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产品名称：N-[4-[(4-乙基-1-哌嗪基)甲基]-3-(三氟甲基)苯基]-4-甲基-3-(1H-吡咯并[2,3-B]吡啶-4-基氧基)苯甲酰胺
产品别名：NG25

生物活性：

Description	NG25 is a potent dual TAK1 and MAP4K2 inhibitor, with IC50s of 149 nM and 21.7 nM, respectively.				
IC_{50} & Target	MAP4K2	TAK1	LYN	GSK	ABL,ARG
	21.7 nM (IC_{50})	149 nM (IC_{50})	12.9 nM (IC_{50})	56.4 nM (IC_{50})	75.2 nM (IC_{50})
	FER	SRC	Eph B2	ZAK	Eph A2
	82.3 nM (IC_{50})	113 nM (IC_{50})	672 nM (IC_{50})	698 nM (IC_{50})	773 nM (IC_{50})
	Eph B4	ZC1/HGK	RAF1		
	999 nM (IC_{50})	3250 nM (IC_{50})	7590 nM (IC_{50})		
In Vitro	NG25 is a potent dual TAK1 and MAP4K2 inhibitor, with IC_{50}s of 149 nM and 21.7 nM, respectively. NG25 also potently suppresses several kinases such as LYN, CSK, FER, p38α, ABL,ARG and SRC, with IC_{50}s of 12.9, 56.4, 82.3, 102, 75.2, and 113 nM, respectively[1]. NG25 is very potent suppressor of CpG B- or CpG A-stimulated secretion of IFNα and CL097-stimulated secretion of IFNβ, with complete inhibition by 400 nM[2]. NG25 treatment reduces cell viability of all tested breast cancer cell lines in a dose dependent manner. NG25 (2 μM) enhances the cytotoxic effect of Dox on breast cancer cells[3].				

Solvent&Solubility

In Vitro:

DMSO : ≥ 100 mg/mL (186.02 mM)

H_2O : < 0.1 mg/mL (insoluble)

* "≥" means soluble, but saturation unknown.

	Solvent / Mass / Concentration	1 mg	5 mg	10 mg
Preparing Stock Solutions	1 mM	1.8602 mL	9.3009 mL	18.6019 mL
	5 mM	0.3720 mL	1.8602 mL	3.7204 mL
	10 mM	0.1860 mL	0.9301 mL	1.8602 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: ≥ 2.5 mg/mL (4.65 mM); Clear solution

此方案可获得 ≥ 2.5 mg/mL (4.65 mM，饱和度未知) 的澄清溶液。

	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。 2.请依序添加每种溶剂： 10% DMSO →90% corn oil Solubility: ≥ 2.5 mg/mL (4.65 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (4.65 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]. Tan L, et al. Discovery of type II inhibitors of TGFβ-activated kinase 1 (TAK1) and mitogen-activated protein kinase kinase kinase kinase 2 (MAP4K2). J Med Chem. 2015 Jan 8;58(1):183-96. [2]. Pauls E, et al. Essential role for IKKβ in production of type 1 interferons by plasmacytoid dendritic cells. J Biol Chem. 2012 Jun 1;287(23):19216-28. [3]. Wang Z, et al. TAK1 inhibitor NG25 enhances doxorubicin-mediated apoptosis in breast cancer cells. Sci Rep. 2016 Sep 7;6:32737.
实验参考：	
Cell Assay	3.5×10^5 Gen2.2 cells or Flt3-DCs are incubated for 1 h in 96-well plates without or with the indicated concentrations of inhibitor, then stimulated with 1 μM CpG (type A or B) or 1 μg/mL of CL097 or R848. After 5 or 12 h the cell culture supernatants are collected, clarified by centrifugation, and frozen at −80°C until cytokine levels are analyzed. For cell viability assays, unstimulated cells are incubated for 12 h in the absence or presence of inhibitors. Cells are then fixed and the percentage of live cells analyzed by flow cytometry. [2]
Kinase Assay	IRF7 is expressed in Escherichia coli as a glutathione S-transferase (GST) fusion protein with a PreScission proteinase cleavage site between the GST and the IRF7. The GST-IRF7 is captured on glutathione-Sepharose and IRF7 released from GST and glutathione-Sepharose by digestion with PreScission proteinase. His6-tagged IKKβ and TBK1 are expressed in their active phosphorylated forms in insect Sf21 cells and purified by affinity chromatography on nickel nitrilotriacetate-agarose. Active GST-IKKα is purchased from Millipore and assayed. [2]
References	[1]. Tan L, et al. Discovery of type II inhibitors of TGFβ-activated kinase 1 (TAK1) and mitogen-activated protein kinase kinase kinase kinase 2 (MAP4K2). J Med Chem. 2015 Jan 8;58(1):183-96. [2]. Pauls E, et al. Essential role for IKKβ in production of type 1 interferons by plasmacytoid dendritic cells. J Biol Chem. 2012 Jun 1;287(23):19216-28. [3]. Wang Z, et al. TAK1 inhibitor NG25 enhances doxorubicin-mediated apoptosis in breast cancer cells. Sci Rep. 2016 Sep 7;6:32737.