

Cell Navigator™ Cell Plasma Membrane Staining Kit *Red Fluorescence*

Catalog number: 22681 Unit size: 500 Tests

Component	Storage	Amount
Component A: Cellpaint™ Deep Red	Freeze (< -15 °C), Minimize light exposure	1 vial
Component B: Assay Buffer	Freeze (< -15 °C)	1 bottle (50 mL)
Component C: DMSO	Freeze (< -15 °C)	1 vial (200 μL)

OVERVIEW

The Cell Navigator™ Plasma Membrane Stain Kit provides a fast and uniform labeling of the plasma membrane without the cell-type differences exhibited by lectins. It may be used as a segmentation tool for HCS (high-content screening), as well as to stain cellular plasma membranes for standard fluorescence microscopy. The stain used in the kit survives fixation, but not permeabilization, so it is not suitable for experiments that also involve probing internal targets via antibodies.

AT A GLANCE

Protocol Summary

- 1. Prepare cells in growth medium
- Incubate cells with Cellpaint™ Deep Red working solution at 37°C for 10 - 20 minutes
- Analyze the cells under fluorescence microscope at Ex/Em = 640/660 nm (Cy5 filter set)

Important Thaw all the kit components at room temperature before starting the experiment.

KEY PARAMETERS

Fluorescence microscope

Excitation Cy5 filter Emission Cy5 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.

Cellpaint™ Deep Red stock solution (500X)

Add 100 μL of DMSO (Component C) into the vial of Cellpaint TM Deep Red (Component A) to make 500X Cellpaint TM Deep Red stock solution. *Note: Protect from light. For storage, seal tubes tightly.*

PREPARATION OF WORKING SOLUTION

Add 20 µL of 500X Cellpaint™ Deep Red stock solution into 10 mL of Assay Buffer (Component B), and mix well to make Cellpaint™ Deep Red working solution. This Cellpaint™ Deep Red working solution is stable for at least 8 hours at room temperature. Protect from light. Note: 20 µL of 500X Cellpaint™ Deep Red 500X stock solution is enough for one 96-well plate.

SAMPLE EXPERIMENTAL PROTOCOL

- Add 100 µL/well (96-well plate) or 50 µL/well (384-well plate) of Cellpaint™ Deep Red working solution in the cell plate.
- Incubate the cells at 37°C for 10 20 minutes, protected from light.
 Note: The optimal concentration of the cell membrane probe varies depending on the specific application. The staining conditions may be modified according to the particular cell type and the permeability of the cells or tissues to the probe.
- 3. Remove Cellpaint™ Deep Red working solution in each well.
- Wash cells with physiological buffer (such as HHBS, PBS or buffer of your choice) for three times.
- Fix cells after staining (Optional). Fix the cells with 4% formaldehyde for 15 - 30 minutes. Wash cells with physiological buffer for three times.
- Observe the fluorescence signal in cells using a fluorescence microscope with Cy5 filter set (Ex/Em =640/660 nm).

EXAMPLE DATA ANALYSIS AND FIGURES

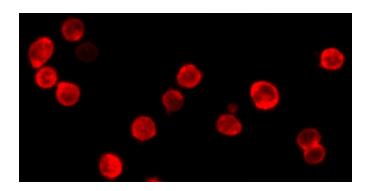


Figure 1. Fluorescence images of HL-60 cells stained with Cell Navigator™ Cell Plasma Membrane Staining Kit *Red Fluorescence* in a 96-well black wall/clear bottom plate. The cells were imaged using a fluorescence microscope equipped with a Cy5 filter set.

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