

BioTracker™ 664 NIR Ca²⁺ Dye

Live Cell Dye

Cat. # SCT022

pack size:1mg

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

Store at -20°C



Data Sheet

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Background

Calcium (Ca²⁺) play a vital role in the physiology and biochemistry of the cell. Calcium ions play an important role in signal transduction pathways, neurotransmitter release from neurons, contraction of all muscle cell types, and in fertilization. Extracellular calcium is also important for maintaining the potential difference across excitable cell membranes, as well as proper bone formation.

The BioTracker™ 664 NIR Ca²⁺ Dye is a live cell far-red fluorescent calcium indicator. The dye changes fluorescent intensity greatly (1000-fold) when it binds to calcium. Multicolor imaging is possible between the dye and Hoechst, Fluorescein, Rhodamine, GFP, YFP and RFP etc., which are fluorescent probes or fluorescent proteins having fluorescent wavelength from UV region to visible area. The advantages of long wavelength region are greater tissue penetration and low phototoxicity.

BioTracker™ 664 NIR Ca²⁺ Dye can be introduced by microinjection, patch clamp and electroporation. BioTracker™ 664 NIR Ca²⁺ AM Dye is an acetoxymethyl ester version and can permeate cell membranes.

Storage

Store BioTracker 664 NIR Ca²⁺ Dye at -20°C, desiccate and protect from light

Note: Centrifuge vial briefly to collect contents at bottom of vial before opening.

Spectral Properties

Absorbance: 650nm

Emission: 664nm

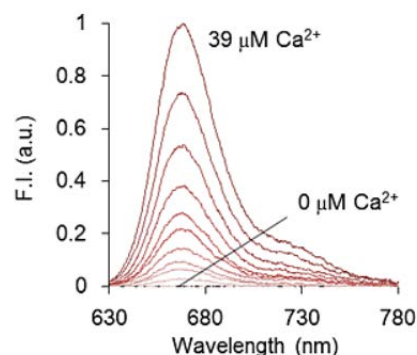


Figure 1. Fluorescent spectra of BioTracker 664 NIR Ca²⁺ Dye was measured (ex: 620nm) in the presence of various concentrations of Ca²⁺. When Ca²⁺ concentration was fluctuated from 0 μM to 39 μM, fluorescent intensity raised more than 1000-fold.

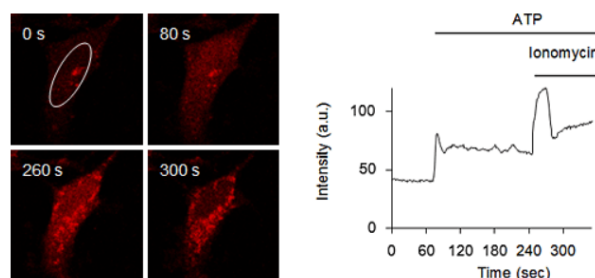


Figure 2. Live cell imaging of Ca²⁺ ions. BioTracker 664 NIR Ca²⁺ Dye was loaded to HeLa cells and stimulated by ATP and ionomycin. Although a part of probe localized to lysosome, it was possible enough to visualize Ca²⁺ concentration change.

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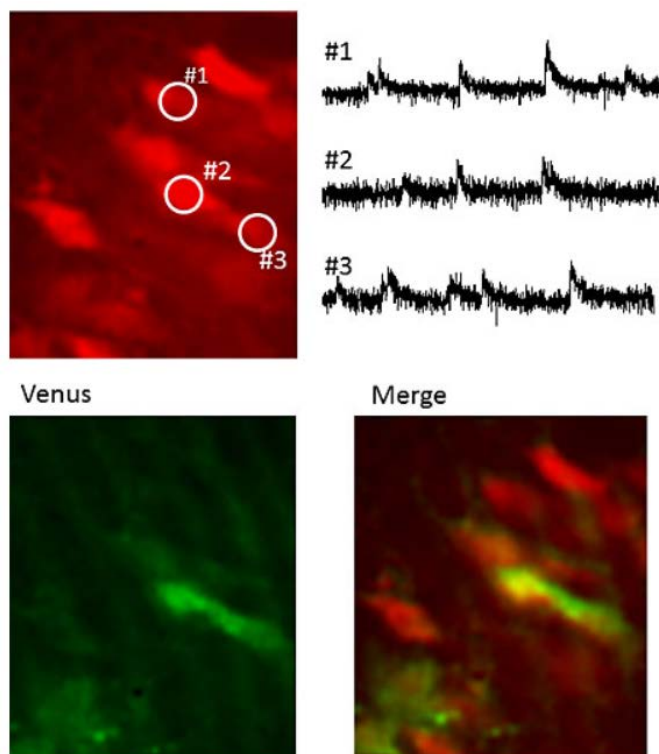


Figure 3. Ca²⁺ imaging of mouse cranial nerve. BioTracker 664 NIR Ca²⁺ AM Dye was added to a mouse brain slice in which a fraction of neurons expressed Venus (a mutant of YFP) and live cell Ca²⁺ imaging was analyzed. Ca²⁺ is shown as red color and Venus is shown green.

Protocol

Reagent Preparation

1. Before opening the vial, spin down the solid to the bottom by a microcentrifuge or by a desktop centrifuge.
2. Dissolve 50µg of dye in 46 µL of DMSO to achieve a 1 mM concentration. To improve the induction efficiency and inhibit localization of the probe, the addition of Pluronic F-127 is recommended.

Staining Protocol of Cultured Cells

1. Dilute an aliquot of stock dye solution to a final concentration of 1-10 µM in appropriate loading medium or buffer such as HBSS. Final concentration of Pluronic F-127 is around 0.01-0.05 %.
2. Remove the culture medium from cell culture dish and wash with loading medium.
4. Add stain solution to the dish and incubate 10 to 60 minutes under 37°C, 5% CO₂ conditions.
5. After staining, remove the stain solution from the dish and wash 2 or 3 times by medium or buffer which is not contain dye. Replace to HBSS buffer and observe the changes of intracellular fluorescence intensity using a fluorescence microscopy.