

产品名称：**PF-04449913**  
产品别名：**Glasdegib**

生物活性：				
Description	Glasdegib (PF-04449913) is a potent and orally bioavailable smoothened inhibitor. Glasdegib (PF-04449913) binds to human SMO (amino acids 181-787) with an IC50 of 4 nM.			
IC <sub>50</sub> & Target	IC50: 4 nM (Smo)[1]			
In Vitro	Glasdegib (PF-04449913) inhibits sonic hedgehog (Shh) stimulated luciferase expression in mouse embryonic fibroblasts with an IC <sub>50</sub> of 6.8 nM; and significantly reduces medulloblastoma growth in a Ptch1 <sup>+/+</sup> p53 <sup>-/-</sup> allograft model at doses that decreased murine Shh target gene expression. In stromal co-culture experiments, FACS analysis demonstrates a significant reduction in BC LSC by Glasdegib (PF-04449913) when compared with normal progenitors. Importantly, human BC LSC engrafted RAG2 <sup>-/-</sup> γC <sup>-/-</sup> mice treated daily with Glasdegib (PF-04449913) compared with vehicle treated controls have a significant spleen weight reduction (p=0.006). This reduction in leukemic burden corresponded with decreased GLI2 protein expression, as determined by both nanoproteomic analysis of FACS purified human progenitors and GLI2 confocal fluorescence microscopic analysis of splenic sections[1].			
In Vivo	Human BC LSC engrafted RAG2 <sup>-/-</sup> γC <sup>-/-</sup> mice treated daily with Glasdegib (PF-04449913) compared with vehicle treated controls had a significant spleen weight reduction (p=0.006). When CD34 <sup>+</sup> cord blood engrafted NSG mice are treated with Glasdegib (PF-04449913), the frequency of human CD45 <sup>+</sup> cells, progenitors and both myeloid and lymphoid cell fate commitment remained comparable to vehicle treated controls indicating that unlike LSC, normal human HSC cell fate decisions are Hh pathway independent. These results highlight the important niche dependent effects of selective SMO inhibition that induce GLI2 downregulation in a cell type and context specific manner[1].			
Solvent&Solubility	<b>In Vitro:</b> <b>DMSO : ≥ 83.33 mg/mL (222.55 mM)</b> <small>* "≥" means soluble, but saturation unknown.</small>			
		<div><div>Solvent</div><div>Mass</div><div>Concentration</div></div>	1 mg	5 mg
	Preparing	1 mM	2.6707 mL	13.3533 mL
	Stock Solutions	5 mM	0.5341 mL	2.6707 mL
		10 mM	0.2671 mL	1.3353 mL
<b>*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。</b> <b>储备液的保存方式和期限</b> -80℃, 6 months; -20℃, 1 month。-80℃ 储存时，请在 6 个月内使用，-20℃ 储存时，请在 1 个月内使用。 <b>In Vivo:</b> 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 <b>In Vitro</b> 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 <div>1.请依序添加每种溶剂： 10% DMSO→40% PEG300 →5% Tween-80 → 45% saline</div> <div>Solubility: ≥ 2.08 mg/mL (5.55 mM); Clear solution</div> <div>此方案可获得 ≥ 2.08 mg/mL (5.55 mM, 饱和度未知) 的澄清溶液。</div>				

	<p>以 1 mL 工作液为例，取 100 <math>\mu</math>L 20.8 mg/mL 的澄清 DMSO 储备液加到 400 <math>\mu</math>L PEG300 中，混合均匀向上述体系中加入 50 <math>\mu</math>L Tween-80，混合均匀；然后继续加入 450 <math>\mu</math>L 生理盐水定容至 1 mL。</p> <p>2.请依序添加每种溶剂： 10% DMSO→ 90% (20% SBE-<math>\beta</math>-CD in saline) Solubility: <math>\geq</math> 2.08 mg/mL (5.55 mM); Clear solution 此方案可获得 <math>\geq</math> 2.08 mg/mL (5.55 mM，饱和度未知) 的澄清溶液。 以 1 mL 工作液为例，取 100 <math>\mu</math>L 20.8 mg/mL 的澄清 DMSO 储备液加到 900 <math>\mu</math>L 20% 的 SBE-<math>\beta</math>-CD 生理盐水水溶液中，混合均匀。</p> <p>3.请依序添加每种溶剂： 10% DMSO →90% corn oil Solubility: <math>\geq</math> 2.08 mg/mL (5.55 mM); Clear solution 此方案可获得 <math>\geq</math> 2.08 mg/mL (5.55 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 <math>\mu</math>L 20.8 mg/mL 的澄清 DMSO 储备液加到 900 <math>\mu</math>L 玉米油中，混合均匀。</p>
References	<p>[1]. Sadarangani A, et al. <u>GLI2 inhibition abrogates human leukemia stem cell dormancy</u>. J Transl Med. 2015 Mar 21;13:98.</p>
实验参考：	
Cell Assay	<p>Normal or BC CML CD34<sup>+</sup> cells are plated on confluent mitomycin-C treated SL/M2 cells with vehicle, Glasdegib (PF-04449913) (1 <math>\mu</math>M), Dasatinib (50 nM), or combination treatment. Mouse bone marrow stromal cell lines, M2-10B4 (M2) and SL/SL (SL) are treated with mitomycin-C (1 mg/mL) and plated in a 1:1 mixture at a total concentration of 100,000 cells/mL one day prior to co-culture with 10,000-20,000 CD34<sup>+</sup> BC CML or normal progenitors. After 1 week of culture, progenitors are FACS sorted into hematopoietic progenitor assays and colonies are scored at 14 days. To assess survival of normal human hematopoietic stem and progenitor cells, irradiated (20 Gray) OP9 (M2 clone) stromal cells are co-cultured with 50,000 human CD34<sup>+</sup> cord blood cells, vehicle or Glasdegib (PF-04449913) in AlphaMEM with 20% Hyclone FBS, 1% pen strep glutamine and supplemented with 50 ng/mL SCF, 10 ng/mL thrombopoietin, and 10 ng/mL Flt3 and quantified by weekly FACS analysis[1]</p>
Animal Administration	<p>Mice[1] RAG2<sup>+/+</sup>c<sup>-/-</sup> mice are transplanted intrahepatically with equal numbers of normal progenitors or BC LSC. Upon detection of human CD45<sup>+</sup> cell peripheral blood engraftment, mice are treated daily by oral gavage with vehicle (50% 1,2 Propandiol, 50% HBSS or methylcellulose), Glasdegib (100 mg/kg), Dasatinib (50 mg/kg), or the combination for 14 days followed by FACS to quantify human engraftment in hematopoietic niches. To assess effects on normal HSC function, 7 to 10 week old NOD. Cg-PrkdcSCID IL2R1Wjl/SzJ mice are sublethally irradiated, transplanted retro-orbitally with 100,000 CD34<sup>+</sup> human cord blood cells and treated 8 weeks later with vehicle or Glasdegib (100 mg/kg) for 14 days followed by FACS engraftment analysis.</p>
References	<p>[1]. Sadarangani A, et al. <u>GLI2 inhibition abrogates human leukemia stem cell dormancy</u>. J Transl Med. 2015 Mar 21;13:98.</p>