产品名称：**VX-11e**
产品别名：**VX-11e**

生物活性：

Description	VX-11e is a potent, selective, and orally bioavailable inhibitor of ERK with K_i < 2 nM.			
IC_{50} & Target	ERK2	GSK3	AURA	CDK2
	2 nM (Ki)	395 (Ki)	540 (Ki)	852 (Ki)
In Vitro	VX-11e is active in the HT29 cell proliferation assay (IC50=48 nM)[1].			
In Vivo	VX-11e is orally bioavailable in both rat and mice[1]. VX-11e (50 mg/kg, p.o.) results in robust inhibition of pRSK, and inhibits tumor growth in NSG mice bearing human melanoma RPDX tumors. VX-11e with BKM120 significantly improves tumor growth inhibition[2].			

Solvent&Solubility

In Vitro:

DMSO : ≥ 100 mg/mL (**199.86 mM**)

H_2O : < 0.1 mg/mL (insoluble)

Preparing Stock Solutions	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
	1 mM	1.9986 mL	9.9930 mL	19.9860 mL
	5 mM	0.3997 mL	1.9986 mL	3.9972 mL
	10 mM	0.1999 mL	0.9993 mL	1.9986 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: ≥ 3.25 mg/mL (6.50 mM); Clear solution

此方案可获得 ≥ 3.25 mg/mL (6.50 mM，饱和度未知) 的澄清溶液。

以 1 mL 工作液为例，取 100 μL 32.5 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% corn oil

Solubility: ≥ 3.25 mg/mL (6.50 mM); Clear solution

此方案可获得 ≥ 3.25 mg/mL (6.50 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 32.5 mg/mL 的澄清 DMSO 储备液加到 900 μL 玉米油中，混合均匀。

[1]. Aronov, Alex M., et al. Structure-Guided Design of Potent and Selective Pyrimidylpyrrole Inhibitors of Extracellular Signal-Regulated Kinase (ERK) Using Conformational Control. Journal of Medicinal

References	Chemistry (2009), 52(20), 6362-6368. [2]. Krepler C, et al. Personalized Preclinical Trials in BRAF Inhibitor-Resistant Patient-Derived Xenograft Models Identify Second-Line Combination Therapies. Clin Cancer Res. 2016 Apr 1;22(7):1592-602.

实验参考：

Cell Assay	Cell proliferation is measured by ^{3}H-thymidine incorporation. The cells are plated at a concentration of 10,000 cells/well in a 96-well plate using growth media, RPMI 1640 containing 10% FBS. Serially diluted compounds are added. The cells and compounds are incubated for 48 hours at 37°C incubator. After 48 hours, 0.4 μCi of ^{3}H-thymidine is added to each wells for 8 hours and returned to the 37°C incubator. The cells are harvested using a Tomtec 96-well cell harvester and the CPM is determined using the Wallac 1205 BETAPLATE liquid scintillation counter. [1]
Animal Administration	Human melanoma RPDX tumors are expanded in vivo using NSG mice prior to the therapy experiments. Pooled tumor chunks banked from early mouse passages are implanted into 50 NSG mice (1:10 expansion). These tumors are harvested when reaching the maximum volume allowed on the protocol (1,000 mm^3), digested, and banked as live cells. The larger part of this stock is retained as a master bank, and the other part is implanted at a 1:5 ratio into NSG mice to use in the therapy experiments. The expansion phase is under continuous drug pressure with PLX4720 200 ppm chemical additive diet at approximately clinical plasma levels. The plasma levels of PLX4720 (103.7 μg/mL ±3.2 after 7 days) are similar to steady-state levels in patients treated with vemurafenib 960 mg twice a day (130.6 μg/mL±71.78). When tumors have reached 200 mm^3 per caliper measurement, animals are randomized into treatment groups followed by a 3-day ishout phase. Tumor size is assessed twice weekly per caliper measurement. Mice are sacrificed after two weeks of treatment or when necessary for animal welfare. Dosing is prolonged when tumor control is achieved as indicated. Tumor tissue is conserved in formalin (for FFPE) and snap-frozen in liquid N2 for protein extraction. Treatment groups are sacrificed 4 hours after last dose. [2]
Kinase Assay	Compounds are assayed for the inhibition of ERK2 by a spectophotometric coupled-enzyme assay. In this assay, a fixed concentration of activated ERK2 (10 nM) is incubated with various concentrations of the compounds in DMSO (2.5%) for 10 min. at 30°C in 0.1 mol/L HEPES buffer, pH=7.5, containing 10 mM $MgCl_2$, 2.5 mM phosphoenolpyruvate, 200 μM NADH, 150 μg/mL pyruvate kinase, 50 μg/mL lactate dehydrogenase and 200 μM erktide peptide. The reaction is initiated by the addition of 65 μM ATP. The rate of decrease of absorbance at 340 nM is monitored. [1]
References	[1]. Aronov, Alex M., et al. Structure-Guided Design of Potent and Selective Pyrimidylpyrrole Inhibitors of Extracellular Signal-Regulated Kinase (ERK) Using Conformational Control. Journal of Medicinal Chemistry (2009), 52(20), 6362-6368. [2]. Krepler C, et al. Personalized Preclinical Trials in BRAF Inhibitor-Resistant Patient-Derived Xenograft Models Identify Second-Line Combination Therapies. Clin Cancer Res. 2016 Apr 1;22(7):1592-602.