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产品名称：CVT-313
产品别名：Cdk2 Inhibitor III

生物活性：

Description	CVT-313 (Cdk2 Inhibitor III) is a potent, selective, reversible, and ATP-competitive inhibitor of CDK2 with IC50 of 0.5 μM.		
IC_{50} & Target	cdk2/cyclin A	Cdk1/cyclin B	Cdk4/cyclin D1
	0.5 μM (IC_{50})	4.2 μM (IC_{50})	215 μM (IC_{50})
In Vitro	CVT-313 (Cdk2 Inhibitor III) has been shown to inhibit other kinases, but at much higher IC50 values, i.e., CDK1 (IC50=4.2 μM), CDK4 D1 (IC50=215 μM), and MAPK/PKA/PKC (IC50>1.25 mM), compared to CDK2 (IC50=0.5 μM). CVT-313 has been shown to have profound effects on cell proliferation at concentrations of 5-20 μM[1]. CVT-313 is a potent CDK2 inhibitor, which is identified from a purine analog library with an IC50 of 0.5 μM in vitro. Inhibition is competitive with respect to ATP (Ki=95 nM), and selective CVT-313 has no effect on other, nonrelated ATP-dependent serine/threonine kinases. When added to CDK1 or CDK4, a 8.5- and 430-fold higher concentration of CVT-313 is required for half-maximal inhibition of the enzyme activity. Using normal and tumor human/murine cell lines, the effects of CVT-313 on cell proliferation is measured. The IC50 for growth inhibition ranged from 1.25 to 20 μM[2].		

Solvent&Solubility

In Vitro:

DMSO： ⩾ 100 mg/mL (249.71 mM)

* "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	2.4971 mL	12.4853 mL	24.9707 mL
	5 mM	0.4994 mL	2.4971 mL	4.9941 mL
	10 mM	0.2497 mL	1.2485 mL	2.4971 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 (6.24 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (6.24 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 (6.24 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (6.24 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 (6.24 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (6.24 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]. Graub R, et al. Cell cycle-dependent phosphorylation of human CDC5 regulates RNA processing. Cell Cycle. 2008 Jun 15;7(12):1795-803. [2]. Brooks EE, et al. CVT-313, a specific and potent inhibitor of CDK2 that prevents neointimal proliferation. J Biol Chem. 1997 Nov 14;272(46):29207-11.
实验参考：	
Cell Assay	MRC-5 cells are grown in Dulbecco's modified Eagle's medium containing 5% fetal calf serum. CVT313 (0, 5, 10, 15 μM) is added to exponentially growing cells in tissue culture. Cell population is measured. Proliferation assays are carried out using the nonradioactive CellTiter 96 kit after 48-h exposure. For FACS analysis of DNA content, cells are trypsinized, fixed in 70% ice-cold ethanol, and treated with 0.1 mg/mL RNase A and 40 μg/mL propidium iodide for 1 h at 37°C[2].
Kinase Assay	For kinase assays, purified CDC5L(295-795)-His6 is mixed with [γ-^{32}P]ATP, COS-7 cell extract, and incubated in 100 μL 20 mM HEPES, pH 7.5, 50 mM NaCl, 2 mM $MnCl_2$, 10 mM $MgCl_2$, 0.5% NP-40, 0.5 mM PMSF, 5 mM benzamidine hydrochloride, 5 mM NaF, 1 mM $NaVO_3$ and the specific inhibitor at 30°C for 10 minutes. Cell extract as a source of kinase activity is prepared from subconfluent, serum-stimulated COS-7 cells lysed in 20 mM HEPES-NaOH, pH 7.5, 50 mM NaCl, 1% Triton X-100, 10% glycerol, protease and phosphotase inhibitors. Phosphorylated proteins are separated by electrophoresis in 15% polyacrylamide-SDS gels. Specific inhibitors included 20 μM staurosporine, 10 μM genistein, 1 μM CVT-313, 10 μM Rp-MB-cAMPS and 50 μM PD98059[1].
References	[1]. Graub R, et al. Cell cycle-dependent phosphorylation of human CDC5 regulates RNA processing. Cell Cycle. 2008 Jun 15;7(12):1795-803. [2]. Brooks EE, et al. CVT-313, a specific and potent inhibitor of CDK2 that prevents neointimal proliferation. J Biol Chem. 1997 Nov 14;272(46):29207-11.