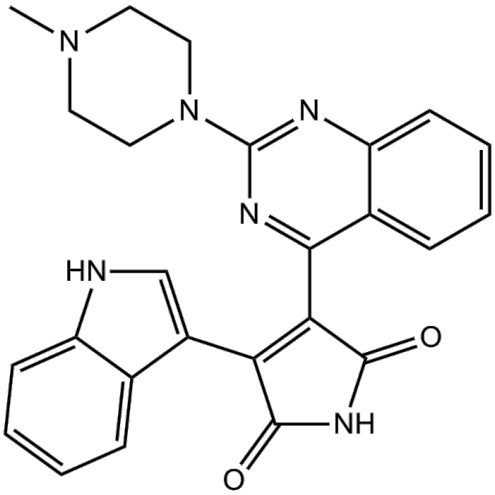
Product Data Sheet

Product Name: Sotrastaurin (AEB071)
Cat. No.: GC15520

Chemical Properties

Cas. No.	425637-18-9		
Chemical Name	3-(1H-indol-3-yl)-4-[2-(4-methylpiperazin-1-yl)quinazolin-4-yl]pyrrole-2,5-dione		
SMILES	CN1CCN(CC1)C2=NC3=CC=CC=C3C(=N2)C4=C(C(=O)NC4=O)C5=CNC6=CC=CC=C65		
Formula	$C_{25}H_{22}N_6O_2$	M.Wt	438.48
Solubility	≥ 21.9mg/mL in DMSO	Storage	Store at -20°C
General tips	For obtaining a higher solubility , please warm the tube at 37 °C and shake it in the ultrasonic bath for a while.Stock solution can be stored below -20°C for several months.		
Shipping Condition	Evaluation sample solution : ship with blue ice All other available size: ship with RT , or blue ice upon request.		

Structure



Protocol

Cell experiment [1]:

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Cell lines	SUDHL-4 and OCI-LY8 DLBCL cells
Preparation Method	The OCI-LY8 cell line was cultured in Iscove's modified Dulbecco's medium supplemented with 10% fetal bovine serum and 1% penicillin–streptomycin, and the SUDHL-4 cell line was cultured in RPMI 1640 containing 10% FBS and 1% penicillin–streptomycin. Cells were incubated in a humidified atmosphere at 37°C in 5% CO_2. Sotrastaurin (40mM) was dissolved in dimethyl sulfoxide (DMSO) and stored at -20°C. SUDHL-4 and OCI-LY8 cells (0.1ml) were seeded into wells of 96- well plates at a density of 2×10^5cells/ml, treated with various concentrations of Sotrastaurin (5, 10, 15, 20, 25, 30, or 40μM) for 24, 48 or 72h in a 37°C incubator. At the end of the incubation, CCK-8 assay was used for cell proliferation detection. Cell apoptosis was measured using the Annexin-V/PI apoptosis detection kit. Cell cycle was assessed by flow cytometry. Protein was extracted for western blot analysis.
Reaction Conditions	5-40μM; 24-72h
Applications	Treatment of SUDHL-4 and OCI-LY8 DLBCL cells with Sotrastaurin blocked cell proliferation, induced apoptosis, and triggered G1 arrest accompanied by down-regulation of MCT-1, cyclin D1, p-ERK and p-AKT and up-regulation of p53 and cleaved caspases-3/8/9.

Animal experiment [2]:

Animal models	C57BL/6 mice

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Preparation Method	A total of seventy-two 8-week-old C57BL/6 mice, 36 females and 36 males, were raised to the age of three months. Mice were then randomly assigned to one of six groups according to sex (n=6 mice each), including two groups of Sham-operated (intragastric administration of PBS only), two groups of medial meniscus (DMM) surgery and two groups of DMM+Sotrastaurin treatment (intragastric administration of 10mg/kg Sotrastaurin). For DMM surgery, mice were anaesthetized with chloral hydrate and then subjected to surgical transection of the medial meniscotibial ligament of the right knee. Sham mice received the same surgical procedure without the transection of the medial meniscotibial ligament. Two days post-surgery, mice were intragastrically administered with Sotrastaurin (treatment groups) or equivalent volume of PBS (Sham and DMM groups only) every other day for 4 or 8 weeks. No adverse reactions or animal fatalaties were recorded during the treatment period, and all mice exhibited normal behavioural activity. Mice were killed at the respective endpoints and the knee joints were excised, cleaned of soft tissues, fixed in 4% PFA, and then processed for micro-computed tomography (micro-CT) and histological assessments.
Dosage form	10mg/kg; i.g.; every other day for 4 or 8 weeks
Applications	Sotrastaurin significantly reduced subchondral bone loss and articular cartilage degeneration, decreased $TRAP^+$ osteoclast numbers, and improved OARSI scores in DMM-induced OA mice.

Caution: Product has not been fully validated for medical applications. For research use only.
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References:
[1] Chang G, Zheng J, Xiao W, et al. PKC inhibition of sotrastaurin has antitumor activity in diffuse large B-cell lymphoma via regulating the expression of MCT-1. *Acta Biochim Biophys Sin (Shanghai)*. 2018;50(4):399-407.
[2] Pang C, Wen L, Qin H, Zhu B, Lu X, Luo S. Sotrastaurin, a PKC inhibitor, attenuates RANKL-induced bone resorption and attenuates osteochondral pathologies associated with the development of OA. *J Cell Mol Med*. 2020;24(15):8452-8465.

Background

Sotrastaurin (AEB071) is a potent and orally-active pan-protein kinase C (PKC) inhibitor, with Ki values of 0.22nM, 0.64nM, 0.95nM, 1.8nM, 2.1nM and 3.2nM for PKCθ, PKCβ, PKCα, PKCη, PKCδ and PKCε, respectively[1]. PKC is a family of Ca^{2+}- or diacylglycerol-dependent serine/threonine kinases that regulate cell proliferation, differentiation, apoptosis and immune responses by phosphorylating diverse downstream substrates[2]. Sotrastaurin is usually used in the research of autoimmune diseases and tumors[3].

In vitro, Sotrastaurin (1μM; 7 days) blocks PKC-driven proliferation and IL-17A/IFN-γ secretion while preserving $Foxp3^{+}CD25^{+}$ suppressive Treg phenotype in human $CD4^{+}$ T cells[4]. Treatment of SUDHL-4 and OCI-LY8 DLBCL cells with Sotrastaurin (5-40μM; 48h) blocked cell proliferation, induced apoptosis, and triggered G1 arrest accompanied by down-regulation of MCT-1, cyclin D1, p-ERK and p-AKT and up-regulation of p53 and cleaved caspases-3/8/9[5].

In vivo, Sotrastaurin (10mg/kg; i.g.; every other day for 4–8 weeks) significantly reduced subchondral bone loss and articular cartilage degeneration, decreased $TRAP^{+}$ osteoclast numbers, and improved OARSI scores in DMM-induced OA mice[6]. Sotrastaurin (30mg/kg; i.g.; twice daily for 3 days) reversed 30-h cold ischemia-induced hepatocellular necrosis and apoptosis, reduced serum ALT, and elevated elevating 14-day survival in syngeneic Sprague–Dawley rat orthotopic liver transplantation model[7].

References:
[1] Evenou JP, Wagner J, Zenke G, et al. The potent protein kinase C-selective inhibitor AEB071

(sotrastaurin) represents a new class of immunosuppressive agents affecting early T-cell activation. *J Pharmacol Exp Ther*. 2009;330(3):792-801.
[2] Deka SJ, Trivedi V. Potentials of PKC in Cancer Progression and Anticancer Drug Development. *Curr Drug Discov Technol*. 2019;16(2):135-147.
[3] Sommerer C, Zeier M. AEB071--a promising immunosuppressive agent. *Clin Transplant*. 2009;23 Suppl 21:15-18.
[4] He X, Koenen HJPM, Smeets RL, et al. Targeting PKC in human T cells using sotrastaurin (AEB071) preserves regulatory T cells and prevents IL-17 production. *J Invest Dermatol*. 2014;134(4):975-983.
[5] Chang G, Zheng J, Xiao W, et al. PKC inhibition of sotrastaurin has antitumor activity in diffuse large B-cell lymphoma via regulating the expression of MCT-1. *Acta Biochim Biophys Sin (Shanghai)*. 2018;50(4):399-407.
[6] Pang C, Wen L, Qin H, Zhu B, Lu X, Luo S. Sotrastaurin, a PKC inhibitor, attenuates RANKL-induced bone resorption and attenuates osteochondral pathologies associated with the development of OA. *J Cell Mol Med*. 2020;24(15):8452-8465.
[7] Kamo N, Shen XD, Ke B, Busuttil RW, Kupiec-Weglinski JW. Sotrastaurin, a protein kinase C inhibitor, ameliorates ischemia and reperfusion injury in rat orthotopic liver transplantation. *Am J Transplant*. 2011;11(11):2499-2507.