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产品名称：VH-298
产品别名：VH-298

生物活性：

Description	VH-298 is a highly potent inhibitor of the VHL:HIF-α interaction with a K_d value of 80 to 90 nM, used in PROTACtechnology.
IC_{50} & Target	Kd: 80 to 90 nM (VHL:HIF-α)[1]
In Vitro	VH-298 is a potent, cell permeable and non-toxic chemical probe that triggers the hypoxic response by blocking the VHL. VH-298 is a highly potent inhibitor of the VHL:HIF-α interaction with K_d values of 90 and 80 nM in isothermal titration calorimetry and competitive fluorescence polarization assay. VH-298 binds with VHL complex very fast and dissociates slowly. VH-298 at 50 μM concentration exhibits negligible off-target effects *in vitro* against more than 100 tested cellular kinases, GPCRs and ion channels. VH-298 is cell permeable and not toxic to cells. The measured permeability of VH-298 is found to be 19.4 nm s^{-1}. VH-298 induces concentration- and time-dependent on-target specific accumulation of hydroxylated HIF-α in human cell lines, including HeLa cancer cells and renal cell carcinoma 4 (RCC4) cells. VH-298 increases mRNA levels of EPO by 2.5-fold in RCC4-HA-VHL, but not in VHL-null RCC4-HA, indicating that pharmacological inhibition of VHL is able to stimulate endogenous EPO synthesis. VH-298 proves as effective as hypoxia in raising PHD2 and HK2 protein levels, however in HFF the BNIP3 protein level increases more with VH-298 treatment than hypoxia treatment[1].
Solvent&Solubility	***In Vitro:***

DMSO : ≥ 83.3 mg/mL (159.08 mM)

* "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	1.9097 mL	9.5484 mL	19.0967 mL
	5 mM	0.3819 mL	1.9097 mL	3.8193 mL
	10 mM	0.1910 mL	0.9548 mL	1.9097 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 (4.77 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (4.77 mM，饱和度未知) 的澄清溶液。

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 (4.77 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (4.77 mM，饱和度未知) 的澄清溶液。 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 (4.77 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (4.77 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]. Frost J, et al. Potent and selective chemical probe of hypoxic signalling downstream of HIF-α hydroxylation via VHL inhibition. Nat Commun. 2016 Nov 4;7:13312.

实验参考：

Cell Assay	Death of CTLs is analyzed by staining with 4′,6-diamidino-2-phenylindole (DAPI). Cells are plated in 96-well plates at 1×10^6 and treated with VHL inhibitors (VH-298) and respective non-binding cis-analogues for 24 h. Cells are spun down and resuspended in HBSS containing DAPI to identify dead and dying populations[1].
Kinase Assay	VH-298 is screened at 50 μM concentration against a panel of 50 kinases. The remaining kinase activity is recorded in the end of the assay. The data is reported as average % activity remaining of assay duplicates for each kinase tested[1].
References	[1]. Frost J, et al. Potent and selective chemical probe of hypoxic signalling downstream of HIF-α hydroxylation via VHL inhibition. Nat Commun. 2016 Nov 4;7:13312.