产品名称：**FRAX597**
产品别名：**FRAX597**

生物活性：

Description	FRAX597 is a potent group I p21-activated Kinases (**PAK**s) inhibitor with **IC_{50}** of 8, 13 and 19 nM for **PAK1**, **2** and **3**.

IC_{50} & Target	PAK1	PAK2	PAK3
	8 nM (IC_{50})	13 nM (IC_{50})	19 nM (IC_{50})

In Vitro	FRAX597 is determined to be a potent, ATP-competitive inhibitor of group I PAKs (PAK 1-3), with biochemical IC50 values as follows: PAK1 IC50=8 nM, PAK2 IC50=13 nM, PAK3 IC50=19 nM. The IC50 toward PAK4, a member of group II PAKs is >10 μM. At a concentration of 100 nM FRAX597 displays a significant (>80% inhibition) inhibitory capacity toward YES1 (87%), RET (82%), CSF1R (91%), TEK (87%), PAK1 (82%), and PAK2 (93%). When measured using the Kinase Glo Assay in the presence of 20 nM protein and 1 μM ATP, FRAX597 displayed an IC50 value of 48 nM against wild type PAK1, while IC50 values against the V342F and V342Y PAK1 mutants are higher than 3 μM and 2 μM, respectively[1].
In Vivo	Analysis of the flux reading for the animals in the two cohorts demonstrates a significantly slower tumor growth rate in FRAX597-treated mice compared with control mice. After 14 days of treatment the animals are sacrificed and the tumors excised and weighed. FRAX597-treated cohort shows significantly lower average tumor weight compared with the control cohort (0.55 g versus 1.87 g, p=0.0001)[1].

Solvent&Solubility

In Vitro:

DMSO : 14.29 mg/mL (**25.60 mM;** Need ultrasonic)

H_2O : < 0.1 mg/mL (insoluble)

Preparing Stock Solutions	Solvent / Mass / Concentration	**1 mg**	**5 mg**	**10 mg**
	1 mM	1.7918 mL	8.9590 mL	17.9179 mL
	5 mM	0.3584 mL	1.7918 mL	3.5836 mL
	10 mM	0.1792 mL	0.8959 mL	1.7918 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: 1.43 mg/mL (2.56 mM); Suspended solution; Need ultrasonic

此方案可获得 1.43 mg/mL (2.56 mM)的均匀悬浊液，悬浊液可用于口服和腹腔注射。

以 1 mL 工作液为例，取 100 μL 14.299999 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: 1.43 mg/mL (2.56 mM); Suspended solution; Need ultrasonic 此方案可获得 1.43 mg/mL (2.56 mM)的均匀悬浊液，悬浊液可用于口服和腹腔注射。 以 1 mL 工作液为例，取 100 μL 14.299999 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: ≥ 1.43 mg/mL (2.56 mM); Clear solution 此方案可获得 ≥ 1.43 mg/mL (2.56 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 14.299999 mg/mL 的澄清 DMSO 储备液加到 900 μL 玉米油中，混合均匀。
References	[1]. Licciulli S, et al. FRAX597, a small molecule inhibitor of the p21-activated kinases, inhibits tumorigenesis of neurofibromatosis type 2 (NF2)-associated Schwannomas. J Biol Chem. 2013 Oct 4;288(40):29105-14.

实验参考：

Cell Assay	30,000 SC4 cells/well are plated in 12-well dishes in triplicate. Cell growth media with or without FRAX597 (1 μM) is replaced daily. At indicated time points, cells from individual wells are trypsinized and counted using a Coulter counter. Statistical analysis is performed using a Student's t test. For cell cycle analysis, cells are harvested, washed once with PBS and fixed in cold 70% ethanol. Fixed cells are resuspended in propidium iodide (PI) buffer (50 μg/mL PI, 250 mg/mL RNase A in PBS) and incubated overnight at 4°C in the dark. Cell cycle distribution is evaluated using Coulter Epics XL flow cytometer. Data are analyzed using WinMDI software[1].
Animal Administration	Mice[1] Nf2-/- SC4 Schwann cells are transduced by lentiviruses carrying pLuc-mCherry and sorted by FACS. 5×104 cells are transplanted into the sciatic nerve sheath of NOD/SCID mice (8 weeks of age) by intraneural injection. Tumor progression is monitored weekly by bioluminescence imaging (BLI) on an IVIS-200 system. The representative images from bioluminescence imaging (BLI) of mice carrying orthotopic tumors treated with FRAX597 (100 mg/kg) or vehicle control at day 14 of treatment. NOD/SCID mice are injected intraneurally with 5×104 SC4/pLuc-mCherry cells and are enrolled into treatment after 10 days. Mice are treated daily for 14 days and imaged every 3 days to follow tumor development.
References	[1]. Licciulli S, et al. FRAX597, a small molecule inhibitor of the p21-activated kinases, inhibits tumorigenesis of neurofibromatosis type 2 (NF2)-associated Schwannomas. J Biol Chem. 2013 Oct 4;288(40):29105-14.