Product Name: DiO

Catalog Number: RA20005

Basic Information

Product Name	DiO
Size	10mg
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 DMSO, anhydrous DMF or EtOH , with a concentration of 1-5 mM . The solubility of DiO in anhydrous DMSO and anhydrous DMF is higher than that in EtOH .

Note: a. Unused storage solution should be stored in aliquots at -20℃ to avoid repeated freezing and thawing;

b. 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 μM. The most commonly used working solution concentration is 5-10 μM.

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 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 1×10^{6}/mL.

(2) 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.

(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) Adherent cells were cultured on sterile coverslips.
(2) Remove the coverslip from the medium, aspirating any excess, but leaving the surface wet.
(3) Add 100 µL 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 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.
(5) Aspirate the dye working solution, wash the coverslip with culture medium 2-3 times, cover all cells with pre-warmed culture medium each time, incubate for 5-10 min, and then aspirate the culture medium. However, keep the surface moist.

DiO excitation/emission wavelength: 484/501nm

Note: This reagent is for scientific research use only!