Cell Explorer™ Live Cell Tracking Kit *Deep Red Fluorescence*

Catalog number: 22624
Unit size: 200 Tests

Component	Storage	Amount
Component A: Track It™ Deep Red	Freeze (<-15 °C), Minimize light exposure	1 vial (50 µL) (500X DMSO stock solution)
Component B: Assay Buffer	Refrigerate (2-8 °C)	1 bottle (20 mL)

OVERVIEW

Our Cell Explorer™ fluorescence imaging kits are a set of tools for labeling cells for fluorescence microscopic investigations of cellular functions. The effective labeling of cells provides a powerful method for studying cellular events in a spatial and temporal context. This particular kit is designed to uniformly label live cells in deep red fluorescence for the studies that require the fluorescent tag molecules retained inside cells for relatively longer time. The kit uses a fluorescent dye that carries a cell-retaining moiety. The dye becomes trapped inside live cells to give a stable fluorescence signal for relatively long time. The dye is a hydrophobic compound that easily permeates intact live cells. The labeling process is robust, requiring minimal hands-on time. It can be readily adapted for a wide variety of fluorescence platforms such as microplate assays, immunocytochemistry and flow cytometry. It is useful for a variety of studies, including cell adhesion, chemotaxis, multidrug resistance, cell viability, apoptosis and cytotoxicity. The kit provides all the essential components with an optimized cell-labeling protocol.

AT A GLANCE

Protocol summary

1. Prepare samples
2. Add 10X Track It™ Deep Red working solution (10 µL/well)
3. Stain the cells at 37 °C for 15 minutes to 1 hour
4. Wash the cells
5. Examine the specimen under fluorescence microscope with Cy5 filter or flow cytometer with 660/20 nm filter (APC channel)

Important Thaw all the components to room temperature. Centrifuge the component A briefly before opening.

KEY PARAMETERS

Instrument:	Fluorescence microscope
Excitation:	Cy5 filter
Emission:	Cy5 filter
Recommended plate:	Black wall/clear bottom
Instrument:	Flow cytometer
Excitation:	640 nm laser
Emission:	660/20 nm filter
Instrument specification(s):	APC channel

PREPARATION OF WORKING SOLUTION

Dilute 500X Track It™ Deep Red DMSO stock solution (Component A) into Assay Buffer (Component B) to make a 10 to 25X Track It™ Deep Red working solution. The working solution should be prepared enough for all the wells at 10 µL/well with the appropriate concentration. For example, to get a 10X final concentration of Track It™ Deep Red for one 96-well microplate, dilute 20 µL of 500X Track It™ Deep Red DMSO stock solution into 1 mL of Assay Buffer (Component B) to make 1 mL of 10X Track It™ Deep Red working solution.

Note The final concentration of the Track It™ Deep Red working solution should be empirically determined for different cell types and/or experimental conditions. It is recommended to test at the concentrations at least over a ten fold range.

Note The unused portion of the Track It™ Deep Red stock solution should be stored at -20 °C. Avoid repeated freeze/thaw cycles.

PREPARATION OF CELL SAMPLES

For guidelines on cell sample preparation, please visit
https://www.aatbio.com/resources/guides/cell-sample-preparation.html

SAMPLE EXPERIMENTAL PROTOCOL

1. Add 10X Track It™ Deep Red working solution to the cell wells which should be equal to 1/10 of the volume of cell culture medium. For example, for a 96-well plate, add 10 µL/well of 10X Track It™ Deep Red working solution into the cells.
2. Incubate the cells in a 37°C, 5% CO_2 incubator for 15 minutes to 1 hour.
3. Wash cells with Hanks and 20 mM Hepes buffer (HHBS) or an appropriate buffer.
4. Fill the cell wells with growth medium.
5. Analyze the cells using a fluorescence microscope with Cy5 filter or flow cytometer with 660/20 nm filter (APC channel).

EXAMPLE DATA ANALYSIS AND FIGURES

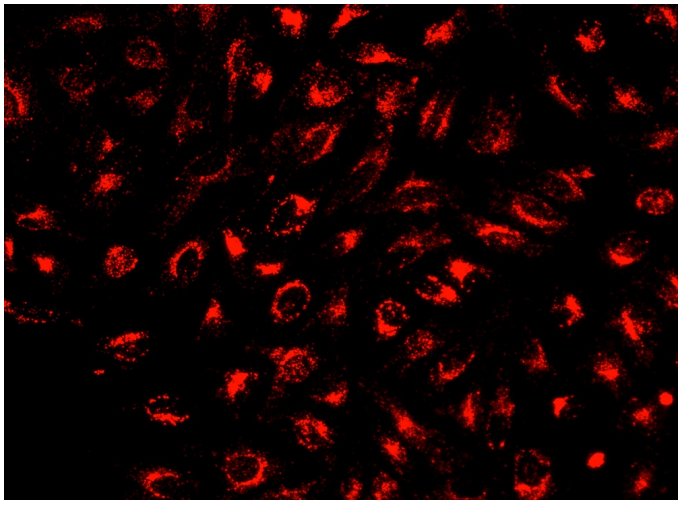

Figure 1. Image of HeLa cells stained with Cell Explorer™ Live Cell Tracking Kit (Cat#22624)in a Costar black wall/clear bottom 96-well plate. Cells were stained with Track It™ Deep Red for 15 minutes and image was aquired with fluorescence microscope using Cy5 filter.

DISCLAIMER

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