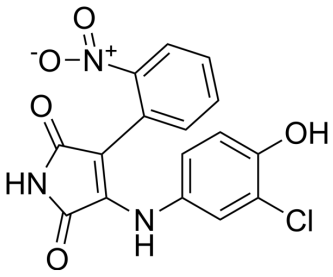
SB 415286

Cat. No.:	HY-15438
CAS No.:	264218-23-7
Molecular Formula:	$C_{16}H_{10}ClN_3O_5$
Molecular Weight:	359.72
Target:	GSK-3; Apoptosis
Pathway:	PI3K/Akt/mTOR; Stem Cell/Wnt; Apoptosis
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 (277.99 mM; Need ultrasonic)

Preparing Stock Solutions

Solvent / Mass / Concentration	1 mg	5 mg	10 mg
1 mM	2.7799 mL	13.8997 mL	27.7994 mL
5 mM	0.5560 mL	2.7799 mL	5.5599 mL
10 mM	0.2780 mL	1.3900 mL	2.7799 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: ≥ 2.5 mg/mL (6.95 mM); Clear solution
2. Add each solvent one by one: 10% DMSO >> 90% (20% SBE-β-CD in saline)
   Solubility: ≥ 2.5 mg/mL (6.95 mM); Clear solution

BIOLOGICAL ACTIVITY

Description

SB 415286 is a potent and selective cell permeable inhibitor of GSK-3α, with an IC_{50} of 77.5 nM, and a K_i of 30.75 nM; SB 415286 is equally effective at inhibiting human GSK-3α and GSK-3β.

IC_{50} & Target

hGSK-3α	hGSK-3β
77.5 nM (IC_{50})	77.5 nM (IC_{50})

In Vitro

SB 415286 (SB-415286) inhibits human GSK-3α with an IC_{50} of 77.5 nM, and a K_i of 30.75 nM. SB-415286 stimulates glycogen synthesis in the Chang human liver cell line with EC_{50} of 2.9 μM. SB-415286 stimulates glycogen synthase activity in Chang human liver cells. SB-415286 induces transcription of a β-catenin-LEF/TCF regulated reporter gene in HEK293 cells[1]. SB 415286 (SB-415286, 5-44 μM) attenuates B65 cell loss mediated by 1 mM H_2O_2. SB-415286 (5-44 μM) causes a significant

dose-dependent decrease in the fluorescence intensity of DCF, and attenuates B65 ROS production as mediated by 1 mM H_2O_2. SB-415286 (5-44 μM) also attenuates ROS production in CGN mediated by 1 mM H_2O_2[2]. SB-415286 (50 μM) induces a substantial suppression of immunoprecipitated GSK3 activity by 97%[3].

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

PROTOCOL

Kinase Assay [1]

GSK-3 kinase activity is measured, in the presence or absence of SB-216763 or SB-415286, in a reaction mixture containing final concentrations of: 1 nM human GSK-3α or rabbit GSK3α; 50 mM MOPS pH 7.0; 0.2 mM EDTA; 10 mM Mg-acetate; 7.5 mM β-mercaptoethanol; 5% (w/v) glycerol; 0.01% (w/v) Tween-20; 10% (v/v) DMSO; 28 μM GS-2 peptide substrate. The GS-2 peptide sequence corresponds to a region of glycogen synthase that is phosphorylated by GSK-3. The assay is initiated by the addition of 0.34 μCi [^{33}P]γ-ATP (IC_{50} determinations) or 2.7 μCi [^{33}P]γ-ATP (K_i determinations). The total ATP concentration is 10 μM (IC_{50} determinations) or ranges from 0 to 45 μM (K_i determinations). Following 30 min incubation at room temperature the assay is stopped by the addition of one third assay volume of 2.5% (v/v) H_3PO_4 containing 21 mM ATP. Samples are spotted onto P30 phosphocellulose mats and these are washed six times in 0.5% (v/v) H_3PO_4. The filter mats are sealed into sample bags containing Wallac betaplate scintillation fluid. ^{33}P incorporation into the substrate peptide is determined by counting the mats in a Wallac microbeta scintillation counter[1].

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

Cell Assay [2]

B65 cells are used after 24 h of in vitro culture. CGN are used after 7-8 days in vitro. Lithium and SB-415286 are dissolved in culture media and DMSO, respectively, and added to the neuronal preparation at the precise concentrations, 1 h before addition H_2O_2 (50 μM to 1 mM). To assess the loss in cell viability, we use the MTT [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium] method. MTT is added to the cells at a final concentration of 250 μM and incubated for 1 h, allowing the reduction in MTT to produce a dark blue formazan product. Media are then removed, and cells are dissolved in dimethylsulfoxide. Formazan production is measured by the absorbency change at 595 nm using a microplate reader. Viability results are expressed as percentages. The absorbency measured from non-treated cells is taken to be 100%[2].

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

CUSTOMER VALIDATION

- Brain Behav Immun. 2021 Jan 4;S0889-1591(20)32487-9.
- Front Pharmacol. 2022 Sep 26;13:925489.
- Food Biosci. 7 May 2022, 101766.
- Food Biosci. April 2022, 101571.
- Oncotarget. 2017 Jul 7;8(47):82174-82184.

See more customer validations on www.MedChemExpress.com

REFERENCES

[1]. Coghlan MP, et al. Selective small molecule inhibitors of glycogen synthase kinase-3 modulate glycogen metabolism and gene transcription. Chem Biol. 2000 Oct;7(10):793-803.

[2]. Pizarro JG, et al. Neuroprotective effects of SB-415286 on hydrogen peroxide-induced cell death in B65 rat neuroblastoma cells and neurons. Int J Dev Neurosci. 2008 May-Jun;26(3-4):269-76.

[3]. MacAulay K, et al. Use of lithium and SB-415286 to explore the role of glycogen synthase kinase-3 in the regulation of glucose transport and glycogen synthase. Eur J Biochem. 2003 Sep;270(18):3829-38.

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

Tel: 609-228-6898 Fax: 609-228-5909 E-mail: tech@MedChemExpress.com
Address: 1 Deer Park Dr, Suite Q, Monmouth Junction, NJ 08852, USA