Pam3CSK4

Synthetic triacylated lipoprotein; TLR2/TLR1 ligand

Catalog code: tlrl-pms, tlrl-pms-5 https://www.invivogen.com/pam3csk4

For research use only

Version 25A30-MM

PRODUCT INFORMATION

Contents: Pam3CSK4 is available in two quantities:

- tlrl-pms: 1 mg
- tlrl-pms-5: 5 mg (5 x 1 mg)
- endotoxin-free water; 1.5 ml with tlrl-pms and 5 x 1.5 ml with tlrl-pms-5 **Storage and stability**
- Pam3CSK4 is shipped at room temperature. Upon receipt, store at 4°C.
- Upon resuspension, prepare aliquots of Pam3CSK4 and store at 4°C for 1 month or at -20°C for 6 months at -20°C.

Quality control

- Purity: ≥95% (UHPLC)
- The TLR2 activity has been tested using HEK-Blue™ TLR2 cells.
- The absence of endotoxins has been confirmed using HEK-Blue™ TLR4 cells.

DESCRIPTION

Pam3CSK4 is a synthetic triacylated lipopeptide (LP) and a TLR2/TLR1 ligand. It is a potent activator of the pro-inflammatory transcription factor NF- κ B^{1,2}. Pam3CSK4 mimics the acylated amino terminus of bacterial LPs. Bacterial LPs are a family of pro-inflammatory cell wall components found in both Gram-positive and Gram-negative bacteria. The stimulatory activity of these LPs resides in their acylated amino terminus. The TLR2/TLR1 heterodimer recognizes LPs with three fatty acids, a structural characteristic of bacterial LPs. 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- κ B³.

1. Brandt K.J. et al., 2013. TLR2 Ligands Induce NF-кB activation from endosomal compartments of human monocytes PLoS One. 8(12):e80743. 2. Aliprantis A.O. et al., 1999. Cell activation and apoptosis by bacterial lipoproteins through toll-like receptor-2. Science 285(5428):736-9. 3. 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.

CHEMICAL PROPERTIES

Chemical name: N-Palmitoyl-S-[2,3-bis(palmitoyloxy)-(2RS)-propyl]-

[R]-cysteinyl-[S]-seryl-[S]-lysyl-[S]-lysyl-[S]-lysine

Solubility: 2 mg/ml in water CAS number: 112208-01-2

Formula: $C_{81}H_{156}N_{10}O_{13}S \bullet 3TFA$ Molecular weight: 1852.33~g/mol Structure: $C_{15}H_{31} + O_{15}H_{31} + O_{1$

MFTHODS

Preparation of stock solution (1 mg/ml)

- 1. Add 1 ml of endotoxin-free water (provided) to 1 mg vial.
- 2. Vortex until completely dissolved. Use immediately or store at 4°C or at -20°C. Prepare aliquots before freezing.
- 3. Prepare further dilutions by adding the appropriate amount of endotoxin-free water.

Working concentration: 0.1-10 ng/ml

TLR2 stimulation using Pam3CSK4

Pam3CSK4 can be used to activate TLR2 in HEK-Blue™ TLR2 cells that were designed to study TLR2 stimulation by monitoring NF-кB activation. Stimulation of HEK-Blue™ TLR2 cells with a TLR2 agonist activates NF-кB which induces the production of SEAP (secreted embryonic alkaline phosphatase). Levels of SEAP can be easily determined using a SEAP detection medium, such as HEK-Blue™ Detection.

For more information visit: https://www.invivogen.com/hek-blue-tlr2.

- 1. Dispense 20 μ l of Pam3CSK4 (0.1-10 ng/ml final concentration) per well of a 96-well plate.
- 2. Prepare a suspension of HEK-Blue™ TLR2 cells in HEK-Blue™ Detection medium.
- 3. Immediately add 180 μl of the cell suspension to each Pam3CSK4-containing well.
- 4. Incubate the plate at 37°C in a CO₂ incubator for 16-24 hours.
- 5. Determine SEAP levels using a spectrophotometer at 620-655 nm.

RELATED PRODUCTS

Product	Description	Cat. Code
FSL-1	TLR2/6 ligand	tlrl-fsl
HEK-Blue™ Detection	SEAP detection medium	hb-det2
HEK-Blue™ hTLR2 cells	Human TLR2 reporter cells	hkb-htlr2
HEK-Blue™ mTLR2 cells	Murine TLR2 reporter cells	hkb-mtlr2
Pam2CSK4	TLR2/6 ligand	tlrl-pm2s-1
Pam3CSK4 Biotin	Biotinylated Pam3CSK4	tlrl-bpms



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