

# MPLAs

## Synthetic Monophosphoryl Lipid A; TLR4 ligand

Catalog code: tlr1-mpls

<https://www.invivogen.com/mplas>

For research use only

Version 23G13-MM

## PRODUCT INFORMATION

### Contents

- 1 mg Synthetic Monophosphoryl Lipid A (MPLAs)

### Storage and stability

- MPLAs is provided as a clear, lipidic film and shipped at room temperature. Upon receipt, store at -20°C.
- Upon resuspension, prepare aliquots of MPLAs and store at -20°C. Resuspended product is stable for 6 months when properly stored. Avoid repeated freeze-thaw cycles.

### Quality control

- Biological activity has been tested using HEK-Blue™ TLR4 cells.
- The absence of bacterial contamination (e.g. lipoproteins) has been confirmed using HEK-Blue™ TLR2 cells.

## BACKGROUND

MPLA is a low-toxicity derivative of lipopolysaccharide (LPS) that retains the immunologically active lipid A portion of the parent molecule. Both LPS and MPLA are TLR4 agonists, but they signal through different adaptors, MyD88 and TRIF, respectively<sup>1,2</sup>. The reduced toxicity of MPLA is attributed to the preferential recruitment of TRIF upon TLR4 activation, resulting in decreased induction of inflammatory cytokines. MPLA is widely used as a vaccine adjuvant due to its potent immunomodulatory properties and low inflammatory toxicity<sup>1,2</sup>.

## DESCRIPTION

Synthetic lipid A from *E. coli*, serotype R515 (MPLAs), is a monophosphoryl lipid A compound produced by chemical synthesis. MPLAs is a potent inducer of TLR4 but does not activate TLR2 even at high concentrations reflecting its high purity. It is structurally similar to natural MPLA except that it contains 6 fatty acyl groups while MPLA purified from bacteria contains a mixture of 5, 6, and 7 acyl lipid A. The number of fatty acids is a major determinant of the immunogenicity of LPS<sup>3</sup>. The most active form of lipid A contains 6 fatty acyl groups and is found in pathogenic bacteria such as *E. coli* and *Salmonella* species. This product may be useful for immunomodulatory studies as it is a pure monophosphoryl lipid A containing 6 fatty acyl groups.

1. Sastry M. *et al.*, 2017. Adjuvants and the vaccine response to the DS-Cav1-stabilized fusion glycoprotein of respiratory syncytial virus. *PLoS One*. 12(10):e0186854. 2. Cui W. *et al.*, 2014. TLR4 ligands lipopolysaccharide and monophosphoryl lipid A differentially regulate effector and memory CD8+ T Cell differentiation. *J Immunol*. 192(9):4221-32. 3. Steimle A. *et al.*, 2017. Structure and function: Lipid A modifications in commensals and pathogens. *Int J Med Microbiol*. 306(5):290-301.

## CHEMICAL PROPERTIES

### Structure:

CAS Number: 1246298-63-4

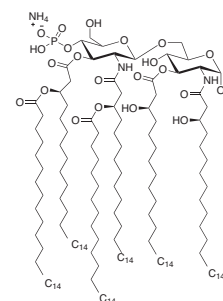
Formula: C<sub>96</sub>H<sub>184</sub>N<sub>3</sub>O<sub>22</sub>P

Molecular weight: 1763.47

Endotoxin level: 1 x 10<sup>6</sup> EU/mg

Solubility: 1 mg/ml DMSO

Working concentration: 300 pg-100 ng/ml



## METHODS

### Preparation of stock suspension (1 mg/ml)

- Add 1 ml of DMSO and vortex until completely resuspended, then sonicate.
- Use immediately or store aliquots at -20°C.
- Prepare dilutions with water.

### Notes:

- The suspension may appear to contain floating fine particles. Difficulties may be encountered for resuspension at higher concentrations.
- Alternatively, MPLAs can be resuspended in DMSO containing 0.2% triethylamine.

### TLR4 activation using MPLAs

MPLAs can be used to activate TLR4 in HEK-Blue™ TLR4 cells. These cells express TLR4, MD-2 and CD14 co-receptor genes, and an NF-κB-inducible SEAP (secreted embryonic alkaline phosphatase) reporter gene. Levels of SEAP can be easily determined using a SEAP detection medium, such as QUANTI-Blue™.

For more information visit: <https://www.invivogen.com/hek-blue-trl4>.

- Distribute 20 µl of MPLAs (300 pg-100 ng/ml final concentration) in a well of a 96-well plate.
- Add 180 µl of HEK-Blue™ TLR4 cell suspension per well.
- Incubate the plate for 16-24 h at 37°C, 5% CO<sub>2</sub>.
- Collect 20 µl of supernatant and add to a well of a 96-well plate containing 180 µl of QUANTI-Blue™.
- Incubate the plate at 37°C for 1-3 h.
- Determine SEAP levels using a spectrophotometer at 620-655 nm.

## RELATED PRODUCTS

Product	Catalog Code
CRX-527	tlr1-crx527
HEK-Blue™ hTLR4 Cells (human TLR4)	hkb-htrl4
HEK-Blue™ mTLR4 Cells (mouse TLR4)	hkb-mtrl4
LPS-EB Ultrapure ( <i>E. coli</i> O111:B4)	tlr1-3pelps
LPS-EK Ultrapure ( <i>E. coli</i> K12)	tlr1-peklps
MPLA-SM*	tlr1-mpla2

## TECHNICAL SUPPORT

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