产品名称：U0126-EtOH
产品别名：U0126

生物活性：

Description	U0126 is a potent and non-ATP competitive MEK1 and MEK2 inhibitor, with IC50s of 70 nM and 60 nM, respectively.			
IC_{50} & Target	MEK2	MEK1	Autophagy	Mitophagy
	60 nM (IC_{50})	70 nM (IC_{50})		
In Vitro	Treatment with U0126 efficiently reduces progeny virus titers of all tested strains in A549 cells. While nM concentrations of U0126 are efficient to reduce H1N1v and H5N1 (MB1), μM concentrations of U0126 are required to reduce the virus titer of H5N1 (GSB) and H7N7. The EC_{50} values for U0126 against H1N1v are 1.2±0.4 μM in A549 cells and 74.7±1.0 μM in MDCKII cells[2].Rat hepatocarcinoma cells (FAO) stimulated by fetal calf serum (FCS) exhibits a significant proportion in S phase (32.62%) whereas U0126 strongly decreases the proportion of cells in S phase (9.92%) and increases the proportion of cells in G_0-G_1 phase and to a lesser extent in G_2/M[3].			
In Vivo	Mice are treated daily with U0126 (i.p., 10.5 mg/kg). In control experiment, tumor sizes are constant or slightly increase all over the kinetic. At the opposite, in all U0126 experiments, engraftment and early tumor growth are significantly decreased. Furthermore, a 60-70% reduction in the volume of tumors treated with U0126 is obtained 9 days after injection and thereafter[3]. Rats are subjected to 120?minutes transient middle cerebral artery occlusion (tMCAO) and thereafter treated with the U0126 (i.p., 30 mg/kg) at 0 and 24 hours of reperfusion. After treatment with U0126, the vasoconstriction to S6c is markedly reduced[4].			
Solvent&Solubility	***In Vitro:*** **DMSO : ≥ 49 mg/mL** (**114.87 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	2.3443 mL	11.7217 mL	23.4434 mL
	5 mM	0.4689 mL	2.3443 mL	4.6887 mL
	10 mM	0.2344 mL	1.1722 mL	2.3443 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: ≥ 3.33 mg/mL (7.81 mM); Clear solution

此方案可获得 ≥ 3.33 mg/mL (7.81 mM，饱和度未知) 的澄清溶液。

	以 1 mL 工作液为例，取 100 μL 33.3 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% corn oil Solubility: 3.33 mg/mL (7.81 mM); Suspended solution; Need ultrasonic and warming 此方案可获得 3.33 mg/mL (7.81 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 33.3 mg/mL 的澄清 DMSO 储备液加到 900 μL 玉米油中，混合均匀。 3.请依序添加每种溶剂： 2% DMSO →40% PEG300 →5% Tween-80 →53% saline Solubility: ≥ 1 mg/mL (2.34 mM); Clear solution
References	[1]. Duncia JV, et al. MEK inhibitors: the chemistry and biological activity of U0126, its analogs, and cyclization products. Bioorg Med Chem Lett. 1998, 8(20), 2839-2844. [2]. Droebner K, et al. Antiviral activity of the MEK-inhibitor U0126 against pandemic H1N1v and highly pathogenic avian influenza virus in vitro and in vivo. Antiviral Res. 2011, 92(2), 195-203. [3]. Bessard A, et al. RNAi-mediated ERK2 knockdown inhibits growth of tumor cells in vitro and in vivo. Oncogene. 2008 Sep 11;27(40):5315-25. [4]. Ahnstedt H, et al. U0126 attenuates cerebral vasoconstriction and improves long-term neurologic outcome after stroke in female rats. J Cereb Blood Flow Metab. 2015 Mar;35(3):454-60.

实验参考：

Cell Assay	A549 and MDCK II cells are seeded in 96-well culture plates at a density of 8×10^4 cells per well in minimal essential medium (MEM) containing 10% heat-inactivated fetal calf serum (FCS), 100 U/mL Penicillin, 100 mg/mL Streptomycin. Cells are incubated at 37°C with 5% CO_2 overnight. Thereafter, cells are washed twice with PBS. MEM containing different concentrations of U0126 (0.001-1000 μM) is added to the cells. After addition of U0126, cells are incubated further for 48 h at 37°C and 5% CO_2. Then, cells are fixed by incubation for 30 min at 4°C with 100 μL 4% paraformaldehyde (PFA). Adding 100 μL crystal violet for 30 min at room temperature stained viable cells. After staining, plates are washed and dried. For the extraction of crystal violet from viable cells 100 μL of 100% methanol is added to each well. After incubation for 30 min at room temperature, the extinction is measured with an enzyme-linked immunosorbent assay (ELISA) reader at OD=490 nm. The percentage of cell viability after treatment with the antiviral compound is calculated[2]
Animal Administration	Mice[3] Athymic female nude mice (SWISS, nu/nu) are used. Prior to injection, FI cells are labeled with a stable fluorescent dye molecule, DiA at 10 μg/mL for 5 h at 37°C. After washing to remove free DiA, cells are trypsinized for inoculation (U0126 experiments) or transfection (RNAi experiments). Biliary epithelial cells are injected subcutaneously, at the indicated times, into the tibia of nude mice. In the chemical experiments, 3 h after inoculation, mice are treated with U0126 (10.5 mg/kg) daily by intraperitoneal injection. The length and width of each tumor are measured every day by using a caliper. The following formula is used to calculate tumor volumes=$width^2\times length/2$. Mice are killed at the end of experiment. Tumors are immediately frozen in liquid nitrogen. Rats[4] Twelve-week-old female Wistar rats (250 to 265 g) are used U0126 (30 mg/kg intraperitoneally) is injected at 0 and 24 hours of reperfusion after tMCAO based on the previous evaluation of the drug

	in male rats. Animals in study II are administered U0126 or vehicle and are killed 14 days after tMCAO. Experimental group assignments are randomized and blinded to the surgical experimenter.
References	[1]. Duncia JV, et al. MEK inhibitors: the chemistry and biological activity of U0126, its analogs, and cyclization products. Bioorg Med Chem Lett. 1998, 8(20), 2839-2844. [2]. Droebner K, et al. Antiviral activity of the MEK-inhibitor U0126 against pandemic H1N1v and highly pathogenic avian influenza virus in vitro and in vivo. Antiviral Res. 2011, 92(2), 195-203. [3]. Bessard A, et al. RNAi-mediated ERK2 knockdown inhibits growth of tumor cells in vitro and in vivo. Oncogene. 2008 Sep 11;27(40):5315-25. [4]. Ahnstedt H, et al. U0126 attenuates cerebral vasoconstriction and improves long-term neurologic outcome after stroke in female rats. J Cereb Blood Flow Metab. 2015 Mar;35(3):454-60.