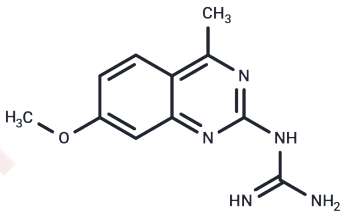
Data Sheet (Cat.No.T5340)

TargetMol

MPO-IN-28

Chemical Properties



CAS No. : 37836-90-1

Formula: C11H13N5O

Molecular Weight: 231.25

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	MPO-IN-28 is a novel potent, irreversible Myeloperoxidase (MPO) inhibitor with IC50 of 44 nM.
Targets(IC50)	Glutathione Peroxidase
In vitro	5 μM of MPO-IN-28 was added to MPO in the presence of 30 μM of guaiacol. In the absence of H2O2, there was no inhibition of MPO, clearly underlining that MPO-IN-28 is an irreversible mechanism-based inhibitor. MPO-IN-28 inhibits the growth of normal human dermal fibroblast (NHDF) at a concentration (IC50) of 17 μM, which is about 400 times higher than the concentration of the MPO inhibitory effect (0.044 μM).
Kinase Assay	The assay is based on the production of taurine chloramine produced by the MPO/H2O2/Cl? system in the presence of a selected inhibitor at defined concentration. The reaction mixture contained the following reagents in a final volume of 200 μL: 10 mM phosphate buffer (pH 7.4, 300 mM NaCl), 15 mM taurine, compound to be tested (up to 20 μM), and a fixed amount of recombinant MPO (6.6 μL of MPO batch solution diluted 2.5 times, 40 nM). When necessary, the volume was adjusted with water. This mixture was incubated at 37 °C, and the reaction was initiated with 10.0 μL of H2O2 (100 μM). After 5 min, the reaction was stopped by the addition of 10 μL of catalase (8 units/μL). To determine the amount of taurine chloramine produced, 50 μL of 1.35 mM solution of thionitrobenzoic acid was added and the volume was adjusted to 300 μL with water. Then the absorbance of the solutions was measured at 412 nm with a microplate reader, and the curve of absorbance as a function of inhibitor concentration was plotted. IC50 values were then determined by standard procedures by taking into account the absence of hydrogen peroxide as 100% inhibition and the absence of inhibitors as 0% inhibition.
Cell Research	To evaluate whether selected compounds exhibited toxicity at the cellular level at concentrations ranging from 0.005 to 50 μM, the colorimetric assay MTT was performed58 in normal human dermal fibroblasts (NHDF). Briefly, cell line was cultured in cell culture flasks, grown, and maintained at 37 °C, 95% humidity, 5% CO2 in fibroblast medium FBM supplemented with 2% fetal bovine serum, 0.1% insulin, rhFGF-B, and gentamicin and amphotericin. NHDF cells were chemically detached with trypsin and seeded in 96-well plates and left to attach for 24 h. Prior to treatment, compounds were dissolved in DMSO at a concentration of 10 mM, and cells were treated with the different concentrations of the compounds diluted in culture medium (5 nM to 50 μM) or left

Cell Research	untreated for 72 h. The amount of viable cells was determined through the MTT mitochondrial reduction into formazan by living cells according to previously described. 58 The optical density (OD) was measured in a Biorad 680RX plate reader at 570 nm (reference 630 nm), and the OD of the untreated control was normalized as 100% of viable cells, allowing determination of the concentration that inhibited their growth by 50% (IC50).

Solubility Information

Solubility	DMSO: 22.73 mg/mL (98.29 mM),Sonication is recommended. (< 1 mg/ml refers to the product slightly soluble or insoluble)
In vivo Formulation	10% DMSO+40% PEG300+5% Tween 80+45% Saline: 2 mg/mL (8.65 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	4.3243 mL	21.6216 mL	43.2432 mL
5 mM	0.8649 mL	4.3243 mL	8.6486 mL
10 mM	0.4324 mL	2.1622 mL	4.3243 mL
50 mM	0.0865 mL	0.4324 mL	0.8649 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

Soubhye J, et al. Discovery of Novel Potent Reversible and Irreversible Myeloperoxidase Inhibitors Using Virtual Screening Procedure. J Med Chem. 2017 Aug 10;60(15):6563-6586.