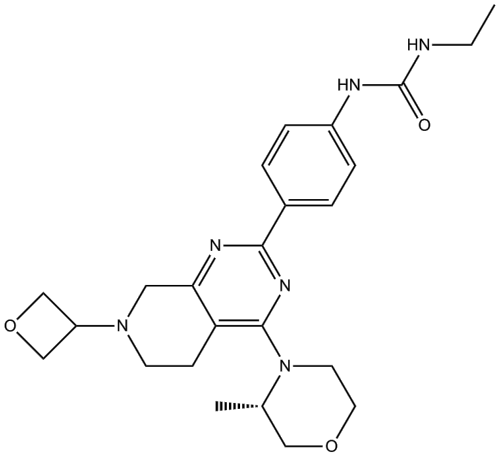
Product Data Sheet

Product Name: GDC-0349
Cat. No.: GC12021

Chemical Properties

Cas. No.	1207360-89-1		
Chemical Name	1-ethyl-3-[4-[4-[(3S)-3-methylmorpholin-4-yl]-7-(oxetan-3-yl)-6,8-dihydro-5H-pyrido[3,4-d]pyrimidin-2-yl]phenyl]urea		
SMILES	CCNC(=O)NC1=CC=C(C=C1)C2=NC3=C(CCN(C3)C4COC4)C(=N2)N5CCOCC5C		
Formula	$C_{24}H_{32}N_6O_3$	M.Wt	452.55
Solubility	≥ 4.55mg/mL in DMSO	Storage	Store at -20°C
General tips	For obtaining a higher solubility , please warm the tube at 37 °C and shake it in the ultrasonic bath for a while.Stock solution can be stored below -20°C for several months.		
Shipping Condition	Evaluation sample solution : ship with blue ice All other available size: ship with RT , or blue ice upon request.		
Structure			

Protocol

Product Data Sheet

Kinase experiment: The kinase activity of mTOR enzyme is assessed by incubating purified recombinant enzyme (mTOR(1360-2549)+GBL, prepared in-house) in a reaction mixture containing ATP, MnCl2, and a fluorescently labeled mTOR substrate, e.g., GFP-4E-BP1. The reaction is stopped by an addition of a Terbium-labeled phospho-specific antibody, e.g., Tb-labeled anti-p4E-BP1 T37/T46, EDTA, and TR-FRET buffer solution. Product formation is detected by way of time-resolved fluorescence resonance energy transfer (TR-FRET), which occurs when the phosphorylated substrate and labeled antibody are in close proximity due to phosphospecific binding. Enzymatic activity is measured as an increase in TR-FRET signal using a Perkin Elmer Envision plate reader. The assay is performed in a 384-well Proxiplate Plus using the following protocol: Compound activity is tested in 10 point dose curves starting at the highest final concentration of 10 uM. They are serially diluted in 100% DMSO prior to further dilution with assay buffer. The reaction mixture (8μL) containing 0.25 nM mTOR+GBL enzyme, 400 nM GFP-4E-BP1, 8 uM ATP, 50 mM Hepes pH 7.5, 0.01% Tween 20, 10 mM MnCl2, 1 mM EGTA, 1 mM DTT, 1% DMSO (±compound) is incubated at room temperature for 30 minutes. 8 μL of solution containing 2 nM Tb-anti-p4E-BP1 antibody & 10 mM EDTA diluted TR-FRET buffer is then added and incubated for 30 minutes to stop the reaction. The plate is scanned with the Envision plate reader. Ki values are calculated in Assay Explorer using the Morrison ATP-competitive tight binding equation for Ki apparent determination[1].

Product Data Sheet

Animal experiment: Mice[1]Human breast cancer cells (MCF7 neo/HER2; modified ATCC variant) are implanted subcutaneously into the mammary fat pad of female NCR nude mice (5×106 cells/100 uL of 1:1 mixture of Hank's Balanced Salt Solution (HBSS)/Matrigel). To support estrogen dependent growth, recipient animals are pre-implanted with 0.36 mg estrogen pellets. Tumors are monitored until they reached a mean tumor volume of approximately 200-225 mm3, then similarly sized tumors are randomly assigned to treatment cohorts (n=5-10). Human 786-O renal adenocarcinoma cells are implanted subcutaneously into the right hind flank of female nu/nu mice (1×107 cells/200 uL in 1:1 PBS/Matrigel). Tumors are monitored until they reached a mean tumor volume of approximately 205 mm3, then similarly sized tumors are randomly assigned to treatment cohorts (n=10). Human prostate cancer NCI-PC3 cells are resuspended in Hank's Balanced Salt Solution and implanted subcutaneously into the right hind flanks of 120 female NCR nude mice. Each mouse is injected with 5×106 cells. Tumors are monitored until they reached a mean tumor volume of approximately 200-250 mm3. The dimesylate salt of GDC-0349 is dosed daily or every third day by oral gavage (100 uL dose /25 gm animal) for 14-21 days.Tumor volume and body weight measurements are collected twice weekly. Tumor volumes are calculated.

References:

[1]. Pei Z, et al. Discovery and Biological Profiling of Potent and Selective mTOR Inhibitor GDC-0349. ACS Med Chem Lett. 2012 Nov 29;4(1):103-7.

Background

GDC-0349 is a potent, selective, ATP-competitive inhibitor of (Mammalian target of rapamycin) mTOR with Ki value of 3.8nM [1].

GDC-0349 is a remarkably selective mTOR inhibitor, with less than 25% inhibition of 266 kinases, including all isoforms of PI3K. GDC-0349 inhibited mTOR downstream markers , involving phospho-Akt(s473) and phospho-4EBP1 in an in vivo PK/PD study in mouse, consistent with an inhibition of mTORC1/mTORC2 complexes [1].

Product Data Sheet

GDC-0349 inhibited tumor growth in a dose-dependent manner when dosed orally once daily in athymic mice in a MCF7-neo/Her2 tumor xenograft model (PI3K mutation). GDC-0349 was also efficacious in other xenograft models, including PC3 (PTEN null) and 786-0 (VHL mutant). However, GDC-0349 showed only modest tumor growth delay in the A549 mouse xenograft model (KRas mutant, LKB1 deficient) [1].

References:

[1] Pei, Z.; Blackwood, E.; Liu, L.; Malek, S.; Belvin, M.; Koehler, M. F. T.; Ortwine, D. F.; Chen, H.; Cohen, F.; Kenny, J. R.; Bergeron, P.; Lau,K.; Ly, C.; Zhao, X.; Estrada, A. A.; Truong, T.; Epler, J. A.; Nonomiya, J.; Trinh, L.; Sideris, S.; Lesnick, J.; Bao, L.; Vijapurkar, U.; Mukadam,S.; Tay, S.; Deshmukh, G.; Chen, Y.-H.; Ding, X.; Friedman, L.; Lyssikatos, J. P.Discovery and biological profiling of potent and selective mTOR inhibitor GDC-0349 ACS Med. Chem. Lett. 2013, 4, 103– 107

Caution: Product has not been fully validated for medical applications. For research use only.

Tel: (909) 407-4943 Fax: (626) 353-8530 E-mail: tech@glpbio.com
Address: 10292 Central Ave. #205, Montclair, CA, USA