

BioTracker™ 510 Green C18(FM3-25) Synaptic Dye

Cat. # SCT134

FOR RESEARCH USE ONLY.
NOT FOR USE IN DIAGNOSTIC PROCEDURES.
NOT FOR HUMAN OR ANIMAL CONSUMPTION.

pack size: 5 mg

Store at 2-8°C



Data Sheet

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Background

BioTracker™ synaptic dyes are a series of fluorescent cationic styryl dyes developed to follow synaptic activities at neuromuscular junctions or synapses. These dyes typically have a lipophilic tail at one end and a highly hydrophilic at the other end. In the presence of cells or tissue preparations, the dyes partition between the aqueous phase, where the dyes are virtually non-fluorescent, and the outer leaflet of the cell surface membranes, where the dyes insert the lipophilic end into the membranes and become intensely fluorescent. During endocytosis following nerve stimulation, the dyes become trapped inside the vesicles. Thus, after washing off the dyes on the cell surface, the fluorescent signal is proportional to the number of newly formed vesicles. On the other hand, during exocytosis, the dyes are released from the vesicles along with neurotransmitters, causing a decrease in fluorescent signal. As a result, the change in fluorescent intensity reflects the amount of endocytosis/exocytosis or synaptic activity.

BioTracker™ 510 Green C18(FM3-25) Synaptic Dye has the longest lipophilic tail (C18). Because of its large size, it is used as a control for ion channel permeability of nerve terminal dyes.

Storage

Store BioTracker™ 510 Green C18(FM3-25) Synaptic Dye at 2-8°C, protected from light. Stock solutions can be prepared at 10 mM and stored at 4°C or -20°C for six months or longer.

Spectral Properties

Absorbance (MeOH): 510 nm
Emission (MeOH): 625 nm

Absorbance (Membranes): 480 nm
Emission (Membranes): 600 nm

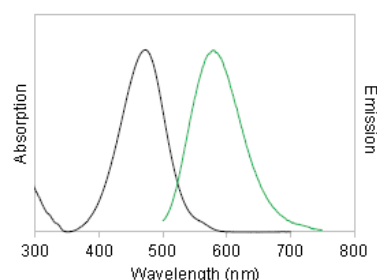


Figure 1. Absorption and emission spectra of BioTracker™ 510 Green C18(FM3-25) Synaptic Dye in liposomes.

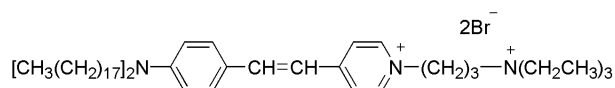


Figure 2. Chemical structure of the BioTracker™ 510 Green C18(FM3-25) Synaptic Dye.

Assay Protocol

The following is an example of a protocol for nerve terminal staining of cultured neurons on coverslips. Nerve terminal dyes also can be used to label endocytic vesicles in non-neuronal cell types. Staining can be performed at 4°C for selective labeling of the plasma membrane; at room temperature or 37° C, endocytosis of the dye generally occurs within 10 minutes. Buffers other than Tyrode solution may be used. The addition of the sodium channel blocker tetrodotoxin (TTX) is optional, its purpose is to block action potentials and prevent synaptic vesicle release after staining. Optimal protocols for specific applications may need to be determined by the user.

1. Dilute BioTracker™ synaptic dye to a final concentration of 4 μ M in 50 mM Tyrode solution. Place the coverslip with your cells in this solution for 1 minute at room temperature. Use enough solution to completely submerge the cells.
2. Transfer the coverslip to Tyrode + 0.5 μ M tetrodotoxin (TTX) solution for 1 minute at room temperature.
3. Wash the coverslip several times in Tyrode + TTX at room temperature.

Note: to reduce background, 1 mM ADVASEP-7 can be added to the wash solution. Alternatively, SCAS can be used to quench background without repeated washes. Incubate the coverslip for 4 minutes at room temperature in Tyrode + TTX + 0.5 mM SCAS.

4. Mount the coverslip in Tyrode + TTX and image.

Note: 50 μ M sulforhodamine 101 can be included during mounting to quench extracellular fluorescence.

Note: BioTracker™ synaptic dyes are not fixable.

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EMD Millipore Corporation, 28820 Single Oak Drive, Temecula, CA 92590, USA 1-800-437-7500

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