Cell Explorer™ Live Cell Tracking Kit *Blue Fluorescence*

Catalog number: 22620
Unit size: 200 Tests

Component	Storage	Amount
Component A: Track It™ Blue	Freeze (< -15 °C), Minimize light exposure	2 vials
Component B: DMSO	Freeze (< -15 °C)	1 vial (0.2 mL)
Component C: Assay Buffer	Freeze (< -15 °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 blue fluorescence for the studies that require the fluorescent tag molecules retained inside cells for relatively longer time. The kit uses a weakly fluorescent dye that carries a cell-retaining moiety. The dye becomes strongly fluorescent upon entering into live cells, and 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 1X Track It™ Blue working solution (100 µL/well)
3. Stain the cells at 37°C for 15 to 60 minutes
4. Wash the cells
5. Examine the specimen under fluorescence microscope with DAPI filter or flow cytometer with 450/40 nm filter (Pacific Blue channel)

Important Thaw all the components at room temperature before opening.

KEY PARAMETERS

Flow cytometer

Excitation	350 nm or 405 nm laser
Emission	450/40 nm filter
Instrument specification(s)	Pacific Blue channel

Fluorescence microscope

Excitation	DAPI filter
Emission	DAPI filter
Recommended plate	Black wall/clear bottom

CELL PREPARATION

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

PREPARATION OF STOCK SOLUTIONS

Unless otherwise noted, all unused stock solutions should be divided into single-use aliquots and stored at -20 °C after preparation. Avoid repeated freeze-thaw cycles.

Track It™ Blue stock solution (2 mM)

Add 25 µL of DMSO (Component B) into one vial of Track It™ Blue (Component A) and mix well to make 2 mM Track It™ Blue stock solution.

PREPARATION OF WORKING SOLUTION

Track It™ Blue working solution
Dilute 2 mM Track It™ Blue stock solution into Assay Buffer (Component C) to make 5 to 50 µM Track It™ Blue working solution.

Note This Track It™ Blue working solution should be prepared enough for all the wells at 100 µL/well with the appropriate concentration. For example, to get Track It™ Blue at the final concentration of 20 µM for one 96-well microplate, dilute 10 µL of the Track It™ Blue stock solution into 1 mL of Assay Buffer (Component C) to make 1 mL of 20 µM (1X) Track It™ Blue working solution.

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

Note We found that 2 µM final in well concentration is sufficient for most of cell lines.

SAMPLE EXPERIMENTAL PROTOCOL

1. Remove Growth medium, wash cells with PBS once.
2. Add 100 µL Track It™ Blue working solution (1X) to each well.
3. Incubate the cells in a 37°C, 5% CO2 incubator for 15 to 60 minutes.
4. Wash cells with Hanks and 20 mM Hepes buffer (HHBS) or an appropriate buffer.
5. Fill the cell wells with Assay Buffer or an appropriate buffer.
6. Analyze the cells using a fluorescence microscope with DAPI filter or flow cytometer with 450/40 nm filter (Pacific Blue channel).

EXAMPLE DATA ANALYSIS AND FIGURES

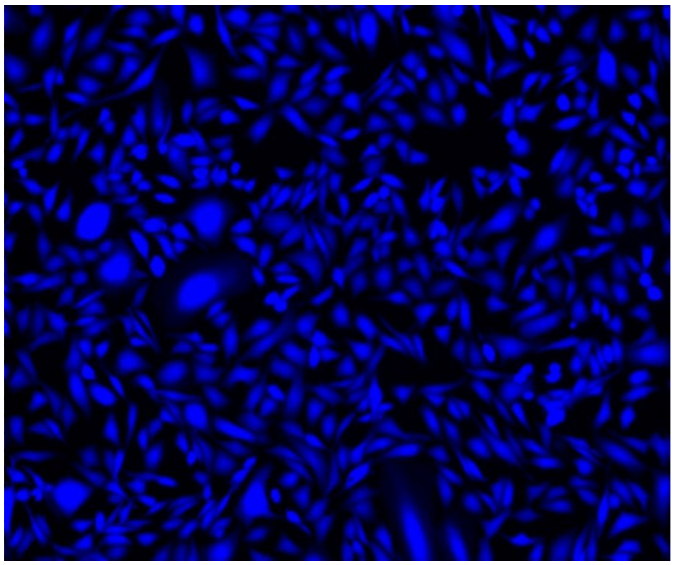

Figure 1. Image of HeLa cells stained with Cell Explorer™ Live Cell Tracking Kit (Cat#22620) in a Costar black wall/clear bottom 96-well plate. Cells were stained with Track It™ Blue for 15 minutes and image was aquired with fluorescence microscope using DAPI filter.

DISCLAIMER

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