Product datasheet

TMRE-Mitochondrial Membrane Potential Assay Kit
ab113852

Overview

Product name: TMRE-Mitochondrial Membrane Potential Assay Kit
Detection method: Fluorescent
Sample type: Adherent cells, Suspension cells
Assay type: Cell-based (qualitative)
Assay time: 0h 35m

Product overview

TMRE-Mitochondrial Membrane Potential Assay Kit ab113852 is used for quantifying changes in mitochondrial membrane potential in live cells by flow cytometry, microplate spectrophotometry and fluorescent microscopy.

ab113852 uses TMRE (tetramethylrhodamine, ethyl ester) to label active mitochondria. TMRE is a cell permeant, positively-charged, red-orange dye that readily accumulates in active mitochondria due to their relative negative charge. Depolarized or inactive mitochondria have decreased membrane potential and fail to sequester TMRE.

The TMRE protocol also uses FCCP (carbonyl cyanide 4-(trifluoromethoxy) phenylhydrazone), which is a ionophore uncoupler of oxidative phosphorylation. Treating cells with FCCP eliminates mitochondrial membrane potential and TMRE staining. TMRE is suitable for the labeling of mitochondria in live cells and is not compatible with fixation.

TMRE protocol summary:
- add FCCP to appropriate control cell samples and incubate for 10 min
- incubate with TMRE for 15-30 min, pellet (suspension cells) / remove media (adherent cells) and wash with PBS / 0.2% BSA
- analyze with micro-plate reader at Ex/Em 549/575 nm, flow cytometer using 488nm laser for excitation and at emission 575 nm, or fluorescent microscope.

Notes

TMRE is only suitable for use with live (not fixed) cells.

Related assays

Review the cell health assay guide to learn about kits to perform a cell viability assay, cytotoxicity assay and cell proliferation assay.

Review the metabolism assay guide to learn about assays for metabolites, metabolic enzymes, mitochondrial function, and oxidative stress, and also about how to assay metabolic function in
live cells using your plate reader.

**Platform**

Microplate reader, Fluor. microscope, Flow cyt.

**Properties**

**Storage instructions**

Store at -20°C. Please refer to protocols.

<table>
<thead>
<tr>
<th>Components</th>
<th>200 tests</th>
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<tbody>
<tr>
<td>1 mM TMRE (in DMSO)</td>
<td>1 x 40μl</td>
</tr>
<tr>
<td>50 mM FCCP (in DMSO)</td>
<td>1 x 10μl</td>
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**Relevance**

Mitochondrial Membrane Potential is an important parameter of mitochondrial function used as an indicator of cell death. The collapse of the mitochondrial Membrane potential coincides with the opening of the mitochondrial permeability transition pores, leading to the release of cytochrome c into the cytosol, which in turn triggers other downstream events in the apoptotic cascade.

**Images**

P19 neurons (750 cells/mm²) were exposed to MDMA on days 7–9 in serum-free medium for 10 min up to 48 hours. The positive control FCCP (carbonyl cyanide-p-trifluoromethoxyphenylhydrazone), an uncoupler of mitochondrial oxidative phosphorylation, was applied at the concentration of 5 μM for 10 min. The cells were incubated with 500 μM TMRE for 30–45 min at 37°C, 5% CO₂, followed by washing once with 100 μl of HBSS containing 0.2% bovine serum albumin. A volume of 200 μL of HBSS containing 0.2% bovine serum albumin was added to each well, and the fluorescence was measured with excitation/emission: 544/590 nm.

**A:** HeLa cells (adherent) were cultured on coverslips and stained with ab113852 (200nM TMRE) for 20 minutes in media, washed briefly with PBS and immediately imaged. **B:** Jurkat cells (suspension) were stained and washed as above and then transferred to a slide and immobilized under a coverslip for imaging.
Analysis of TMRE staining using a fluorescent plate reader and a microplate.

Chart showing mean fluorescent intensity +/- standard deviation from quadruplicate measurements of 400 nM TMRE stained Jurkat cells in a 96-well microplate +/- treatment with FCCP.

Flow Cytometry histogram of Jurkat cells stained with ab113852 (100nM TMRE) with (blue) or without (red) treatment with 100µM FCCP.

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