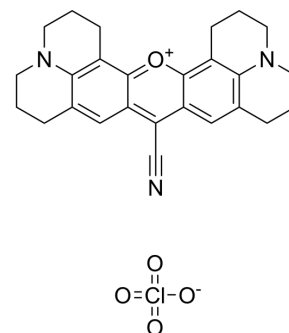


## Rhodamine 800

<b>Cat. No.:</b>	HY-101876
<b>CAS No.:</b>	137993-41-0
<b>Molecular Formula:</b>	C <sub>26</sub> H <sub>26</sub> ClN <sub>3</sub> O <sub>5</sub>
<b>Molecular Weight:</b>	495.95
<b>Target:</b>	Fluorescent Dye
<b>Pathway:</b>	Others
<b>Storage:</b>	4°C, sealed storage, away from moisture and light * In solvent : -80°C, 6 months; -20°C, 1 month (sealed storage, away from moisture and light)



### SOLVENT & SOLUBILITY

#### In Vitro

DMSO : 83.33 mg/mL (168.02 mM; Need ultrasonic)

Concentration	Solvent	Mass		
		1 mg	5 mg	10 mg
Preparing Stock Solutions	1 mM	2.0163 mL	10.0817 mL	20.1633 mL
	5 mM	0.4033 mL	2.0163 mL	4.0327 mL
	10 mM	0.2016 mL	1.0082 mL	2.0163 mL

Please refer to the solubility information to select the appropriate solvent.

### BIOLOGICAL ACTIVITY

#### Description

Rhodamine dyes are membrane-permeable cationic fluorescent probes that specifically recognize mitochondrial membrane potentials, thereby attaching to mitochondria and producing bright fluorescence, and at certain concentrations, rhodamine dyes have low toxicity to cells, so they are commonly used to detect mitochondria in animal cells, plant cells, and microorganisms<sup>[1]</sup>.

#### In Vitro

1. Preparation of Rhodamine 800 working solution
  - 1.1 Preparation of the stock solution  
Dissolve 1 mg Rhodamine 800 in 525 µL DMSO to obtain 5 mM of stock solution.
  - 1.2 Preparation of Rhodamine 800 working solution  
Dilute the stock solution in serum-free cell culture medium or PBS to obtain 1-20 µM of working solution.  
Note: Please adjust the concentration of Rhodamine 800 working solution according to the actual situation.
2. Cell staining
  - 2.1 Suspension cells (6-well plate)
    - a. Centrifuge at 1000 g at 4°C for 3-5 minutes and then discard the supernatant. Wash twice with PBS, 5 minutes each time. The cell density is 1×10<sup>6</sup>/mL.
    - b. Add 1 mL of working solution, and then incubate at room temperature for 5-30 minutes.

- c. Centrifuge at 400 g at 4°C for 3-4 minutes and then discard the supernatant.
- d. Wash twice with PBS, 5 minutes each time.
- e. Resuspend cells with serum-free cell culture medium or PBS. Observation by fluorescence microscopy or flow cytometry.

#### 2.2 Adherent cells

- a. Culture adherent cells on sterile coverslips.
- b. Remove the coverslip from the medium and aspirate excess medium.
- c. Add 100 µL of working solution, gently shake it to completely cover the cells, and then incubate at room temperature for 30-60 minutes.
- d. Wash twice with medium, 5 minutes each time. Observation by fluorescence microscopy or flow cytometry.

Note: If detection by flow cytometry, cells need to be resuspended before staining.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

## PROTOCOL

### Kinase Assay [2]

Rhodamine 800 (Rh800) and indocyanine green (ICG) are dissolved in PBS and their concentrations are set to 40 nM. To 3 mL of Rhodamine 800 solution (40 nM/PBS), 100 µL of p-sulfonatocalix[n]arenes (S[n]) (1 mM) is added. To this solution, microliter aliquots of acetylcholine (ACh) (350 mM) are added under stirring. After 2 min stirring, fluorescence spectra are measured. For the competitive fluorophore displacement by other neurotransmitters, amino acids, ammonium chloride, and choline, similar procedure as the above method is applied<sup>[2]</sup>.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

## REFERENCES

- [1]. Sakanoue J, et al. Rhodamine 800 as a probe of energization of cells and tissues in the near-infrared region: a study with isolated rat liver mitochondria and hepatocytes. *J Biochem.* 1997 Jan;121(1):29-37.
- [2]. Sakanoue J, et al. Rhodamine 800 as a probe of energization of cells and tissues in the near-infrared region: a study with isolated rat liver mitochondria and hepatocytes. *J Biochem.* 1997 Jan;121(1):29-37.
- [3]. Jin T. et al. Near-infrared fluorescence detection of acetylcholine in aqueous solution using a complex of rhodamine 800 and p-sulfonatocalix[8]arene. *Sensors (Basel).* 2010;10(3):2438-49.

**Caution: Product has not been fully validated for medical applications. For research use only.**

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