

Product Data Sheet

Product Name: Monochlorobimane (Chlorobimane)

Cat. No.: GC30550

Chemical Properties

Cas. No. 76421-73-3

SMILES O=C1N(C(C(C)=C2C)=O)N2C(CCl)=C1C

Formula $C_{10}H_{11}ClN_2O_2$ M.Wt 226.66

Solubility DMSO : 50 mg/mL (220.59 mM; Storage Store at -20°C, protect from light
Need ultrasonic)

General tips For obtaining a higher solubility , please warm the tube at 37 °C and shake it in the ultrasonic bath for a while. Stock solution can be stored below -20°C for several months.

Shipping Condition Evaluation sample solution : ship with blue ice All other available size: ship with RT , or blue ice upon request.

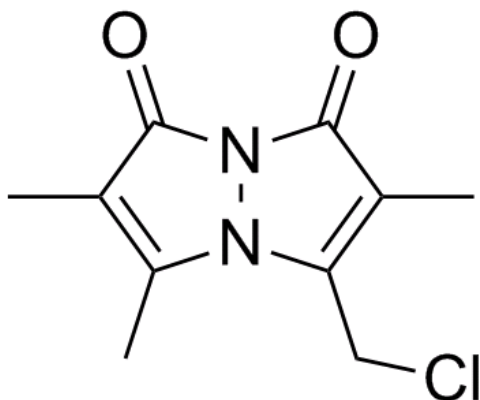
Caution: Product has not been fully validated for medical applications. For research use only.

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Structure



Protocol

Evaluation of GSH content using Monochlorobimane

This protocol only provides a guideline, and should be modified according to your specific needs

1. Stock and dilute solution preparation for DHE staining of cells

Dilute the Monochlorobimane stock solution (50mM in DMSO) with PBS to a working solution with a final concentration of 1mM. NOTE: The working solution must be prepared fresh on the day of the experiment.

2. Cells cell harvesting

Algal cells were harvested in the exponential phase of growth by centrifugation (2,500×g; 15min) and suspended in fresh culture medium at 1×10^6 cells/ml within EP tube.

3. Cell staining

(1) Remove the media from EP tube and wash gently once with either 1×PBS.

(2) Add an appropriate amount of the medium containing 20μM Monochlorobimane to the EP tube,

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mix well, then aliquot 200µl onto a 96-well microplate and incubate in the dark at 25°C for 30 minutes.

(3) After 30min incubation period, remove the Monochlorobimane-containing media and wash gently three times with 1×PBS.

(4) Move the plate to the microplate reader (Ex/Em=380/480nm) for measuring the fluorescence intensity.

4. Experimental Precautions

(1) Monochlorobimane should be stored in the dark throughout the experiment.

(2) This protocol and all reagents are for scientific research use only.

References:

[1] Machado M D, Soares E V. Assessment of cellular reduced glutathione content in *Pseudokirchneriella subcapitata* using monochlorobimane[J]. *Journal of Applied Phycology*, 2012, 24(6): 1509-1516.

Background

Monochlorobimane (Chlorobimane) is a non-fluorescent cell-permeant probe at 380nm wavelength excitation and 470nm wavelength emission^[1]. Once inside the cell, Monochlorobimane reacts with reduced glutathione (GSH) to form fluorescent bimane–glutathione (B-SG) adducts, in a reaction catalyzed by glutathione S-transferase^[2]. Monochlorobimane has been widely used for detecting GSH levels and screening bacterial populations that express glutathione transferase (GST)^[3].

In vivo, intracranially inject 10µg/5µl of Monochlorobimane at a rate of 5µl/3min for 30 minutes, which significantly reduced the staining of cytochrome c oxidase (COX) and succinate dehydrogenase (SDH) in the hippocampus of rats induced by microsphere embolism (ME)^[4].

References:

[1] Kamencic H, Lyon A, Paterson P G, et al. Monochlorobimane fluorometric method to measure tissue glutathione[J]. *Analytical biochemistry*, 2000, 286(1): 35-37.

[2] Machado M D, Soares E V. Assessment of cellular reduced glutathione content in *Pseudokirchneriella subcapitata* using monochlorobimane[J]. *Journal of Applied Phycology*, 2012, 24(6): 1509-1516.

[3] Eklund B I, Edalat M, Stenberg G, et al. Screening for recombinant glutathione transferases active with monochlorobimane[J]. *Analytical biochemistry*, 2002, 309(1): 102-108.

[4] Oshikawa S, Miyake-Takagi K, Takagi N, et al. Effects of monochlorobimane on cerebral

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ischemia-induced damage to mitochondria[J]. Biological and Pharmaceutical Bulletin, 2004, 27(2): 166-169.

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