PRODUCT INFORMATION



LysoBrite™ NIR

Item No. 25155

Ex./Em. Max: 636/650 nm Supplied as: A solution in DMSO

-20°C Storage: Stability: ≥1 year

Information represents the product specifications. Batch specific analytical results are provided on each certificate of analysis.

Description

LysoBrite™ reagents are fluorogenic probes that can be used to label lysosomes within live cells. They are hydrophobic and easily cross live cell membranes to accumulate in lysosomes likely via the lysosomal pH gradient. LysoBrite™ fluorescence is enhanced in the acidic environment within the lysosome, and this fluorescence can be measured using fluorescence microscopy, microplate fluorometry, or flow cytometry. LysoBrite[™] NIR exhibits excitation/emission maxima of 636/650 nm, respectively.

Assay Protocol

1. Prepare lysosome-staining solution

- a. Warm the LysoBrite™ NIR solution to room temperature.
- b. Dilute 20 µl of 500X LysoBrite™ NIR into 10 mL of Hanks balanced salt solution (HBSS) or buffer of your choice.

Note 1: 20 µL of 500X LysoBrite™ NIR yields a volume of lysosome-staining solution sufficient for one 96-well plate. Aliquot and store unused LysoBrite™ NIR stock solution at less than -15°C. LysoBrite™ NIR is light sensitive. Light exposure and repeated freeze-thaw cycles should be avoided

Note 2: The optimal working concentration is application specific. Staining conditions may be modified according to cell type and/or permeability.

2. Prepare and stain the cells

a. Adherent cells:

- Grow cells in a 96-well black wall/clear bottom plate (100 µL/well) or on coverslips inside a petri dish filled with appropriate culture medium.
- When cells reach the desired confluency, add 100 µL of the lysosome-staining solution prepared in step 1b.
- iii. Incubate cells at 37°C, 5% CO₂ for 30 minutes to 2 hours.
- iv. Wash cells twice with pre-warmed (37°C) HBSS or buffer of your choice, then fill wells with buffer or growth medium.
- Observe cells using fluorescence technique of choice.

b. Suspension cells*:

- Add and equal volume of lysosome-staining solution prepared in step 1b to cells.
- iii. Incubate cells at 37°C, 5% CO₂ for 30 minutes to 2 hours.
- iv. Wash cells twice with pre-warmed (37°C) HHBS or buffer of your choice, then fill wells with buffer or growth medium.
- Observe cells using fluorescence technique of choice.

*Note 3: Suspension cells may be attached to coverslips that have been treated with BD Cell-Tak $^{ ext{ iny R}}$ (BD Biosciences) and stained as adherent cells.

Note 4: If cells are not sufficiently stained it is recommended to increase either the labeling concentration or the incubation time to increase cellular dye accumulation.

WARNING
THIS PRODUCT IS FOR RESEARCH ONLY - NOT FOR HUMAN OR VETERINARY DIAGNOSTIC OR THERAPEUTIC USE.

This material should be considered hazardous until further information becomes available. Do not ingest, inhale, get in eyes, on skin, or on clothing. Wash thoroughly after handling. Before use, the user must review the complete Safety Data Sheet, which has been sent via email to your institution.

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