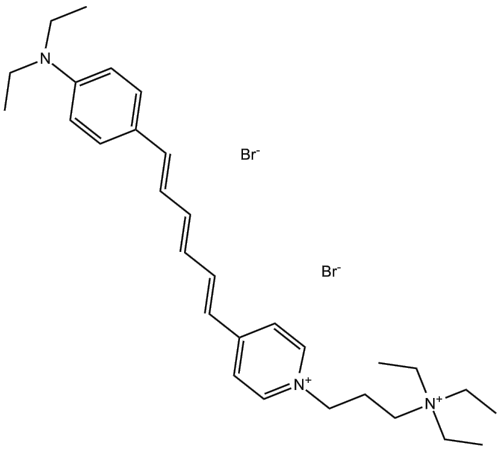
Product Data Sheet

Product Name: SynaptoRedTM C2
Cat. No.: GC14835

Chemical Properties

Cas. No.	162112-35-8		
Chemical Name	4-((1E,3E,5E)-6-(4-(diethylamino)phenyl)hexa-1,3,5-trien-1-yl)-1-(3-(triethylammonio)propyl)pyridin-1-ium bromide		
SMILES	CC[N+](CC)(CC)CCC[N+]1=CC=C(/C=C/C=C/C=C/C2=CC=C(C=C2)N(CC)CC)C=C1.[Br-].[Br-]		
Formula	$C_{30}H_{45}Br_2N_3$	M.Wt	607.51
Solubility	DMSO : 50 mg/mL (82.30 mM; Need ultrasonic)	Storage	Store at -20°C
General tips	For obtaining a higher solubility , please warm the tube at 37 °C and shake it in the ultrasonic bath for a while.Stock solution can be stored below -20°C for several months.		
Shipping Condition	Evaluation sample solution : ship with blue ice All other available size: ship with RT , or blue ice upon request.		

Structure



Protocol

This plan only provides a guide, please modify it to meet your specific needs.

1. Solution preparation

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(1) Stock solution: Prepare SynaptoRedTM C2 stock solution with DMSO or EtOH at a concentration of 1-5mM.

Note: After unused stock solution is aliquoted, store it at -20°C or -80°C away from light to avoid repeated freezing and thawing.

(2) Working solution: Dilute the stock solution with a suitable buffer (such as serum-free culture medium, HBSS or PBS) to prepare SynaptoRedTM C2 working solution with a concentration of 5-20μM.

Note: The working solution must be prepared and used immediately. The final concentration of the working solution needs to be optimized according to different cell lines and experimental systems. It is recommended to start from the recommended concentration and explore the optimal concentration in a 10-fold range.

2. Suspended cell staining

(1) Suspended cells are centrifuged at 4°C and 1000rpm for 3-5min and the supernatant is discarded. Wash twice with PBS for 5min each time.

(2) Add 1mL of SynaptoRedTM C2 working solution and incubate at room temperature in the dark for 5-30min. The optimal culture time varies for different cells.

(3) After incubation, centrifuge at 1000-1500rpm for 5min, remove the supernatant, and wash 2-3 times with PBS, 5min each time.

(4) Resuspend the cells in pre-warmed serum-free cell culture medium or PBS. Observe by fluorescence microscopy or flow cytometry.

3. Staining of adherent cells

(1) Culture adherent cells on sterile coverslips.

(2) Remove the coverslip from the culture medium, aspirate the excess culture medium, and place the coverslip in a humid environment.

(3) Add 100μL SynaptoRedTM C2 working solution from one corner of the coverslip and gently shake to evenly cover all cells with the dye.

(4) Incubate at room temperature in the dark for 5-30min. The optimal culture time varies for different cells.

(5) Aspirate the dye working solution and wash the coverslip 2-3 times with culture medium. Observe by fluorescence microscopy.

Note: All fluorescent dyes have quenching problems. Please try to avoid light to slow down fluorescence quenching. For your safety and health, please wear a lab coat and disposable gloves when operating.

Background

SynaptoRedTM C2 is a membrane-selective red fluorescent dye and a highly lipophilic water-soluble styrene dye that can specifically bind to cell membranes and endomembranous organelles to produce fluorescence with an excitation wavelength of 530-560nm and an emission wavelength of 590-650nm[1, 2]. SynaptoRedTM C2 can be used as a marker for endocytic and exocytic membrane structures and is also used to track synaptic activity by staining synaptic vesicles at synapses or neuromuscular junctions[3]. SynaptoRedTM C2 can be used to stain bacteria (Gram-positive or Gram-negative) and yeast, but is rapidly internalized into the vacuole membrane in yeast[4, 5].

References:
[1] Betz W J, Mao F, Bewick G S. Activity-dependent fluorescent staining and destaining of living vertebrate motor nerve terminals[J]. Journal of Neuroscience, 1992, 12(2): 363-375.
[2] Kang S Y, Pokhrel A, Bratsch S, et al. Engineering Bacillus subtilis for the formation of a durable living biocomposite material[J]. Nature communications, 2021, 12(1): 7133.
[3] Fischer-Parton S, Parton R M, Hickey P C, et al. Confocal microscopy of FM4-64 as a tool for analysing endocytosis and vesicle trafficking in living fungal hyphae[J]. Journal of microscopy, 2000, 198(3): 246-259.
[4] Wang H Y, Hua X W, Jia H R, et al. Universal cell surface imaging for mammalian, fungal, and bacterial cells[J]. ACS Biomaterials Science & Engineering, 2016, 2(6): 987-997.
[5] Fischer-Parton S, Parton R M, Hickey P C, et al. Confocal microscopy of FM4-64 as a tool for analysing endocytosis and vesicle trafficking in living fungal hyphae[J]. Journal of microscopy, 2000, 198(3): 246-259.