling buffer (use a slightly acidic buffer for labeling).

#### References:

Protein crosslinking reviews: a) Brunner, J. (1993) *Annu. Rev. Biochem.* **62**, 485–514. b) Freedman, R. B. (1979) *Trends Biochem. Sci.* 193–197. c) Herrmann, J. M., Westermann, B., Neupert, W. (2001) *Methods Cell Biol.* **65**, 217–230. d) Fancy, D. A. (2000) *Curr. Opin. Chem. Biol.* **4**, 28–32. e) Fasold, H., Klappenberger, J., Meyer, C., Remold, H. (1971) *Angew. Chem. Internat. Edit.* **10**, 795–801. f) Bayley, H. Photogenerated Reagents in Biochemistry and Molecular Biology, Vol. 12. Elsevier, Amsterdam, Neth, 1983.

3-Trifluoromethyl-3-phenyldiazirine reference: Brunner, J., Senn, H. & Richards, F. M. (1980) J. Bio. Chem. **255**, 3313–3318.



**Figure 1**. oxLink™ workflow using water-soluble Phenyldiazirine oxLink™. oxLink™ is tethered to a target biomolecule of interest (M1) containing an artificially introduced ketone or aldehyde group via its aminooxy functional group. The resulting oxime bond can be hydrolyzed, releasing the photocrosslinked biomolecule (M2) with a free aminooxy group. M2 can then react with any aldehyde- or ketone-containing molecule, such as a solid phase, biotin, or fluorescent dye, for further purification, detection, and identification.

# **Important Notes & Contact Information**

#### READ BEFORE USING ANY RESOURCES PROVIDED HEREIN

The information provided in this document and the methods included in this package are for information purposes only. CellMosaic provides no warranty of performance or suitability for the purpose described herein.

Sample data are provided for illustrative and example purposes only and represent a small dataset used to verify kit performance in the CellMosaic laboratory. Information about the chemicals and reagents used in the kit are provided as necessary.

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