产品名称：**LY2857785**
产品别名：**LY2857785**

## 生物活性：

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| **Description** | LY2857785 is a type I reversible and competitive ATP kinase inhibitor against CDK9 ($IC_{50}$ 11 nM) and other transcription kinases CDK8 ($IC_{50}$ 16 nM), and CDK7 ($IC_{50}$ 246 nM). | | |
| **$IC_{50}$ & Target** | CDK9 | CDK8 | CDK7 |
| | 0.011 μM ($IC_{50}$) | 0.016 μM ($IC_{50}$) | 0.246 μM ($IC_{50}$) |
| **In Vitro** | LY2857785 shows good selectivity against a panel of 114 protein kinases, with only 5 other protein kinases inhibited with potency ($IC_{50}$) less than 0.1 μM, and a total of 14 kinases less than 1 μM. At the cellular level, LY2857785 inhibits CTD P-Ser2 and CTD P-Ser5 in U2OS cells at $IC_{50}$s 0.089 (n=13) and 0.042 (n=1) μM, respectively. However, LY2857785 only induces a moderate $G_2$-M DNA content increase, from 35% to 55%, with $EC_{50}$ 0.135 μM. LY2857785 shows potent compound exposure- and time-dependent cell proliferation inhibition in MV-4-11, RPMI8226, and L363 cells. When incubated between 4 to 24 hours, the cell growth inhibition potency reaches a maximal effect at 8 hours with $IC_{50}$s 0.04, 0.2, and 0.5 μM for MV-4-11, RPMI8226, and L363 cells, respectively. LY2857785-induced cancer cell apoptosis is also time dependent, reaching maximal potency at 8 hours with $IC_{50}$0.5 μM in L363 cells[1]. | | |
| **In Vivo** | In HCT116 xenograft tumor-bearing mice, LY2857785 demonstrates dose-dependent RNAP II CTD P-Ser2 inhibition potently with TED50 of 4.4 mg/kg and TEC50 of 0.36 μM. LY2857785 also shows significant duration of CTD P-Ser2 inhibition for 3 to 6 hours at TED70 (8 mg/kg) in HCT116 and MV-4-11 nude mice xenograft models. In the nude rat MV-4-11 xenograft model, LY2857785 similarly shows dose-dependent CTD P-Ser2 inhibition for 8 hours at TED70 (7 mg/kg) and TED90 (10 mg/kg). LY2857785 demonstrates the most dramatic tumor regression in the AML MV-4-11 xenograft tumor model either by i.v. bolus in mice or i.v. infusion in rats[1]. | | |
| **Solvent&Solubility** | ***In Vitro:***<br>**DMSO : 10 mg/mL (22.29 mM;** Need ultrasonic) | | |

| Preparing Stock Solutions | Solvent / Mass / Concentration | 1 mg | 5 mg | 10 mg |
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| | 1 mM | 2.2292 mL | 11.1458 mL | 22.2916 mL |
| | 5 mM | 0.4458 mL | 2.2292 mL | 4.4583 mL |
| | 10 mM | 0.2229 mL | 1.1146 mL | 2.2292 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: ≥ 1 mg/mL (2.23 mM); Clear solution

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| | 此方案可获得 ≥ 1 mg/mL (2.23 mM，饱和度未知) 的澄清溶液。<br>以 1 mL 工作液为例，取 100 μL 10.0 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。<br><br>2.请依序添加每种溶剂： 10% DMSO→ 90% (20% SBE-β-CD in saline)<br>Solubility: ≥ 1 mg/mL (2.23 mM); Clear solution<br>此方案可获得 ≥ 1 mg/mL (2.23 mM，饱和度未知) 的澄清溶液。<br>以 1 mL 工作液为例，取 100 μL 10.0 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。<br><br>3.请依序添加每种溶剂： 10% DMSO →90% corn oil<br>Solubility: ≥ 1 mg/mL (2.23 mM); Clear solution<br>此方案可获得 ≥ 1 mg/mL (2.23 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。<br>以 1 mL 工作液为例，取 100 μL 10.0 mg/mL 的澄清 DMSO 储备液加到 900 μL 玉米油中，混合均匀。 |
| **References** | [1]. Yin T, et al. A novel CDK9 inhibitor shows potent antitumor efficacy in preclinical hematologic tumor models. Mol Cancer Ther. 2014 Jun;13(6):1442-56. Mol Cancer Ther. 2014 Jun;13(6):1442-56. |

**实验参考：**

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| **Cell Assay** | Solid tumor cells are plated in poly-D-lysine coated and hematologic cell lines are seeded in noncoated 96-well plates overnight before being treated with compounds (e.g, LY2857785). Solid tumor cells are fixed with Prefer for 20 minutes at room temperature and permeated with 0.1% Triton X-100 in PBS for 15 minutes. Caspase-3 expression is measured by immunofluorescence with antiactivated caspase-3. Terminal deoxynucleotidyl transferase-mediated dUTP nick end labeling (TUNEL) activity is measured with In Situ Cell Death Detection Kit. Both assays are analyzed on Acumen Explorer laser-scanning fluorescence microplate cytometer. Hematologic tumor cells are assayed for cell viability with CellTiter-Glo Luminescent Cell Viability Assay[1]. |
| **Animal Administration** | Mice and Rats[1]<br>For xenograft models, human cancer cells U87MG, MV-4-11, A375, and HCT116 are implanted into female nude rats or athymic nude female mice. The animals are dosed with saline, Rapamycin, or LY2857785, respectively. MV-4-11 xenografts in nude mice are treated by LY2857785 (4, 8, and 18 mg/kg) i.v. bolus. MV-4-11 xenografts in nude rats are treated with LY2857785 (3, 6, and 9 mg/kg) 4-hour i.v. infusion. An untreated vehicle control group is administered saline i.v. every 3 days. Flow cytometry analysis is conducted using Beckman Coulter's CXP software. Statistical significance of the effect of LY2857785 and/or control compounds is assessed by Dunnett method, one-way ANOVA. |
| | CDK7 and CDK9 reaction mixtures contain 10 mM Tris-HCl (pH 7.4), 10 mM HEPES, 5 mM DTT, 10 μM ATP, 0.5 μCi 33p-ATP, 10 mM $MnCl_2$, 150 mM NaCl, 0.01% Triton X-100, 2% DMSO, 0.05 mM CDK7/9ptide, and 2 nM CDK7/Mat1/cyclin H, or 2 nM CDK9/cyclin T1, respectively. CDK8/cyclin C reaction is performed in HEPES 30 mM, DTT 2 mM, $MgCl_2$ 5 mM, 0.015% Triton X-100, 5 μM ATP, and 400 nM of RBER-CHKStide containing 20 nM of enzyme. LY2857785 in DMSO is diluted serially 1:3 for dose response. Reactions are carried out in 96-well polystyrene plates. The reactions are incubated at room temperature for 60 minutes and followed by termination with 10% H3PO4 or 10% trichloroacetic acid (TCA). For the filter binding assay, reactions are transferred to 96-well filter |

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| **Kinase Assay** | plates and measured by Microbeta scintillation counter. For ADP Transcreener Fluorescent Polarization Assays, reactions are quenched with ADP detection mix, incubated 2 hours at room temperature and then FP is measured at $\lambda_{ex}$=610 nm, $\lambda_{em}$=670 nm on a Tecan Ultra 384 plate reader. The concentration of ADP product is calculated from millipolarization (μP) using a prepared ADP/ATP dilution series as a standard curve. Kinase profiling are carried out in 96-well polystyrene plates. Briefly, in a final volume of 25 μL the enzyme is incubated with the appropriate buffer, peptide substrate, and the diluted LY2857785. Reactions are initiated by the addition of ATP/[$^{33}$P] and the ATP mix is incubated at room temperature for 40 minutes. Reactions are quenched with the addition 5 μL of 3% phosphoric acid, 10 μL of the reaction are spotted onto a filtermat, washed 3 times for 5 minutes in 75 mM phosphoric acid and once in methanol. Once the filters are dry, they are submitted to scintillation counting[1]. |
| **References** | [1]. Yin T, et al. A novel CDK9 inhibitor shows potent antitumor efficacy in preclinical hematologic tumor models. Mol Cancer Ther. 2014 Jun;13(6):1442-56. Mol Cancer Ther. 2014 Jun;13(6):1442-56. |