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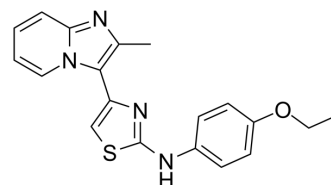
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JK184

Cat. No.:	HY-13307
CAS No.:	315703-52-7
Molecular Formula:	C ₁₉ H ₁₈ N ₄ OS
Molecular Weight:	350.44
Target:	Hedgehog
Pathway:	Stem Cell/Wnt
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 : ≥ 50 mg/mL (142.68 mM)
 * "≥" means soluble, but saturation unknown.

	Solvent Concentration	Mass	1 mg	5 mg	10 mg
Preparing Stock Solutions	1 mM		2.8536 mL	14.2678 mL	28.5356 mL
	5 mM		0.5707 mL	2.8536 mL	5.7071 mL
	10 mM		0.2854 mL	1.4268 mL	2.8536 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 (7.13 mM); Clear solution
- Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)
Solubility: ≥ 2.5 mg/mL (7.13 mM); Clear solution
- Add each solvent one by one: 10% DMSO >> 90% corn oil
Solubility: ≥ 2.5 mg/mL (7.13 mM); Clear solution

BIOLOGICAL ACTIVITY

Description	JK184 is a potent Hedgehog (Hh) pathway inhibitor with IC ₅₀ of 30 nM in mammalian cells.
IC ₅₀ & Target	IC ₅₀ : 30 nM (Hedgehog) ^[1]
In Vitro	JK184 is designed to antagonize Hh signaling by inhibiting glioma (Gli)-dependent transcriptional activity in a dose dependent manner. JK184 significantly inhibits proliferation of HUVECs with IC ₅₀ of 6.3 μg/mL after three days incubation.

To evaluate anti-tumor effect of JK184, MTT assay is conducted in Panc-1 and BxPC-3 cells after administration with indicated concentrations of compounds, half maximal inhibitory concentration (IC₅₀) of JK184 (23.7 ng/mL in anc-1 and 34.3 ng/mL in BxPC-3)^[1]. Claudin-low cell lines are more sensitive to JK184 treatment than are MCF10a, MTSV1-7, or HMLE-shGFP and HMLE-pBP cells, and JK184 induced a dose-dependent decrease in glioma-associated oncogene homolog 1 (GLI1) transcript and protein levels in these cells. Treatment with the IC₅₀ dose of JK184 enhances the proportion of HMLE-shEcad cells that stained with Annexin-V, but are negative for propidium iodide (PI) (P<0.0001, t test)^[2]. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

In Vivo

JK184 (5 mg/kg, injected intravenously) exhibits good anti-proliferative activity in subcutaneous Panc-1 and BxPC-3 tumor models, and is a good candidate as antitumor drug targeted Hh signaling. However, JK184 has a poor pharmacokinetic profile and bioavailability^[1]. MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL

Cell Assay ^[1]

The Shh-LIGHT2 cells are seeded in 96-well plates and grown to confluency. The Shh-LIGHT2 cells are treated with various concentrations of JK184 micelles or free JK184 or micelles in DMEM containing 0.5% CS, 0.1 mg/mL streptomycin, 100 U/mL penicillin, 5% Shh-N conditioned medium obtained from Shh-N-producing HEK293 cells. The treated cells are cultured further for 60 h, and firefly and Renilla luciferase activities are measured using a dual luciferase kit. Proliferation assay or apoptosis evaluation of HUVECs is measured using MTT method or FCM analysis, respectively. HUVECs are treated with a series concentration of free JK184, JK184 micelles, or blank MPEG-PCL micelles for 48 h, respectively. The mean percentage of cell inhibition or apoptosis is calculated^[1].

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

Animal Administration ^[1]

Mice^[1]

Five-week-old female athymic (nu/nu) mice are used. BxPC-3 and Panc-1 tumors are established by s.c. injection of 1×10^7 cells. Mice bearing tumors around 100 mm³ are selected and randomized into treatment groups (5 mice per group). Mice are injected intravenously every day for 30 days with 100 μ L of NS (control), blank micelles, free JK184 (5 mg/kg body weight), or JK184 micelles (5 mg/kg body weight), respectively. Tumor length and width are determined every 3 days and tumor volume (TV) is calculated using the following formula: $TV = 0.5 \times \text{length} \times \text{width}^2$. At the end of experiment, mice are sacrificed. Solid tumors are removed and processed for immunohistochemical analysis and terminal deoxynucleotidyl transferase-mediated dUTP nick end labeling (TUNEL) assay.

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

CUSTOMER VALIDATION

- Aging (Albany NY). 2020 Jul 20;12(16):16270-16293.
- J Oncol. 2020 Jun 2;2020:1657896.

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REFERENCES

- [1]. Zhang N, et al. Biodegradable polymeric micelles encapsulated JK184 suppress tumor growth through inhibiting Hedgehog signaling pathway. Nanoscale. 2015 Feb 14;7(6):2609-24.
- [2]. Colavito SA, et al. Significance of glioma-associated oncogene homolog 1 (GLI1) expression in claudin-low breast cancer and crosstalk with the nuclear factor kappa-light-chain-enhancer of activated B cells (NFkB) pathway. Breast Cancer Res. 2014 Sep 25;16

Caution: Product has not been fully validated for medical applications. For research use only.

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