

MitoDNA™ Red 610

Catalog number: 22687
Unit size: 1 mg

Component	Storage	Amount (Cat No. 22687)
MitoDNA™ Red 610	Freeze (< -15 °C), Minimize light exposure	1 mg

OVERVIEW

There are very few probes that can be effectively used to detect mitochondrial deoxyribonucleic acid (mtDNA). The common fluorescent DNA probes (such as DAPI, Hoechst or SYBR® Green) lack the specificity to target mitochondria. They predominantly stain nuclei. MitoDNA™ Red 610 is a cell permeable dye that specifically stains mtDNA in live cells. It provides an efficient way of labeling mtDNA for the dynamic imaging of mtDNA in live cells. MitoDNA™ Red 610 has a large Stokes Shift that gives great signal-to-noise ratio and can be easily used for multiplex staining in cells with other fluorescent imaging probes. mtDNA is a small circular DNA found within mitochondria present in the cytoplasm of a cell. This DNA is supplementary to the nucleic acid material found in the nucleus of each cell. The mtDNA codes for 37 genes that promote the proper functioning of some cells. The mitochondria synthesize adenosine triphosphate (ATP) through oxidative phosphorylation and encode information for the synthesis of enzymes, transfer ribonucleic acid (tRNA), and ribosomal RNA (rRNA). Disorders of mtDNA and mutations in its genes can predispose to health problems like age-related hearing loss, diabetes, and brain, heart, and liver failure, among other conditions. Moreover, mtDNA and its associated mitochondrial disorders can predispose people to different types of cancers including lymphomas, leukemias, and breast, intestine, liver and kidney tumors etc.

AT A GLANCE

Important Note

Before using MitoDNA™ Red 610 for the first time, allow it to thaw at room temperature. Then, briefly centrifuge it to collect the dried pellet.

Protocol Summary

1. Prepare cells in a growth medium.
2. Stain cells with MitoDNA™ Red 610 working solution.
3. Incubate samples for 5 to 15 minutes at 37 °C.
4. Monitor fluorescence intensity at Ex/Em = 490/610 nm.

KEY PARAMETERS

Fluorescence microscope

Emission	610 nm
Excitation	490 nm
Recommended plate	Black wall/clear bottom

PREPARATION OF STOCK SOLUTIONS

Unless otherwise noted, all unused stock solutions should be divided into single-use aliquots and stored at -20 °C after preparation. Avoid repeated freeze-thaw cycles

MitoDNA™ Red 610 Stock Solution

1. Prepare a 5 to 10 mM MitoDNA™ Red 610 stock solution in DMSO. For example, add 290 µL of DMSO to the MitoDNA™ Red 610 vial to create a 10 mM stock solution.

Note: Prepare a single aliquot of the unused MitoDNA™ Red 610 stock solution and store it at ≤ -20 °C, protected from light. Avoid repeated freeze-thaw cycles.

PREPARATION OF WORKING SOLUTION

MitoDNA™ Red 610 Working Solution

1. Prepare a 5 to 10 µM working solution by diluting the MitoDNA™ Red 610 stock solution in Hanks' solution with 20 mM HEPES buffer (HHBS).

Note: For optimal results, use this solution within a few hours of preparation.

Note: Cover the working solution with foil or store it in a dark place to protect it from light.

SAMPLE EXPERIMENTAL PROTOCOL

1. Plate the cells in a 96-well plate with black walls and a clear bottom.
2. Remove the cell culture medium and add 100 µL of MitoDNA™ Red 610 working solution directly to the cells.
3. Incubate the cells at 37°C for 5-15 minutes, protected from light.
4. Remove the dye working solution and wash the cells twice with HHBS buffer.
5. Add HHBS buffer and analyze the cells using a fluorescence microscope with excitation/emission settings of 490/610 nm.

Note: The concentration and incubation time of MitoDNA™ Red 610 may vary depending on the cell line. Test different concentrations to determine the optimal dose.

EXAMPLE DATA ANALYSIS AND FIGURES

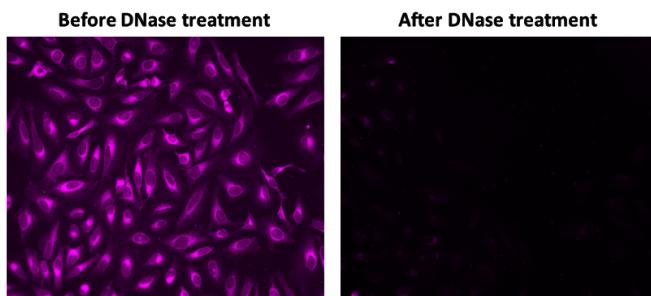


Figure 1. The fluorescence response of MitoDNA™ Red 610 (5 μ M) in HeLa cells was assessed before and after DNase treatment. Fluorescence intensities were monitored using fluorescence microscopy.

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