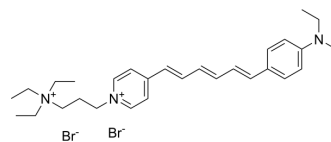


FM4-64

Cat. No.:	HY-103466
CAS No.:	162112-35-8
Molecular Formula:	C ₃₀ H ₄₅ Br ₂ N ₃
Molecular Weight:	607.51
Target:	Fluorescent Dye
Pathway:	Others
Storage:	-20°C, sealed storage, away from moisture and light * In solvent : -80°C, 6 months; -20°C, 1 month (sealed storage, away from moisture and light)



SOLVENT & SOLUBILITY

In Vitro	DMSO : 50 mg/mL (82.30 mM; Need ultrasonic)					
	Preparing Stock Solutions	Solvent	Mass	1 mg	5 mg	10 mg
		Concentration				
		1 mM		1.6461 mL	8.2303 mL	16.4606 mL
		5 mM		0.3292 mL	1.6461 mL	3.2921 mL
	10 mM		0.1646 mL	0.8230 mL	1.6461 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 mg/mL (1.65 mM); Clear solution 2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline) Solubility: ≥ 1 mg/mL (1.65 mM); Clear solution					

BIOLOGICAL ACTIVITY

Description	FM4-64 is a very lipophilic, water-soluble styrene dyes, can specifically bind to cell membranes and inner membrane organelles to produce fluorescence. FM4-64 is widely used in endocytic and exospic membrane structure markers.
In Vitro	1. Preparation of FM working solution 1.1 Preparation of the stock solution Dissolve FM in DMSO to obtain 5 mM of FM. Note: It is recommended to store the stock solution at -20°C or -80°C away from light and avoid repetitive freeze-thaw cycles. 1.2 Preparation of FM working solution Dilute the stock solution in HBSS to obtain 5-20 μM of FM working solution. Note: Please adjust the concentration of FM working solution according to the actual situation. 2. Cell staining

2.1 Suspension cells (6-well plate)

- a. Centrifuge at 1000 g at 4°C 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
- b. Add 1 mL of working solution, and then incubate at room temperature for 5-30 minutes.
- c. Centrifuge at 400 g at 4°C for 3-4 minutes and then discard the supernatant.
- d. Wash twice with PBS, 5 minutes each time.
- e. Resuspend cells with serum-free cell culture medium or PBS. Observation by fluorescence microscopy or flow cytometry.

2.2 Adherent cells

- a. Culture adherent cells on sterile coverslips.
- b. Remove the coverslip from the medium and aspirate excess medium.
- c. Add 100 µL of working solution, gently shake it to completely cover the cells, and then incubate at room temperature for 5-30 minutes.
- d. Wash twice with medium, 5 minutes each time. Observation by fluorescence microscopy or flow cytometry.

Storage

-20°C, 1 year.

Protect from light.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

CUSTOMER VALIDATION

- ACS Nano. 2023 Mar 27.

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REFERENCES

- [1]. J E Gale, et al. FM1-43 dye behaves as a permeant blocker of the hair-cell mechanotransducer channel. *J Neurosci.* 2001 Sep 15;21(18):7013-25.
- [2]. Zal T, et al. Spectral shift of fluorescent dye FM4-64 reveals distinct microenvironment of nuclear envelope in living cells. *Traffic.* 2006;7(12):1607-1613.
- [3]. Stenovec M, et al. Distinct labelling of fusion events in rat lactotrophs by FM 1-43 and FM 4-64 is associated with conformational differences. *Acta Physiol (Oxf).* 2007;191(1):35-42.
- [4]. Bolte S, et al, Satiat-Jeunemaitre B. FM-dyes as experimental probes for dissecting vesicle trafficking in living plant cells. *J Microsc.* 2004;214(Pt 2):159-173.

Caution: Product has not been fully validated for medical applications. For research use only.

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