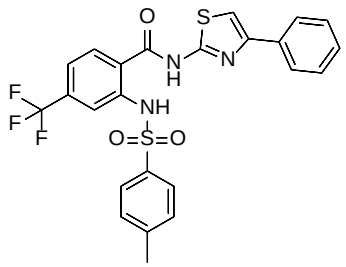
Product data sheet

MedKoo Cat#: 407457 Name: ML364 CAS: 1991986-30-1 Chemical Formula: $C_{24}H_{18}F_3N_3O_3S_2$ Exact Mass: 517.0742 Molecular Weight: 517.5412	
Product supplied as:	Powder
Purity (by HPLC):	≥ 98%
Shipping conditions	Ambient temperature
Storage conditions:	Powder: -20°C 3 years; 4°C 2 years.
	In solvent: -80°C 3 months; -20°C 2 weeks.

1. Product description:
ML364 is a small molecule inhibitor of the deubiquitinase USP2 with potential anticancer activity. ML364 has an IC50 of 1.1 μm in a biochemical assay using an internally quenched fluorescent di-ubiquitin substrate. Direct binding of ML364 to USP2 was demonstrated using microscale thermophoresis. ML364 induced an increase in cellular cyclin D1 degradation and caused cell cycle arrest as shown in Western blottings and flow cytometry assays utilizing both Mino and HCT116 cancer cell lines.

2. CoA, QC data, SDS, and handling instruction
SDS and handling instruction, CoA with copies of QC data (NMR, HPLC and MS analytical spectra) can be downloaded from the product web page under "QC And Documents" section. Note: copies of analytical spectra may not be available if the product is being supplied by MedKoo partners. Whether the product was made by MedKoo or provided by its partners, the quality is 100% guaranteed.

3. Solubility data

Solvent	Max Conc. mg/mL	Max Conc. mM
DMF	30.0	57.97
DMSO	37.67	72.78
DMSO:PBS (pH 7.2) (1:2)	0.33	0.64
Ethanol	22.5	43.47

4. Stock solution preparation table:

Concentration / Solvent Volume / Mass	1 mg	5 mg	10 mg
1 mM	1.93 mL	9.66 mL	19.32 mL
5 mM	0.39 mL	1.93 mL	3.86 mL
10 mM	0.19 mL	0.97 mL	1.93 mL
50 mM	0.04 mL	0.19 mL	0.39 mL

5. Molarity Calculator, Reconstitution Calculator, Dilution Calculator
Please refer the product web page under section of "Calculator"

6. Recommended literature which reported protocols for in vitro and in vivo study
In vitro study

1. Hashimoto M, Saito N, Ohta H, Yamamoto K, Tashiro A, Nakazawa K, Inanami O, Kitamura H. Inhibition of ubiquitin-specific protease 2 causes accumulation of reactive oxygen species, mitochondria dysfunction, and intracellular ATP decrement in C2C12 myoblasts. Physiol Rep. 2019 Jul;7(14):e14193. doi: 10.14814/phy2.14193. PMID: 31353872; PMCID: PMC6661303.
2. Davis MI, Pragani R, Fox JT, Shen M, Parmar K, Gaudiano EF, Liu L, Tanega C, McGee L, Hall MD, McKnight C, Shinn P, Nelson H, Chattopadhyay D, D'Andrea AD, Auld DS, DeLucas LJ, Li Z, Boxer MB, Simeonov A. Small Molecule Inhibition of the Ubiquitin-specific Protease USP2 Accelerates cyclin D1 Degradation and Leads to Cell Cycle Arrest in Colorectal Cancer and Mantle Cell Lymphoma Models. J Biol Chem. 2016 Nov 18;291(47):24628-24640. doi: 10.1074/jbc.M116.738567. Epub 2016 Sep 28. PMID: 27681596; PMCID: PMC5114414.

In vivo study

Product data sheet

1. Zhang Y, Dong L, Sun L, Hu X, Wang X, Nie T, Li X, Wang P, Pang P, Pang J, Lu X, Yao K, You X. ML364 exerts the broad-spectrum antivirulence effect by interfering with the bacterial quorum sensing system. Front Microbiol. 2022 Dec 22;13:980217. doi: 10.3389/fmicb.2022.980217. PMID: 36619997; PMCID: PMC9813848.
2. Hashimoto M, Fujimoto M, Konno K, Lee ML, Yamada Y, Yamashita K, Toda C, Tomura M, Watanabe M, Inanami O, Kitamura H. Ubiquitin-Specific Protease 2 in the Ventromedial Hypothalamus Modifies Blood Glucose Levels by Controlling Sympathetic Nervous Activation. J Neurosci. 2022 Jun 8;42(23):4607-4618. doi: 10.1523/JNEUROSCI.2504-21.2022. Epub 2022 May 3. PMID: 35504726; PMCID: PMC9186793.

7. Bioactivity

Biological target:

ML364 is a selective ubiquitin specific peptidase 2 (USP2) inhibitor (IC_{50}=1.1 μM).

In vitro activity

Treatment with this dose of ML364 for 5 days caused > 70% decrease in the incorporation of BrdU to the cells; thus, ML364 decreased the proliferation of C2C12 cells (Fig. 7B). On the other hand, treatment with ML364 for 8 h also decreased the intracellular level of ATP to 64.9% of vehicle-treated cells (Fig. 7C). Moreover, treatment with ML364 markedly decreased the potential of the mitochondrial membrane in C2C12 cells (0.37-fold difference vs. that of vehicle-treated cells, $P < 0.001$; Fig. 7D).

Reference: Physiol Rep. 2019 Jul;7(14):e14193. https://pubmed.ncbi.nlm.nih.gov/31353872/

In vivo activity

In the P. aeruginosa PAO1 or the CRPA 16-2 infection mouse model, ML364 treatment could significantly improve the survival rate from 0% to 70% or from 30% to 90%, respectively. The survival rates of mice infected with S. aureus ATCC 29213 or MRSA 08-50 could be increased from 20% to 50% or even from 10% to 70%, by the administration of ML364 (Figure 5C). In Figure 5D, ML364 was able to significantly reduce the bacterial loads in the organs of mice ($p \leqslant 0.01$), when the mice were systemically infected with CRPA 16-2 or MRSA 08-50.

Reference: Front Microbiol. 2022 Dec 22;13:980217. https://pubmed.ncbi.nlm.nih.gov/36619997/