

# Product Name: Dilinoleyl Dil Membrane Probe (Orange-Red) Product number: RA20002

# **Basic Information**

| Product name | Dilinoleyl Dil Membrane Probe (Orange-Red) |
|--------------|--|
| Size         | 5 mg                                       |
| Storage      | Store at 4°C, away from light              |
| Shipping     | Shipped with ice pack                      |
| Validity     | 12 months                                  |

# **Reagent preparation**

### Staining solution preparation

(1) Preparation of stock solution: The stock solution is prepared with anhydrous DMSO or EtOH at a concentration of 1~10 mM.

Note: Unused stock solution should be stored in aliquots at -20°C to avoid repeated freezing and thawing.

(2) Preparation of working solution: Dilute the stock solution with a suitable buffer (e.g. serum-free culture medium, HBSS or PBS) to prepare a working solution with a concentration of 1-10  $\mu$ M.

Note: The final concentration of the working solution is recommended to be optimized according to different cell lines and experimental systems. It is recommended to start exploring the optimal concentration within a range of 10 times the recommended concentration.

## **Experimental procedures**

#### Suspension cell staining

(1) Add an appropriate volume of staining working solution to resuspend the cells to a density of  $1 \times 10^6$  cells/mL. (2) Incubate the cells at 37°C for 5 to 20 minutes. The optimal incubation time varies for different cells. You can use 20 minutes as the initial incubation time, and then optimize the system to obtain a uniform labeling effect.

(3) At the end of incubation, centrifuge at 1000-1500 rpm for 5 min. Pour off the supernatant and slowly add 37°C preheated growth medium to resuspend the cells.

(4) Repeat step (3) two more times.

### Adherent cell staining

(1) Culture adherent cells on sterile glass coverslips.

(2) Remove the coverslip from the medium, aspirating any excess, but leaving the surface wet.

(3) Add 100  $\mu$ L of dye working solution to one corner of the coverslip and gently shake to allow the dye to evenly cover all cells.

- (4) Incubate the cells at 37°C for 5 to 20 minutes. The optimal incubation time varies for different cells. You can use 20 minutes as the initial incubation time, and then optimize the system to obtain a uniform labeling effect.
- (5) Aspirate the dye working solution, wash the coverslip 2-3 times with culture medium, cover all cells with prewarmed culture medium each time, incubate for 5-10 min, and then aspirate the culture medium, but keep the surface moist.

Dilinoleyl Dil excitation/emission wavelength: 549/565 nm

## Note: This reagent is for scientific research use only!