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DiD perchlorate

Cat. No.: HY-D1028

CAS No.: 127274-91-3

Molecular Formula: $C_{61}H_{99}ClN_2O_4$

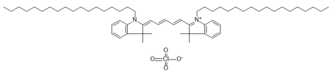
Molecular Weight: 959.9

Target: Fluorescent Dye

Pathway: Others

Storage: 4°C, protect from light, stored under nitrogen

* The compound is unstable in solutions, freshly prepared is recommended.



SOLVENT & SOLUBILITY

In Vitro

DMSO : 25 mg/mL (26.04 mM; ultrasonic and warming and heat to 60°C)

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	1.0418 mL	5.2089 mL	10.4178 mL
	5 mM	0.2084 mL	1.0418 mL	2.0836 mL
	10 mM	0.1042 mL	0.5209 mL	1.0418 mL

Please refer to the solubility information to select the appropriate solvent.

In Vivo

1. Add each solvent one by one: 10% DMSO >> 40% PEG300 >> 5% Tween-80 >> 45% saline
   Solubility: ≥ 1.67 mg/mL (1.74 mM); Clear solution
2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)
   Solubility: ≥ 1.67 mg/mL (1.74 mM); Clear solution
3. Add each solvent one by one: 10% DMSO >> 90% corn oil
   Solubility: ≥ 1.67 mg/mL (1.74 mM); Clear solution

BIOLOGICAL ACTIVITY

Description

DiD is a long-chain carbocyanine dye. Carbocyanine dyes are widely used as Di to label cells, organelles, liposomes, viruses and lipoproteins[2].

In Vitro

General Protocol

1. Preparing Stain Solutions of Di

1) Prepare DMF, DMSO or ethanol stock solutions: The stock solutions should be prepared in dimethyl formamide (DMF), dimethylsulfoxide (DMSO, or ethanol DMSO at 1-5 mM. DMF is preferable to ethanol as a solvent for Di. The stock solution should be used promptly. Any unused solution need to be aliquoted and refrozen at least -20℃. Avoid repeated freeze/thaw cycle. The solution can be stored for 6 months.

	2) Prepare working solutions: Dilute the stock solutions into a suitable buffer such as serum-free culture medium, HBSS or PBS to make 1 to 5 μM working solutions. We do not recommend storing the aqueous solution for more than one day. Note: The final concentration of the working solution should be empirically determined for different cell types and/or experimental conditions. 2. Suspension cells 1) Centrifuge at 1000 g at 4℃ for 3-5 minutes and then discard the supernatant. Wash twice with PBS, 5 minutes each time.The cell density is 1×10^6/mL 2) Add 1 mL of Di working solution, and then incubate at room temperature for 5-30 minutes. 3) Centrifuge at 400 g at 4℃ for 3-4 minutes and then discard the supernatant. 4) Wash twice with PBS, 5 minutes each time. 5) Resuspend cells with serum-free cell culture medium or PBS.Observation by fluorescence microscopy or flow cytometry. 3. Adherent cells 1) Culture adherent cells on sterile coverslips. 2) Remove the coverslip from the medium and aspirate the excess medium. 3) Add 100 μL of working solution, gently shake it to completely cover the cells,and then incubate at room temperature for 5-30 minutes. 4) Wash twice with medium, 5 minutes each time. Observation by fluorescence microscopy or flow cytometry. MCE has not independently confirmed the accuracy of these methods. They are for reference only.
In Vivo	Two weeks after injection of stained cells, a single, bright, DiD perchlorate (DiD)-positive cells located between 15 to 40 μm from the endosteum is found. Results reveal a progressive appearance of cell clusters of decreased dye intensity, consistent with the partitioning of DiD perchlorate label on cell division[3]. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL

Animal Administration [1]	1 to 5×10^5 cells per mL HSPCs are stained with 5 μM DiD perchlorate (DiD) or 7.5 μM DiI in PBS without serum for 10 min at 37°C, washed once in PBS and immediately injected into the tail vein of recipient mice. Unless stated differently, each imaged CD45.2 mouse receive 8,000 to 15,000 labelled CD45.1 HSPCs together with 3 to 5×10^5 CD45.2 supportive whole bone-marrow mononuclear cells to ensure survival[1]. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

CUSTOMER VALIDATION

- Chem Eng J. 2024 Feb 16, 149761.
- J Control Release. 2021 Jun 2;S0168-3659(21)00283-2.
- Discov Nano. 2024 Jan 4;19(1):4.
- Research Square Preprint. 2023 Aug 7.

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REFERENCES

[1]. Gan WB, et al. Multicolor "DiOlistic" labeling of the nervous system using lipophilic dye combinations. Neuron. 2000 Aug;27(2):219-25.

[2]. Lo Celso C, et al. Live-animal tracking of individual haematopoietic stem/progenitor cells in their niche.

[3]. Kenji Yumoto, et al. A novel method for monitoring tumor dormancy using fluorescent dye DiD. Cytometry A. 2014 Jun; 85(6): 548–555.

[4]. Meng Li, et al. In Vivo Tracking of Human Adipose-derived Mesenchymal Stem Cells in a Rat Knee Osteoarthritis Model with Fluorescent Lipophilic Membrane Dye. J Vis Exp. 2017; (128): 56273.

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