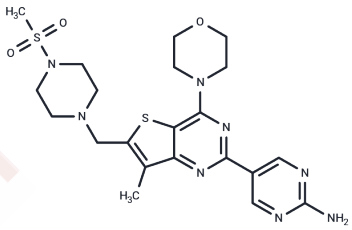
Data Sheet (Cat.No.T3692)

TargetMol

GNE-477

Chemical Properties



CAS No. : 1032754-81-6

Formula: C21H28N8O3S2

Molecular Weight: 504.63

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	GNE-477 is a potent and efficacious dual (PI3K/mTOR) inhibitor.
Targets(IC50)	mTOR,PI3K
Kinase Assay	Fluorescence Polarization Based Assays for XIAP, cIAP1, and cIAP2 BIR3 Proteins: FL-AT-406 (the fluorescently tagged AT-406) is employed to develop a set of new FP assays for determination of the binding affinities of Smac mimetics to XIAP, cIAP-1, and cIAP-2 BIR3 proteins. The Kd value of FL-AT-406 to each IAP protein is determined by titration experiments using a fixed concentration of FL-AT-406 and different concentrations of the protein up to full saturation. Fluorescence polarization values are measured using an Infinite M-1000 plate reader in Microfluor 2 96-well, black, round-bottom plates. To each well, FL-AT-406 (2, 1, and 1 nM for experiments with XIAP BIR3, cIAP-1 BIR3, and cIAP-2 BIR3, respectively) and different concentrations of the protein are added to a final volume of 125 µL in the assay buffer (100 mM potassium phosphate, pH 7.5, 100 µg/mL bovine γ-globulin, 0.02% sodium azide, with 4% DMSO). Plates are mixed and incubated at room temperature for 2-3 hours with gentle shaking. The polarization values in millipolarization units (mP) are measured at an excitation wavelength of 485 nm and an emission wavelength of 530 nm. Equilibrium dissociation constants (Kd) are then calculated by fitting the sigmoidal dose-dependent FP increases as a function of protein concentrations using Graphpad Prism 5.0 software. In competitive binding experiments for XIAP3 BIR3, AT-406 is incubated with 20 nM XIAP BIR3 protein and 2 nM FL-AT-406 in the assay buffer (100 mM potassium phosphate, pH 7.5; 100 µg/mL bovine γ-globulin; 0.02% sodium azide). In competitive binding experiments for cIAP1 BIR3 protein, 3 nM protein and 1 nM FL-AT-406 are used. In competitive binding experiments for cIAP2 BIR3, 5 nM protein and 1 nM FL-AT-406 are used. For each competitive binding experiment, polarization values are measured after 2-3 hours of incubation using an Infinite M-1000 plate reader.The IC50 value, the inhibitor concentration at which 50% of the bound tracer is displaced, is determined from the plot using nonlinear least-squares analysis. Curve fitting is performed using the PRISM software. A Ki value for AT-406 is calculated.

Solubility Information

Solubility	DMSO: 6.88 mg/mL (13.63 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: 1 mg/mL (1.98 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	1.9816 mL	9.9082 mL	19.8165 mL
5 mM	0.3963 mL	1.9816 mL	3.9633 mL
10 mM	0.1982 mL	0.9908 mL	1.9816 mL
50 mM	0.0396 mL	0.1982 mL	0.3963 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

Heffron TP, et al. Bioorg Med Chem Lett. 2010 Apr 15;20(8):2408-11.