

Product Name: DiA Membrane Probe (Green)

Product number: RA20001

Basic Information

Product name	DiA Membrane Probe (Green)
Size	50 mg
Storage	Store at 4°C, away from light
Shipping	Shipped with ice pack
Validity	12 months
Ex/Em	456/590 nm

Product Introduction

DiA (4-Di-16-ASP) is a lipophilic green fluorescent dye commonly used for labeling and imaging cell membranes. DiA exhibits weak fluorescence in aqueous solutions, but its fluorescence intensity significantly increases when it is incorporated into cell membranes or bound to lipophilic biomolecules. DiA is frequently used to label cell membranes and other hydrophobic structures, and it is suitable for staining both live and fixed cells. It can be used in combination with Dil for dual-color labeling, which facilitates the simultaneous observation of different cells or cellular structures.

Reagent preparation

(1) Preparation of stock solution: The stock solution is prepared with anhydrous DMSO, anhydrous DMF or EtOH, with a concentration of 1-5 mM. The solubility of DiA in anhydrous DMSO and anhydrous DMF is higher than that in EtOH.

Note:

- Unused stock solution should be stored in aliquots at -20°C to avoid repeated freezing and thawing;
 - If the solution is difficult to dissolve, it can be heated appropriately and ultrasonicated to promote dissolution.
- (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 1-30 μ M. The most commonly used working solution concentration is 5-10 μ 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

- Add an appropriate volume of staining working solution to resuspend the cells to a density of 1×10^6 cells/mL.
- Incubate the cells at 37°C for 2-20 min. The optimal incubation time varies for different cells. You can use 20 min as the initial incubation time, and then optimize the system to obtain a uniform labeling effect.
- 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.
- Repeat step (3) two more times.

Adherent cell staining

- Culture adherent cells on sterile glass coverslips.
- Remove the coverslip from the medium, aspirating any excess, but leaving the surface wet.
- Add 100 μ L of dye working solution to one corner of the coverslip and gently shake to allow the dye to evenly cover all cells.
- Incubate the cells at 37°C for 2-20 min. The optimal incubation time varies for different cells. You can use 20 min as the initial incubation time, and then optimize the system to obtain a uniform labeling effect.
- Aspirate the dye working solution, wash the coverslip 2-3 times with culture medium, cover all cells with pre-warmed culture medium each time, incubate for 5-10 min, and then aspirate the culture medium, but keep

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the surface moist.

Note: This reagent is for scientific research use only!