Pinometostat

Cat. No.: HY-15593

CAS No.: 1380288-87-8

Molecular Formula: $C_{30}H_{42}N_8O_3$

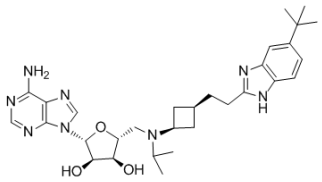
Molecular Weight: 562.71

Target: Histone Methyltransferase

Pathway: Epigenetics

Storage:

Powder	-20°C	3 years
	4°C	2 years
In solvent	-80°C	6 months
	-20°C	1 month



SOLVENT & SOLUBILITY

In Vitro

DMSO : ≥ 47.8 mg/mL (84.95 mM)

* "≥" means soluble, but saturation unknown.

Preparing Stock Solutions

Solvent / Mass / Concentration	1 mg	5 mg	10 mg
1 mM	1.7771 mL	8.8856 mL	17.7711 mL
5 mM	0.3554 mL	1.7771 mL	3.5542 mL
10 mM	0.1777 mL	0.8886 mL	1.7771 mL

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

In Vivo

1. Add each solvent one by one: 10% DMSO >> 40% PEG300 >> 5% Tween-80 >> 45% saline
   Solubility: ≥ 1.67 mg/mL (2.97 mM); Clear solution
2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)
   Solubility: ≥ 1.67 mg/mL (2.97 mM); Clear solution
3. Add each solvent one by one: 10% DMSO >> 90% corn oil
   Solubility: ≥ 1.67 mg/mL (2.97 mM); Clear solution

BIOLOGICAL ACTIVITY

Description

Pinometostat (EPZ-5676) is a potent DOT1L histone methyltransferase inhibitor with a K_i of 80 pM.

IC_{50} & Target

Ki: < 80 pM (DOT1L histone methyltransferase)

In Vitro

Pinometostat (EPZ-5676) inhibits H3K79me2 with IC_{50} values of 3 nM and 5 nM in MV4-11 and HL60 cells, respectively. Pinometostat (EPZ-5676) is a potent inhibitor of MV4-11 proliferation with an IC_{50} value of 3.5 nM[1]. Pinometostat (EPZ-

	5676) induces a synergistic and durable antiproliferative effect, increases expression of differentiation markers and apoptosis as single agent, and demonstrates combination benefit in combination with AML standard of care drugs in MLL-r cells[2]. MCE has not independently confirmed the accuracy of these methods. They are for reference only.
In Vivo	Pinometostat (EPZ-5676) (70 mg/kg, i.p.) causes complete and sustained regression in a rat xenograft model of MLL-rearranged leukemia. Pinometostat (EPZ-5676) (70, 35 mg/kg, i.v.) reduces HOXA9 and MEIS1 mRNA levels of tumors taken from rats, and reduces MLL-fusion target gene expression in vivo[1]. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL

Cell Assay [1]	To analyse inhibition of histone methylation in MV4-11 cells following Pinometostat treatment, extracted histones (400 ng) are fractionated on a 10-20% Tris HCl gels with Tris-Glycine SDS running buffer under denaturing conditions and transferred to nitrocellulose filters. Filters are cut into strips and incubated for 1 hour in blocking buffer at room temperature (RT) and then incubated overnight at 4°C in blocking buffer. Filters are washed 3 times for 5 minutes with wash buffer (Phosphate buffered saline (PBS) including 0.01% Tween 20 (PBST)) and incubated with infrared tagged secondary antibody at RT for 1 hour. Filters are washed in PBST and reprobed for 1 hour at RT with the appropriate total histone antibody control (mouse anti-histone H3 (1:20,000), CST 3638, or mouse anti-histone H4 (1:10,000), CST 2935). Filters are washed again in PBST and incubated with infrared tagged secondary antibody (IRDye 800Cw donkey-anti-mouse IgG (1:20,000), Li-Cor 926-32212) at RT for 1 hour. After a final ish in PBST, filters are scanned using the Odyssey infared imager (Li-cor). To analyse inhibition of H3K79 methylation in peripheral blood mononuclear cells (PBMCs) from rats dosed with Pinometostat (EPZ-5676), 20 μL of PBMC whole cell lysate is fractionated on denaturing gels and analysed by immunoblotting with antibodies to H3K79me2 or total H3. Signal intensities specific for the H3K79me2 antibody and total histone H3 control antibody are quantified using Odyssey software. The H3K79me2 signal intensity is normalized by dividing it by the total histone H3 control signal intensity in the same lane. MCE has not independently confirmed the accuracy of these methods. They are for reference only.
Animal Administration [1]	0.2 mL of a MV4-11 cell suspension (1×10^7 cells) in PBS is injected subcutaneously into female athymic nude mice (Crl:NU(Ncr)-Foxn1nu). Tumors are measured by calipers and mice are randomized according to tumor size into treatment groups (n=10) before the initiation of dosing with Pinometostat (EPZ-5676) when tumor volumes reache approximately 100 mma^3. Pinometostat is administered intraperitoneally three times daily for 28 days at 10 and 20 mg/kg in 10% ethanol in saline. Mice are weighed and tumors measured with calipers twice weekly until the end of the study. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

CUSTOMER VALIDATION

- Cell Syst. 2018 Apr 25;6(4):424-443.e7.
- Cell Death Dis. 2018 Jan 26;9(2):129.
- BMC Cancer. 2016 Aug 31;16:700.
- bioRxiv. 2020 Jun.
- bioRxiv. 2019 Nov.

See more customer validations on www.MedChemExpress.com

REFERENCES

[1]. Daigle SR, et al. Potent inhibition of DOT1L as treatment for MLL-fusion leukemia. Blood. 2013 Jun 25. [Epub ahead of print]

[2]. Klaus CR, et al. DOT1L inhibitor EPZ-5676 displays synergistic antiproliferative activity in combination with standard of care drugs and hypomethylating agents in MLL-rearranged leukemia cells. J Pharmacol Exp Ther. 2014 Sep;350(3):646-56.

India Contact:
Life Technologies (India) Pvt. Ltd.
306, Aggarwal City Mall, Opposite M2K Pitampura, Delhi – 110034 (INDIA). Ph: +91-11-42208000, 42208111, 42208222, Mobile: +91-9810521400, Fax: +91-11-42208444
Email: customerservice@lifetechindia.com Website: www.lifetechindia.com