LIJTF500025: A Chemical Probe for LIMK1/2

Version 1.0 (17th July 203)

Web link for more details: https://www.thesgc.org/chemical-probes/th257

Overview

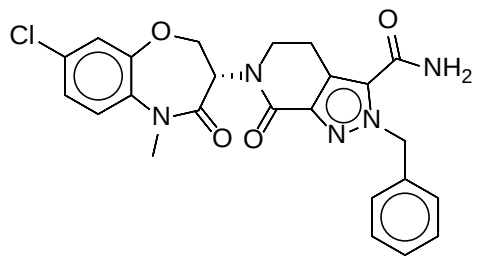
The LIM kinases are Ser/Thr kinases of the TKL family. LIMK1 and LIMK2 share 50% sequence identity (> 70% in KD). The LIM kinases are activated via different pathways such as the MK-2; Rho-ROCK; Rha-PAK pathway or also via the BMP-signalling pathway. The most prominent substrate for the LIM kinases is cofilin, that is inactivated via phosphorylation at Ser3. The LIMKs are involved in different cellular functions such as they promote actin turnover or are responsible for microtubule disassembly.

Summary

Chemical Probe Name	LIJTF500025
Negative control compound	LIJTF500120
Target(s) (synonyms)	LIMK1 / LIMK2; LIM domain kinase 1/2
Recommended cell assay concentration	Use at concentration of 1 µM; use with negative control LIJTF500120 + additional TP-030 for best interpretation of data.
Suitability for *in* vivo use and recommended dose	LIJTF500025 was not tested in vivo.
Publications	
Orthogonal chemical probes	
In vitro assay(s) used to characterise	ITC
Cellular assay(s) for target-engagement	NanoBRET™

Chemical Probe & Negative Control Structures and Use

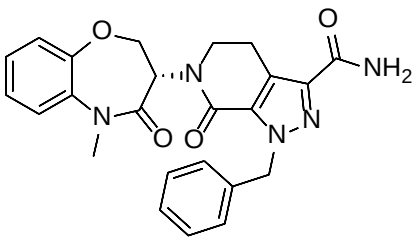
LIJTF500025 Chemical Probe



SMILES:
CN1C([C@H](COc2c1ccc(Cl)c2)N3CCc4c(C(N)=O)n(Cc5ccccc5)nc4C3=O)=O

InChiKey: KIBQDIDFPAQGOU-SFHVURJKSA-N
Molecular weight: 479.92
Storage Stable as solid in the dark at -20°C. NB making aliquots rather than freeze-thawing is recommended
Dissolution: Soluble in DMSO up to 10 mM; use only 1 freeze/thaw cycle per aliquot

LIJTF500120 Negative Control



SMILES:
CN1C2=CC=CC=C2OC[C@@H](C1=O)N3CCC4=C(C3=O)N(N=C4C(N)=O)CC5=CC=CC=C5
InChiKey: NQMLLMFMFLFBAI-SFHVURJKSA-N
Molecular weight: 445,48
Storage: Stable as solid in the dark at -20°C. NB making aliquots rather than freeze-thawing is recommended
Dissolution: Soluble in DMSO up to 10 mM; use only 1 freeze/thaw cycle per aliquot

Chemical Probe Profile

In vitro Potency & Selectivity:

LIJTF500025 had an K_D of 37 nM on LIMK1 determined by ITC. The selectivity of the compound was confirmed by DSF in the SGC-FFM panel against 107 kinases and by scanMAX Kinase Assay Panel from Eurofins(Discovery) against a panel of 468 kinases at a screening concentration of 1 µM. The selectivity score S35 (1µM) was 0.007 with the top hits being LIMK1 (6.8% of control), RIPK1 (23%) and LIMK1 (30%).

Potency in Cells and Cellular Target Engagement:

LIJTF500025 displayed an EC_{50} of 82 nM, 52 nM and 6 nM on LIMK1, LIMK2 and RIPK1 respectively in intact cells in the NanoBRET assay. LIJTF500025 was able to inhibit the phosphorylation of Cofilin in a dose dependent manner in LN229 cells. LIJTF500025 showed no cytotoxicity up to a concentration of 10 µM in three different cell lines (HEK293T; U2OS and MRC-9).