产品名称：
(S)-5-chloro-N2-(1-(5-fluoropyrimidin-2-yl)ethyl)-N4-(5-methyl-1H-pyrazol-3-yl)pyrimidine-2,4-diamine
产品别名：**AZD-1480**

生物活性：

Description	AZD-1480 is an ATP-competitive inhibitor of JAK1 and JAK2 with IC_{50}s of 1.3 and <0.4 nM, respectively.	
IC_{50} & Target	JAK2	JAK1
	0.4 nM (IC_{50})	1.3 nM (IC_{50})
In Vitro	AZD1480 (5μM) induces G2/M arrest and cell death by inhibiting Aurora kinases[1]. AZD1480 is a potent JAK2 inhibitor that can suppress growth, survival, as well as FGFR3 and STAT3 signaling and downstream targets including Cyclin D2 in human multiple myeloma cells. At low micromolar concentrations, AZD1480 blocks cell proliferation and induces apoptosis of myeloma cell lines[2]. AZD1480 effectively blocks constitutive and stimulus-induced JAK1, JAK2, and STAT-3 phosphorylation in both human and murine glioma cells, and leads to a decrease in cell proliferation and induction of apoptosis[3]. AZD1480 is a potent, competitive small-molecule inhibitor of JAK1/2 kinase, and that it is capable of inhibiting STAT3 phosphorylation and tumor growth in a STAT3-dependent manner. AZD1480 inhibits tumor angiogenesis and metastasis in part by affecting the tumor microenvironment[4].	
In Vivo	AZD1480 inhibits the STAT3 phosphorylation in an xenograft model of human solid tumors and multiple myeloma[1]. In vivo, AZD1480 inhibits the growth of subcutaneous tumors and increases survival of mice bearing intracranial glioblastoma (GBM) tumors by inhibiting STAT-3 activity, indicating that pharmacologic inhibition of the JAK/STAT-3 pathway by AZD1480 should be considered for study in the treatment of patients with GBM tumors[3]. AZD1480 blocks lung infiltration of myeloid cells and formation of pulmonary metastases in both mouse syngeneic experimental and spontaneous metastatic models. Furthermore, AZD1480 reduces angiogenesis and metastasis in a human xenograft tumor model[4]. AZD1480 suppresses the growth of human solid tumor xenografts harboring persistent Stat3 activity[5].	

Solvent&Solubility

In Vitro:

DMSO : 50 mg/mL (143.36 mM; Need ultrasonic)

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	2.8672 mL	14.3361 mL	28.6722 mL
	5 mM	0.5734 mL	2.8672 mL	5.7344 mL
	10 mM	0.2867 mL	1.4336 mL	2.8672 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 (7.17 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (7.17 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 (7.17 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (7.17 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 (7.17 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (7.17 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]. Derenzini E, et al. The JAK inhibitor AZD1480 regulates proliferation and immunity in Hodgkin lymphoma. Blood Cancer J. 2011 Dec;1(12):e46. [2]. Scuto A, et al. The novel JAK inhibitor AZD1480 blocks STAT3 and FGFR3 signaling, resulting in suppression of human myeloma cell growth and survival. Leukemia. 2011 Mar;25(3):538-50. [3]. McFarland BC, et al. Therapeutic potential of AZD1480 for the treatment of human glioblastoma. Mol Cancer Ther. 2011 Dec;10(12):2384-93. [4]. Xin H, et al. Antiangiogenic and antimetastatic activity of JAK inhibitor AZD1480. Cancer Res. 2011 Nov 1;71(21):6601-10. [5]. Hedvat M, et al. The JAK2 inhibitor AZD1480 potently blocks Stat3 signaling and oncogenesis in solid tumors. Cancer Cell. 2009 Dec 8;16(6):487-9 [6]. Ni J, et al. Tyrosine receptor kinase B is a drug target in astrocytomas. Neuro Oncol. 2017 Jan;19(1):22-30.

实验参考：

Cell Assay	Renca or 786-O cells are suspended in DMEM medium with 5% FBS, and seeded in 96-well plates (5×10^3 per well) to allow adhesion and then treated with DMSO or AZD1480 for 48 hours. Cell viability is determined by MTS assay. Absorbance at 490 nm is measured with Mikrotek Laborsysteme. Mouse endothelial cells and splenic CD11b+/c- myeloid cells are enriched from tumor-bearing mice,and cultured in 5% FBS RPMI-1640 medium. HUVECs are cultured on collagen 1-coated plates in complete medium. All cells are treated with DMSO and AZD1480 at various doses for 24 hours. Cell viability is determined by counting cell number manually. All the experiments are repeated 3 times.［4］
	For subcutaneous (s.c.) tumor model, 2.5×10^6 Renca or 786-O cells suspended in 100 μL PBS are injected into the flank of BALB/c or nude mice, respectively. When average tumor volume reaches approximately 100-150 mm^3, AZD1480 or vehicle is administered by oral gavage either once a day at the dose of 50 mg/kg, or twice daily at 30 mg/kg, as indicated. Tumor size is measured by caliper every other day. For experimental lung metastasis model, 0.1×10^6 Renca or 1×10^6 786-O cells

Animal Administration	suspended in 500 μL PBS are injected via tail vein to BALB/c or nude mice, respectively. Three days later, mice are orally treated with AZD1480 (50 mg/kg/d) or vehicle for 21 days for Renca tumors and 60 days for 786-O tumors respectively. For the Calu-6 model, 3×10^6 tumor cells in matrigel are implanted s.c. into the flanks of nude mice, randomized into vehicle (twice daily, BID) and drug treatment (AZD1480, 30 mg/kg BID) groups, and dosed orally daily for 19 days. For spontaneous lung metastasis model, 2×10^5 4T1 cells suspended in 100 μL PBS are injected in the mammary gland of female BALB/c mice by gently penetrating the skin. AZD1480 (50 mg/kg/d) or vehicle is given orally for 21 days. [1]
Kinase Assay	Inhibition studies of AZD1480 are performed using recombinant Jak1, Jak2, or Jak3 under buffer conditions of 50 mM HEPES pH 7.3, 1 mM DTT, 0.01% Tween-20, 50 mM/mL BSA, and 10 mM $MgCl_2$. Jak3 enzyme is expressed as N-terminal GST fusion in insect cells and purified by glutathione-affinity and size-exclusion chromatographies. Enzymes are assayed in the presence of AZD1480 (10 point dose response, in triplicate, from 8.3 μM to 0.3 nM in half-log dilution steps) using 1.5 μM peptide substrate (Jak1: FITC-C6-KKHTDDGYMPMSPGVA-NH2, Jak2 and Jak3: FAM-SRCtide) and screened under their respective ATP K_m (Jak1: 55 μM, Jak2: 15 μM, Jak3: 3 μM) and approximated physiological ATP concentration of 5 mM. Phosphorylated and unphosphorylated peptides are separated and quantified by a Caliper LC3000 system for calculating percent inhibition. [5]
References	[1]. Derenzini E, et al. The JAK inhibitor AZD1480 regulates proliferation and immunity in Hodgkin lymphoma. Blood Cancer J. 2011 Dec;1(12):e46. [2]. Scuto A, et al. The novel JAK inhibitor AZD1480 blocks STAT3 and FGFR3 signaling, resulting in suppression of human myeloma cell growth and survival. Leukemia. 2011 Mar;25(3):538-50. [3]. McFarland BC, et al. Therapeutic potential of AZD1480 for the treatment of human glioblastoma. Mol Cancer Ther. 2011 Dec;10(12):2384-93. [4]. Xin H, et al. Antiangiogenic and antimetastatic activity of JAK inhibitor AZD1480. Cancer Res. 2011 Nov 1;71(21):6601-10. [5]. Hedvat M, et al. The JAK2 inhibitor AZD1480 potently blocks Stat3 signaling and oncogenesis in solid tumors. Cancer Cell. 2009 Dec 8;16(6):487-9 [6]. Ni J, et al. Tyrosine receptor kinase B is a drug target in astrocytomas. Neuro Oncol. 2017 Jan;19(1):22-30.