

Pam3CSK4

Synthetic triacylated lipoprotein - TLR1/2 ligand

Catalog # tlr1-pms

For research use only

Version # 12A10-MM

PRODUCT INFORMATION

Content:

- 1 mg Pam3CSK4
- 1.5 ml endotoxin-free water

Storage and stability:

- Pam3CSK4 is provided lyophilized and shipped at room temperature. Store at 4°C. Lyophilized product is stable 1 year when properly stored.
- Upon resuspension, prepare aliquots of Pam3CSK4 and store at 4°C for short term storage or -20°C for long storage. Resuspended product is stable 1 month at 4°C and 6 months at -20°C when properly stored.

DESCRIPTION

Bacterial lipoproteins are a family of proinflammatory cell wall components found in both Gram positive and Gram negative bacteria. The stimulatory activity of bacterial lipoproteins resides in their acylated amino terminus. Pam3CSK4 is a synthetic tripalmitoylated lipopeptide that mimicks the acylated amino terminus of bacterial lipoproteins. Pam3CysSerLys4 (Pam3CSK4) is a potent activator of the proinflammatory transcription factor NF-κB¹. Recognition of Pam3CSK4 is mediated by TLR2 which cooperates with TLR1 through their cytoplasmic domain to induce the signaling cascade leading to the activation of NF-κB³.

1. Aliprantis AO *et al.*, 1999. Cell activation and apoptosis by bacterial lipoproteins through toll-like receptor-2. *Science*.285(5428):736-9. 2. Ozinsky A. *et al.*, 2000. The repertoire for pattern recognition of pathogens by the innate immune system is defined by cooperation between toll-like receptors. *PNAS*. 97(25):13766-71. 3. Schindler U. & Baichwal VR., 1994. Three NF-κB binding sites in the human E-selectin gene required for maximal tumor necrosis factor alpha-induced expression. *Mol Cell Biol*, 14(9):5820-5831.

CHEMICAL PROPERTIES

Chemical name: N-Palmitoyl-S-[2,3-bis(palmitoyloxy)-(2RS)-propyl]-[R]-cysteinyll-[S]-seryll-[S]-lysyll-[S]-lysyll-[S]-lysyll-[S]-lysine

CAS number: 112208-00-1

Formula: C₈₁H₁₅₆N₁₀O₁₃S

Molecular weight: 1509.6

Endotoxin level: <0.001 EU/μg

METHODS

Preparation of stock solution (1 mg/ml)

Stimulation of TLR2 can be achieved with 1-300 ng/ml Pam3CSK4.

- Add 1 ml endotoxin-free water (provided) and vortex for 30 seconds or until complete solubilization.
- Prepare further dilutions by adding the appropriate amount of endotoxin-free water.

Pam3CSK4 stimulation

- Transfect your cell line with an NF-κB-inducible reporter plasmid, i.e. a plasmid carrying a reporter gene, such as SEAP or luciferase, under the control of an NF-κB-inducible ELAM-1 (E-selectin) promoter³.

Note: InvivoGen provides pNiFty, a family of NF-κB-inducible reporter plasmids that can be transfected transiently (pNiFty) or stably (pNiFty2). pNiFty plasmids are available either with the SEAP or luciferase reporter genes (see Related Products). If your cell line does not naturally express TLR2 and TLR1, cotransfect with a plasmid co-expressing both TLRs, such as pDUO-TLR1/TLR2 (see Related Products).

Note: Alternatively, evaluate TLR2 activation using reporter cells, such as InvivoGen's HEK-Blue™ hTLR2 cells which express the human TLR2 and SEAP reporter genes. NF-κB production in these cells can be easily quantified using a SEAP detection medium, such as QUANTI-Blue™ or HEK-Blue™ Detection.

- Twenty-four to forty-eight hours after transfection, stimulate cells with 1- 300 ng/ml Pam3CSK4 for 6 hours to 24 hours.
- Determine Pam3CSK4 stimulation on TLR2 by assessing reporter gene expression using the appropriate detection system.

RELATED PRODUCTS

Product	Catalog Code
HEK-Blue™ hTLR2 cells	hkb-htlr2
pNiFty2-Luc (Zeo®)	pnifty2-luc
pNiFty2-SEAP (Zeo®)	pnifty2-seap
pDUO-hTLR1/2 (human genes)	pduo-htlr12
pDUO-mTLR1/2 (mouse genes)	pduo-mtlr12

TECHNICAL SUPPORT

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