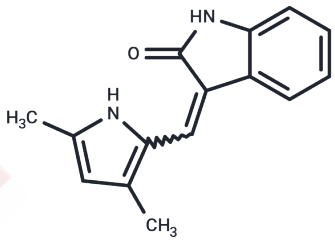
Data Sheet (Cat.No.T2064)

TargetMol

Semaxinib

Chemical Properties



CAS No. : 204005-46-9

Formula: C15H14N2O

Molecular Weight: 238.28

Storage: Powder: -20°C for 3 years | In solvent: -80°C for 1 year
Actual storage temperature shall be subject to the COA.

Biological Description

Description	Semaxinib (SU5416) is a potent and selective VEGFR2 inhibitor (IC50: 1.23 μM), exhibiting a 20-fold greater selectivity for VEGFR2 over PDGFRβ, with no activity against InsR, EGFR, and FGFR. Semaxinib reversibly inhibits ATP binding to the tyrosine kinase domain of VEGFR2, potentially inhibiting VEGF-stimulated endothelial cell migration and proliferation, thereby reducing tumor microvasculature.
Targets(IC50)	VEGFR
In vitro	SU5416 inhibits VEGF-driven mitogenesis in a dose-dependent manner with an IC50 of 0.04±0.02 μM (n=3). In contrast, SU5416 blocked FGF-dependent mitogenesis of HUVECs with an IC50 of 50 μM (n=10). The selective activity of SU5416 on Flk-1 is supported by the fact that testing of SU5416 using NIH 3T3 cells overexpressing either the EGF or insulin receptors indicated a complete lack of activity (IC50>100 μM). This observation is confirmed by immunoblotting after ligand stimulation. An IC50 of 20.26±5.2 μM (n=7), which is about 20-fold less in potency on PDGF-dependent autophosphorylation, is observed when SU5416 is tested in NIH 3T3 cells overexpressing the human PDGF receptor β[1].
In vivo	Administering SU5416 daily (i.p., 3 mg/kg/day) effectively inhibits C6 tumor growth in the colon, achieving a growth inhibition of 62% by day 16 (P=0.001), comparable to the 54% inhibition in the hindflank by day 18 (P=0.001). This demonstrates SU5416's potential to suppress tumor development in various locations, possibly due to differences in preexisting vasculature. Further, a higher dose of SU5416 (25 mg/kg) significantly reduces tumor growth rates, with tumors being only 8% the size of those in control animals by day 22, primarily through a substantial decrease in the area covered by newly formed glioma microvasculature in treated animals, suggesting a minimized initial tumor vascularization.
Kinase Assay	Solubilized membranes from 3T3 Flk-1 cells are added to polystyrene ELISA plates that has been precoated with a monoclonal antibody that recognizes Flk-1. After an overnight incubation with lysate at 4°C, serial dilutions of SU5416 are added to the immunolocalized receptor. To induce autophosphorylation of the receptor, various concentrations of ATP are added to the ELISA plate wells containing serially diluted solutions of SU5416. The autophosphorylation is allowed to proceed for 60 min at room temperature and then stopped with EDTA. The amount of phosphotyrosine present on the Flk-1 receptors in the individual wells is determined by incubating the immunolocalized receptor with a biotinylated monoclonal antibody directed against

Kinase Assay	phosphotyrosine. After removal of the unbound anti-phosphotyrosine antibody, avidin-conjugated horseradish peroxidase H is added to the wells. A stabilized form of 3,3′,5,5′ -tetramethyl benzidine dihydrochloride and Water2 is added to the wells. The color readout of the assay is allowed to develop for 30 min, and the reaction is stopped with H2SO4. Parallel biochemical kinase assays are performed to measure autophosphorylation on EGFR and fibroblast growth factor receptor[1].
Cell Research	SU5416 is dissolved in DMSO and stored,and then diluted with appropriate media (DMSO<0.5%) before use[1] 3T3Her2 and 488 g2M2 are NIH3T3 fibroblast cell lines engineered to overexpress Her2 and to express human PDGF-BB and human PDGF receptor β.Both cell lines are cultured in DMEM supplemented with 2% CS and 2 mM L-glutamine.C6,Calu 6,A375,A431,and SF767T are plated in their respective growth medium at 2×103 cells/100 μL/well in 96-well,flat-bottomed plates.SU5416 is serially diluted in media containing DMSO (<0.5%) and added to cultures of tumor cells 1 day after the initiation of culture.Cell growth is measured after 96 h using the sulforhodamine B method.IC50s are calculated by curve fitting using four-parameter analysis[1].

Solubility Information

Solubility	DMSO: 10 mg/mL (41.97 mM),Sonication is recommended. (< 1 mg/ml refers to the product slightly soluble or insoluble)
In vivo Formulation	10% DMSO+40% PEG300+5% Tween 80+45% Saline: 1 mg/mL (4.2 mM),Suspension. *Please add the solvents sequentially, clarifying the solution as much as possible before adding the next one. Dissolve by heating and/or sonication if necessary. Working solution is recommended to be prepared and used immediately. The formulation provided above is for reference purposes only. In vivo formulations may vary and should be modified based on specific experimental conditions.*

Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	4.1967 mL	20.9837 mL	41.9674 mL
5 mM	0.8393 mL	4.1967 mL	8.3935 mL
10 mM	0.4197 mL	2.0984 mL	4.1967 mL
50 mM	0.0839 mL	0.4197 mL	0.8393 mL

Please select the appropriate solvent to prepare the stock solution, according to the solubility of the product in different solvents. Please use it as soon as possible.

Note: The dilution table applies only to solid products. For liquid products, please calculate the stock solution based on the stated concentration and/or density.

Reference

Fong TA, et al. SU5416 is a potent and selective inhibitor of the vascular endothelial growth factor receptor (Flk-1/KDR) that inhibits tyrosine kinase catalysis, tumor vascularization, and growth of multiple tumor types. Cancer Res, 1999, 59(1), 99-106.

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Huang X, Zhu J, Jiang Y, et al. SU5416 attenuated lipopolysaccharide-induced acute lung injury in mice by modulating properties of vascular endothelial cells. Drug Design, Development and Therapy. 2019, 13: 1763

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