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Technical Information

MitoFlamma® Green

Overview

MitoFlamma® Green is a mitochondria-selective green fluorescent dye that allows to detect mitochondrial morphology in living cells. MitoFlamma® Green appears to preferentially accumulate in mitochondria regardless of mitochondrial membrane potential in certain cell types, making it a possible tool for determining mitochondrial mass. MitoFlamma® Green enables researchers to observe mitochondrial activity, localization and abundance as well as monitoring the effect of drugs or other external stimuli on the mitochondrial function. MitoFlamma® Green probes are retained in the mitochondria during the fixation step and after permeabilization with some detergents during subsequent processing steps. After fixation, labeled samples can be applied in a variety of experiments such as immunocytochemistry, *in situ* hybridization, microplate-based analysis, etc.

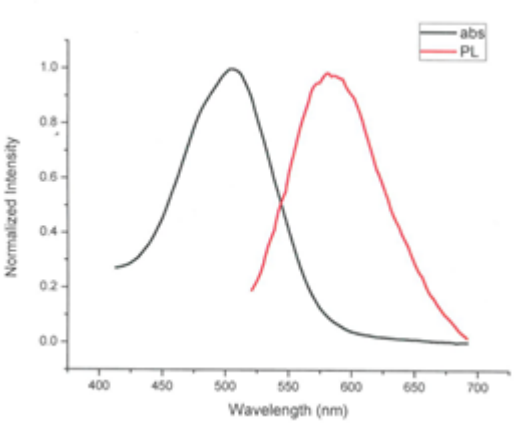

Figure 1. Excitation/emission spectra of MitoFlamma® Green

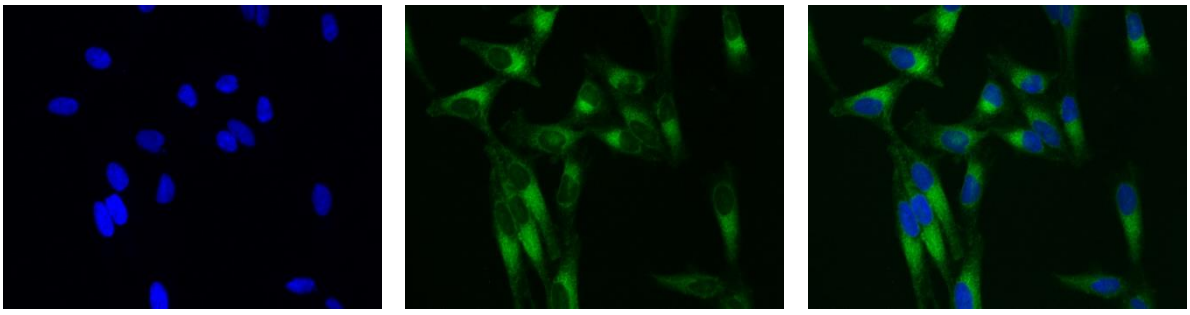

Figure 2. HeLa cell staining with MitoFlamma® Green
Left: DAPI, middle: MitoFlamma Green, right: merged image

Before beginning of mitochondrial labeling Experiment

Materials and equipment required but not provided

- DMSO
- PBS buffer or suitable growth medium for live cell imaging
- Aldehyde based fixatives such as paraformaldehyde for cell fixation
- Aldehyde based detergents such as Trition® X-100
- Micropipette
- Fluorescence microscope
- 37 ℃ incubator

Experimental protocols

Labeling of mitochondria in fixed or suspended cells with MitoFlamma® Green

1. Prepare the fixed/suspended cells by seeding appropriate cells onto confocal dishes 6 hours before.
2. Adjust the temperature of the media to 37 ℃.
3. Prepare a 1 μM labeling MitoFlamma® Green solution by diluting original 1 mM DMSO solution with the media from step 3, *protect from light.
4. Aspirate the culture medium from the cells grown dishes (for suspended cells, centrifuge the dishes in advance), and add 1 μM labeling MitoFlamma® Green solution to the dishes and incubate for 1 h at 37 ℃ under 5% CO_2 in humid condition. *the incubation may be varied for optimal labeling condition.
5. Aspirate the labeling media, and rinse the cells twice with PBS at 37 ℃.
6. Analyze stained cells with appropriate equipment

Fixation (continuing from step 5 in labeling experiment)

1. Fix the cells for 15 min in 4%parnformaldehyde at 37 ℃.
2. Aspirate the fixative, and rinse the cell twice with PBS.
3. View the cells using FITC filter set.

Permeabilization (optional)

- When permeabilization is needed for subsequent steps such as immunocytochemistry, incubate fixed cells in buffer containing detergent such as 0.2% Triton® X-100.
- Following permeabilization, rinse the cells in buffer and proceed with immunocytochemistry procedure.
- Alternatively, the cells may be permeabilized by incubating in ice-cold acetone for 5 minutes, and then washed in PBS.
- Even when cells are not going to be labeled with an antibody, this acetone-permeabilization step may be useful in improving the signal-to background ratio.

Custom Labeling Service

Based on accumulated know-how and technologies, BioActs provide a wide range of custom services such as protein fluorescence labeling, organic synthesis, oligonucleotide synthesis upon customers' request. Our reliable technology has acknowledged by our clients from domestic and overseas universities, institutions, in vitro diagnostic and pharmaceutical companies and has enabled to steadily conduct their requirements. In addition, we can introduce fluorescent materials to many other compounds such as organic and inorganic compounds, drugs, hormones, polymer, peptides, proteins, antibodies, etc. We also can provide chemical and optical analytical data, along with cell and animal experiments.


Nucleic acid


Peptide/Protein


Antibody


Small molecules /Polymer

Technical Support

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SDS (Safety Data Sheets) You can find SDS at www.bioacts.com, the official website of BioActs.
CoA (Certificate of Analysis) provides detailed quality information of each product. To see CoA, check the lot number written on each product's page at www.bioacts.com, when having trouble with check, contact to our technical support team