THNAN69: A Chemical Probe for LIMK2

Version 1.0 (23rd June 2025)

Web link for more details: https://www.sgc-ffm.uni-frankfurt.de/#!specificprobeoverview/THNAN69

Overview

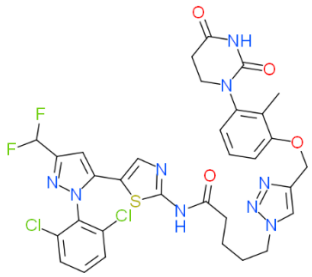
LIMK2 is phosphorylated and activated by ROCK, a downstream effector of Rho. Active LIMK2 phosphorylates cofilin, inhibiting its actin-depolymerizing activity. This relieves the levering stress on actin and allows polymerization to occur. Actin rearrangement is essential in regulating cell cycle progression, apoptosis, and migration. LIMK2 is ubiquitously expressed.

Summary

Chemical Probe Name	THNAN69 (degrader)
Negative control compound	THNAN69-NC
Target(s) (synonyms)	LIMK2
Recommended *in vitro* assay concentration	Use at concentration between 10 to 100 nM for THNAN69 and THNAN69-NC; use with negative control for best interpretation of data
Suitability for *in* vivo use and recommended dose	Not tested in vivo
Publications	None at time of writing
In vitro assay(s) used to characterise	
Cellular assay(s) for target-engagement	NanoBRET, EGFP-LIMK2 depletion assay

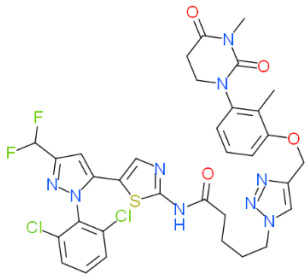
Chemical Probe & Negative Control Structures and Use

THNAN69 Chemical Probe



SMILES:
Cc1c(cccc1OCc1cn(CCCCC(Nc2ncc(c3cc(C(F)F)nn3c3c(cccc3[Cl])[Cl])s2)=O)nn1)N1CCC(NC1=O)=O
InChiKey: UJDFQDPAVQIQLB-UHFFFAOYSA-N
Molecular weight: 743.14 g/mol
Storage: As a dry powder or as DMSO stock solutions (10 mM) at -20 °C. DMSO stocks beyond 3-6 months or 2 freeze/thaw cycles should be tested for activity before use
Dissolution: Soluble in DMSO up to 10 mM; use only 1 freeze/thaw cycle per aliquot

THNAN69-NC Negative Control



SMILES
Cc1c(cccc1OCc1cn(CCCCC(Nc2ncc(c3cc(C(F)F)nn3c3c(cccc3[Cl])[Cl])s2)=O)nn1)N1CCC(N(C)C1=O)=O
InChiKey: ZBAWXHCJKJRWSY-UHFFFAOYSA-N
Molecular weight: 757.16 g/mol
Storage: As a dry powder or as DMSO stock solutions (10 mM) at -20 °C. DMSO stocks beyond 3-6 months or 2 freeze/thaw cycles should be tested for activity before use
Dissolution: Soluble in DMSO up to 10 mM; use only 1 freeze/thaw cycle per aliquot

Chemical Probe Profile

In vitro Potency & Selectivity:

The selectivity of THNAN69 for LIMK2 was confirmed by whole-cell proteomics. The LIMK1 NanoBRET EC_{50} is 412 nM, the DSF deltaTM = 6.9 °C and SPR binding KD = 249 nM.

Potency in Cells and Cellular Target Engagement:

For LIMK2 the NanoBRET EC_{50} is 80 nM. In the EGFP-LIMK2 depletion assay DC_{50} = 1 nM. THNAN69 degrades LIMK2 at 10 nM with a D_{max} of ~90%.