产品名称：**TAK-733**
产品别名：**TAK-733**

生物活性：

Description	TAK-733 is a potent and selective MEK allosteric site inhibitor with an IC_{50} of 3.2 nM.
IC_{50} & Target	MEK
	3.2 nM (IC_{50})
In Vitro	TAK-733 exhibits potent enzymatic and cell activity with an IC_{50} of 3.2 nM against constitutively active MEK enzyme and an EC_{50} of 1.9 nM against ERK phosphorylation in cells. TAK-733 does not inhibit any other kinases, receptors or ion channels that are tested with inhibitor concentrations up to 10 μM. TAK-733 is found to bind plasma protein moderately (ca. 97% for human and 96% for mouse), and exhibits high permeability and high microsomal stability across species. It does not inhibit P450s up to 30 μM[1]. TAK-733 demonstrates broad activity in most melanoma cell lines with relative resistance observed at IC_{50} >0.1 μM in vitro. Thirty-four melanoma cell lines are exposed in vitro to increasing concentrations of TAK-733 for 72 hours. Of the 34 cell lines, 27 are $BRAF^{V600E}$ mutant and 7 are wild-type. SRB proliferation assays are performed and the resulting IC_{50} concentrations allowed stratification of cell lines into three categories: relatively resistant, intermediate, and highly sensitive. Relatively resistant and highly sensitive lines are assigned based on an IC_{50} that differ by at least 10 fold[2].
In Vivo	The pharmacokinetics of TAK-733 is evaluated in nude mouse, rat, dog and monkey. Low clearance and high oral bioavailability are observed in all species. TAK-733 demonstrates broad antitumor activity in mouse xenograft models of human cancer including models of melanoma, colorectal, NSCLC, pancreatic and breast cancer[1]. Daily oral administration of 1, 3, 10, and 30 mg/kg of TAK-733 for 14 days (Days 10 to 23) results in tumor growth delay in the A375 cell-implanted mice (5/group). TAK-733 (35, 70, 100, and 160 mg/kg) also significantly inhibits tumor growth on an intermittent dosing schedule of 3 days per week for 2 weeks (Days 10, 13, 15, 17, 20, and 22). Three partial regressions (PR), a 60% response rate, are observed in mice administered with 30 mg/kg of TAK-733 daily and in mice administered with 160 mg/kg of TAK-733 intermittently. Responses, CR (complete regression) and partial regressions (PR) are also observed in mice administered with 70, 100, and 160 mg/kg of TAK-733 intermittently. The tumor regression rate is more pronounced with the intermittent administration regimen; the greatest reduction in tumor volume is observed at 160 mg/kg (57.29%), versus a maximum reduction of 46.97% at 30 mg/kg once daily. By the last day of administration, tumor growth is significantly ($p<0.05$ for %T/C, Student's t-test) inhibited in mice administered 3, 10, and 30 mg/kg once daily or 35, 70, 100, and 160 mg/kg intermittently[2].
Solvent&Solubility	***In Vitro:***
	DMSO : ≥ 33 mg/mL (65.45 mM)
	* "≥" means soluble, but saturation unknown.

Preparing Stock Solutions	Solvent / Mass / Concentration	**1 mg**	**5 mg**	**10 mg**
	1 mM	1.9832 mL	9.9161 mL	19.8322 mL
	5 mM	0.3966 mL	1.9832 mL	3.9664 mL
	10 mM	0.1983 mL	0.9916 mL	1.9832 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 →90% corn oil Solubility: ≥ 2.5 mg/mL (4.96 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (4.96 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]. Dong Q, et al. Discovery of TAK-733, a potent and selective MEK allosteric site inhibitor for the treatment of cancer. Bioorg Med Chem Lett. 2011 Mar 1;21(5):1315-9. [2]. Micel LN, et al. Antitumor activity of the MEK inhibitor TAK-733 against melanoma cell lines and patient-derived tumor explants. Mol Cancer Ther. 2015 Feb;14(2):317-25.

实验参考：

Cell Assay	The cutaneous melanoma cell lines in logarithmic growth phase are transferred to 96 well flat bottom plates with lids. One hundred μL cell suspensions containing 2000-3000 viable cells are plated into each well and incubated overnight prior to exposure with increasing concentrations of TAK-733 (10, 20, 30, 40, 50, 60, 70, 80, 90, and 100 nM) for 72 hours. Post drug administration, media is removed and cells are fixed with cold 10% trichloroacetic acid for 30 min. at 4°C. Cells are then washed with water and stained with 0.4% SRB for 30 min at room temperature, washed again with 1% acetic acid, followed by stain solubilization with 10 mM tris at room temperature. The absorbance at 565 nm is measured on a plate reader. Cell proliferation curves are derived from the raw absorbance (OD) data. Statistical analyses and graphical representation of data are using GraphPad Prism version 5.00[2].
Animal Administration	Mice[2] Five to six-week-old female athymic nude mice are used. A375 human melanoma xenograft tumors are generated by harvesting cells from mid-log phase cultures using Trypsin-EDTA. Approximately 5×10^6 cells suspended in Hanks' balanced salt solution (HBSS) are injected sc into the right flank of 6-8-week-old mice. Oral administration of TAK-733 (1 mg/kg or 10 mg/kg) is initiated when tumors in all mice in each experiment range in size from 100 to 200 mm^3 for antitumor efficacy studies and from 300 to 500 mm3 for pharmacodynamic (PD) studies[2].
References	[1]. Dong Q, et al. Discovery of TAK-733, a potent and selective MEK allosteric site inhibitor for the treatment of cancer. Bioorg Med Chem Lett. 2011 Mar 1;21(5):1315-9. [2]. Micel LN, et al. Antitumor activity of the MEK inhibitor TAK-733 against melanoma cell lines and patient-derived tumor explants. Mol Cancer Ther. 2015 Feb;14(2):317-25.