

# **Product Information**

## Cell Tracker CM-Dil

Catalog Number: C4060

Product Size: 20 µg, 1 mg

Application Scope: Cell tracing, long-term tracking, cellular imaging

### **Parameters**

Appearance: Purple solid soluble in DMSO, DMF or EtOH

Ex/Em: 553/570 nm

CAS NO.: 180854-97-1

Molecular Formula: C68H105Cl2N3O

Molecular Weight: 1051.5

Molecular Structure:

### **Storage**

Store at -20°C and protect from light. When stored as directed, product is stable for at least 12 months.

# **Description**

Cell Tracker CM-DiI is a derivative of DiI and has better solubility in water than DiI. Cell Tracker CM-DiI fluorescent dye has been designed to freely pass through cell membranes into cells, where it is transformed into cell membrane-impermeant reaction products. Cell Tracker CM-DiI dye is retained in living cells through several generations. The dye is transferred to daughter cells, but not adjacent cells in a population. Cell Tracker CM-DiI dye is designed to display fluorescence for at least 72 hours, and the dye exhibits ideal tracking properties: it is stable, nontoxic at working

concentrations, well retained in cells, and brightly fluorescent at physiological pH. Additionally, the excitation and emission spectra of Cell Tracker CM-Dil dye are well separated from GFP (green fluorescent protein) spectra allowing for multiplexing.

Cell Tracker CM-DiI incorporates a mildly thiol-reactive chloromethyl substituent that confers aldehyde fixability via conjugation to thiol-containing peptides and proteins. Unlike other cell membrane dyes, Cell Tracker CM-DiI is retained in cells throughout fixation, permeabilization, and paraffin embedding procedures.

## **Protocol**

## 1. Dyeing liquid preparation

(1) Preparation of stock solution: Use DMSO or DMF to make a stock solution with a concentration of 1-2 mg/mL.

Note: It is recommended to store the storage solution at -20 °C, and aliquot it in small quantities to avoid repeated freeze-thaw cycles.

(2) Preparation of working solution: Dilute the storage solution with a suitable buffer (such as serum-free medium, HBSS or PBS) to prepare a working solution with a concentration of 1 to  $2~\mu M$ .

Note: You may need to optimize the staining procedure for each particular cell type by varying the dye concentration, staining volume, labeling time, or wash steps.

#### 2. Cell staining





We suggest incubating cells in the working solution for 5 minutes or less at 37°C, and then for an additional 15 minutes at 4°C. Incubation at this lower temperature appears to allow the dye to label the plasma membrane but slows down endocytosis, thus reducing dye localization into cytoplasmic vesicles. After labeling, wash cells with phosphate-buffered saline (PBS) and resuspend in fresh medium. For adherent cells, labeling in culture while attached results in improved viability compared to labeling after dissociation.

Cell Tracker CM-DiI stained cells are compatible with

subsequent fixation and permeabilization:

- a. Fixed. Fix with 3.7% paraformaldehyde in PBS for 10 min at room temperature.
- b. Permeability. Permeabilize with acetone at -20°C for 10 min.

# **Notes**

- 1. There are quenching problems with fluorescent dyes. Please avoid light to slow down the fluorescence quenching.
- 2. For your safety and health, please wear lab coats and disposable gloves.