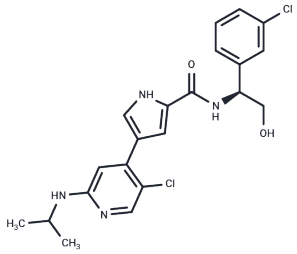
Data Sheet (Cat.No.T7005)

TargetMol

Ulixertinib

Chemical Properties



CAS No. : 869886-67-9

Formula: C21H22Cl2N4O2

Molecular Weight: 433.33

Appearance: no data available

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

Biological Description

Description	Ulixertinib (VRT752271) (BVD-523, VRT752271) is an effective and reversible ERK1/ERK2 inhibitor. The IC50 of Ulixertinib is less than 0.3 nM for ERK2.
Targets(IC50)	ERK
In vitro	In an A375 melanoma cell line containing a b-RAFV600E mutation, Ulixertinib reduces the levels of phosphorylated ERK2 (pERK) and of the phosphorylation of the downstream kinase RSK (pRSK) with IC50 of 4.1/0.14 μM, respectively. Ulixertinib also inhibits A375 cell proliferation with IC50 of 180 nM. [1]
Kinase Assay	ERK2 Rapidfire Mass Spectrometry Inhibition of Catalysis Assay: MEK U911-activated ERK2 protein is expressed and purified in-house. Enzyme and substrate solutions are made up in assay buffer consisting of 50 mM Tris (pH 7.5), 10 mM MgCl2, 0.1 mM EGTA, 10 mM DTT and 0.01% (v/v) CHAPS. 1.2 nM ERK2 protein is prepared in assay buffer and 10 μL is dispensed into each well of a polypropylene, 384-well plate containing test and reference control compounds. The compound plates had previously been dosed with a 12 point range from 100 μM down to 0.1 nM in order to calculate compound IC50s, with a total DMSO concentration in the assay of 1%. Following a 20 minute pre-incubation of enzyme and compound at room temperature, 10 μL of substrate solution is added consisting of 16 μM Erktide (IPTTPITTTYFFFK) and 120 μM ATP (measured Km) in assay buffer. The reaction is allowed to progress for 20 minutes at room temperature before being quenched by the addition of 80 μl 1% (v/v) formic acid. The assay plates are then run on the RapidFire Mass Spectrometry platform to measure substrate (unphosphorylated Erktide) and product (phosphorylated Erktide) levels.
Cell Research	A375 cells are cultured in cell media composed of DMEM, 10% (v/v) Foetal Calf Serum and 1% (v/v) L-Glutamine. After harvesting, cells are dispensed into black, 384-well Costar plates to give 200 cells per well in a total volume of 40 μL cell media, and are incubated overnight at 37°C, 90% relative humidity and 5% CO2 in a rotating incubator. Test compounds and reference controls are dosed directly into the cell plates, into the inner 308 wells, using a Labcyte Echo 555 acoustic dispenser. The cells are dosed over a 12 point range from 30 μM down to 0.03 nM in order to calculate compound IC50s, with a total DMSO concentration in the assay of 0.3%. The cell plates are then incubated for 72 hours at 37°C. Cells were fixed and stained by the addition of 20 μL 12% formaldehyde in PBS/A (4% final concentration) and 1:2000 dilution of Hoechst 33342, with a 30 minute room temperature incubation, and then washed with PBS/A. A cell

count is performed on the stained cell plates using a Cellomics ArrayScanTM VTI imaging platform. A Day 0 cell plate is also fixed, stained and read to generate a cell count baseline for determining compound cytotoxic effects as well as anti-proliferative effects. (Only for Reference)

Solubility Information

Solubility	H2O: < 1 mg/mL (insoluble or slightly soluble), DMSO: 70 mg/mL (161.54 mM),Sonication is recommended. 10% DMSO+40% PEG300+5% Tween 80+45% Saline: 8 mg/mL (18.46 mM),Solution. Ethanol: 12 mg/mL (27.69 mM),Sonication is recommended. (< 1 mg/ml refers to the product slightly soluble or insoluble)

Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	2.3077 mL	11.5386 mL	23.0771 mL
5 mM	0.4615 mL	2.3077 mL	4.6154 mL
10 mM	0.2308 mL	1.1539 mL	2.3077 mL
50 mM	0.0462 mL	0.2308 mL	0.4615 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

Ward RA, et al. J Med Chem. 2015, 58(11), 4790-4801.
Caiola E, Iezzi A, Tomanelli M, et al. LKB1 deficiency renders non-small-cell lung cancer cells sensitive to ERK inhibitors.: ERK inhibitors in LKB1 mutated NSCLC. Journal of Thoracic Oncology. 2019
Caiola E, Iezzi A, Tomanelli M, et al. LKB1 deficiency renders non-small-cell lung cancer cells sensitive to ERK inhibitors.: ERK inhibitors in LKB1 mutated NSCLC[J]. Journal of Thoracic Oncology. 2019.
Caiola E, Iezzi A, Tomanelli M, et al. LKB1 Deficiency Renders NSCLC Cells Sensitive to ERK Inhibitors. Journal of Thoracic Oncology. 2020, 15(3): 360-370