

# Synthetic diacylated lipoprotein; TLR2/TLR6 ligand

Catalog code: tlrl-fsl <a href="https://www.invivogen.com/fsl1">https://www.invivogen.com/fsl1</a>

# For research use only

Version 22D13-NJ

#### 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  $4^{\circ}\text{C}.$
- $\bullet$  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.
- The absence of endotoxins has been confirmed using HEK-Blue™ TLR4 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 $^3$ . FSL-1 stimulation induces a MyD88-dependent signaling cascade leading to AP-1 and NF- $\kappa$ B activation and the subsequent cytokine production $^3$ .

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

Chemical name: (S,R)-(2,3-bispalmitoyloxypropyl)-Cys-Gly-Asp-Pro-

Lys-His-Pro-Lys-Ser-Phe
Solubility: 10 mg/ml in water
CAS number: 322455-70-9
Formula: C<sub>84</sub>H<sub>140</sub>N<sub>14</sub>O<sub>18</sub>S
Molecular weight: 1666.2 g/mol

#### **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: 1-100 ng/ml

## TLR2 stimulation using FSL-1

FSL-1 can be used to activate TLR2 in HEK-Blue™ TLR2 cells that were designed to study TLR2 stimulation by monitoring NF-κB activation. Stimulation of HEK-Blue™ 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™ Detection.

For more information visit: <a href="https://www.invivogen.com/hek-blue-tlr2">https://www.invivogen.com/hek-blue-tlr2</a>.

- 1. Dispense 20  $\mu$ l of FSL-1 (1-100 ng/ml final concentration) per well of a 96-well plate.
- 2. Prepare a suspension of HEK-Blue<sup>™</sup> TLR2 cells in HEK-Blue<sup>™</sup> Detection medium.
- 3. Immediately add 180  $\mu l$  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	hb-det2
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



InvivoGen USA (Toll-Free): 888-457-5873 InvivoGen USA (International): +1 (858) 457-5873 InvivoGen Europe: +33 (0) 5-62-71-69-39 InvivoGen Asia: +852 3622-3480

E-mail: info@invivogen.com

