MN714: A Chemical Probe for SOCS2

Version 1.0 (18th October 2023)

Web link for more details: https://doi.org/10.1038/s41467-023-41894-3

Overview

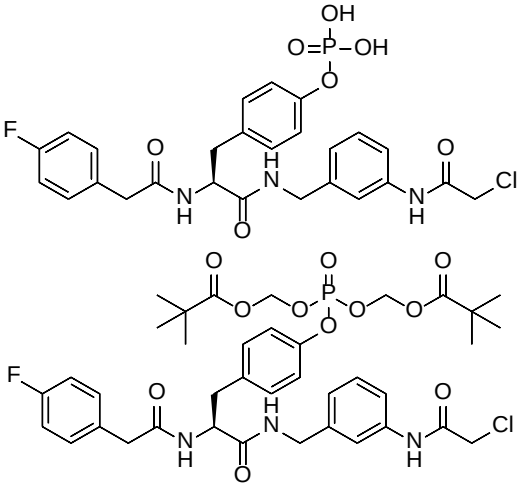
Suppressor of cytokine signalling 2 (SOCS2) is a substrate recognition subunit of a Cullin RING E3 ligase and is involved in JAK-STAT signalling pathways. The $CRL5^{SOCS2}$ ligase complex binds phosphotyrosine residues *via* a conserved SH2 domain and targets these post-translationally modified proteins for proteasomal regulation. Dysregulation of SOCS2 also plays an important role in cancer progression and inflammatory diseases. **MN714** is a POM-protected prodrug, releasing the active species **MN551** with 40 minutes half-life time.

Summary

Chemical Probe Name	MN551/MN714
Negative control compound	RC292
Target(s) (synonyms)	Suppressor of cytokine signalling 2 (SOCS2)
Recommended *in vitro* assay concentration	Use at concentration up to 10 µM for **MN551** and **RC292**; use with control for best interpretation of data
Suitability for *in vivo* use and recommended dose	Below 10 µM in cellular assays. Not tested in animal models.
Publications	PMID: 37816714
Orthogonal chemical probes	-
In vitro assay(s) used to characterise	ITC, SPR, FP, competitive ^{19}F NMR, intact MS
Cellular assay(s) for target-engagement	Live cell HiBiT CETSA, SOCS2 pull-down/proteomics

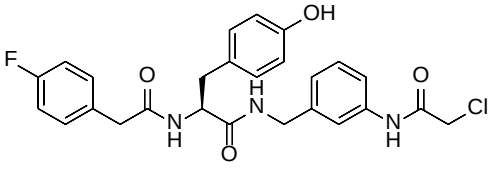
Chemical Probe & Negative Control Structures and Use

Chemical Probe (MN551, top), prodrug (MN714, bottom)



SMILES:
MN551:
O=C(N[C@@H](CC1=CC=C(OP(O)(O)=O)C=C1)C(NCC2=CC=CC(NC(CCl)=O)=C2)=O)CC3=CC=C(C=C3)F
MN714:
FC1=CC=C(CC(N[C@H](C(NCC2=CC=CC(NC(CCl)=O)=C2)=O)CC3=CC=C(OP(OOC(C(C)(C)C)=O)(OCOC(C(C)(C)C)=O)=O)C=C3)=O)C=C1
InChiKey:
MN551: WSEFGYSEXKXLRO-QHCPKHFHSA-N
MN714: VVIUEMCRCUOKIE-HKBQPEDESA-N
Molecular weight: *MN551:* 577.93 g/mol. *MN714:* 806.22 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.

Negative Control



SMILES:
FC1=CC=C(CC(N[C@H](C(NCC2=CC=CC(NC(CCl)=O)=C2)=O)CC3=CC=C(O)C=C3)=O)C=C1

InChiKey: COTZHXZLSPZSAU-QHCPKHFHSA-N

Molecular weight: 497.95 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:

MN714 does not bind SOCS2 directly in vitro. The active species **MN551** shows potent activity on SOCS2 (k_{inact}/K_I = 58 M^{-1} s^{-1}) and no activity on $SOCS2^{C111S}$, SOCS4. Covalent modification of SOCS2 by **MN551** blocks the interaction with phosphorylated GHR peptides in vitro.

Potency in Cells and Cellular Target Engagement:

The EC_{50} for increase of T_{agg} in live cell split-NanoLuc CETSA was 2.52 ±0.42 μM after 8 hours treatment in HeLa cells. **MN714** treatment abrogates SOCS2 pulldown from K562 cells with phosphorylated GHR peptide with EC_{50} of 5.9 ±2.6 μM.