LPS-EB

Standard lipopolysaccharide from *E. coli* 0111:B4 strain; TLR4 and TLR2 ligand

Catalog code: tlrl-eblps

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

For research use only

Version 23F20-MM

PRODUCT INFORMATION

Contents

- 5 mg LPS-EB (lipopolysaccharide from *E. coli* 0111:B4)
- 1.5 ml endotoxin-free water

Storage and stability

- LPS-EB is shipped at room temperature. Upon receipt, store product at -20 °C.
- Resuspended LPS-EB may be stored for 1 month at 4 °C or for 6 months when aliquoted and stored at -20 °C. 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

Lipopolysaccharide (LPS) is the principal component of Gram-negative bacteria that activates the innate immune system through its recognition by Toll-like receptor 4 (TLR4). This leads to a signaling cascade that ultimately results in the activation of NF-κB and the production of proinflammatory cytokines[1]. LPS-EB is a preparation of smooth (s)-form LPS purified from the Gram-negative *E. coli* 0111:B4, a pathogenic serotype of *E. coli* known to cause significant gastric disease[2,3].

LPS-EB is a standard lipopolysaccharide (LPS) preparation extracted by a phenol-water mixture. LPS-EB contains other bacterial components, such as lipoproteins, and therefore stimulates both TLR4 and TLR2.

1. Kuzmich N.N. *et al.*, 2017. TLR4 Signaling pathway modulators as potential therapeutics in inflammation and sepsis. Vaccines (Basel) 5(4):34. **2. Coleman W.G., Jr. *et al.*, 1977.** Genetic analysis of Escherichia coli O111:B4, a strain of medical and biochemical interest. J Bacteriol 130:656-60. **3. Viljanen M.K. *et al.*, 1990.** Outbreak of diarrhea due to *Escherichia coli* O111:B4 in schoolchildren and adults: association of Vi antigen-like reactivity. Lancet 336:831-4.

PRODUCT PROPERTIES

Species: *Escherichia coli*
Specificity: TLR4 and TLR2 agonist
Working concentration: 10 ng-10 µg/ml
Solubility: 5 mg/ml in water

METHODS

Preparation of stock solution (5 mg/ml)

1. Add 1 ml of endotoxin-free water (provided) and homogenize.

Notes:

- *This product can be reconstituted by injecting water through the rubber cap using a needle and syringe.*
- *LPS-EB stock solution may appear cloudy.*

TLR4 activation using LPS-EB

LPS-EB 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: https://www.invivogen.com/hek-blue-htlr4.

1. Add 20 µl of LPS-EB at 10 ng-10 µg/ml in a well of a 96-well plate.
2. Prepare a cell suspension ~140,000 cells per ml in HEK-Blue™ Detection.
3. Add 180 µl of the cell suspension (~25,000 cells) to each LPS-EB-containing well.
4. Incubate the plate for 6-24 h at 37°C, 5% CO_2.
5. 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 *E. coli 0111:B4*	tlrl-3pelps
LPS-SM Ultrapure	LPS from *S. minnesota*	tlrl-smlps
MPLAs	Synthetic MPLA	tlrl-mpls

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