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TraKine™ Cell Plasma Membrane Staining Kit (Green Fluorescence)

Cat #: KTC4001 Size: 100 T/500 T/2000 T

	Cell Plasma Membrane Staining Kit (Green Fluorescence)		
REF	**Cat #:** KTC4001	LOT	**Lot #:** Refer to product label
	Fluorescence excitation/emission: 484 nm/501 nm		
	Storage: Stored at -20℃ for 6 months		

Assay Principle

The cell membrane (plasma membrane) is a thin semi-permeable membrane, consisting of a lipid bilayer with embedded proteins that separates the interior of all cells from the environment. The basic function of the cell membrane is to protect the cell from its surroundings. The cell membrane controls the movement of substances in and out of cells and organelles. In this way, it is selectively permeable to ions and organic molecules. Cell membranes are involved in a variety of cellular processes such as cell adhesion, ion conductivity and cell signaling and serve as the attachment surface for several extracellular structures, including the cell wall, glycocalyx, and intracellular cytoskeleton. TraKine™ Cell Plasma Membrane Staining Kit (Green Fluorescence) is a set of fluorescence imaging tool for rapid staining of plasma membranes in living and fixed suspended or attached cells depending on the cell type and experimental conditions. The kit uses a proprietary lipophilic carbocyanine dye (Ex/Em= 484/501 nm) that weakly fluorescent in water but highly fluorescent and quite photostable when incorporated into membranes.

Materials Supplied and Storage Conditions

Kit components	Size			Storage conditions
	100 T	500 T	2000 T	
CPM™ Green (1000×)	50 μL	250 μL	1 mL	-20℃, protect from light
Assay Buffer (10×)	5 mL	25 mL	100 mL	4℃

Materials Required but Not Supplied

- Centrifuge
- Pipettes and pipette tips
- Deionized water
- Fluorescence Microscopy or Flow Cytometer
- 24-well plate for cell culture
- Phosphate-buffered saline (PBS)

Reagent Preparation

CPM™ Green (1000×): Warm to room temperature. Aliquot and store unused CPM™ Green (1000×) stock solutions at -20℃. Protect from light and avoid repeated freeze-thaw cycles.

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1×Assay Buffer: Prepare 1×Assay Buffer by dilute 10×Assay Buffer with deionized water. Warm to 37℃ before use.

Staining Solution: Mix 1 μL of CPM™ Green (1000×) in each 1 mL 1×Assay Buffer. Scale up accordingly for larger numbers of assays.

Assay Procedure

Note: The optimal concentration of the CPM™ Green and incubation time varies depending on the specific application. The staining conditions may need modified according to the particular cell type.

A. Quantification by Flow Cytometry

1.Treat cells with the desired method.

Note: We recommend keeping unstained control cells (i.e. without CPM™ Green) suspended in Assay Buffer for both treated and untreated samples to set up the flow cytometer instrument.

2.For non-adherent cells, Collect 1-5×10^5 cells by centrifugation (4℃, 300 g, 5 min). Wash with ice-cold PBS twice and discard the PBS. For adherent cells, using Trypsin (EDTA free) to digest cells firstly and then centrifugation.

3.Resuspend the cells pellet in 500 μL Staining Solution.

4.Incubate the cells at 37℃ for 5-20 min in the dark.

5.Centrifuge cells at 500 g and discard supernatant.

6.Wash cell pellet with PBS and repeat step 5.

7.Resuspend cell pellet in 0.5 mL of the pre-warmed PBS and analyze the cells by flow cytometry using FITC channel (usually FL1).

B. Detection by Fluorescence Microscopy

1.For suspension cells: Follow the protocol for flow cytometry from step1 to step4 and place the cell suspension from Step A.4 on a glass slide. Cover the cells with a glass coverslip. Analyze cells by fluorescence microscopy using the appropriate filters as soon as possible.

2.For adherent cells: the suggested protocol is as below.

2.1. Grow cells directly on a coverslip in 24 well dish. Incubate in a CO_2 Incubator at 37℃ for at least 24 h before treatment.

2.2. Wash cells with PBS twice.

2.3. Add 0.5 mL of Staining solution to cells and incubate at 37℃ for 5-10 min in the dark.

2.4. Wash cells with pre-warm PBS twice.

2.5. Fix cells after staining (Optional): Fix the cells with 4% paraformaldehyde for 15-30 min. Other fixatives, particularly lutaraldehyde, tend to produce unacceptably high levels of background fluorescence.

2.6. Invert coverslip on a glass slide and visualize cells fluorescence microscopy using the appropriate filters as soon as possible (Ex/Em=484/501 nm).

Recommended Products

Catalog No.	Product Name
KTC4002	TraKine™ Cell Plasma Membrane Staining Kit (Orange Fluorescence)
KTC4003	TraKine™ Mitochondrion Staining Kit (Green Fluorescence)
KTC4004	TraKine™ Mitochondrion Staining Kit (Orange Fluorescence)
KTC4005	TraKine™ Mitochondrion and Nuclear Staining Kit
KTC4008	TraKine™ F-actin Staining Kit (Green Fluorescence)
KTC4009	TraKine™ F-actin Staining Kit (Orange Fluorescence)

Disclaimer

The reagent is only used in the field of scientific research, not suitable for clinical diagnosis or other purposes.

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