产品名称：PD0332991 HCl
产品别名：帕布昔利布盐酸盐；PD 0332991 hydrochloride；Palbociclib hydrochloride

生物活性：

Description	Palbociclib hydrochloride is a highly selective CDK4/6 inhibitor with IC_{50}s of 11 nM and 16 nM, respectively.				
IC_{50} & Target [1]	Cdk4/cyclin D3	Cdk4/cyclin D1	Cdk6/cyclin D2	DYRK1A	MAPK
	9 nM (IC_{50})	11 nM (IC_{50})	16 nM (IC_{50})	2000 nM (IC_{50})	8000 nM (IC_{50})
In Vitro	The IC50 of Palbociclib (PD 0332991) for reduction of retinoblastoma (Rb) phosphorylation at Ser^{780} in MDA-MB-435 breast carcinoma cells is 66 nM. Palbociclib is equally effective at reducing Rb phosphorylation at Ser^{795} in this tumor with an IC50 of 63 nM, and similar effects on both Ser^{780} and Ser^{795} phosphorylation are obtained in the Colo-205 colon carcinoma[1]. The MP-MRT-AN (AN), KP-MRT-RY (RY), G401, and KP-MRT-NS (NS) cell lines are effectively inhibited by Palbociclib (PD) in a concentration-dependent manner in a WST-8 assay. The IC50s are 0.01 μM, 0.01 μM, 0.06 μM, and 0.6 μM, respectively. In contrast, the KP-MRT-YM (YM) cell line is resistant to Palbociclib (IC50>10 μM). The flow cytometry results show that Palbociclib at concentrations between 0 to 1.0 μM induces G1 arrest in the AN, RY, G401 and NS cell lines in a concentration-dependent manner, but has no effect on YM cells. The BrdU incorporation results are consistent with the WST-8 and flow cytometry results: PD reduces BrdU incorporation (indicating G1 arrest) in the AN, RY, G401 and NS cell lines, but not in the YM cell line. Palbociclib, even at a concentration of 0.05 μM, significantly reduces BrdU incorporation in the AN, RY, and G401 cell lines ($p<0.05$)[2].				
In Vivo	Palbociclib (PD 0332991) exhibits significant antitumor efficacy against multiple human tumor xenograft models. In mice bearing Colo-205 colon carcinoma xenografts (p16 deleted), daily p.o. dosing for 14 days with Palbociclib (150 or 75 mg/kg) produces rapid tumor regressions and a corresponding tumor growth delay of ~50 days with >1 log of tumor cell kill at the highest dose tested. At 37.5 mg/kg, the tumor slowly regressed during treatment. Even at doses as low as 12.5 mg/kg, a 13-day growth delay is obtained indicating a 90% inhibition of tumor growth rate. Likewise, robust antitumor activity is seen in the MDA-MB-435 breast carcinoma (p16 deleted) where complete tumor stasis is apparent at 150 mg/kg and some cell kill is evident at the highest dose[1].				

In Vitro:

DMSO : 5.4 mg/mL (**11.16 mM;** Need ultrasonic)

H_2O : 4.9 mg/mL (**10.12 mM;** Need ultrasonic and warming)

Preparing Stock Solutions	Solvent / Mass / Concentration	**1 mg**	**5 mg**	**10 mg**
	1 mM	2.0662 mL	10.3308 mL	20.6616 mL
	5 mM	0.4132 mL	2.0662 mL	4.1323 mL
	10 mM	0.2066 mL	1.0331 mL	2.0662 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

Solvent&Solubility	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: ≥ 0.54 mg/mL (1.12 mM); Clear solution 此方案可获得 ≥ 0.54 mg/mL (1.12 mM，饱和度未知) 的澄清溶液。 以 1 mL 工作液为例，取 100 μL 5.4 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: ≥ 0.54 mg/mL (1.12 mM); Clear solution 此方案可获得 ≥ 0.54 mg/mL (1.12 mM，饱和度未知) 的澄清溶液。 以 1 mL 工作液为例，取 100 μL 5.4 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： Lactic acid buffer (50 mM, pH 4.0) Solubility: 33.33 mg/mL (68.87 mM); Clear solution; Need ultrasonic
References	[1]. Fry DW, et al. Specific inhibition of cyclin-dependent kinase 4/6 by PD 0332991 and associated antitumor activity in human tumor xenografts. Mol Cancer Ther. 2004 Nov;3(11):1427-38. [2]. Katsumi Y, et al. Sensitivity of malignant rhabdoid tumor cell lines to PD 0332991 is inversely correlated with p16 expression. Biochem Biophys Res Commun, 2011, 413(1), 62-68. [3]. Hsieh FS, et al. Palbociclib induces activation of AMPK and inhibits hepatocellular carcinoma in a CDK4/6-independent manner. Mol Oncol. 2017 Aug;11(8):1035-1049.

实验参考：

Cell Assay	MRT cell lines, G401, MP-MRT-AN (AN), KP-MRT-RY (RY), KP-MRT-NS (NS), and KP-MRT-YM (YM) cell lines are seeded in normal growth medium into 96-well cell plates. After 24 h, the culture medium is replaced with culture medium containing Palbociclib (0.05 or 1 μM) or DMSO. Cells are cultured and treated in triplicate. Cell proliferation is determined 8 days after the treatment by WST-8 assay using a Cell Counting Kit-8. [1]
Animal Administration	Mice (18-22 g) are randomized and then implanted s.c. with tumor fragments (30 mg) into the region of the right axilla. Treatment is initiated when tumors reach 100 to 150 mg. PD 0332991 (150 or 75 mg/kg, p.o.) is given according to the schedule and dose indicated in the table and figure legends by gavage as a solution in sodium lactate buffer (50 mM, pH 4.0) based on mean group body weight. In all experiments, there are 12 mice in the control group and 8 mice each in the treated groups. Additional details for each experiment are given in the table legends. [1]
Kinase Assay	CDK assays are performed in 96-well filter plates. All CDK-cyclin kinase complexes are expressed in insect cells through baculovirus infection and purified. The substrate for the assays is a fragment (amino acids 792-928) of pRb fused to GST (GST·RB-Cterm). The total volume in each well is 0.1 mL containing a final concentration of 20 mM Tris-HCl, pH 7.4, 50 mM NaCl, 1 mM dithiothreitol, 10 mM $MgCl_2$, 25 μM ATP (for CDK4-cyclin D1, CDK6-cyclin D2, and CDK6-cyclin D3) or 12 μM ATP (for CDK2-cyclin E, CDK2-cyclin A, and CDC2-cyclin B) containing 0.25 μCi of [γ-^{32}P]ATP, 20 ng of enzyme, 1 μg of GST·RB-Cterm, and Palbociclib (0.001-0.1μM). All components except the

	[γ-^{32}P]ATP are added to the wells, and the plate is placed on a plate mixer for 2 min. The reaction is started by adding the [γ-^{32}P]ATP and the plate is incubated at 25°C for 15 min. The reaction is terminated by addition of 0.1 mL of 20% trichloroacetic acid and the plate is kept at 4°C for at least 1 hour to allow the substrate to precipitate. The wells are then washed 5 times with 0.2 mL of 10% trichloroacetic acid and radioactive incorporation is determined with a β plate counter. [1]
References	[1]. Fry DW, et al. Specific inhibition of cyclin-dependent kinase 4/6 by PD 0332991 and associated antitumor activity in human tumor xenografts. Mol Cancer Ther. 2004 Nov;3(11):1427-38. [2]. Katsumi Y, et al. Sensitivity of malignant rhabdoid tumor cell lines to PD 0332991 is inversely correlated with p16 expression. Biochem Biophys Res Commun, 2011, 413(1), 62-68. [3]. Hsieh FS, et al. Palbociclib induces activation of AMPK and inhibits hepatocellular carcinoma in a CDK4/6-independent manner. Mol Oncol. 2017 Aug;11(8):1035-1049.