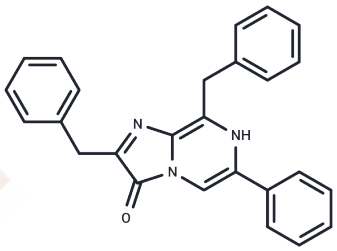
Data Sheet (Cat.No.T78389)

TargetMol

Coelenteramine 400a

Chemical Properties



CAS No. : 70217-82-2

Formula: C26H21N3O

Molecular Weight: 391.46

Storage: Store at low temperature
Powder: -20°C for 3 years | In solvent: -80°C for 1 year
Actual storage temperature shall be subject to the COA.

Biological Description

Description	Coelenteramine 400a (Coelenterazine 400a), a Coelenterazine derivative, serves as a substrate for Renilla luciferase (RLuc), facilitating the emission of blue light at 395 nm [1] [2]. This compound alters the bioluminescence color of RLuc by substituting the sulfur and oxygen heteroatoms in the methylene bridge. Coelenteramine 400a enhances signal resolution, proving useful in bioluminescence resonance energy transfer (BRET) research [3].
Targets(IC50)	Others
In vitro	Luminescent reaction I. Solution preparation 1. Mother solution preparation: Use ethanol to prepare Coelenteramine 400a into a mother solution of appropriate concentration (1-10mM). The concentration of the mother solution can be adjusted according to the specific experimental conditions. 2. Working solution preparation: Dilute it with water/PBS/DMEM to the required working solution when using. II. Operation steps 1. Mix 10 μL of Coelenteramine 400a solution with a concentration of 1.0-5.0 mM and 10 μL of Renilla luciferase (RLuc8.6-53) solution with a concentration of 2.01 mg/mL with 200 μL buffer. 2. Use a multi-channel spectrometer equipped with a liquid nitrogen-cooled CCD detector to record the bioluminescence spectrum of the mixed solution. Note: The absolute spectral sensitivity of the spectrometer needs to be calibrated in advance with a 500 W spectral irradiance standard lamp. 3. A custom-made photometer equipped with a photomultiplier tube (PMT, H11890-01, Hamamatsu Photonics, Japan) was used to measure the relevant parameters of the bioluminescent reaction. Note: The absolute response of the photometer needs to be calibrated with an integrating sphere spectrometer and is determined by plotting a linear graph between the relative counts measured by the photometer and the absolute values measured by the integrating sphere. 4. In a test tube, 5-15 μL of a 10 nM (50-150 fmol in total) Coelenteramine 400a solution was pre-placed in the photometer, and then 150 μL of a 20 μg/mL luciferase solution (in 0.1 M GTA buffer, pH 6.0, 7.0, and 8.0) was injected to start the reaction. The reaction was monitored until the luminescent reaction was complete, and the quantum yield of

In vitro	the bioluminescent reaction was calculated from the total number of photons obtained and the number of luciferin molecules. 5. The Michaelis constant (Km) of the substrate coelenteramine 400a was calculated using the Lineweaver-Burk plot. The catalytic constant (kcat) was calculated. The fluorescence spectrum and absolute fluorescence quantum yield of the corresponding oxidation product coelenteramide 400a (CTMD 400a) were measured using Quantaurus-QY with the excitation light set to 300 nm. The concentration of CTMD 400a solution in DMSO was 20 μM during the measurement. [4] The above information is based on published literature. Experimental procedures should be appropriately modified to meet specific research demands.

Solubility Information

Solubility	Methanol: 1 mg/mL (2.55 mM),DMSO inactivates the activity of Coelenteramine 400a. Ethanol: < 0.5 mg/mL(insoluble or slightly soluble) (< 1 mg/ml refers to the product slightly soluble or insoluble)

Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	2.5545 mL	12.7727 mL	25.5454 mL
5 mM	0.5109 mL	2.5545 mL	5.1091 mL
10 mM	0.2555 mL	1.2773 mL	2.5545 mL
50 mM	0.0511 mL	0.2555 mL	0.5109 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

Levi J, et al. Bisdeoxycoelenterazine derivatives for improvement of bioluminescence resonance energy transfer assays. J Am Chem Soc. 2007 Oct 3;129(39):11900-1.
Bertrand L, et al. The BRET2/arrestin assay in stable recombinant cells: a platform to screen for compounds that interact with G protein-coupled receptors (GPCRS). J Recept Signal Transduct Res. Feb-Nov 2002;22(1-4):533-41.
Yuan M L, et al. Luminescence of coelenterazine derivatives with C-8 extended electronic conjugation[J]. Chinese Chemical Letters, 2016, 27(4): 550-554.