产品名称：4-(3-Chloroanilino)-6,7-Dimethoxyquinazoline Hydrochloride
产品别名：AG-1478

生物活性：

Description	AG-1478 is a selective **EGFR** tyrosine kinase inhibitor with **IC_{50}** of 3 nM.
IC_{50} & Target	EGFR
	3 nM (IC_{50})
In Vitro	AG-1478 (AG1478) is irreversible for growth regulation of human lung (A549) and prostate (DU145) cancer cell lines, cultured in chemically defined DMEM/F12 medium. AG-1478 seems to be more effective at lower concentrations, but is unable to completely inhibit growth of A549 cells[1]. Inhibition of EGFR by specific tyrosine kinase inhibitor AG-1478 (AG1478) significantly decreases the angiotensin II-mediated synthesis of TGF-β and fibronectin by cardiac fibroblasts. EGFR is pharmacologically inhibited by small-molecule inhibitor AG-1478 with IC50 of 4 nM[2]. Both Polyfect (PF) and Superfect (SF) treatment lead to increased apoptosis in HEK 293 cells to a similar extent as assessed by flow cytometry. The antioxidant, tempol, significantly reduced dendrimer-mediated apoptosis for both PF and SF. AG-1478 (AG1478), at a 10-fold higher dose (100 μM) than used in signaling studies, is used as a positive control and significantly induced apoptosis in HEK 293 cells[3].
In Vivo	Administration of AG-1478 (AG1478) significantly reduces myocardial inflammation, fibrosis, apoptosis, and dysfunction in both two obese mouse models. ApoE-/- mice are first fed with HFD for 8 weeks (ApoE-HFD), and then administrated with AG-1478 (10 mg/kg/day) or 542 (10 mg/kg/day) for another 8 weeks by oral gavage. AG-1478 or 542 treatment blocks HFD induced cardiac EGFR phosphorylation in vivo, without affecting the plasma level of low density lipoprotein (LDL) and total triglyceride (TG)[2]. Administration of EGF (10 nM) leads to a robust and reproducible elevation in EGFR phosphorylation that can be blocked by AG-1478 (AG1478), a known inhibitor of EGFR phosphorylation. Increasing doses of Polyfect (PF) result in a significant reduction in EGF-induced EGFR phosphorylation ($p<0.05$) but this is to a lesser extent than observed with AG1478[3].
Solvent&Solubility	***In Vitro:*** **DMSO : ≥ 100 mg/mL (316.71 mM)** * "≥" means soluble, but saturation unknown.

Preparing Stock Solutions

Solvent / Mass / Concentration	1 mg	5 mg	10 mg
1 mM	3.1671 mL	15.8353 mL	31.6706 mL
5 mM	0.6334 mL	3.1671 mL	6.3341 mL
10 mM	0.3167 mL	1.5835 mL	3.1671 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 (7.92 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (7.92 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% (20% SBE-β-CD in saline) Solubility: ≥ 2.5 mg/mL (7.92 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (7.92 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 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: ≥ 2.5 mg/mL (7.92 mM); Clear solution 此方案可获得 ≥ 2.5 mg/mL (7.92 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]. Bojko A, et al. The effect of tyrphostins AG494 and AG1478 on the autocrine growth regulation of A549 and DU145 cells. Folia Histochem Cytobiol. 2012 Jul 5;50(2):186-95. [2]. Li W, et al. EGFR Inhibition Blocks Palmitic Acid-induced inflammation in cardiomyocytes and Prevents Hyperlipidemia-induced Cardiac Injury in Mice. Sci Rep. 2016 Apr 18;6:24580. [3]. Akhtar S, et al. Cationic Polyamidoamine Dendrimers as Modulators of EGFR Signaling In Vitro and In Vivo. PLoS One. 2015 Jul 13;10(7):e0132215.

实验参考：

Cell Assay	DU145 (HTB-81) and A549 (CCL-185) cells are seeded on 96-well plates at concentrations of 4×10^3 cells/well in MEM (DU145 cells) or DMEM (A549 cells), supplemented with 100 IU/mL penicillin and 100 μg/mL streptomycin in the presence of 10% FBS. Following 24 h of incubation, the culture medium is replaced with serum-free DMEM/F12 (1:1) supplemented with Transferrin (5 mg/mL), sodium selenite (2 ng/mL) and albumin (0.5 mg/mL) [DMEM/F12+]. After an additional 24 h of incubation (Day 0), the medium is replaced by serum-free DMEM/F12+ medium containing tyrosine kinase inhibitors: AG494, AG-1478 respectively in concentration ranges 1-20 μM and 0.1-8 μM. The incubation is continued for the next 24 h at 37°C in a humidified atmosphere. The modified crystal violet staining method (CV) and MTT assay are used to determine the influence of the tyrphostins on the proliferation of target cells. The absorbance is measured using a Tecan multiscan plate recorder[1].
	Mice[2] 28 C57BL/6 or ApoE-/- mice are randomly divided into four weight-matched groups. 7 mice are fed with standard animal low-fat diet containing 10 kcal.% fat, 20 kcal.% protein and 70 kcal.% carbohydrate serve as a normal control group (Control or ApoE-LF), while the remaining 21 mice are fed with high-fat diet containing 60 kcal.% fat, 20 kcal.% protein and 20 kcal.% carbohydrate for 16 weeks. Since 9^{th} week, AG-1478 or 542 are given daily by oral gavage at a dose of 10 mg/kg/day for the next 8 weeks. Mice in the Control and HFD groups are gavaged with vehicle (1% CMC-Na solution) only. At the day before the sacrifice of ApoE-/- mice, doppler analysis is performed to

Animal Administration	determine the pathologic cardiac hypertrophy. Rats[3] Male Wistar rats weighing about 300g are used in this study and divided into the following groups (N=5). Group 1: Non-diabetic (Control, C) animals, Group 2: C+PF (10mg/kg administered as a single intraperotoneal (i.p) injection) Group 3: C+SF (10mg/kg i.p); Group 4: C+AG-1478 (1 mg/kg i.p). Group 5: Rats bearing 4 weeks of diabetes (D) induced by a single i.p. injection of streptozotocin (55 mg/kg body weight); Group 6: D+PF (10 mg/kg i.p) Group 7: D+SF (10 mg/kg i.p) and Group 8: D+AG-1478 (1 mg/kg i.p). AG-1478 and dendrimer treatments are administered as single dose for 24h prior to sacrifice. Rat body weight and basal glucose levels are assessed before and after treatments just before sacrificing the animals. An automated blood glucose analyzer is used to assess blood glucose concentrations and rats with a blood glucose concentration above 250 mg/dL (approx. 14 mM) are declared diabetic as in previous studies.
References	[1]. Bojko A, et al. The effect of tyrphostins AG494 and AG1478 on the autocrine growth regulation of A549 and DU145 cells. Folia Histochem Cytobiol. 2012 Jul 5;50(2):186-95. [2]. Li W, et al. EGFR Inhibition Blocks Palmitic Acid-induced inflammation in cardiomyocytes and Prevents Hyperlipidemia-induced Cardiac Injury in Mice. Sci Rep. 2016 Apr 18;6:24580. [3]. Akhtar S, et al. Cationic Polyamidoamine Dendrimers as Modulators of EGFR Signaling In Vitro and In Vivo. PLoS One. 2015 Jul 13;10(7):e0132215.