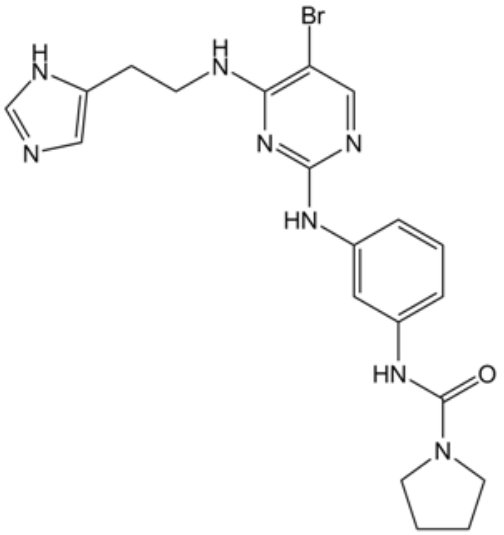
Product Data Sheet

Product Name: BX-912
Cat. No.: GC16818

Chemical Properties

Cas. No.	702674-56-4		
Chemical Name	N-[3-[[5-bromo-4-[2-(1H-imidazol-5-yl)ethylamino]pyrimidin-2-yl]amino]phenyl]pyrrolidine-1-carboxamide		
SMILES	C1CCN(C1)C(=O)NC2=CC=CC(=C2)NC3=NC=C(C(=N3)NCCC4=CN=CN4)Br		
Formula	$C_{20}H_{23}BrN_8O$	M.Wt	471.35
Solubility	≥ 23.55mg/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: PDK1 is assayed in a direct kinase assay and a coupled assay format measuring PDK1 and PtdIns-3,4-P2 mediated activation of AKT2. For the coupled assay, the final assay mixture (60 μL) contains: 15 mM MOPS, pH 7.2, 1 mg/mL bovine serum albumin, 18 mM β-glycerol phosphate, 0.7 mM dithiothreitol, 3 mM EGTA, 10 mM MgOAc, 7.5 μM ATP, 0.2 μCi of [γ-33P]ATP, 7.5 μM biotinylated peptide substrate (biotin-ARRRDGGGAQPFRPRAATF), 0.5 μL of PtdIns-3,4-P2-containing phospholipid vesicles, 60 pg of purified recombinant human PDK1, and 172 ng of purified recombinant human AKT2. After incubation for 2 h at room temperature, the biotin-labeled peptide is captured from 10 μL of the assay mixture on Streptavidin-coated SPA beads, and product formation is measured by scintillation proximity in a Wallac MicroBeta counter. The product formed is proportional to the time of incubation and to the amount of PDK1 and inactive AKT2 added. PDK1 is added at suboptimal levels so that the assay can sensitively detect inhibitors of AKT2 activation as well as direct inhibitors of PDK1 or AKT2[1].

Cell experiment: The cell lines MDA-468, MDA-453, HCT-116, U87-MG, U2OS, PC-3, B16F10, and MiaPaCa; LOX amelanotic human melanoma cells; and HeLa cells seeded at a low density (1,500-3,000 cells/well, 0.1 mL/well, 96-well plates) are incubated overnight. Compound treatments are made by adding 10 μL/well of BX-912 (1, 10, 100 and 1000 nM) in 1% DMSO and growth medium (final concentration of DMSO, 0.1%), followed by brief shaking. Treated cells are incubated for 72 h, and viability is measured by the addition of 10 μL of the metabolic dye WST-1. The WST-1 signal is read in a plate reader at 450 nm, and a no cell, or zero time cell, background is subtracted to calculate the net signal[1].

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

Product Data Sheet

References:

[1]. Feldman RI, et al. Novel small molecule inhibitors of 3-phosphoinositide-dependent kinase-1. J Biol Chem. 2005 May 20;280(20):19867-74.

Background

BX-912 is a potent and selective inhibitor of 3-phosphoinositide-dependent kinase 1 (PDK1) with IC50 value of 26nM [1]. BX-912 has shown 9-fold selectivity for PDK1 relative to PKA and 105-fold selectivity against PKC [1].

BX-912 has been revealed to suppress the MDA-468 cells (one kind of breast tumor cell expressing high levels of activated Akt) growth and increase caspase-3/7 activity. On the contrary, BX-912 had no effect on caspase-3/7 activation in normal primary PrECs and primary HMECs. Moreover, BX-912 has been demonstrated to induce cell cycle arrest of MDA-468 cells at the G2/M phase [1].

References:
[1] Feldman RI1, Wu JM, Polokoff MA, Kochanny MJ, Dinter H, Zhu D, Biroc SL, Alicke B, Bryant J, Yuan S, Buckman BO, Lentz D, Ferrer M, Whitlow M, Adler M, Finster S, Chang Z, Arnaiz DO. Novel small molecule inhibitors of 3-phosphoinositide-dependent kinase-1. J Biol Chem. 2005 May 20;280(20):19867-74. Epub 2005 Mar 16.