

Product Data Sheet

Product Name: GW4869

Cat. No.: GC19186

Chemical Properties

Cas. No. 6823-69-4

Chemical Name 3,3'-(1,4-phenylene)bis[N-[4-(4,5-dihydro-1H-imidazol-2-yl)phenyl]-dihydrochloride-2-pro penamide

SMILES O=C(NC1=CC=C(C2=NCCN2)C=C1)/C=C/C3=CC=C(/C=C/C(NC4=CC=C(C5=NCCN5)C=C4)=O)C=C3.[H]Cl.[H]Cl

Formula $C_{30}H_{30}Cl_2N_6O_2$

M.Wt 577.5

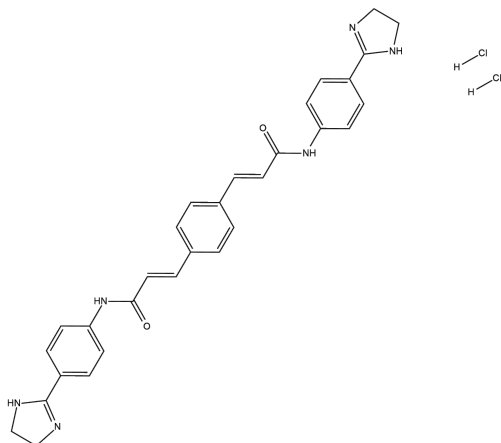
Solubility Water : < 0.1 mg/mL (insoluble);
DMSO : 2 mg/mL

Storage Store at -20°C, ready to use

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 MCF7 human breast cancer cells

Preparation method GW-4869 was routinely stored at -80°C as a 1.5 mm stock suspension in DMSO. Right before use, the suspension was solubilized by the addition of 5% methane sulfonic acid (MSA) (2.5 μl of 5% MSA in sterile double-distilled H_2O were added to 50 μl of GW4869 stock suspension; therefore, the concentration of the GW-4869 stock solution at the time of the experiments was 1.43 mm). The suspension was mixed and warmed up at 37°C until clear.

Reaction
Conditions 30 min

Applications GW4869 is a noncompetitive neutral sphingomyelinase (N-SMase) inhibitor with an IC_{50} of 1 μM . GW4869 (10 μM) partially inhibited TNF-induced sphingomyelin (SM) hydrolysis, and 20 μM of the compound was protected completely from the loss of SM. The action of GW4869 occurred downstream of the drop in glutathione, which was shown previously to occur upstream of the activation of N-SMase. GW4869 does not interfere with other key TNF-mediated signaling effects. GW4869 was able, in a dose-dependent manner, to significantly protect from cell death as measured by nuclear condensation, caspase activation, PARP degradation, and trypan blue uptake.

Animal experiment [2]:

Animal models Male wild-type C57BL/6 mice

Dosage form GW 4869, dissolved in DMSO (0.005%), was intraperitoneally (i.p.) injected at one dose of 2.5 $\mu\text{g/g}$ before sham or CLP surgery.

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Applications

Pre-treatment with GW 4869 significantly impaired release of both exosomes and pro-inflammatory cytokines (TNF- α , IL-1 β , IL-6) in RAW264.7 macrophages. WT mice pretreated with GW4869 displayed lower amounts of exosomes and pro-inflammatory cytokines in the serum than control PBS-injected mice. Accordingly, GW4869 treatment diminished the sepsis-induced cardiac inflammation, attenuated myocardial depression and prolonged survival.

References:

- [1]. Luberto, Chiara, et al. "Inhibition of tumor necrosis factor-induced cell death in MCF7 by a novel inhibitor of neutral sphingomyelinase." *Journal of Biological Chemistry* 277.43 (2002): 41128-41139.
- [2]. Essandoh, Kobina, et al. "Blockade of exosome generation with GW4869 dampens the sepsis-induced inflammation and cardiac dysfunction." *Biochimica et Biophysica Acta (BBA)-Molecular Basis of Disease* 1852.11 (2015): 2362-2371.

Background

GW4869 is a noncompetitive neutral sphingomyelinase inhibitor with an IC₅₀ of 1 μ M. GW4869 (10 μ M) partially inhibits TNF-induced sphingomyelin (SM) hydrolysis, and 20 μ M of the compound is protected completely from the loss of SM. The addition of 10-20 μ M GW4869 completely inhibits the initial accumulation of ceramide, whereas this effect is partially lost at later time points (24 h). The action of GW4869 occurs downstream of the drop in glutathione. GW4869 is able, in a dose-dependent manner, to significantly protect from cell death. These protective effects of gw4869 are accompanied by significant inhibition of cytochrome c release from mitochondria and caspase 9 activation, therefore localizing N-SMase activation upstream of mitochondrial dysfunction[1].

Pre-treatment with GW4869 significantly impairs the release of both exosomes and pro-inflammatory cytokines (TNF- α , IL-1 β , IL-6) in RAW264.7 macrophages. At 12 h after LPS treatment or CLP surgery, WT mice pretreated with GW4869 display lower amounts of exosomes and pro-inflammatory cytokines in the serum than control PBS-injected mice. Accordingly, GW4869 treatment diminishes sepsis-induced cardiac inflammation, attenuates myocardial depression, and prolongs survival[2].

References:

- [1]. Luberto C, et al. Inhibition of tumor necrosis factor-induced cell death in MCF7 by a novel

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inhibitor of neutral sphingomyelinase. J Biol Chem. 2002 Oct 25;277(43):41128-39.

[2]. Essandoh K, et al. Blockade of exosome generation with GW4869 dampens the sepsis-induced inflammation and cardiac dysfunction. Biochim Biophys Acta. 2015 Nov;1852(11):2362-71.

[3]. Nakamura H, et al. Sphingomyelin Regulates the Activity of Secretory Phospholipase A2 in the Plasma Membrane. J Cell Biochem. 2015 Sep;116(9):1898-907.

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