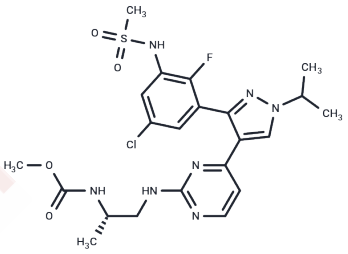
Data Sheet (Cat.No.T6487)

TargetMol

Encorafenib

Chemical Properties



CAS No. : 1269440-17-6

Formula: C22H27ClFN7O4S

Molecular Weight: 540.01

Storage: Keep away from direct sunlight,Keep away from moisture,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	Encorafenib (LGX818) is an orally available mutated BRaf V600E inhibitor(IC50=0.3 nM) with potential antineoplastic activity.
Targets(IC50)	Raf
In vitro	In the A375 (BRAFV600E) human melanoma cell line Encorafenib suppresses phospho-ERK (EC50 = 3 nM) leading to potent inhibition of proliferation (EC50 = 4 nM). No significant activity is observed against a panel of 100 kinases (IC50 > 900 nM) and Encorafenib does not inhibit proliferation of > 400 cell lines expressing wild-type BRAF. Contributing to the high potency of Encorafenib is the extremely slow off-rate from BRAFV600E which is not observed with other RAF inhibitors. In biochemical assays the dissociation half-life is >24 hours which translated into sustained target inhibition in cells following drug wash-out. [1]
In vivo	Encorafenib treatment at oral doses as low as 6 mg/kg resulted in strong (75%) and sustained (>24 hours) decrease in phospho-MEK, even following clearance of drug from circulation in single dose PK/PD studies in human melanoma xenograft models (BRAFV600E). Encorafenib induces tumor regression in multiple BRAF mutant human tumor xenograft models grown in immune compromised mice and rats at doses as low as 1 mg/kg. Consistent with the in vitro data, Encorafenib is inactive against BRAF wild-type tumors at doses up to 300 mg/kg bid, with good tolerability and linear increase in exposure. Efficacy is also achieved in a more disease-relevant spontaneous metastatic melanoma and a model of melanoma brain metastasis. Encorafenib is a potent and selective RAF kinase inhibitor with unique biochemical properties that contribute to an excellent pharmacological profile. [1]
Kinase Assay	The Raf kinase activity reaction is started by the addition of 10 μL per well of 2×ATP diluted in assay buffer. After 3 hours (bRaf(V600E)) or 1 hour (c-Raf), the reactions are stopped with the addition of 10 μL of stop reagent (60 mM EDTA). Phosphorylated product is measured using a rabbit anti-p-MEK antibody and the Alpha Screen IgG (ProteinA) detection Kit, by the addition of 30 μL to the well of a mixture of the antibody (1:2000 dilution) and detection beads (1:2000 dilution of both beads) in bead buffer (50 mM Tris, pH 7.5, 0.01% Tween20). The additions are carried out under dark conditions to protect the detection beads from light. A lid is placed on top of the plate and incubated

Kinase Assay	for 1 hour at room temperature, then the luminescence is read on a PerkinElmer Envision instrument. The concentration of each compound for 50% inhibition (IC50) is calculated by non-linear regression using XL Fit data analysis software
Cell Research	LGX818 is dissolved in DMSO. A375 is a melanoma cell line that harbors the B-Raf V600E mutation. A375-luc cells engineered to express luciferase is plated to 384-well white clear bottom plates as 1,500 cells/50 μL/well in DMEM containing 10% FBS. Test compounds, dissolved in 100% DMSO at appropriate concentrations, are transferred to the cells by a robotic Pin Tool (100 mL). The cells are incubated for 2 days at 25°C, then 25 μL of BrightGloTM is added to each well and the plates are read by luminescence. The concentration of each compound for 50% inhibition (IC50) is calculated by non-linear regression using XL Fit data analysis software. wild type and V600E B-Raf.

Solubility Information

Solubility	H2O: < 1 mg/mL (insoluble or slightly soluble), Ethanol: 93 mg/mL (172.22 mM),Sonication is recommended. DMSO: 257 mg/mL (475.92 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: 9.3 mg/mL (17.22 mM),Suspension. *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	1.8518 mL	9.2591 mL	18.5182 mL
5 mM	0.3704 mL	1.8518 mL	3.7036 mL
10 mM	0.1852 mL	0.9259 mL	1.8518 mL
50 mM	0.037 mL	0.1852 mL	0.3704 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

Darrin D Stuart, et al. Cancer Res, 2012, 72(8 Supplement): 3790
Li B, Ming H, Qin S, et al.HSPA8 Activates Wnt/β-Catenin Signaling to Facilitate BRAF V600E Colorectal Cancer Progression by CMA-Mediated CAV1 Degradation.Advanced Science.2023: 2306535.