

CLI-095

TLR4 Signaling Inhibitor

Catalog # tlr1-cli95

For research use only

Version # 09D03-MM

PRODUCT INFORMATION

Content:

- 1 mg CLI-095

Storage and stability:

- CLI-095 is provided as a solid and shipped at room temperature. Store at -20°C. Solid product is stable 1 year at -20°C when properly stored.
- Upon resuspension, CLI-095 should be aliquoted and stored at -20°C. Avoid repeated freeze-thaw cycles. Resuspended product is stable 3 months at -20°C when properly stored.

DESCRIPTION

CLI-095, also known as TAK-242, is a novel cyclohexene derivative that suppresses specifically TLR4 signaling, inhibiting the production of NO and pro-inflammatory cytokines¹. It acts by blocking the signaling mediated by the intracellular domain of TLR4, but not the extracellular domain. It potently suppresses both ligand-dependent and -independent signaling of TLR4².

1. Li M. *et al.*, 2006. A Novel Cyclohexene Derivative, Ethyl (6R)-6-[N-(2-Chloro-4-fluorophenyl)sulfamoyl]cyclohex-1-ene-1-carboxylate (TAK-242), Selectively Inhibits Toll-Like Receptor 4-Mediated Cytokine Production through Suppression of Intracellular Signaling. *Mol. Pharmacol.*, 69:1288-1295. 2. Kawamoto T. *et al.*, 2008. TAK-242 selectively suppresses Toll-like receptor 4-signaling mediated by the intracellular domain. *Eur J Pharmacol.* 584(1):40-8.

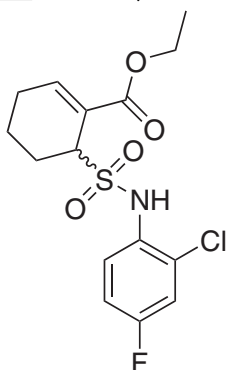
CHEMICAL PROPERTIES

Formula: C₁₅H₁₇ClFNO₄S

Molecular weight: 361.82

Solubility: DMSO (1 mg/ml)

Working concentration: 50 nM- 5 μM



METHODS

Preparation of CLI-095 stock solution (100 μg/ml)

1. Add 1 ml DMSO to 1 mg CLI-095 vial (1 mg/ml).
2. Vortex until complete solubilization.
3. Take 100 μl of solubilized CLI-095 (1 mg/ml), add 900 μl sterile culture media to obtain a stock solution of 100 μg/ml.
4. Aliquote and store stock solution at -20°C.

TLR4 signaling using HEK-Blue™ -4 cells:

HEK-Blue™-4 cells are engineered HEK293 cells that stably express an NF-κB-inducible SEAP (secreted embryonic alkaline phosphatase) reporter gene, and the TLR4, MD2 and CD14 genes. LPS-induced TLR4 activation in HEK-Blue™-4 cells induces the activation of NF-κB and the subsequent release of SEAP in the supernatant, which is readily detected by using QUANTI-Blue™, a colorimetric enzyme assay which turns blue in the presence of SEAP.

- 1- Prepare a cell suspension at 150,000 cells/ml.
- 2- Add 160 μl of cell suspension per well of a 96-well plate.
- 3- Add 20 μl of CLI-095 at the appropriate concentration to reach a final concentration of 1 μg/ml (3 μM).
- 4- Incubate for 6 hours at 37°C.
- 5- Add 20 μl LPS-EK Ultrapure (10 ng/ml final concentration).
- 6- Incubate overnight at 37°C.
- 7- Add 20 μl supernatant to 180 μl QUANTI-Blue™ in a 96-well plate.
- 8- Incubate 1-3 hours at 37°C
- 9- Assess SEAP levels with the naked eye or spectrophotometrically by reading the OD at 655 nm.

RELATED PRODUCTS

Product	Catalog Code
HEK-Blue™ LPS Detection Kit	rep-lps
HEK-Blue™ Selection	hb-sel
Normocin™	ant-nr-1
HEK-Blue™ Detection (2 pouches)	hb-det1
HEK-Blue™-4 Cells	hb4-cells
LPS-EK Ultrapure	tlr1-eklps
QUANTI-Blue™	rep-qbl

TECHNICAL SUPPORT

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