Data Sheet

BioTracker™ 488 Green Mitochondria Dye

Live Cell Dye

SCT136

Pack Size 20 x 50 µg

Store at -20 °C

FOR RESEARCH USE ONLY

Not for use in diagnostic procedures. Not for Human or Animal Consumption.

Background

Mitochondria are organelles found in large numbers in most cells, in which the biochemical processes of respiration and energy production occurs. In metabolically active cells, mitochondria produce a membrane potential by maintaining a proton gradient across the inner and outer membranes. Loss of mitochondrial membrane potential is a hallmark for apoptosis and poor cell health.

BioTracker™ Mitochondria dyes are fluorogenic stains for staining mitochondria in live cells. The dyes are membrane permeable and become brightly fluorescent upon accumulation in the mitochondrial membrane. Staining depends on mitochondrial mass, and not on mitochondrial membrane potential. Signal is lost as mitochondrial membrane integrity is lost during cell death allowing monitoring of cell viability.

The BioTracker[™] 488 Green Mitochondria Dye can be detected using the green FITC/GFP channel. Also, the green staining is not dependent on membrane potential, and can be used to stain both live and fixed cells.

Kit Components

20 vials of 50 µg lyophilized dye

Spectral Properties

Absorbance: 490 nm Emission: 523 nm



1

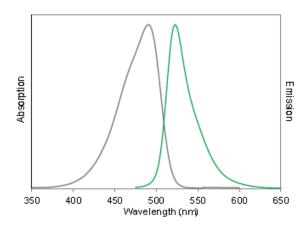


Figure 1. Excitation and emission spectra of BioTracker[™] 488 Green Mitochondria Dye.

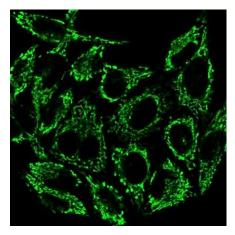


Figure 2. Hela cells stained with BioTracker[™] 488 Green Mitochondria Dye.

Storage and Handling

Store BioTracker™ 488 Green Mitochondria Dye at -20 °C. Protect from Light.

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

Protocols

Reconstitution

Prepare 200 μ M stock solution, dissolve one 50 μ g vial of lyophilized dye in 400 μ L anhydrous DMSO or DMF. Store at -20 °C.

Live Cell Staining

 When cells are at appropriate confluence, remove the medium and add pre-warmed medium containing 100 nM BioTracker™ Mitochondria dye. For suspension cells, pellet the cells and resuspend in medium containing diluted BioTracker™ Mitochondria dye.

Note: The optimal staining concentration may vary by cell type and application. We recommend performing an initial test with the dyes at concentrations between 20-200 nm. At higher concentrations, other structures may be stained.

Note: Alternatively, the dye can be added directly to the culture medium. We recommend making a dilute stock solution in culture medium to avoid exposing the cells to a transient high concentration of dye. For example, dilute dye to 10 times the final desired concentration in culture medium, and then add 1/10 volume of the dilute stock to the medium on the cells and mix well by gently pipetting up and down.

2. Incubate cells for 15 minutes or longer at 37 °C. Washing is not required before imaging.

Note: Longer staining times may result in brighter staining. BioTracker[™] Mitochondria dyes show no obvious toxicity at 100 nM in MCF-7 cells with incubation times up to 72 hours, but toxicity may vary by cell type.

3. Analyze fluorescence by fluorescence microscopy or flow cytometry using the appropriate excitation/emission settings or detection channel (see Spectral Properties).

Note: BioTracker[™] Mitochondria dyes are not well-retained after fixation. For fixed cell staining with BioTracker[™] Mitochondria Green, we recommend fixation before staining (see below). Other BioTracker[™] Mitochondria dyes cannot be use in fixed cells.

Staining of Fixed Cells

- 1. Fix cells in 4% formaldehyde in PBS for 10 minutes at room temperature.
- 2. Following fixation, rinse cells in PBS and incubate with Biotracker™ 488 Green Mitochondria dye.
- 3. Rinse cells with PBS before imaging.

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