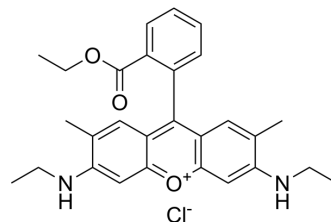


Rhodamine 6G

Cat. No.:	HY-D0309
CAS No.:	989-38-8
Molecular Formula:	C ₂₈ H ₃₁ ClN ₂ O ₃
Molecular Weight:	479.01
Target:	Fluorescent Dye
Pathway:	Others
Storage:	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 : 25 mg/mL (52.19 mM; Need ultrasonic)				
	H ₂ O : 10 mg/mL (20.88 mM; ultrasonic and warming and heat to 60°C)				
	Preparing Stock Solutions	<div>Solvent Concentration</div> <div>Mass</div>	1 mg	5 mg	10 mg
		1 mM	2.0876 mL	10.4382 mL	20.8764 mL
		5 mM	0.4175 mL	2.0876 mL	4.1753 mL
10 mM		0.2088 mL	1.0438 mL	2.0876 mL	
Please refer to the solubility information to select the appropriate solvent.					
In Vivo	1. Add each solvent one by one: PBS Solubility: 4 mg/mL (8.35 mM); Clear solution; Need ultrasonic and warming and heat to 60°C				
	2. Add each solvent one by one: 10% DMSO >> 40% PEG300 >> 5% Tween-80 >> 45% saline Solubility: 2.5 mg/mL (5.22 mM); Suspended solution; Need ultrasonic				
	3. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline) Solubility: 2.5 mg/mL (5.22 mM); Suspended solution; Need ultrasonic				

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 ^[1] .
In Vitro	<ol style="list-style-type: none"> Preparation of Rhodamine 6G working solution Preparation of the stock solution

	<p>Dissolve 1 mg Rhodamine 6G in 525 μL DMSO to obtain 5 mM of stock solution.</p> <p>1.2 Preparation of Rhodamine 6G working solution</p> <p>Dilute the stock solution in serum-free cell culture medium or PBS to obtain 1-20 μM of working solution.</p> <p>Note: Please adjust the concentration of Rhodamine 6G working solution according to the actual situation.</p> <p>2. Cell staining</p> <p>2.1 Suspension cells (6-well plate)</p> <p>a. Centrifuge at 1000 g at 4$^{\circ}$C for 3-5 minutes and then discard the supernatant. Wash twice with PBS, 5 minutes each time. The cell density is 1×10^6/mL.</p> <p>b. Add 1 mL of working solution, and then incubate at room temperature for 5-30 minutes.</p> <p>c. Centrifuge at 400 g at 4$^{\circ}$C for 3-4 minutes and then discard the supernatant.</p> <p>d. Wash twice with PBS, 5 minutes each time.</p> <p>e. Resuspend cells with serum-free cell culture medium or PBS. Observation by fluorescence microscopy or flow cytometry.</p> <p>2.2 Adherent cells</p> <p>a. Culture adherent cells on sterile coverslips.</p> <p>b. Remove the coverslip from the medium and aspirate excess medium.</p> <p>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.</p> <p>d. Wash twice with medium, 5 minutes each time. Observation by fluorescence microscopy or flow cytometry.</p> <p>Note: If detection by flow cytometry, cells need to be resuspended before staining.</p> <p>MCE has not independently confirmed the accuracy of these methods. They are for reference only.</p>
In Vivo	<p>Melanoma-transplanted mice receiving Rhodamine 6G demonstrate prolonged survival, improved clinical parameters, inhibited tumor growth and metastases count, compared to their untreated counterparts. Twice-a-week 10-6M Rhodamine 6G regimen yield the most prominent results^[2]. The Rhodamine-6G enters the circulatory system and labels leukocytes. It is possible to monitor changes in the interactions between leukocytes and the endothelium by determining the numbers of rolling and adhering leukocytes as well as the total flux of these cells^[3].</p> <p>MCE has not independently confirmed the accuracy of these methods. They are for reference only.</p>

PROTOCOL

Cell Assay ^[2]	<p>Malignant cells and normal control cultures are seeded in equal (protein adjusted) cell amounts into 6-well tissue culture plates. The cells are pulsed with 25 μCi/mL of 3H-Thymidine and immediately treated with Rhodamine 6G at the fixed concentration of 1 μM for 24h, 48h, 72h or 5 days (120h). Following 24h, 48h, 72h or 5 days, the excessive radioactive material is washed out with PBS. The cell samples are transferred into polystyrene vials containing 4 ml scintillation liquid, and their radioactivity counted in a β-counter. Total cell protein is assessed by Bradford's assay^[2].</p> <p>MCE has not independently confirmed the accuracy of these methods. They are for reference only.</p>
Animal Administration ^[2]	<p>Mice: C57Bl mice are implanted with B16-F10 melanoma and treated with Rhodamine 6G (1, 0.1, 0.01 μM) at different dosage/time regimens. Viability and proliferation of cultured tumor cells are analyzed^[2].</p> <p>MCE has not independently confirmed the accuracy of these methods. They are for reference only.</p>

REFERENCES

- [1]. Kutushov M, et al. Low concentrations of Rhodamine 6G selectively destroy tumor cells and improve survival of melanoma transplanted mice. *Neoplasma*. 2013;60(3):262-73.
- [2]. Zehentbauer FM, et al. Fluorescence spectroscopy of Rhodamine 6G: concentration and solvent effects. *Spectrochim Acta A Mol Biomol Spectrosc*. 2014;121:147-51.
- [3]. Kutushov M, et al. Low concentrations of Rhodamine 6G selectively destroy tumor cells and improve survival of melanoma transplanted mice. *Neoplasma*. 2013;60(3):262-73.

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

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