MitoStain™ Mitochondria Stain

Catalog #	Description
135-1320	**MitoStain Lime Green 399/550 Mitochondria Stain**
135-1321	**MitoStain Green 498/520 Mitochondria Stain**
135-1322	**MitoStain Orange 545/575 Mitochondria Stain**
135-1323	**MitoStain Red 575/600 Mitochondria Stain**
135-1324	**MitoStain Deep Red 640/659 Mitochondria Stain**

For research purposes only.

Description

MitoStain is an easy-to-use probe-based solution for visualizing mitochondria in mammalian cells by fluorescence microscopy.

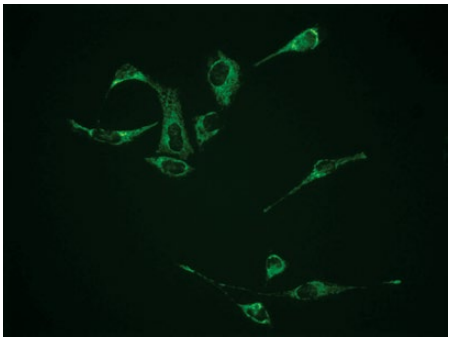
Mitochondria are small organelles (0.5–1 µm) found in most eukaryotic cells. Though the primary function of mitochondria is to produce energy through respiration, these structures are also involved in many other essential activities, such as cell signaling, cellular differentiation, control of the cell cycle, cell growth, and cell death. The number of mitochondria in a cell can vary by organism and tissue type, and the proper identification of these organelles is fundamental to biological research.

Bio-Rad's MitoStain takes advantage of a fluorogenic probe that permeates the plasma membranes of live cells and selectively accumulates in mitochondria (Figure 1). The probe includes a cell-retaining group that traps the fluorescence signal within the mitochondria, thereby significantly increasing staining efficiency.

The MitoStain family of products can be used with both suspension and adherent cells, as well as proliferating and nonproliferating cells.

MitoStain Mitochondria Stains are available in five different excitation/emission wavelength combinations (Table 1) to enable multicolor experiments using the most common fluorescence cell imagers and microscopes.

A



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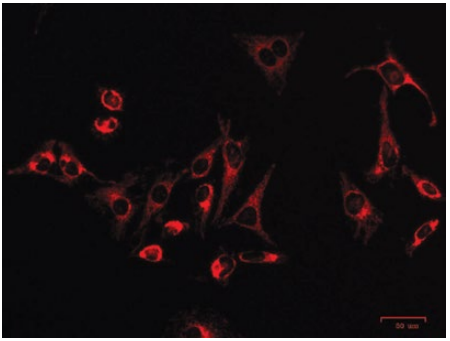

Fig. 1. Staining of mitochondria with 1x MitoStain Green 498/520 (A) and 1x MitoStain Red 575/600 (B). HeLa cells were stained with MitoStain and visualized with the ZOE™ Fluorescent Cell Imager.

Contents and Storage

Each box contains five vials of MitoStain (20 µl of 500x stain per vial). Store MitoStain at –20°C. Protect from light and avoid repeated freeze-thaw cycles.

Ordering Information

Use Table 1 to select the appropriate MitoStain for any given cell imaging experiment.

Table 1. Ordering information.

Catalog Number	Description	Optimal Channel on ZOE Fluorescent Cell Imager	Excitation, nm	Emission, nm	Fixability
135-1320	MitoStain Lime Green 399/550	—	399	550	—
135-1321	MitoStain Green 498/520	Green	498	520	—
135-1322	MitoStain Orange 545/575	Red	545	575	+
135-1323	MitoStain Red 575/600	Red	575	600	+
135-1324	MitoStain Deep Red 640/659	—	640	659	+

Protocol

This protocol provides guidelines and should be modified according to your specific needs.

Preparing the Working Solution

1. Warm MitoStain to room temperature and centrifuge briefly before opening.
2. Prepare a working solution of 1x MitoStain by diluting the stock solution 1:500 in cell growth medium.

Note 1: One vial (20 µl) of MitoStain makes 10 ml of 1x working solution, which is sufficient for one 96-well plate.

Note 2: Optimal concentration of MitoStain varies depending on the specific application. Staining conditions may be modified according to the particular cell type.

Preparing and Staining Adherent Cells

1. Grow the cells according to your experimental procedure. When cells reach the desired confluence, replace the cell growth medium with an equal volume of 1x MitoStain (see Preparing the Working Solution section).
2. Incubate the cells in a 37°C, 5% CO_2 incubator for 30 min.
3. Wash the cells twice with prewarmed (37°C) phosphate buffered saline (PBS) or buffer of your choice. Keep the cells in growth medium.
4. Observe the cells using a ZOE Fluorescent Cell Imager or any fluorescence microscope equipped with an appropriate filter set and light source.

Preparing and Staining Suspension Cells

1. Spin down the cells and replace the cell growth medium with an equal volume of 1x MitoStain (see Preparing the Working Solution section). Resuspend the cells gently and thoroughly.
2. Incubate the cells in a 37°C, 5% CO_2 incubator for 30 min.
3. Wash the cells twice with prewarmed (37°C) PBS or buffer of your choice.
4. Keep the cells in growth medium; mount an aliquot on a slide under a coverslip if desired.
5. Observe the cells using a ZOE Fluorescent Cell Imager or any fluorescence microscope equipped with an appropriate filter set and light source.

Visit **bio-rad.com/web/MitoStain** for more information.

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