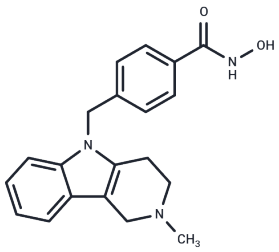
Data Sheet (Cat.No.T1966)

TargetMol

Tubastatin A

Chemical Properties



CAS No. : 1252003-15-8

Formula: C20H21N3O2

Molecular Weight: 335.4

Storage: 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	Tubastatin A (Tubastatin A BASE) is an effective and specific HDAC6 inhibitor (IC50: 15 nM, in a cell-free assay). Its selectivity is 1000-fold against all the other isozymes except HDAC8.
Targets(IC50)	Apoptosis,Antibacterial,HDAC,Autophagy
In vitro	Tubastatin A (5 μM) dose-dependently protected neuronal cells from homocysteine-induced death, which was completely avoided at 10 μM. Tubastatin A (10 μM) slightly induced histone acetylation. When applied to cholangiocarcinoma cell lines, Tubastatin A (10 μM) induced an increase in the level of acetylated α-tubulin and the restoration of primary cilia expression, which correlated with the down-regulation of Hedgehog and MAPK signaling pathways, and a decrease in the rate of cell proliferation (50% on average) and infiltration (40%). Tubastatin A significantly inhibited TNF-α and IL-6 when acting on lipopolysaccharide-stimulated human THP-1 macrophages (IC50: 272/712 nM). When acting on mouse Raw 264.7 macrophages, Tubastatin A dose-dependently inhibited nitric oxide secretion (IC50: 4.2 μM).
In vivo	Tubastatin A (5 μM) dose-dependently protected neuronal cells from homocysteine-induced death, which was completely avoided at 10 μM. Tubastatin A (10 μM) slightly induced histone acetylation. When applied to cholangiocarcinoma cell lines, Tubastatin A (10 μM) induced an increase in the level of acetylated α-tubulin and the restoration of primary cilia expression, which correlated with the down-regulation of Hedgehog and MAPK signaling pathways, and a decrease in the rate of cell proliferation (50% on average) and infiltration (40%). Tubastatin A significantly inhibited TNF-α and IL-6 when acting on lipopolysaccharide-stimulated human THP-1 macrophages (IC50: 272/712 nM). When acting on mouse Raw 264.7 macrophages, Tubastatin A dose-dependently inhibited nitric oxide secretion (IC50: 4.2 μM).
Kinase Assay	HDAC enzymatic assays: Tubastatin A is dissolved and diluted in assay buffer (50 mM HEPES, pH 7.4, 100 mM KCl, 0.001% Tween-20, 0.05% BSA, and 20 μM tris(2-carboxyethyl)phosphine) to 6-fold of the final concentration. HDAC enzymes are diluted to 1.5-fold of the final concentration in assay buffer and pre-incubated with Tubastatin A for 10 minutes before the addition of the substrate. The amount of FTS (HDAC1, HDAC2, HDAC3, and HDAC6) or MAZ-1675 (HDAC4, HDAC5, HDAC7, HDAC8, and HDAC9) used for each enzyme is equal to the Michaelis constant (Km), as determined by a titration curve. FTS or MAZ-1675 is diluted in assay buffer to 6-fold the final concentration with 0.3 μM sequencing grade trypsin. The substrate/trypsin mix is added

Kinase Assay	to the enzyme/compound mix and the plate is shaken for 60 seconds and then placed into a SpectraMax M5 microtiter plate reader. The enzymatic reaction is monitored for release of 7-amino-4-methoxy-coumarin over 30 minutes, after deacetylation of the lysine side chain in the peptide substrate, and the linear rate of the reaction is calculated.
Cell Research	Anchorage-independent growth is assessed by growing cells in soft agar. About 25,000 cells suspended in 0.4% agar in culture media are layered over a 1% agar layer in a 6-well plate. Media are added twice a week and pictures are taken after 21 days of incubation. The number and size of colonies are analyzed using the Gel-Pro software. (Only for Reference)

Solubility Information

Solubility	DMSO: 35 mg/mL (104.35 mM),Sonication is recommended. Ethanol: < 1 mg/mL (insoluble or slightly soluble), H2O: < 1 mg/mL (insoluble or slightly soluble), (< 1 mg/ml refers to the product slightly soluble or insoluble)
In vivo Formulation	10% DMSO+40% PEG300+5% Tween 80+45% Saline: 1 mg/mL (2.98 mM),Sonication is recommended. *Please add the solvents sequentially, clarifying the solution as much as possible before adding the next one. Dissolve by heating and/or sonication if necessary. Working solution is recommended to be prepared and used immediately. The formulation provided above is for reference purposes only. In vivo formulations may vary and should be modified based on specific experimental conditions.*

Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	2.9815 mL	14.9076 mL	29.8151 mL
5 mM	0.5963 mL	2.9815 mL	5.963 mL
10 mM	0.2982 mL	1.4908 mL	2.9815 mL
50 mM	0.0596 mL	0.2982 mL	0.5963 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

Butler KV, et al. J Am Chem Soc, 2010, 132(31), 10842-10846.
Gradilone SA, et al. Cancer Res, 2013, 73(7), 2259-2270.
Vishwakarma S, et al. Int Immunopharmacol, 2013, 16(1), 72-78.
Santo L, et al. Blood, 2012, 119(11), 2579-2589.