Product Information

DiO (DiOC18(3))

Catalog Number: D4007

Product Size: 10 mg

Application Scope: Labeling cytoplasmic membrane and intracellular membrane, cell tracing

Parameters

Appearance: Yellow solid

Solubility: DiO is soluble in absolute ethanol, DMSO and DMF, and the solubility in DMSO is about 10 mg / ml. When it is difficult to dissolve, you can heat or sonicate appropriately.

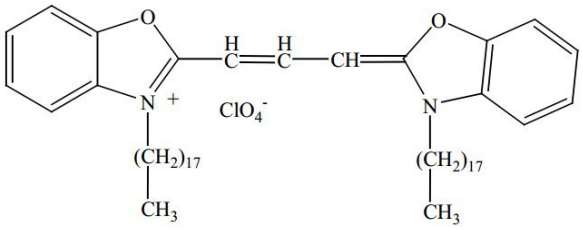
Ex/Em (MeOH): 484/501 nm

CAS No.: 34215-57-1

Molecular Formula: $C_{53}H_{85}ClN_2O_6$

Molecular Weight: 882

Molecular Structure:



Storage

Store at -20℃ and protect from light. When stored as directed, product is stable for at least 12 months.

Description

DiO, also called DiOC18(3), is a green fluorescent, lipophilic carbocyanine dye that is widely used as a lipophilic tracer. It is weakly fluorescent in water but highly fluorescent and quite photostable when incorporated into membranes.

DiO is widely used as anterograde and retrograde neuronal tracers in living and fixed tissues and cells. DiI labeling does not appreciably affect cell viability.

In addition to fluorescent labeling of cell membranes, DiO can also be used to detect cell fusion and adhesion, cell migration during development or transplantation, detect lipid diffusion in cell membranes by FRAP (Fluorescence Recovery After Photobleaching), detect cytotoxicity, and label lipoproteins.

Cells can be fixed with formaldehyde either before or after staining, but not recommended for permeabilization. DiO staining is usually less intense than that of DiI, and occasionally fails completely in fixed tissues.

Protocol

1. Dyeing liquid preparation

(1) Stock solution: Use DMSO, DMF or EtOH to make a stock solution with a concentration of 1 to 5 mM.

Note: It is recommended to store the storage solution at -20 ℃, and aliquot it in small quantities to avoid repeated freeze-thaw cycles.

(2) Preparation of working solution: Dilute the storage solution with a suitable buffer (such as serum-free medium, HBSS or PBS) to prepare a working solution with a concentration of 1 to 30 μM. The most common working solution concentration is 5-10 μM.

Note: You may need to optimize the staining procedure for each particular cell type by varying the dye concentration, staining volume, labeling time, or wash steps.

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2. Suspension cell staining

(1) Suspend cells at a density of 1×10^6/mL in working solution.

(2) Incubate for 20 minutes at 37°C. The optimal incubation time will vary depending on cell type. Start with 20 minutes and optimize as needed for uniform labeling.

(3) Pellet the cells by centrifugation at 1000～1500 rpm for 5 minutes.

(4) Remove the supernatant and wash the cells by gently resuspending them in warm (37°C) medium.

(5) Repeat the centrifugation and wash steps (Steps 3 and 4) two more times.

(6) Image fluorescence. Cells can be imaged in culture medium.

3. Adherent cell staining

(1) Remove growth medium from the cells.

(2) Add enough working solution to completely cover the cells.

(3) Incubate the cells at 37°C. The optimal incubation time will vary depending on the cell type. Start with 20 minutes and optimize as needed for uniform labeling.

(4) Remove the working solution.

(5) Wash the cells by adding fresh warm growth medium and incubating at 37°C for 5 minutes. Repeat this wash step two more times.

(6) Image fluorescence. Cells can be imaged in culture medium.

Notes

1. When DiO stains fixed cell or tissue samples, the samples should be fixed with 4% paraformaldehyde in PBS. The use of other improper fixing solutions will result in a high fluorescence background.

2. There are quenching problems with fluorescent dyes. Please avoid light to slow down the fluorescence quenching.

3. For your safety and health, please wear lab coats and disposable gloves.

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