产品名称： **2-环戊基-4-(5-苯基-1H-吡咯并[2,3-B]吡啶-3-基)苯甲酸**
产品别名： **GSK 650394**

生物活性：

Description	GSK 650394 is a novel SGK inhibitor with IC50 of 62 nM and 103 nM for SGK1 and SGK2 in the SPA assay respectively.
IC_{50} & Target	IC50: 62 nM (SGK1), 103 nM (SGK2)
In Vitro	GSK650394 is relatively non-toxic, with LC_{50} values of 41 μM in M1 cells (68 times its activity IC_{50}) and a LC_{50}greater than 100 μM in HeLa cells. GSK650394 inhibits SGK1-mediated epithelial transport with an IC_{50} of 0.6 μM in the SCC assay. GSK650394 inhibits the growth of LNCaP cells with IC_{50} of approximately 1 μM [1] GSK650394A inhibits the insulin-induced phosphorylation of PKB-Ser^{473} at 3 μM, and essentially abolishes this response at 10 μM. GSK650394A (1-10 μM) does not alter the phosphorylation of PRAS40-Ser246 in hormone-deprived cells or prevent the insulin-induced phosphorylation of this residue [2].
In Vivo	GSK650394 (1, 10, and 30 μM, 10 μL/rat, intrathecally) dose-dependently prevents CFA-induced pain behavior and the associates SGK1 phosphorylation, GluR1 trafficking, and protein-protein interactions at 1 day after CFA administration[3]. GSK650394 at concentrations of 10, 30, and 100 nM (10 μL), but not vehicle solution (SNL 3D+Veh and SNL 7D+Veh, respectively), dose-dependently increases the withdrawal latency of the ipsilateral hindpaw at 1-3 and 1-5 h after injection at days 3 and 7 postsurgery (SNL 3D+GSK and SNL 7D+GSK, respectively). GSK650394 (from day 0 to 6 postsurgery; 100 nM, 10 μL, i.t.) administration alleviates SNL-induced allodynia at days 3, 5, and 7 postsurgery in SNL animals[4].

Solvent&Solubility

In Vitro:

DMSO : ≥ 40.7 mg/mL (106.42 mM)

H_2O : < 0.1 mg/mL (insoluble)

* "≥" means soluble, but saturation unknown.

Preparing Stock Solutions

Solvent / Mass / Concentration	1 mg	5 mg	10 mg
1 mM	2.6147 mL	13.0736 mL	26.1472 mL
5 mM	0.5229 mL	2.6147 mL	5.2294 mL
10 mM	0.2615 mL	1.3074 mL	2.6147 mL

*请根据产品在不同溶剂中的溶解度选择合适的溶剂配制储备液；一旦配成溶液，请分装保存，避免反复冻融造成的产品失效。

储备液的保存方式和期限：-80°C, 6 months; -20°C, 1 month。-80°C 储存时，请在 6 个月内使用，-20°C 储存时，请在 1 个月内使用。

In Vivo:

请根据您的实验动物和给药方式选择适当的溶解方案。以下溶解方案都请先按照 **In Vitro** 方式配制澄清的储备液，再依次添加助溶剂：

——为保证实验结果的可靠性，澄清的储备液可以根据储存条件，适当保存；体内实验的工作液，建议您现用现配，当天使用； 以下溶剂前显示的百分比是指该溶剂在您配制终溶液中的体积占比；如在配制过程中出现沉淀、析出现象，可以通过加热和/或超声的方式助溶

1.请依序添加每种溶剂： 10% DMSO→40% PEG300 →5% Tween-80 → 45% saline

Solubility: 2.5 mg/mL (6.54 mM); Suspended solution; Need ultrasonic

此方案可获得 2.5 mg/mL (6.54 mM)的均匀悬浊液，悬浊液可用于口服和腹腔注射。

以 1 mL 工作液为例，取 100 μL 25.0 mg/mL 的澄清 DMSO 储备液加到 400 μL PEG300 中，混合均匀；

	向上述体系中加入 50 μL Tween-80，混合均匀；然后继续加入 450 μL 生理盐水定容至 1 mL。 2.请依序添加每种溶剂： 10% DMSO→ 90% (20% SBE-β-CD in saline) Solubility: ≥ 2.5 mg/mL (6.54 mM); Suspended solution 此方案可获得 ≥ 2.5 mg/mL (6.54 mM，饱和度未知) 的均匀悬浊液，悬浊液可用于口服和腹腔注射。 以 1 mL 工作液为例，取 100 μL 25.0 mg/mL 的澄清 DMSO 储备液加到 900 μL 20% 的 SBE-β-CD 生理盐水水溶液中，混合均匀。 3.请依序添加每种溶剂： 10% DMSO →90% corn oil Solubility: ≥ 2.5 mg/mL (6.54 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (6.54 mM，饱和度未知) 的澄清溶液，此方案不适用于实验周期在半个月以上的实验。 以 1 mL 工作液为例，取 100 μL 25.0 mg/mL 的澄清 DMSO 储备液加到 900 μL 玉米油中，混合均匀。
References	[1]. Sherk AB, et al. Development of a small-molecule serum- and glucocorticoid-regulated kinase-1 antagonist and its evaluation as a prostate cancer therapeutic. Cancer Res. 2008 Sep 15;68(18):7475-83. [2]. Mansley MK, et al. Effects of nominally selective inhibitors of the kinases PI3K, SGK1 and PKB on the insulin-dependent control of epithelial Na+ absorption. Br J Pharmacol. 2010 Oct;161(3):571-88. [3]. Peng HY, et al. Spinal SGK1/GRASP-1/Rab4 is involved in complete Freund's adjuvant-induced inflammatory pain via regulating dorsal horn GluR1-containing AMPA receptor trafficking in rats. Pain. 2012 Dec;153(12):2380-92. [4]. Peng HY, et al. Spinal serum-inducible and glucocorticoid-inducible kinase 1 mediates neuropathic pain via kalirin and downstream PSD-95-dependent NR2B phosphorylation in rats. J Neurosci. 2013 Mar 20;33(12):5227-40.

实验参考：

Cell Assay	The toxicity of GSK650394 to M-1 and HeLa cells is assessed using the Cell Proliferation Kit (XTT) following manufacturer's instructions. Briefly, 10,000 HeLa or M-1 cells/well are plated into 96-well plates in 100μL of the appropriate maintenance media. After 48 h, media is removed and replaced with 100 μL of EMEM with Earle's salts containing 2 mM L-glutamine and 1% antibiotic-antimycotic overnight. M-1 cells are also supplemented with 1 μg/mL insulin, 6.25 μg/mL sodium selenite, and 6.25 μg/mL transferrin. After 24 h, the media is removed and replaced with 100 μL media alone or media containing increasing concentrations of GSK650394. For HeLa cells, 50 μL of activated XTT solution is added after 4 h. For M-1 cells, 50 μL of activated XTT solution is added after 24 h. Following a 2 h incubation, absorbance is measured at 490 nm using a SpectraMAX PLUS spectrophotometer and the data analyzed to obtain IC_{50} values using GraphPad Prism 3 software. [1]
Animal Administration	Briefly, the rats are anesthetized under isoflurane anesthesia (induction 5%, maintenance 2% in oxygen). An incision is made, and the left L5 spinal nerves are carefully isolated and tightly ligated with 6-0 silk sutures 2-5 mm distal to the dorsal root ganglia. GSK650394 (10, 30, and 100 nM, 10 μL) is administered by bolus injection at 3 or 7 d or by daily injection for 7 d (day 0-6) postspinal nerve ligation. A vehicle solution of a volume identical to that of the tested agents is dispensed to serve as a control. [4]
	1]. Sherk AB, et al. Development of a small-molecule serum- and glucocorticoid-regulated kinase-1

References	antagonist and its evaluation as a prostate cancer therapeutic. Cancer Res. 2008 Sep 15;68(18):7475-83. [2]. Mansley MK, et al. Effects of nominally selective inhibitors of the kinases PI3K, SGK1 and PKB on the insulin-dependent control of epithelial Na+ absorption. Br J Pharmacol. 2010 Oct;161(3):571-88. [3]. Peng HY, et al. Spinal SGK1/GRASP-1/Rab4 is involved in complete Freund's adjuvant-induced inflammatory pain via regulating dorsal horn GluR1-containing AMPA receptor trafficking in rats. Pain. 2012 Dec;153(12):2380-92. [4]. Peng HY, et al. Spinal serum-inducible and glucocorticoid-inducible kinase 1 mediates neuropathic pain via kalirin and downstream PSD-95-dependent NR2B phosphorylation in rats. J Neurosci. 2013 Mar 20;33(12):5227-40.