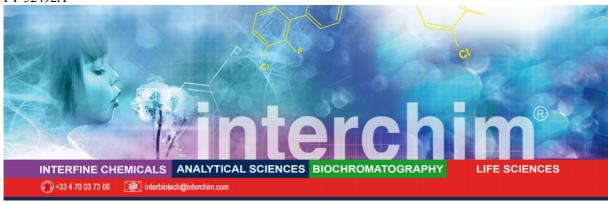
FT-52492A



Dihydroethidium

 O_2^- probes to use in flow cytometry or microplate assays

Product Information

Name: Dihydroethidium (DHE)

also called Hydroethidium, Hydroethidine

Catalog Number: FP-52492A, 25 mg

FP-52492B, 10 x 1 mg *Special Air - free Packaging*

FP-524929, 20 x 50 μg

Structure : $C_{21}H_{21}N_3$ Molecular Weight : MW=315

Solubility: In DMSO or DMF

Absorption / Emission : $\lambda_{\text{exc}} \lambda_{\text{em}}$ (hydrolysed, dsDNA) = 518 / 605 nm

 $\lambda_{\text{exc}} \setminus \lambda_{\text{em}} \text{ (free)} = 355 / \text{none}$

EC (M^{-1} cm⁻¹): (355)= 14 000

Storage: $-20^{\circ}\text{C} > 1 \text{ year.}$ (j). Protect from light and moisture

Introduction

Dihydroethidium is the chemically reduced form of the commonly used DNA dye ethidium bromide. Dihydroethidium can passively cross the membrane of living cells and not bind to nucleic acids. It is itself weakly fluorescent in the blue range in cytoplasm, where it is re-oxidized by intracellular enzymes. Upon DNA intercalation, it becomes red fluorescent. The probe is useful to detect oxidative activities in viable cells, including respiratory burst in phagocytes, oxidation in resting leukocytes, and to detect multidrug-resistant cancer cells. DHE is uncharged and is not selectively accumulated in the mitochondrial matrix. As such, the fluorescent DHE oxidation product reports cytoplasmic and intermembrane space superoxide levels, although DHE may also be oxidized by hydrogen peroxide.

Directions for use

Guidelines for use – fibroblastic type of cells o

The working concentration and time should be depermined for each cell type.

1) Prepare a stock solution by mixing 7 mg of dihydroethidium in 1 ml of DMSO or DMF. Mix 20 μ l of this solution with 10 ml of PBS, then filter it.



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2) Staining of surface adherent cells:

The adherent cells are incubated with the staining soltion for 15 min, at room temperature, in darkness. Then remove the staining solution and wash the cells 3 times with the buffer . Deposit one drop of the buffer on a slide. Remove the coverslip from the petri dish, wipe gently the side opposite to the cells with a tissue and place the coverslip vertically on the slide with the cells facing the drop of buffer. Drop the coverslip and gently remove excess liquid with a tissue.

Staining of cells in suspension:

Prepare the cell suspension and centrifuge to obtain a cell pellet: add the staining solution and suspend gently. Incubate 15 min at room temperature, in darkness. The cells are centrifugated to remove the staining solution and wash the pellet once with buffer. Place a drop of the suspended cells on a slide and cover with a coverslip. Gently remove excess liquid with a tissue.

Note: the stained cells can be placed in medium again and cultured for various periods of time to follow the destaining process and visualize those structures which hold to the dyes more firmly. Be to maintain sterile conditions.

Related products

- Ethidium bromide, <u>FP-06022A</u>
- Ethidium homodimer 1, <u>FP-25810A</u>
- H₂DCFDA, <u>FP-467312</u>

- DCFDA, <u>FP-BB4320</u>
- TMRE, <u>FP-41391A</u>

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Ordering information

Catalog size quantities and prices may be found at http://www.fluoprobes.com Please inquire for higher quantities (availability, shipment conditions).

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