LPS-PG

Standard lipopolysaccharide from Porphyromonas gingivalis; TLR2 and TLR4 ligand

Catalog code: tlrl-pglps

https://www.invivogen.com/lps-pg

For research use only

Version 23G12-MM

PRODUCT INFORMATION

Contents

- 1 mg lipopolysaccharide from Porphyromonas gingivalis (LPS-PG)

- 1.5 ml endotoxin-free water

Storage and stability

- LPS-PG is shipped at room temperature. Upon receipt, store product at -20 $^{\circ}\text{C}.$

- Upon resuspension, prepare aliquots and store at -20 $^\circ \rm C.$ Resuspended product is stable for 6 months when properly stored. Avoid repeated freeze-thaw cycles.

Quality control

Activation of TLR4 has been confirmed using HEK-Blue™ TLR4 cells.
The presence of other bacterial components (e.g. lipoproteins) has been assessed using HEK-Blue™ TLR2 cells.

DESCRIPTION

LPS-PG is a semi-rough (sr)-form of lipopolysaccharide (LPS) from the Gram-negative bacteria *Porphyromonas gingivalis*. LPS-PG is an important virulence factor in the mechanisms of periodontal disease. LPS is the principal component of Gram-negative bacteria that activates the innate immune system.

LPS recognition is predominantly mediated by TLR4¹. The TLR4 response to LPS-PG is dependent on the presence of key accessory molecules, CD14 and MD2². LPS-PG presents a unique and heterogenous chemical structure, which differs from traditionally recognized enteric bacterium-derived LPS. The fact that LPS-PG exhibits activity in C3H/HeJ mice, which are deficient for TLR4, led to a common belief that this LPS is a TLR2 ligand^{3.4}. However, structural and functional studies of LPS-PG have revealed that it activates cells through TLR4. The TLR2 activity of LPS-PG is ascribed to a contaminant lipoprotein⁵. LPS-PG is a standard LPS preparation that activates TLR2 and TLR4.

1. Poltorak A. *et al.*, **1998**. Defective LPS signaling in C3H/HeJ and C57BL/10ScCr mice: mutations in TLR4 gene. Science, 282:2085-8. **2.** Darveau R.P. *et al.*, **2004**. Porphyromonas gingivalis lipopolysaccharide contains multiple lipid A species that functionally interact with both toll-like receptors 2 and 4. Infect Immun. 72(9):5041-51. **3.** Kirikae T. *et al.*, **1999**. Lipopolysaccharides (LPS) of oral black-pigmented bacteria induce tumor necrosis factor production by LPS-refractory C3H/HeJ macrophages in a way different from that of Salmonella LPS. Infect Immun. 67(4):1736-42. **4.** Hirschfeld M. *et al.*, **2001**. Signaling by toll-like receptor 2 and 4 agonists results in differential gene expression in murine macrophages. Infect Immun. 69(3):1477-82. **5.** Ogawa T. *et al.*, **2007**. Chemical structure and immunobiological activity of Porphyromonas gingivalis lipid A. Front Biosci. 12:3795-812.

PRODUCT PROPERTIES

Species: Porphyromonas gingivalis Specificity: TLR2 and TLR4 agonist Solubility: 1 mg/ml in water

Working concentrations:

- TLR4 activity: 100 ng-10 µg/ml
- TLR2 activity: 10 ng/ml 10 µg/ml

METHODS

Preparation of stock solution (1 mg/ml)

- Add 1 ml of endotoxin-free water (provided) and homogenize. *Note: LPS-PG stock solution may appear cloudy.*

TLR4 activation using LPS-PG

LPS-PG can be used to activate TLR4 in HEK-Blue[™] TLR4 cells, that were designed to study TLR4 stimulation by monitoring NF-κB activation. Stimulation of HEK-Blue[™] TLR4 cells with a TLR4 agonist activates NF-κB which induces the production of SEAP (secreted embryonic alkaline phosphatase). Levels of SEAP can be easily determined using HEK-Blue[™] Detection, a cell culture medium that allows the detection of SEAP as the reporter protein is secreted by the cells.

For more information visit: <u>https://www.invivogen.com/hek-blue-tlr4</u>.

- Add 20 μl of LPS-PG at various concentrations (100 ng-10 $\mu g/ml)$ in a well of a 96-well plate.

- Prepare a cell suspension ~140,000 cells per ml in HEK-Blue™ Detection.

- Add 180 μl of the cell suspension (~25,000 cells) to each LPS-PG-containing well.

- Incubate the plate for 6-24 h at 37°C, 5% CO₂.

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

RELATED PRODUCTS

Product	Description	Cat. Code
HEK-Blue™ Detection	SEAP Detection reagent	hb-det2
HEK-Blue™ hTLR4 Cells	Human TLR4 reporter cells	hkb-htlr4
HEK-Blue™ mTLR4 Cells	Murine TLR4 reporter cells	hkb-mtlr4
LPS-EB Ultrapure	LPS from <i>E. coli</i> 0111:B4	tlrl-3pelps
MPLA-SM*	MPLA from <i>S. minnesota</i>	tlrl-mpla2
MPLAs	Synthetic MPLA	tlrl-mpls

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