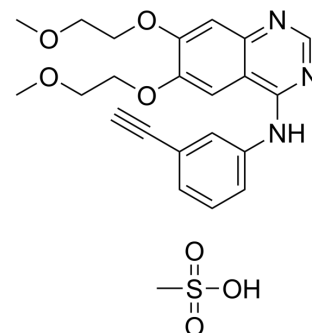


Erlotinib mesylate

Cat. No.:	HY-12008A
CAS No.:	248594-19-6
Molecular Formula:	C ₂₃ H ₂₇ N ₃ O ₇ S
Molecular Weight:	489.54
Target:	EGFR; Autophagy
Pathway:	JAK/STAT Signaling; Protein Tyrosine Kinase/RTK; Autophagy
Storage:	Please store the product under the recommended conditions in the Certificate of Analysis.



BIOLOGICAL ACTIVITY

Description	Erlotinib mesylate (CP-358774 mesylate) inhibits purified EGFR kinase with an IC ₅₀ of 2 nM. Erlotinib (mesylate) is a click chemistry reagent, it contains an Alkyne group and can undergo copper-catalyzed azide-alkyne cycloaddition (CuAAC) with molecules containing Azide groups.
IC ₅₀ & Target	EGFR 2 nM (IC ₅₀)
In Vitro	Erlotinib mesylate (CP-358774 mesylate) is also a potent inhibitor of the recombinant intracellular (kinase) domain of the EGFR, with an IC ₅₀ of 1 nM. The proliferation of DiFi cells is strongly inhibited by Erlotinib with an IC ₅₀ of 100 nM for an 8-day proliferation assay ^[1] . The combination of B-DIM and Erlotinib (2 μM) results in a significant inhibition of colony formation in BxPC-3 cells when compared with either agent alone. The combination of B-DIM and Erlotinib (2 μM) results in a significant induction of apoptosis only in BxPC-3 cells when compare with the apoptotic effect of either agent alone ^[2] . MCE has not independently confirmed the accuracy of these methods. They are for reference only.
In Vivo	There is a 1.49-fold statistically significant difference between AUC _{0-inf} after p.o. administration of Erlotinib (5 mg/kg) comparing Bcrp1/Mdr1a/1b ^{-/-} and WT mice (7,419±1,720 versus 4,957±1,735 ng*h/mL respectively, P=0.01) ^[3] . The administration of Erlotinib (10 mg/kg/day, or 20 mg/kg/day) to Bleomycin (BLM)-treated rats shows no exacerbation of lung injuries in indices such as macroscopic findings, lung weights, histopathological scores (lung lesion density and lung fibrosis score), and pulmonary hydroxyproline (HyP) level. The result suggests that Erlotinib does not have any exacerbating effects on lung injuries induced by BLM in rats ^[4] . MCE has not independently confirmed the accuracy of these methods. They are for reference only.

PROTOCOL

Kinase Assay ^[1]	The kinase reaction is performed in 50 μL of 50 mM HEPES (pH 7.3), containing 125 mM NaCl, 24 mM MgCl ₂ , 0.1 mM Na ₃ VO ₄ , 20 μM ATP, 1.6 μg/mL EGF, and 15 ng of EGFR, affinity purified from A431 cell membranes. The compound in DMSO is added to give a final DMSO concentration of 2.5%. Phosphorylation is initiated by addition of ATP and proceeded for 8 min at room temperature, with constant shaking. The kinase reaction is terminated by aspiration of the reaction mixture and is washed 4 times with wash buffer. Phosphorylated PGT is measured by 25 min of incubation with 50 μL per well HRP-conjugated PY54 antiphosphotyrosine antibody, diluted to 0.2 μg/mL in blocking buffer (3% BSA and 0.05% Tween 20 in PBS). Antibody is removed by aspiration, and the plate is washed 4 times with wash buffer. The colorimetric signal is developed by addition of
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	<p>TMB Microwell Peroxidase Substrate, 50 µL per well, and stopped by the addition of 0.09 M sulfuric acid, 50µL per well. Phosphotyrosine is estimated by measurement of absorbance at 450 nm. The signal for controls is typically 0.6-1.2 absorbance units, with essentially no back ground in wells without ALP, EGFR, or POT and is proportional to the time of incubation for 10 mm^[1].</p> <p>MCE has not independently confirmed the accuracy of these methods. They are for reference only.</p>
Cell Assay ^[2]	<p>To test the viability of cells treated with B-DIM, Erlotinib, or the combination, BxPC-3 and MIAPaCa cells are plated (3,000-5,000 per well) in a 96-well plate and incubated overnight at 37°C. A range of concentrations for both B-DIM (10-50 µM) and Erlotinib (1-5 µM) is initially tested. Based on the initial results, the concentration of B-DIM (20 µM) and Erlotinib (2 µM) are chosen for all assays. The effects of B-DIM (20 µM), Erlotinib (2 µM), and the combination on BxPC-3 and MIAPaCa cells are determined by the standard MTT assay after 72 h and is repeated three times. The color intensity is measured by a Tecan microplate fluorometer at 595 nm. DMSO-treated cells are considered to be the untreated control and assigned a value of 100%. In addition to the above assay, we have also done clonogenic assay for assessing the effects of treatment^[2].</p> <p>MCE has not independently confirmed the accuracy of these methods. They are for reference only.</p>
Animal Administration ^{[3][4]}	<p>Mice^[3]</p> <p>Bcrp1/Mdr1a/1b^{-/-} and WT mice are treated p.o. or i.p. with 5 mg/kg Erlotinib. The i.p. administration is chosen assuming good drug absorption and complete bioavailability. Sampling is done from the tip of the lateral tail vein in three series. During the first series, whole blood samples are collected at 15 min and 0.5, 1.5, 5, and 10 h after injection. Based on the results of this initial group, the sampling times of the two subsequent series are adapted to 5 and 15 min and 0.5, 1.5, 4, and 8 h after injection. After collection, the blood samples are immediately centrifuged and plasma is stored at -20°C until high-performance liquid chromatographic analysis took place.</p> <p>Rats^[4]</p> <p>Seven-week-old male Crl:CD (SD) rats (244-297 g) are used. The animals are treated with Erlotinib (10 mg/kg and 20 mg/kg) orally by gavage.</p> <p>MCE has not independently confirmed the accuracy of these methods. They are for reference only.</p>

CUSTOMER VALIDATION

- Cell Res. 2020 Oct;30(10):833-853.
- Nat Immunol. 2022 Feb;23(2):251-261.
- Bioact Mater. 2021 Nov 4;13:312-323.
- Sci Transl Med. 2018 Jul 18;10(450):eaaq1093.
- Nat Commun. 2023 Apr 24;14(1):2342.

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REFERENCES

- [1]. Moyer JD, et al. Induction of apoptosis and cell cycle arrest by CP-358,774, an inhibitor of epidermal growth factor receptor tyrosine kinase. Cancer Res. 1997 Nov 1;57(21):4838-48.
- [2]. Ali S, et al. Apoptosis-inducing effect of erlotinib is potentiated by 3,3'-diindolylmethane in vitro and in vivo using an orthotopic model of pancreatic cancer. Mol Cancer Ther, 2008, 7(6), 1708-1719.
- [3]. Marchetti S, et al. Effect of the ATP-binding cassette drug transporters ABCB1, ABCG2, and ABCC2 on erlotinib hydrochloride (Tarceva) disposition in in vitro and in vivo pharmacokinetic studies employing Bcrp1^{-/-}/Mdr1a/1b^{-/-} (triple-knockout) and wild-type mice. Mol Cancer Ther. 2008 Aug;7(8):2280-7.
- [4]. Adachi K, et al. Effects of erlotinib on lung injury induced by intratracheal administration of bleomycin (BLM) in rats. J Toxicol Sci. 2010 Aug;35(4):503-14.

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