Product Data Sheet

Product Name: 20-HEDE (WIT 002)
Cat. No.: GC33412

Chemical Properties

Cas. No.	240427-90-1		
SMILES	O=C(O)CCCC/C=C\CCCCCCC/C=C\CCCCO		
Formula	$C_{20}H_{36}O_3$	M.Wt	324.5
Solubility	Soluble in DMSO	Storage	Store at -20°C
General tips	For obtaining a higher solubility , please warm the tube at 37 ℃ and shake it in the ultrasonic bath for a while.Stock solution can be stored below -20℃ 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	20-HEDE (WIT 002)		

Protocol

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Cell experiment:

Human NSCLC cell lines (e.g A549) are seeded onto the upper well of the chamber. Subsequently, serum-free medium with HET0016 (10 μM), WIT002 (10 μM), WIT003 (0.01, 0.1, 1 μM), or ethanol as a control is added to the upper chamber, while the lower well is filled to the top (500 μL) with RPMI-1640 containing 5% fetal calf serum (FCS) as a chemoattractant. Cells are allowed to migrate for 5 h. Cells that have invaded to the bottom surface of the filter are counted with an ocular micrometer in a blinded manner, counting a minimum of 10 high-powered fields (HPF) [2]. Human renal cell adenocarcinoma lines (RPTC) 786-O or 769-P cells are plated and next day (0 hrs) transferred to serum-free medium containing either EGF or ET-1. Cells are exposed either to 10 μM WIT002 or vehicle. Cell counting is performed at the day of transfer to serum free medium (0 hrs) and 24, 48 and 72 hrs thereafter. Medium is changed to fresh containing mitogens and drugs every 24 hrs. Data presented are characteristic experiment from at least two separate experiments, each performed in triplicate[3].

Animal experiment:

Mice[3]Experiments are carried out on 6-week immunodefficient athymic nude mice weighing 20-26 g. Animals are acclimated for 1 week prior to injection with renal cell carcinoma. Immediately before each implantation, the cells are trypsinized, counted and resuspended in 10% serum containing RPMI media. The concentration of cells is adjusted to 40 millions/mL and 4 million cells/animal are injected subcutaneously. The cells are allowed to grow for 7-15 days until the size of the tumors reach approximately 0.1 cm3. The mice then receive daily subcutaneous injection (s.c.) injections of WIT 002 (10 mg/kg/day in 200 μL) in an isotonic $NaPO_4$ buffer (pH 9.0) or vehicle (0.1 M $NaPO_4$ solution pH 9.0). The diameter of the tumor is measured on every 3-4 days for 2 weeks using precision calibers. At the end of the experiment the mice are euthanized with CO_2 and the tumors were excised to confirm the diameter measurements[3].

References:

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[1]. Ming Yua . et al. Effects of a 20-HETE antagonist and agonists on cerebral vascular tone. Eur J Pharmacol. 2004 Feb 23;486(3):297-306.
[2]. Wei Yu, et al. Cytochrome P450 ω-hydroxylase promotes angiogenesis and metastasis by upregulation of VEGF and MMP-9 in non-small cell lung cancer. Cancer Chemother Pharmacol. 2011 Sep; 68(3): 619-29.
[3]. Anna Alexanian, et al. Down-regulation of 20-HETE Synthesis and Signaling Inhibits Renal

Adenocarcinoma Cell Proliferation and Tumor Growth. Anticancer Res. 2009 October ; 29(10): 3819-3824.

Background

WIT 002 is an antagonist of 20-hydroxyeicosatetraenoic acid (20-HETE).

ω-hydroxylation activity toward arachidonic acid is high in A549 cells, thus, A549 cells are treated with HET0016 or WIT 002 in the invasion assays, and both of them significantly decrease invasion[2]. WIT 002 inhibits proliferation of 786-O and 769-P renal adenocarcinoma cells, but HET0016 and WIT 002 fail to inhibit proliferation of normal renal epithelial cells RPTC[2][3].

The effect of the 20-HETE antagonist, WIT 002 on the growth of 786-O clear cell renal carcinoma is assessed in ectopic mouse model of renal tumor. The growth of tumors is significantly suppressed by WIT 002 administered daily to athymic nude mice implanted subcutaneously with cells 786-O. Tumor growth is inhibited by 84%±128%. It is of note that in these experiments WIT 002 treatment start only after the tumor is seeded for 7-14 days and is relatively large 0.1 cm. Thus, WIT 002 is effective at arresting the growth of a fairly advanced tumor[3].

[1]. Ming Yua . et al. Effects of a 20-HETE antagonist and agonists on cerebral vascular tone. Eur J Pharmacol. 2004 Feb 23;486(3):297-306. [2]. Wei Yu, et al. Cytochrome P450 ω-hydroxylase promotes angiogenesis and metastasis by upregulation of VEGF and MMP-9 in non-small cell lung cancer. Cancer Chemother Pharmacol. 2011 Sep; 68(3): 619-29. [3]. Anna Alexanian, et al. Down-regulation of 20-HETE Synthesis and Signaling Inhibits Renal Adenocarcinoma Cell Proliferation and Tumor Growth. Anticancer Res. 2009 October ; 29(10): 3819-3824.

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