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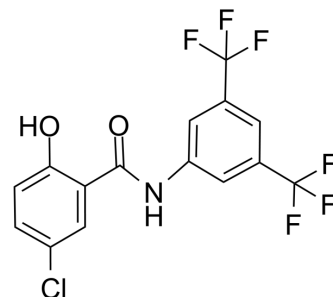
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IMD-0354

Cat. No.:	HY-10172		
CAS No.:	978-62-1		
Molecular Formula:	C ₁₅ H ₈ ClF ₆ NO ₂		
Molecular Weight:	383.67		
Target:	IKK		
Pathway:	NF-κB		
Storage:	Powder	-20°C	3 years
		4°C	2 years
	In solvent	-80°C	2 years
		-20°C	1 year



SOLVENT & SOLUBILITY

In Vitro

DMSO : ≥ 100 mg/mL (260.64 mM)
 H₂O : < 0.1 mg/mL (ultrasonic;warming;heat to 60°C) (insoluble)
 * "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent Concentration	Mass		
		1 mg	5 mg	10 mg
	1 mM	2.6064 mL	13.0320 mL	26.0641 mL
	5 mM	0.5213 mL	2.6064 mL	5.2128 mL
	10 mM	0.2606 mL	1.3032 mL	2.6064 mL

Please refer to the solubility information to select the appropriate solvent.

In Vivo

- Add each solvent one by one: 10% DMSO >> 40% PEG300 >> 5% Tween-80 >> 45% saline
 Solubility: ≥ 2.5 mg/mL (6.52 mM); Clear solution
- Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)
 Solubility: 2.5 mg/mL (6.52 mM); Suspended solution; Need ultrasonic

BIOLOGICAL ACTIVITY

Description

IMD-0354 (IKK2 Inhibitor V) is a selective IKKβ inhibitor which inhibits NF-κB activity^[1]. IMD0354 inhibits TNF-α induced NF-κB transcription activity with an IC₅₀ of 1.2 μM^[2].

IC₅₀ & Target

IKKβ	NF-κB
	1.2 μM (IC ₅₀)

In Vitro

IMD-0354 inhibits NF-κB activity in HMC-1 cells, resulting in complete repression of growth factor-independent proliferation of mast cells. When the DNA-binding activity of NF-κB is inhibited by treatment with IMD-0354, cell proliferation is

completely suppressed. HMC-1 cells are incubated with increasing concentrations of IMD-0354 or STI571 for 24, 48, and 72 hours, and numbers and viability of cells are determined by a dye exclusion test and an MTT assay. IMD-0354 suppresses cell proliferation in a time- and dose-dependent manner. The inhibitory effect of IMD-0354 is remarkable, even at lower concentrations, when compared with that of STI571^[1]. IMD0354 inhibits TNF- α induced NF- κ B transcription activity with an IC₅₀ of 1.2 \pm 0.3 μ M^[2].

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

In Vivo

Daily administration with 5 mg/kg IMD-0354 significantly suppresses tumor expansion in nude mice implanted with established MDA-MB-231 tumors. In mice treated with IMD-0354, tumor progression is restrained^[3]. The number of infiltrating cells in aqueous humor is 53.6 \pm 9.8 \times 10⁵, 72.5 \pm 17.0 \times 10⁵, 127.25 \pm 32.0 \times 10⁵, and 132.0 \pm 25.0 \times 10⁵ cells/mL in rats treated with 30, 10, 3, or 0 mg/kg of IMD-0354, respectively. The total protein concentrations of aqueous humor are 92.6 \pm 3.1 mg/mL, 101.5 \pm 6.8 mg/mL, 112.6 \pm 1.9 mg/mL, and 117.33 \pm 1.8 mg/mL in rats treated with 30, 10, 3, and 0 mg/kg of IMD-0354, respectively^[4].

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL

Cell Assay^[1]

HMC-1 cells (2 \times 10⁵ cells/mL) are incubated with various concentrations of IMD-0354 (0.1, 0.5, 1, 5 and 10 μ M), STI571, or pyrrolidine dithiocarbamate (PDTTC) for the indicated hours, and viable cell numbers are calculated with the use of the trypan blue dye exclusion test at each time point. Cells (2 \times 10⁵ cells/mL) are incubated in phenol red free α -MEM containing 10% FCS (for HMC-1 and IC-2 cells) or 5% FCS (for CBhCMCs), and antibiotics with or without various concentrations of IMD-0354 (0.1, 0.5, 1, 5 and 10 μ M), STI571, or PDTTC. IC-2^{WT} cells and CBhCMCs are incubated in the presence of 100 ng/mL recombinant rat or recombinant human SCF. One hundred microliters of cell suspension is applied to each well of 96-well culture plates and are incubated for 24, 48, and 72 hours. Before 4 hours from the end of the culture, 10 μ L of 5 mg/mL MTT dissolved in PBS is added to each well. The reaction is stopped with the addition of 100 μ L of 10% SDS in 0.01 N HCl. Absorbance is measured at 577 nm with ImmunoMini NJ-2300^[1].

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Animal Administration^{[3][4]}

Mice^[3]

MDA-MB-231 cells are suspended in PBS (5 \times 10⁶ cells/100 μ L mouse) and s.c. injected to the back of female BALB/c nude mice at the age of 4 to 5 weeks. After growth, the tumor is removed surgically and 100 mg of each established tumor is transplanted to the back of other female nude mice at the age of 4 weeks under ether anesthesia. IMD-0354 is suspended in saline and 5 mg/kg body weight IMD-0354 (suspended in 100 μ L/mouse) is given to each mouse by i.p. injection once a day for 28 days after the implantation. Saline is injected in nude mice as a control. Estimated tumor volume (mm³) and tumor weight (mg) are calculated.

Rats^[4]

Eight-week-old male Lewis rats (180-220 g) are used. Endotoxin-induced uveitis (EIU) is induced with subcutaneous injection with 200 μ g LPS from Escherichia coli that has been diluted in 200 μ L PBS. At the same time, the rats are injected intraperitoneally with 30, 10, or 3 mg/kg of IMD-0354, diluted in 500 μ L of 0.5% CMC. Control EIU rats are intraperitoneally administered 500 μ L of CMC alone. Naïve rats are used as controls. All experiments are performed in triplicate with five animals in each group.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

CUSTOMER VALIDATION

- Sci Adv. 2023 Oct 6;9(40):eadi8343.
- Cancer Immunol Res. 2023 Apr 16;OF1-OF14.
- Pharmacol Res. 2022 Dec 7;187:106593.

- Cell Death Dis. 2020 Jun 12;11(6):455.
- Mol Ther Nucleic Acids. May 20, 2022.

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REFERENCES

- [1]. Tanaka A, et al. A novel NF-kappaB inhibitor, IMD-0354, suppresses neoplastic proliferation of human mast cells with constitutively activated c-kit receptors. Blood. 2005 Mar 15;105(6):2324-31.
- [2]. Li YR, et al. Study of the inhibitory effects on TNF- α -induced NF- κ B activation of IMD0354 analogs. Chem Biol Drug Des. 2017 Dec;90(6):1307-1311.
- [3]. Tanaka A, et al. A new IkappaB kinase beta inhibitor prevents human breast cancer progression through negative regulation of cell cycle transition. Cancer Res. 2006 Jan 1;66(1):419-26.
- [4]. Lennikov A, et al. Amelioration of endotoxin-induced uveitis treated with an IkB kinase β inhibitor in rats. Mol Vis. 2012;18:2586-97.
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Caution: Product has not been fully validated for medical applications. For research use only.

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