# 产品名称：AZD3965
# 产品别名：AZD3965

## 生物活性：

| | |
|---|---|
| **Description** | AZD3965 is a selective **MCT1** inhibitor with a $K_i$ of 1.6 nM, showing 6-fold selectivity over MCT2. |
| **$IC_{50}$ & Target** | Ki: 1.6 nM (MCT1)[1] |
| **In Vitro** | AZD3965 is designed to selectively inhibit Monocarboxylate transporter-1 (MCT1) and will therefore be expected to influence the movement of lactate into and out of cells[1]. AZD3965 treatment causes a 3.7 fold increase in intracellular lactate in hypoxic COR-L103 and 3.7 fold and 3.9 fold increases in normoxic and hypoxic NCI-H1048 cells respectively. In all other cases a <1.9 fold increase is observed. These data are consistent with AZD3965 blocking lactate transport in cells where AZD3965 also reduces cell number and is consistent with AZD3965 acting via inhibition of MCT1. When MCT1 is overexpressed the EC50 of NCI-H1048 is increased from 0.14 nM to 10.5 nM in NCI-H1048 cells. This is consistent with AZD3965 acting via MCT1 inhibition[2]. |
| **In Vivo** | COR-L103 tumor bearing mice are treated with 100 mg/kg AZD3965 or vehicle BIDfor 21 days and the tumor volume monitored. Pharmacokinetic analysis demonstrates that 100 mg/kg AZD3965 BID results in concentrations of free AZD3965 predicted to inhibit lactate transport. AZD3965 treatment significantly reduces the growth of COR-L103 tumors, although tumor regression is not seen, consistent with AZD3965 only targeting the hypoxic fraction of the tumor[2]. |

**Solvent&Solubility**

***In Vitro:***

**DMSO : ≥ 36 mg/mL (69.83 mM)**

* "≥" means soluble, but saturation unknown.

**Preparing Stock Solutions**

| Solvent / Mass / Concentration | 1 mg | 5 mg | 10 mg |
|---|---|---|---|
| 1 mM | 1.9398 mL | 9.6991 mL | 19.3983 mL |
| 5 mM | 0.3880 mL | 1.9398 mL | 3.8797 mL |
| 10 mM | 0.1940 mL | 0.9699 mL | 1.9398 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 (4.85 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (4.85 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)<br>Solubility: ≥ 2.5 mg/mL (4.85 mM); Clear solution<br>此方案可获得 ≥ 2.5 mg/mL (4.85 mM，饱和度未知) 的澄清溶液。<br>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。<br><br>3.请依序添加每种溶剂： 10% DMSO →90% corn oil<br>Solubility: ≥ 2.5 mg/mL (4.85 mM); Clear solution<br>此方案可获得 ≥ 2.5 mg/mL (4.85 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 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]. Bola BM, et al. Inhibition of monocarboxylate transporter-1 (MCT1) by AZD3965 enhances radiosensitivity by reducing lactate transport. Mol Cancer Ther. 2014 Dec;13(12):2805-16.<br>[2]. Polanski R, et al. Activity of the monocarboxylate transporter 1 inhibitor AZD3965 in small cell lung cancer. Clin Cancer Res. 2014 Feb 15;20(4):926-37. |

**实验参考：**

| | |
|---|---|
| **Animal Administration** | Mice[2]<br>COR-L103 xenografts are grown by subcutaneous injection of 5×106 cells in 0.2 mL of 1:1 serum-free RPMI:Matrigel into the mid-dorsal flank of 8 to 14-week-old male NOD scid gamma mice. Mice are housed in individually vented caging systems in a 12-hour light/12-hour dark environment and maintained at uniform temperature and humidity. Tumor size is measured twice a week using calipers and the volume calculated as tumor length×tumor width2/2. 30 days after implantation, mice bearing tumors between 150 and 250mm3 are randomized into two groups of six and treated with 100 mg/kg BID AZD3965 in 0.5% hydroxypropyl methyl cellulose, 0.1% tween 80 or vehicle only by oral gavage for 21 days. Measurements are continued 3 times a week for the duration of drug treatment to assess tumor growth kinetics. At sacrifice tumors are collected to determine intra-tumor lactate concentration. |
| **Kinase Assay** | Cells are plated overnight and treated with 100 nM AZD3965 or vehicle for 24 hours. The cells are then washed, once in PBS and twice with lysis buffer (50 mM Mops, 100 mM KCl, 5 mM MgCl2, 1 mM EDTA, 0.1 mM DTT, 1 mM PMSF supplemented with 1× mini complete protease inhibitor cocktail tablets. The cells are harvested by scraping and centrifugation, and the pellet snap frozen without buffer in liquid nitrogen. Frozen aliquots of cells are thawed on ice and re-suspended in lysis buffer. Cells are lysed by 3 rounds of freezing in liquid nitrogen and thawing at 37°C for 2 minutes each. Lysates are subsequently centrifuged (13000 g, 10min, 4°C) until clear and then stored on ice. Enzyme activity in the cell lysates is determined using a micro-plate reader to measure either production or depletion of NADH/NADPH, through its absorbance at 340/10 nm, occurring as a result of direct or coupled enzyme reactions. The 96 well plates used for the assays are pre-heated to 37°C for 10 minutes prior to starting reactions, initiated by the addition of 5 μL cell lysate to 95 μL of reaction buffer (50 mM Mops pH 7.4, 100 mM KCl, 5 mM free magnesium). The standard reaction buffer is supplemented to assay the kinetics of the different enzymes. Absorbance values for controls are subtracted from absorbance of corresponding reactions. Graphpad prism 6 is used to plot V0 values against substrate concentration and determine Vmax and Km values. The Vmax is then normalised to the protein concentration in the cell lysate[1]. |

| | |
|---|---|
| **References** | [1]. Bola BM, et al. Inhibition of monocarboxylate transporter-1 (MCT1) by AZD3965 enhances radiosensitivity by reducing lactate transport. Mol Cancer Ther. 2014 Dec;13(12):2805-16.<br>[2]. Polanski R, et al. Activity of the monocarboxylate transporter 1 inhibitor AZD3965 in small cell lung cancer. Clin Cancer Res. 2014 Feb 15;20(4):926-37. |