产品名称：**PTC-209**
产品别名：**PTC-209**

生物活性：

Description	PTC-209 is a specific BMI-1 inhibitor with an IC_{50} of 0.5 μM.
IC_{50} & Target	IC50: 0.5 μM (BMI-1, in HT1080 tumor cells)[1]
In Vitro	PTC-209 is a recently developed inhibitor of BMI1, in biliary tract cancer (BTC) cells. PTC-209 reduces overall viability in BTC cell lines in a dose-dependent fashion (0.04-20 μM). Treatment with PTC-209 leads to slightly enhanced caspase activity and stop of cell proliferation. Cell cycle analysis reveals that PTC-209 causes cell cycle arrest at the G1/S checkpoint[2]. PTC-209 (100, 200, or 500 nM) decreases BMI1 and increases p16 protein expression in canine OSA cell lines. Compare to vehicle control, BMI1 protein expression decreases by 40% and 25% in the Abrams and D17 cell lines, respectively, following 500 nM PTC-209 treatment. In the Moresco cell line, BMI1 protein expression decreases by 16% and 39% following 200 nM and 500 nM PTC-209 treatment, respectively, as compared to vehicle control. Increases in p16 protein levels can be observed in all cell lines beginning at 100 nM PTC-209 and are highest at the 500 nM PTC-209 dose for Abrams (120% increase) and Moresco (200% increase), but appeares to top out at 200 nM for the D17 cell line (54% increase)[3].
In Vivo	Pharmacokinetic analysis demonstrates that PTC-209 (60 mg/kg, subcutaneously once a day) effectively inhibits BMI-1 production in tumor tissue in vivo. Inhibition of BMI-1 with PTC-209 halts growth of preestablished tumors in vivo[1].
Solvent&Solubility	***In Vitro:*** **DMSO : ≥ 32 mg/mL (64.62 mM)** * "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	2.0194 mL	10.0971 mL	20.1943 mL
	5 mM	0.4039 mL	2.0194 mL	4.0389 mL
	10 mM	0.2019 mL	1.0097 mL	2.0194 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 (5.05 mM); Suspended solution; Need ultrasonic

此方案可获得 2.5 mg/mL (5.05 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 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。
References	[1]. Kreso A, et al. Self-renewal as a therapeutic target in human colorectal cancer. Nat Med. 2014 Jan;20(1):29-36. [2]. Christian Mayr, et al. The BMI1 inhibitor PTC-209 is a potential compound to halt cellular growth in biliary tract cancer cells. Oncotarget. 2016 Jan 5; 7(1): 745-758. [3]. Shahi MH, et al. BMI1 is expressed in canine osteosarcoma and contributes to cell growth and chemotherapy resistance. PLoS One. 2015 Jun 25;10(6):e0131006. [4]. Chen D, et al. Targeting BMI1+ Cancer Stem Cells Overcomes Chemoresistance and Inhibits Metastases in Squamous Cell Carcinoma. Cell Stem Cell. 2017 May 4;20(5):621-634.e6.

实验参考：

Cell Assay	MTT assays are used to assess proliferation of Abrams, D17, and Moresco canine OSA cells following treatment of PTC-209 alone and in combination with Dox or Carbo. 500 cells are seeded in 96 well plates with DMEM/10%FBS and allowed to adhere overnight (16-18 hrs). For single treatment PTC-209 experiments, cells are incubated with drug for 72hrs at final concentrations of 0, 200, 300, 400, 500, and 600nM. For combination treatment experiments, cells are incubated with drug(s) for 72hrs at the following final concentrations: PTC-209 (0, 100, 200, and 500 nM), Dox (0, 3, and 30 nM), Carbo (0, 3, and 30 μM). Vehicle controls include DMSO (PTC-209), 0.9% saline (Dox), and water (Carbo). Additional controls included untreated (UT) cells (no veh or drug) and wells containing media (DMEM/10%FBS) alone (to assess background absorbance). Briefly, MTT solution is added to each well at a final conc. of 0.5mg/mL and incubated at 37°C for 4hrs. 200uL of DMSO is added to dissolve formazin crystals and absorbance is measured at 570nM and 630nM (reference wavelength) using a spectrophotometer (Spectramax 190). 6 wells per group are used for PTC-209 single treatment experiments, and 4 wells per group are used for combination treatment experiments, and all experiments are repeated twice. Statistical analysis is performed using 2-way ANOVA with Tukey's multiple comparisons test. [3]
Animal Administration	Mice[1] For the experiments where mice are dosed with the drug in vivo, tumor cells are injected subcutaneously into nude mice (male, aged 8-10 weeks), and when the tumors reach an approximate 0.2 cm^3 volume, PTC-209 is administered subcutaneously once a day at a dose of 60 mg per kg body weight (control animals received equal volumes of vehicle, 14% DMSO, 36% polyethylene glycol 400 and 50% polypropylene glycol). Tumor volume measurements are recorded every 3-7 d until the endpoint is reached.
References	[1]. Kreso A, et al. Self-renewal as a therapeutic target in human colorectal cancer. Nat Med. 2014 Jan;20(1):29-36. [2]. Christian Mayr, et al. The BMI1 inhibitor PTC-209 is a potential compound to halt cellular growth in biliary tract cancer cells. Oncotarget. 2016 Jan 5; 7(1): 745-758. [3]. Shahi MH, et al. BMI1 is expressed in canine osteosarcoma and contributes to cell growth and chemotherapy resistance. PLoS One. 2015 Jun 25;10(6):e0131006. [4]. Chen D, et al. Targeting BMI1+ Cancer Stem Cells Overcomes Chemoresistance and Inhibits Metastases in Squamous Cell Carcinoma. Cell Stem Cell. 2017 May 4;20(5):621-634.e6.