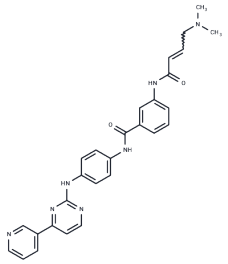
Data Sheet (Cat.No.T3598)

TargetMol

JNK-IN-7

Chemical Properties



CAS No. : 1408064-71-0

Formula: C28H27N7O2

Molecular Weight: 493.56

Appearance: no data available

Storage: Powder: -20°C for 3 years | In solvent: -80°C for 1 year

Biological Description

Description	JNK-IN-7 (JNK inhibitor) is a selective JNK1/2/3 inhibitor (IC50: 1.54/1.99/0.75 nM). It can also inhibit phosphorylation of c-Jun, which is a substrate of JNK kinase.
Targets(IC50)	JNK
In vitro	JNK-IN-7 is a relatively selective JNK inhibitor in cells, binding to JNK1, 2, 3, IRAK1 (IC50=14.1 nM), YSK4 (IC50=4.8 nM), ERK3 (IC50=22 nM), PIK3C3, PIP5K3, and PIP4K2C[1]. In HCT116 cells, TNF stimulation for 24 and 48 hours significantly decreases divalent metal-ion transporter 1 (DMT1) expression, a process that JNK-IN-7 can notably counteract[2].
Kinase Assay	A375 cells are pre-treated with 1 μM JNK-IN-7 for the indicated amounts of time. Remove the medium and wash 3 times with PBS. Resuspend the cell pellet with 1 mL Lysis Buffer (1% NP-40, 1% CHAPS, 25 mM Tris, 150 mM NaCl, Phosphatase Inhibitor Cocktail). Rotate end-to-end for 30 min at 4°C. Lysates are cleared by centrifugation at 14000 rpm for 15 min in the Eppendorf. The cleared lysates gel filtered into Kinase Buffer (0.1% NP-40, 20 mM HEPES, 150 mM NaCl, Phosphatase Inhibitor Cocktail, Protease Inhibitor Cocktail) using Bio-Rad 10DG colums. The total protein concentration of the gel-filtered lysate should be around 5-15 mg/mL. Cell lysate is labeled with the probe at 5 μM for 1 hour. Samples are reduced with DTT, and cysteines are blocked with iodoacetamide and gel filtered to remove excess reagents and exchange the buffer. Add 1 volume of 2X Binding Buffer (2% Triton-100, 1% NP-40, 2 mM EDTA, 2X PBS) and 50 μL streptavidin bead slurry and rotate end-to-end for 2 hours, centrifuge at 7000 rpm for 2 min. Wash 3 times with 1X Binding Buffer and 3 times with PBS. Add 30 μL 1X sample buffer to beads, heat samples at 95°C for 10 min. Run samples on an SDS-PAGE gel at 110V. After transferred, the membrane is immunoblotted with JNK antibody[1].
Cell Research	JNK-IN-7 is prepared in DMSO and stored, and then diluted with appropriate medium before use[2]. Intestinal epithelial cell line (HCT116) is cultured in DMEM medium, supplemented with 10% heat-inactivated fetal bovine serum (FBS), penicillin (100?U/mL) and streptomycin (100?g/mL), 2?mM L-gentamycin, and 50?μM 2-ME. These cells are stimulated with TNF (20?ng/mL), LPS (100?ng/mL), and IFN-γ (20?ng/mL), respectively. After 24 or 48?h of culture, cells are harvested followed by extraction of total RNA, and the levels of DMT1 mRNA are analyzed by qRT-PCR. To determine the mechanisms of TNF involved in regulating DMT1 expression, JNK-IN-7 (1?μM), NF-κB inhibitor (BAY 11-7082, 1?μM), and caspase-3/8 inhibitor (Z-DEVD-FMK, 50?μM) are also added into the culture medium. After 48?h of culture, cells are then collected to detect the expression of

	DMT1 by qRT-PCR[2].

Solubility Information

Solubility	DMSO: 55 mg/mL (111.44 mM),Sonication is recommended. (< 1 mg/ml refers to the product slightly soluble or insoluble)

Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	2.0261 mL	10.1305 mL	20.261 mL
5 mM	0.4052 mL	2.0261 mL	4.0522 mL
10 mM	0.2026 mL	1.013 mL	2.0261 mL
50 mM	0.0405 mL	0.2026 mL	0.4052 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.

Reference

Zhang T, et al. Discovery of Potent and Selective Covalent Inhibitors of JNK. Chem Biol. 2012 Jan 27;19(1):140-54.
Xu P, Zeng S, Xia X, et al. FAM172A promotes follicular thyroid carcinogenesis and may be a marker of FTC. Endocrine-Related Cancer. 2020, 1(aop)
Wu W, et al. Divalent metal-ion transporter 1 is decreased in intestinal epithelial cells and contributes to the anemia in inflammatory bowel disease. Sci Rep. 2015 Nov 17;5:16344.
Xu P, Zeng S, Xia X, et al. FAM172A promotes follicular thyroid carcinogenesis and may be a marker of FTC[J]. Endocrine-Related Cancer. 2020, 1(aop).