产品名称：SLx-2119
产品别名：KD-025

生物活性：

Description	SLx-2119 (KD-025) is a selective inhibitor of ROCK2 with an IC_{50} of 105 nM.	
IC_{50} & Target	ROCK2	ROCK1
	105 nM (IC_{50})	24 μM (IC_{50})
In Vitro	SLx-2119 (40 μM) induces significant down-regulations of Tsp-1 and CTGF mRNA levels in PASMC. The microarray hybridized with aRNA from HMVEC treated with SLx-2119, shows a 5-times higher background than the other arrays[1].	
In Vivo	SLx-2119 (KD-025; 100, 200 or 300 mg/kg, i.p.) dose-dependently reduces infarct volume after transient middle cerebral artery occlusion. SLx-2119 is at least as efficacious in aged, diabetic or female mice, as in normal adult males[2].	

Solvent&Solubility

In Vitro:

DMSO : ≥ 29 mg/mL (64.09 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	2.2099 mL	11.0495 mL	22.0990 mL
	5 mM	0.4420 mL	2.2099 mL	4.4198 mL
	10 mM	0.2210 mL	1.1049 mL	2.2099 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 (5.52 mM); Suspended solution; Need ultrasonic

此方案可获得 2.5 mg/mL (5.52 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 (5.52 mM); Suspended solution; Need ultrasonic

此方案可获得 2.5 mg/mL (5.52 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 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 (5.52 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (5.52 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]. Boerma, M., et al. Comparative gene expression profiling in three primary human cell lines after treatment with a novel inhibitor of Rho kinase or atorvastatin. Blood Coagul Fibrinolysis, 2008. 19(7): p. 709-18. [2]. Lee, J.H., et al. Selective ROCK2 Inhibition In Focal Cerebral Ischemia. Ann Clin Transl Neurol, 2014. 1(1): p. 2-14. [3]. Yang W, et al. Critical role of ROCK2 activity in facilitating mucosal CD4+ T cell activation in inflammatory bowel disease. J Autoimmun. 2018 May;89:125-138. [4]. Chen W, et al. Screening RhoA/ROCK inhibitors for the ability to prevent chronic rejection of mouse cardiac allografts.Transpl Immunol. 2018 Jun 6. pii: S0966-3274(18)30029-7.

实验参考：

Cell Assay	Western blots are used to determine whether HMVEC, NHDF and PASMC express ROCK1 and ROCK2. The cells are incubated for 24 hours in 3 mL culture media containing SLx-2119. All cells are collected at passage 3 and lysed on ice in 25 mM Tris-HCl pH 7.5, 150 mM NaCl, 0.5% tritonX-100, 10% glycerol, 10 mM NaF and a protease inhibitor cocktail. Protein concentration is determined using a BCA protein assay reagent. Cell lysates (35 μg) are separated on 7.5% or 12.5% SDS-PAGE polyacrylamide gels and transferred to PVDF membrane filters. Membranes are blocked in 5% non-fat milk in TBS containing 0.1% Tween 20. Blots are probed with antibodies to ROCK1, ROCK2 or actin and washed well before incubation with HRP-conjugated secondary antibodies and visualization with an enhanced chemiluminescence (ECL) kit.［1］
Animal Administration	Young adult (C57BL/6, 2-3 months old, male 22-30 g, female 16-23 g), aged (C57BL/6, 12 months old, 33-52 g) are used in all experiments. Vehicle (0.4% methylcellulose) or SLx-2119 (100, 200 or 300 mg/kg) is administered every 12 h via orogastric gavage. The dosing paradigm is chosen based on the pharmacokinetic profile after oral administration in mice. Atorvastatin (4 mg/mL) is dissolved in phosphate-buffered saline (pH 7.4) containing 45% 3-hydroxypropyl-B-cyclodextrin and 10% ethanol, and administered at a dose of 20 mg/kg per day as a single daily intraperitoneal injection for 2 weeks. [2]
References	[1]. Boerma, M., et al. Comparative gene expression profiling in three primary human cell lines after treatment with a novel inhibitor of Rho kinase or atorvastatin. Blood Coagul Fibrinolysis, 2008. 19(7): p. 709-18. [2]. Lee, J.H., et al. Selective ROCK2 Inhibition In Focal Cerebral Ischemia. Ann Clin Transl Neurol, 2014. 1(1): p. 2-14. [3]. Yang W, et al. Critical role of ROCK2 activity in facilitating mucosal CD4+ T cell activation in inflammatory bowel disease. J Autoimmun. 2018 May;89:125-138. [4]. Chen W, et al. Screening RhoA/ROCK inhibitors for the ability to prevent chronic rejection of mouse cardiac allografts.Transpl Immunol. 2018 Jun 6. pii: S0966-3274(18)30029-7.