JC-10 (10mM)

Cat #: orb1154752 (manual)

For Research Use Only. Not For Use in Diagnostic Procedures!

Catalog No.	Size
orb1154752-100	100 tests
orb1154752-200	200 tests
orb1154752-500	500 tests

Molecular Weight: ~600

Solvent: DMSO

Storage Condition: Aliquot and store at -20℃. Avoid freeze and thaw. Term of validity is one year since production.

Product introduction

JC-10 is a new cation lipophilic indicator for fluorescence to detect the potential of the membrane of mitochondria in flow cytometry. It is able to enter mitochondria selectively, showing a green fluorescence to red fluorescence reversibly, when the potential of mitochondria increases (more than 80~120mV). The characteristic originates from the reversible form of JC-10 aggregated on the membrane, leading to the emission wavelength changing from 520 nm to 570 nm (520 nm correspond to the monomer of JC-10, and 570 nm correspond to the polymers). When 490 nm activating, the mitochondria polarized, and the color of JC-10 changed from green to red. The two colors usually can be detected by flow cytometers with different channels. FL1 channel can analyze green fluorescence and FL2 channel can analyze green orange fluorescence. Solid color is used to detect for quantitative analysis, which can be applied to two channels of FL1 and FL2. JC-10 is suitable for various detection of cell lines such as neuron and muscle cells. It can also be applied to the procedure such as ATP synthesis, ROS, cell apoptosis. JC-10 is also suitable for fluorescent microscope and fluorescent microplate reader analysis.

JC-10 aggregates in the mitochondria when the membrane potential is low, existing with monomers; With the increase of membrane potential, more JC-10 aggregated in mitochondria matrix aggregate to form J-aggregations, leading to the emission wavelength changed from 520 nm to 570 nm. JC-10 aggregates is a reflection of membrane polarization. The strength of JC-10 fluorescence indicates the depolarization and hyperpolarization situation of mitochondria membrane.

JC-10 have more advantages than JC-1: good solubility, simple operation, time-saving, strong signal, good signal-to-noise ratio, little errors, good repeatability.

Assay Protocol

1. Preparation of JC- 10 working solution

Biorbyt Ltd
7 Signet Court, Swann Road, Cambridge, CB5 8LA. United Kingdom
Email: info@biorbyt.com | Phone: +44 (0)1223 859-353 | Fax: +44 (0)1223 280-240

Biorbyt LLC
68 TW Alexander Drive, Durham, NC 27709. United States
Email: info@biorbyt.com | Phone: +1 (415) 906-5211 | Fax: +1 (415) 651-8558

1. 1 Prepare a conservative solution of 2 ~ 10 mM, anhydrous DMSO. The conservative solution should be used in time or aliquot at -20°C for conservation.

Note: Avoid repeated freeze-thaw cycles, and protect from light.

1.2 Preparation of 1 X JC- 10 working solution: dissolve the JC- 10 solid with DMSO or thaw the aliquoted conservative solution of JC- 10 when you current use it. Prepare 10 ~30 μmol/L 1 X JC- 10 working solution in HHBS buffer or the buffer you need and then mix well by vertexing.

Note: for some cell lines, working solution at pH 8 might prevent JC- 10 leakage.

2. JC- 10 detection by microplate reader:

2. 1 Compounds were used to detect cells to achieve ideal state and reduce cell apoptosis. For example, Jurkat cells use camptothecin to detect 4 ~6 hours. Blank control (medium) was added with the same amount of compound buffer.

2.2 96-well plate were added 100 μL or 384-well each well, and add 25 μL JC- 10 working reagent (see 1.2) to cell culture plate.

2.3 The plate added JC- 10 incubate in the incubator at 37°C,5%CO2 for 15 ~60 min. Note: Appropriate incubation time is determined according to cell type and cell concentration. Every experiment needs to optimize incubation time.

2.4 Remove the JC- 10 working reagent in the culture plate, HHBS or the buffer needed for washing cells. Add 100 μL HHBS buffer per well into 96-well plate or 25 μL per well into 384-well plate.

2.5 Detect the fluorescence change of Ex/Em =490/525 and 490/590, and analyze the value.

3 JC- 10 detection by fluorescence microscope or flow cytometry

3. 1 Compounds can detect cells to achieve ideal state and reduce cell apoptosis (for example, Jurkat cells use camptothecin to detect 4 ~6 hours).

3.2 Counting 1 ~5 x 10^5/ per tube for cell centrifugation;

3.3 Resuspend cells with 500 μL JC- 10 working reagent (see 1.2);

3.4 Incubate at R.T. or 37°C for 10 ~30 mins protect from light.;

3.5 Wash cell with HHBS buffer or the buffer you need. Resuspend cells with 500 μL HHBS buffer, 1 ~5 X105cells for each tube.

3.6 Detect the fluorescence change of Ex/Em =490/525 and 490/590 with fluorescence microscope (FITC channel and TRITC channel) or flow cytometer (FL1 channel and FL2 channel).

Cautions:

1 For your safety and health, please wear protective clothes and disposal gloves.

2 This compound has cell cytotoxicity, potential carcinogenicity and Teratogenicity. Avoid direct contact.