

## Dihydrorhodamine 123

### Chemical Properties

CAS No. : 109244-58-8

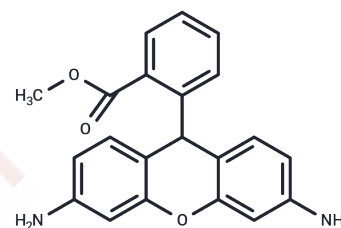
Formula: C<sub>21</sub>H<sub>18</sub>N<sub>2</sub>O<sub>3</sub>

Molecular Weight: 346.38

Storage: Keep away from direct sunlight, Store at low temperature, Keep away from moisture

Powder: -20°C for 3 years

Actual storage temperature shall be subject to the COA.



### Biological Description

Description	Dihydrorhodamine 123 (DHR 123) is a fluorescent probe specific for intracellular reactive oxygen species (ROS) ( $\lambda_{ex}=488$ nm, $\lambda_{em}=525$ nm), exhibiting high permeability to cell membranes and fluorescence activation upon oxidation. Dihydrorhodamine 123 is a classic tool for detecting cellular oxidative stress, reactive oxygen species (ROS) production, and antioxidant screening.
Targets(IC50)	Others
In vitro	<p><b>Methods:</b> Primary human neutrophils were pretreated with 25 <math>\mu</math>M Dihydrorhodamine 123 for 0–60 min, followed by 30 sec UV-C irradiation or PMA (25 nM) stimulation. ROS levels in primary neutrophils were detected using a fluorometer.</p> <p><b>Results:</b> PMA stimulation produced strong signals, while UV irradiation generated only an initial false-positive response. [1]</p>
Cell Research	<p>I. Detection of Reactive Oxygen Species (ROS)</p> <ol style="list-style-type: none"> <li>1. Solution preparation: Dissolve DHR 123 in an appropriate solvent (such as DMSO or PBS) at a concentration of typically 1–10 <math>\mu</math>M.</li> <li>2. Cell staining: Add DHR 123 solution to cultured cells and incubate at 37°C for typically 30–60 min.</li> <li>3. Oxidation process: ROS produced in cells oxidize DHR 123, converting it into fluorescent products.</li> <li>4. Fluorescence measurement: After staining, measure the fluorescence of the oxidation products using a fluorescence spectrophotometer or fluorescence microscope with an excitation wavelength of 488 nm and an emission wavelength of 525 nm.</li> <li>5. Analysis: Analyze ROS levels in cells by fluorescence intensity. Experiments can be performed in real time or at fixed time points.</li> </ol> <p>II. Assessment of mitochondrial function and membrane potential</p> <ol style="list-style-type: none"> <li>1. Cell incubation: Incubate cells as described above.</li> <li>2. Mitochondrial ROS detection: Oxidized DHR 123 will show a fluorescent signal under a fluorescence microscope, indicating the generation of mitochondrial ROS.</li> <li>3. Fluorescence microscopy observation: Observe the fluorescent signal to see the distribution of ROS in specific areas of the cell (especially mitochondria).</li> </ol> <p>III. Flow cytometry for ROS quantitative analysis</p> <ol style="list-style-type: none"> <li>1. Cell staining: Add DHR 123 solution to cells as described above.</li> </ol>

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Cell Research	<p>2. Flow cytometric analysis: After incubation, wash the cells and perform fluorescence analysis using a flow cytometer. The fluorescence intensity is proportional to the ROS level in the sample, which can achieve quantitative analysis.</p> <p>The above information is based on published literature. Experimental procedures should be appropriately modified to meet specific research demands.</p>
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### Solubility Information

Solubility	DMSO: 262.5 mg/mL (757.84 mM),Sonication is recommended. (< 1 mg/ml refers to the product slightly soluble or insoluble)
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### Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	2.887 mL	14.435 mL	28.870 mL
5 mM	0.5774 mL	2.887 mL	5.774 mL
10 mM	0.2887 mL	1.4435 mL	2.887 mL
50 mM	0.0577 mL	0.2887 mL	0.5774 mL

Please select the appropriate solvent to prepare the stock solution, according to the solubility of the product in different solvents. Please use it as soon as possible.

Note: The dilution table applies only to solid products. For liquid products, please calculate the stock solution based on the stated concentration and/or density.

### Reference

Henderson LM, Chappell JB. Dihydrorhodamine 123: a fluorescent probe for superoxide generation? Eur J Biochem. 1993 Nov 1;217(3):973-80.

Zavvar M, et al. Dihydrorhodamine-123 flow cytometry method: time for substantial revision in technical procedure. Lab Med. 2024 Sep 8:lmae076.

Živančević K, et al. ZnO-Induced Cytotoxicity and Mitochondrial Stress in Microglia: Implications of the Protective Role of Immunoglobulin G In Vitro. Balkan Med J. 2024 Sep 6;41(5):348-356.

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