产品名称：AZD1208
产品别名：AZD1208

生物活性：

Description	AZD1208 is a novel, orally bioavailable, highly selective PIM kinases inhibitor.
In Vitro	AZD1208 shows good antiproliferative activity in a megakaryoblastic leukemia cell line, MOLM-16, with GI_{50} values less than 100 nM[1]. AZD1208 (10 μM) inhibits the growth of Ramos cells, and at 1 μM, strongly inhibits PIM kinases in all cell at 1 μM. AZD1208 induces apoptosis, and PIM2 knockdown is mainly associated with an alteration of the cell cycle[2]. The combination of AZD1208 and AZD2014 rapidly activates AMPKα, a negative regulator of translation machinery through mTORC1/2 signaling in AML cells; profoundly inhibits AKT and 4EBP1 activation; and suppresses polysome formation[3].

Solvent&Solubility

In Vitro:

DMSO : 50 mg/mL (131.76 mM; Need ultrasonic)

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	2.6352 mL	13.1759 mL	26.3519 mL
	5 mM	0.5270 mL	2.6352 mL	5.2704 mL
	10 mM	0.2635 mL	1.3176 mL	2.6352 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 (6.59 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (6.59 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 (6.59 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (6.59 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 (6.59 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (6.59 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]. Dakin LA, et al. Discovery of novel benzylidene-1,3-thiazolidine-2,4-diones as potent and selective inhibitors of the PIM-1, PIM-2, and PIM-3 protein kinases. Bioorg Med Chem Lett. 2012 Jul 15;22(14):4599-604. [2]. Kreuz S, et al. Loss of PIM2 enhances the anti-proliferative effect of the pan-PIM kinase inhibitor AZD1208 in non-Hodgkin lymphomas. Mol Cancer. 2015 Dec 8;14:205. [3]. Harada M, et al. The novel combination of dual mTOR inhibitor AZD2014 and pan-PIM inhibitor AZD1208 inhibits growth in acute myeloid leukemia via HSF pathway suppression. Oncotarget. 2015 Nov 10;6(35):37930-47.

实验参考：

Cell Assay	MOLM-16 cells, purchased from DSMZ and cultured in RPMI containing 10% fetal bovine serum (FBS) and 1% L-glutamine, are plated at 20,000 cells per well in 96 well plates overnight. Cells are treated for 72 hours with compound or control vehicle (dimethyl sulfoxide) and cell viability is measured after the addition of Cell Titer-Blue for 4 hours at 37°C and reading of fluorescence on a Tecan Infinite® 200. The GI_{50} is determined by calculating growth at each dose relative to vehicle treated cells and cell viability at the time of treatment. [1]
Kinase Assay	The activity of purified human PIM-1, PIM-2 and PIM-3 enzymes on substrate FL-Ahx-Bad (FITC-(AHX)RSRHSSYPAGT-COOH) is determined using a mobility shift assay on a Caliper LC3000 reader. The PIM-1 assay is performed in a 12 mL reaction containing 50 mM HEPES (pH 7.5), 1 mM DTT, 0.01% Tween 20, 50 mg/mL BSA, 10 mM $MgCl_2$, 1.5 mM FL-Ahx-Bad peptide, 100 mM ATP, 2.5 nM PIM-1 and various amount of inhibitor. The reaction is quenched after 90 minute incubation at 25°C with 5 mL of stop mix consisting of 100 mM HEPES, 121 mM EDTA, 0.8% Coating Reagent 3 and 0.01% Tween 20. The ATP and enzyme concentrations for the PIM-2 assay are 5 mM and 2.5 nM, respectively, while 50 mM of ATP and 0.33 nM of enzyme is used for PIM-3 assays. For high [ATP] screenings, 5 mM ATP is used with 0.67 nM enzyme for both PIM-1 and PIM-2 or 0.11 nM PIM-3. Fluorescence of phosphorylated and unphosphorylated substrate is detected and a ratiometric value is calculated to determine percent turnover. IC_{50} values are determined from dose-response data using IDBS ActivityBase software. [1]
References	[1]. Dakin LA, et al. Discovery of novel benzylidene-1,3-thiazolidine-2,4-diones as potent and selective inhibitors of the PIM-1, PIM-2, and PIM-3 protein kinases. Bioorg Med Chem Lett. 2012 Jul 15;22(14):4599-604. [2]. Kreuz S, et al. Loss of PIM2 enhances the anti-proliferative effect of the pan-PIM kinase inhibitor AZD1208 in non-Hodgkin lymphomas. Mol Cancer. 2015 Dec 8;14:205. [3]. Harada M, et al. The novel combination of dual mTOR inhibitor AZD2014 and pan-PIM inhibitor AZD1208 inhibits growth in acute myeloid leukemia via HSF pathway suppression. Oncotarget. 2015 Nov 10;6(35):37930-47.