产品名称：**ENMD-2076**
产品别名：**ENMD-2076**

## 生物活性：

| | |
|---|---|
| **Description** | ENMD-2076 is a multi-targeted kinase inhibitor with $IC_{50}$s of 1.86, 14, 58.2, 15.9, 92.7, 70.8, 56.4 nM for Aurora A, Flt3, KDR/VEGFR2, Flt4/VEGFR3, FGFR1, FGFR2, Src, PDGFRα, respectively. |

**$IC_{50}$ & Target**

| Aurora A | KDR | Flt-4 | FGFR1 | FGFR2 | PDGFRα |
|---|---|---|---|---|---|
| 14 nM ($IC_{50}$) | 58.2 nM ($IC_{50}$) | 15.9 nM ($IC_{50}$) | 92.7 nM ($IC_{50}$) | 70.8 nM ($IC_{50}$) | 56.4 nM ($IC_{50}$) |

| Flt3 |
|---|
| 1.86 nM ($IC_{50}$) |

**In Vitro**

ENMD-2076 is selective toward Aurora A versus Aurora B ($IC_{50}$=350 nM). ENMD-2076 inhibits HUVEC growth with an $IC_{50}$ value of 0.15 mM. Against 10 human leukemia cell lines, the $IC_{50}$ values range from 0.025 to 0.53 mM. Within this panel, MV4:11 cells are the most sensitive cells by a factor of greater than 4. The lymphoma-derived U937 cell line treated with ENMD-2076 shows that the ENMD-2076 induces a dose-dependent increase in G2-M-phase arrest as well as the induction of apoptosis. ENMD-2076 inhibits cellular Flt3 ligand (FL)-induced Flt3 autophosphorylation in THP-1 cells, which have been shown to express FL-responsive wild-type Flt- 3 (18) with an $IC_{50}$ value of 28 nM. ENMD-2076 inhibits stem cell factor (SCF)-induced Kit autophosphorylation in MO7e cells with an $IC_{50}$ value of 40 nM. ENMD-2076 inhibits VEGFR2/KDR autophosphorylation with an $IC_{50}$ value of 7 nM[1].

**In Vivo**

ENMD-2076 treatment results in statistically significant, dose dependent inhibition of tumor growth or tumor regression. Moreover, there is no correlation between tumor growth rate and antitumor efficacy, which would conceivably be expected for a mitotic kinase inhibitor, as fast growing (e.g., A375 melanoma) and slow-growing (e.g., HT29 colon carcinoma) tumors are similarly inhibited by ENMD-2076. ENMD-2076 is well tolerated at daily doses up to 302 mg/kg (equivalent to 200 mg/kg of the free base), with no weight loss or signs of morbidity noted in any study at this dose with the exception of the A375 model[1].

**Solvent&Solubility**

***In Vitro:***

**DMSO : ≥ 31 mg/mL (82.56 mM)**

**$H_2O$ : < 0.1 mg/mL** (insoluble)

* "≥" means soluble, but saturation unknown.

| Preparing Stock Solutions | Solvent / Mass / Concentration | 1 mg | 5 mg | 10 mg |
|---|---|---|---|---|
| | 1 mM | 2.6633 mL | 13.3166 mL | 26.6333 mL |
| | 5 mM | 0.5327 mL | 2.6633 mL | 5.3267 mL |
| | 10 mM | 0.2663 mL | 1.3317 mL | 2.6633 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<br>1.请依序添加每种溶剂： 10% DMSO→40% PEG300 →5% Tween-80 → 45% saline<br>Solubility: ≥ 2.5 mg/mL (6.66 mM); Clear solution<br>此方案可获得 ≥ 2.5 mg/mL (6.66 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 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。<br>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 (6.66 mM); Suspended solution; Need ultrasonic<br>此方案可获得 2.5 mg/mL (6.66 mM)的均匀悬浊液，悬浊液可用于口服和腹腔注射。<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>3.请依序添加每种溶剂： 10% DMSO →90% corn oil<br>Solubility: ≥ 2.5 mg/mL (6.66 mM); Clear solution<br>此方案可获得 ≥ 2.5 mg/mL (6.66 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]. Fletcher GC, et al. ENMD-2076 is an orally active kinase inhibitor with antiangiogenic and antiproliferative mechanisms of action. Mol Cancer Ther. 2011 Jan;10(1):126-37.<br>[2]. Wang X, et al. Preclinical activity of a novel multiple tyrosine kinase and aurora kinase inhibitor, ENMD-2076, against multiple myeloma. Br J Haematol. 2010 Aug;150(3):313-25. |

**实验参考：**

| | |
|---|---|
| **Cell Assay** | The antiproliferative effect of ENMD-2076 on adherent tumor cell lines is measured by plating 500 cells per well in a 96-well plate and incubating with 9 doses of compound, spanning 0.3 nM to 125 mM, for 96 hours. Cellular proliferation is measured using the sulforhodamine B assay[1] |
| **Animal Administration** | Mice: Cell lines are injected subcutaneously or into the mammary fat pad (MDA-MB-231 only) of 5- to 6-week-old CB.17 SCID or NCr nude mice. Tumors are allowed to grow for 10 to 50 days before drug treatment. All treatments are with ENMD-2076 in water or ENMD-2076 free base in CMC-Tween vehicle (0.075% carboxymethylcellulose, 0.085% Tween 80 in water), administered orally. Percent tumor growth inhibition is calculated[1] |
| **Kinase Assay** | Recombinant Aurora A and B kinase enzymes assays are carried out in kinase assay buffer (50 mM of HEPES, pH 7.5, 10 mM of $MgCl_2$, 5 mM of EGTA, 0.05% Brij-35) supplemented with 2 mM of DTT. Activities are determined at an ATP concentration equivalent to the apparent Km for each enzyme, and an enzyme concentration that results in approximately 30% phosphorylation of the peptide substrate after 1 hour. Dose–response curves of relative enzyme activity versus ENMD-2076 concentration are plotted with Grafit and used to calculate $IC_{50}$ values[1] |
| **References** | [1]. Fletcher GC, et al. ENMD-2076 is an orally active kinase inhibitor with antiangiogenic and antiproliferative mechanisms of action. Mol Cancer Ther. 2011 Jan;10(1):126-37.<br>[2]. Wang X, et al. Preclinical activity of a novel multiple tyrosine kinase and aurora kinase inhibitor, ENMD-2076, against multiple myeloma. Br J Haematol. 2010 Aug;150(3):313-25. |