

TP-030-2: A Chemical Probe for RIPK1

Version 1.0 (25th March 2021)

Web link for more details: <https://www.sgc-ffm.uni-frankfurt.de/#!specificprobeoverview/TP-030-2>

Overview

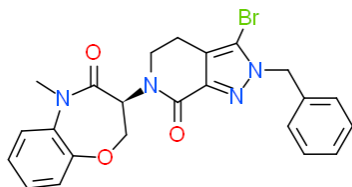
The serine/threonine protein kinase [RIPK1](#) functions in various cellular pathways and is a crucial upstream regulator of necroptosis. It forms a complex with various proteins, e.g. RIPK3 and is associated with a variety of pathologies such as ischemic injury, inflammatory diseases, neurodegenerative diseases.

Summary

Chemical Probe Name	TP-030-2
Negative control compound	TP-030n
Target(s) (synonyms)	RIPK1 (receptor interacting serine/threonine kinase 1, RIP)
Recommended cell assay concentration	Use at concentration of 100 nM for TP-030-2 and TP-030n; use with control and orthogonal probe for best interpretation of data
Suitability for <i>in vivo</i> use and recommended dose	Suitable for <i>in vivo</i> use, preliminary tests were done in mice.
Publications	PMID: 29485864
Orthogonal chemical probes	TP-030-1
<i>In vitro</i> assay(s) used to characterise	TR-FRET
Cellular assay(s) for target-engagement	HT29 necroptosis assay

Chemical Probe & Negative Control Structures and Use

TP-030-2 Chemical Probe



SMILES: CN1C([C@H](COc2ccccc12)N1CCc2c(C1=O)nn(Cc1ccccc1)c2[Br])=O

InChIKey: KHVVJKLNKLQJZ-SFHVURJKSA-N

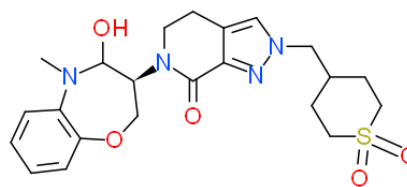
Molecular weight: 480.08

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

TP-030n Negative Control



SMILES:

CN1C([C@H](COc2ccccc12)N1CCc2cn(CC3CCS(CC3)(=O)=O)nc2C1=O)=O

InChIKey: IVVSBEQDCAZLFY-SFHVURJKSA-N

Molecular weight: 458.2

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:

TP-030-2 shows potent activity on human RIPK1 ($K_i = 0.43$ nM, TR-FRET) and mouse ($IC_{50} = 100$ nM). No significant binding was observed at $1 \mu\text{M}$ for 303 kinases tested (Takeda Global Kinase Panel) for TP-030-2 and TP-030n. The only hit found in the Eurofins-Panlabs screen of 68 targets for TP-030-2 was HTR2B with 63% inhibition. The same screen was clean for TP-030n. TP-030n is not active in the TR_FRET assay: hRIPK1 ($K_i = 6.9 \mu\text{M}$), mRIPK1 ($K_i > 10 \mu\text{M}$).

Potency in Cells and Cellular Target Engagement:

TP-030-2 is a highly potent in the HT29 necroptosis assay with $IC_{50} = 1.3$ nM.