**FSL-1** Synthetic diacylated lipoprotein; TLR2/TLR6 ligand

Catalog code: tlrl-fsl

https://www.invivogen.com/fsl1

### For research use only

Version 23K13-MM

## PRODUCT INFORMATION

#### Contents

- 100 µg lipopeptide FSL-1
- 1.5 ml endotoxin-free water

### Storage and stability

- $\bullet\,$  FSL-1 is provided lyophilized and shipped at room temperature. Upon receipt, store at 4°C.
- Upon resuspension, store at 4°C. Resuspended product is stable for 6 months at 4°C.

Note: We do not recommend freezing the resuspended product as it may result in reduced TLR2/TLR6 activity.

### Quality control

- The TLR2 activity has been tested using HEK-Blue<sup>™</sup> TLR2 cells.

## DESCRIPTION

FSL-1 (Pam2CGDPKHPKSF) is a synthetic lipoprotein (LP) that represents the N-terminal part of the 44-kDa lipoprotein LP44 of *Mycoplasma salivarium*<sup>1</sup>. The framework structure of FSL-1 is the same as that of MALP-2, a *Mycoplasma fermentans* derived lipopeptide (LP), but they differ in the amino acid sequence and length of the peptide portion<sup>2</sup>. Mycoplasmal LP, such as FSL-1 and MALP-2, contain a lipolyated N-terminal diacylated cysteine residue, whereas bacterial LP contain a triacylated one. This structural difference plays a crucial role in the initial recognition of microbial LP by the host innate immune system.

Mycoplasmal LP, such as FSL-1, are recognized by TLR2 and TLR6, whereas bacterial LP and Pam3CSK4, a synthetic LP, are recognized by TLR2 and TLR1<sup>3</sup>. FSL-1 stimulation induces a MyD88-dependent signaling cascade leading to AP-1 and NF- $\kappa$ B activation and the subsequent cytokine production<sup>3,4</sup>.

**1.** Shibata Ki. *et al.*, 2000. The N-terminal lipopeptide of a 44-kDa membrane-bound lipoprotein of Mycoplasma salivarium is responsible for the expression of intercellular adhesion molecule-1 on the cell surface of normal human gingival fibroblasts. J. Immunol. 165:6538–44. **2.** Okusawa T. *et al.*, 2004. Relationship between Structures and Biological Activities of Mycoplasmal Diacylated Lipopeptides and Their Recognition by Toll-Like Receptors 2 and 6. Infect Immun. 72(3): 1657-65. **3.** Takeuchi O. *et al.*, 2001. Discrimination of bacterial lipoproteins by Toll-like receptor 6. Int Immunol. 13(7):933-40. **4.** Ahmad R. *et al.*, 2014. FSL-1 induces MMP-9 production through TLR-2 and NF-κB/AP-1 signaling pathways in monocytic THP-1 cells. Cell Physiol Biochem. 34(3):929-42.

# CHEMICAL PROPERTIES

 $\begin{array}{c} \mbox{Chemical name: } (S,R)-(2,3-bispalmitoyloxypropyl)-Cys-Gly-Asp-Pro-Lys-His-Pro-Lys-Ser-Phe} \\ \mbox{Solubility: } 10 mg/ml in water \\ \mbox{CAS number: } 322455-70-9 \\ \mbox{Formula: } C_{84}H_{140}N_{14}O_{18}S \\ \mbox{Molecular weight: } 1666.2 \ g/mol \\ \mbox{Structure: } \\ \mbox{CH}_{3}(CH_{2})_{14} \\ \mbox{O} \\ \mbox{O} \\ \mbox{CH}_{3}(CH_{2})_{14} \\ \mbox{O} \ \mbox{O} \\ \mbox{O} \ \mbox{O} \\ \mbox{O} \ \mbox{O} \ \mbox{O} \\ \mbox{O} \ \mb$ 

### **METHODS**

### Preparation of stock solution (100 µg/ml)

- Add 1 ml endotoxin-free water (provided) and vortex until completely dissolved.

- Prepare further dilutions by adding the appropriate amount of endotoxin-free water.

Working concentration: 10 pg/ml-100 ng/ml

### TLR2 stimulation using FSL-1

FSL-1 can be used to activate TLR2 in HEK-Blue<sup>™</sup> TLR2 cells that were designed to study TLR2 stimulation by monitoring NF-κB activation. Stimulation of HEK-Blue<sup>™</sup> TLR2 cells with a TLR2 agonist activates NF-κB 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<sup>™</sup> Detection. For more information visit: <u>https://www.invivogen.com/hek-blue-tlr2</u>.

1. Dispense 20  $\mu l$  of FSL-1 (10 pg/ml-100 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  $\mu I$  of the cell suspension to each FSL-1-containing well.

4. Incubate the plate at 37°C in a CO<sub>2</sub> incubator for 6-24 hours.

5. Determine SEAP levels using a spectrophotometer at 620-655 nm.

# **RELATED PRODUCTS**

Product	Description	Cat. Code
HEK-Blue <sup>™</sup> Detection	SEAP detection medium	h b - d e t 2
HEK-Blue <sup>™</sup> hTLR2 cells	Human TLR2 reporter cells	hkb-htlr2
HEK-Blue <sup>™</sup> mTLR2 cells	Murine TLR2 reporter cells	hkb-mtlr2
Pam2CSK4	TLR2/6 ligand	tlrl-pm2s-1
Pam3CSK4	TLR2/TLR1 ligand	tlrl-pms

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