

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

 Catalog number: 22680
 Unit size: 500 Tests

Component	Storage	Amount
Component A: Cellpaint™ Orange	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
2. Incubate cells with Cellpaint™ Orange working solution at 37 °C for 10 - 20 minutes
3. Analyze the cells under fluorescence microscope at Ex/Em = 540/590 nm (TRITC filter set)

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

KEY PARAMETERS

Fluorescence microscope

Excitation	TRITC filter
Emission	TRITC 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™ Orange stock solution (500X)

Add 100 µL of DMSO (Component C) into the vial of Cellpaint™ Orange (Component A) to make 500X Cellpaint™ Orange stock solution. *Note: Unused 500X Cellpaint™ Orange stock solution can be stored at ≤ -20 °C for one month if the tubes are sealed tightly. Protect from light.*

PREPARATION OF WORKING SOLUTION

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

SAMPLE EXPERIMENTAL PROTOCOL

1. Add 100 µL/well (96-well plate) or 50 µL/well (384-well plate) of Cellpaint™ Orange working solution in the cell plate.
2. 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™ Orange working solution in each well.
4. Wash cells with physiological buffer (such as HBBS, PBS or buffer of your choice) for three times.
5. Fix cells after staining (Optional). Fix the cells with 4% formaldehyde for 15 - 30 minutes. Wash cells with physiological buffer for three times.
6. Observe the fluorescence signal in cells using a fluorescence microscope with TRITC filter set (Ex/Em = 540/590 nm).

EXAMPLE DATA ANALYSIS AND FIGURES

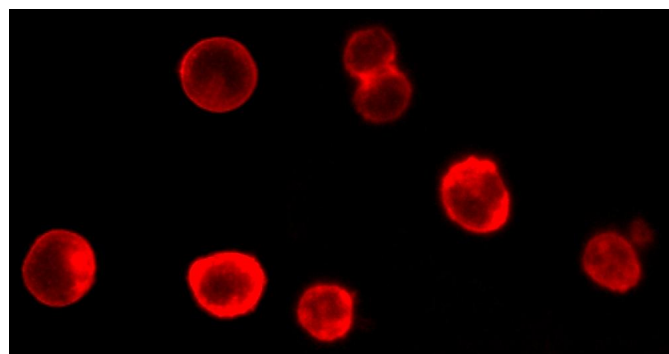


Figure 1. Fluorescence images of HL-60 cells stained with Cell Navigator™ Cell Plasma Membrane Staining Kit *Orange Fluorescence* in a 96-well black wall/clear bottom plate. The cells were imaged using a fluorescence microscope equipped with a TRITC filter.

DISCLAIMER

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