Poly(I:C) LMW

Low Molecular Weight

Synthetic analog of dsRNA; TLR3 ligand

Catalog code: tlrl-picw, tlrl-picw-250 https://www.invivogen.com/polyic-lmw

For research use only

Version 23K09-MM

PRODUCT INFORMATION

Contents

• Poly(I:C) LMW is provided lyophilized and is available in two sizes:

tlrl-picw: 25 mg tlrl-picw-250: 250 mg

• Sterile endotoxin-free physiological water (NaCl 0.9%)

10 ml with catalog code tlrl-picw

2 x 25 ml with catalog code tlrl-picw-250

Storage and stability

- Product is shipped at room temperature. Upon receipt, store at 4°C.
- Upon resuspension, prepare aliquots of Poly(I:C) LMW and store at 4°C or at -20°C. Resuspended product is stable for 1 month at 4°C and 1 year at -20°C. Avoid repeated freeze-thaw cycles.

Quality control:

- Absorbance spectrum
- TLR3 activity has been verified using HEK-Blue™ TLR3 cells
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using cellular assays.

BACKGROUND

Polyinosinic-polycytidylic acid (poly(I:C)) is a synthetic analog of double stranded RNA (dsRNA), a molecular pattern associated with viral infection. Both natural and synthetic dsRNAs are known to induce type I interferons (IFN) and other cytokines production. Poly(I:C) is recognized by Toll-like receptor 3 (TLR3)1.2. Upon poly(I:C) recognition, TLR3 activates the transcription factor interferon regulatory factor 3 (IRF3), through the adapter protein Toll-IL-1 receptor (TIR) domaincontaining adapter inducing IFN- β (TRIF, also known as TICAM-1)³. Activation of IRF3 leads to the production of type I IFNs, especially IFN-β. A second pathway involves the recruitment of TNF receptorassociated factor 6 (TRAF6) or receptor interacting protein 1 (RIP1), with the subsequent activation of the transcription factors NF- κ B and AP-14. Activation of this pathway triggers the production of inflammatory cytokines and chemokines such as TNF- α , IL-6 and CXCL10. Poly(I:C) is also recognized by the cytosolic RNA helicases retinoic acid-inducible protein I (RIG-I) and melanoma differentiationassociate gene 5 (MDA-5)5.

1. Alexopoulou L. et al., 2001. Recognition of double-stranded RNA and activation of NF-kB by Toll-like receptor 3. Nature, 413(6857):732-8. 2. Matsumoto M. et al., 2002. Establishment of a monoclonal antibody against human Toll-like receptor 3 that blocks double-stranded RNA-mediated signaling. Biochem Biophys Res Commun, 293(5):1364-9. 3. Yamamoto M. et al., 2003. Role of Adaptor TRIF in the MyD88-Independent Toll-Like Receptor Signaling Pathway. Science 301: 640. 4. Kawai T & Akira S., 2008. Tollikereceptor and RIG-I-like receptor signaling. Ann N Y Acad Sci. 1143:1-20. 5. Kato H. et al., 2006. Differential roles of MDA5 and RIG-I helicases in the recognition of RNA viruses. Nature. 441(7089):101-5.

PRODUCT DESCRIPTION

Polyinosinic-polycytidylic acid (usually abbreviated as poly(I:C) or poly(rI):poly(rC)) is a synthetic analog of double-stranded RNA (dsRNA), a molecular pattern associated with viral infection. Poly(I:C) activates the antiviral pattern recognition receptors TLR3, RIG-I/MDA5 and PKR, thereby inducing signaling via multiple inflammatory pathways, including NF- κ B and IRF. Low Molecular Weight Poly(I:C) comprises short strands of inosine poly(I) homopolymer annealed to strands of cytidine poly(C) homopolymer.

The average size of Poly(I:C) LMW is 0.2 kb to 1 kb.

METHODS

Preparation of sterile stock solution (20 mg/ml)

Stimulation of TLR3 can be achieved with 30 ng-10 μ g/ml Poly(I:C) LMW. - Add 1.25 ml of the endotoxin-free physiological water to the 25 mg Poly(I:C) LMW vial or 12.5 ml to the 250 mg Poly(I:C) LMW vial to obtain a solution at 20 mg/ml.

- Mix the solution by pipetting up and down until complete solubilization.

TLR3 activation of TLR3 with Poly(I:C) LMW

Poly(I:C)LMW can be used to stimulate hTLR3 in HEK-Blue hTLR3 cells. These cells are designed for studying the stimulation of hTLR3 by monitoring the activation of NF- κ B. Stimulation with a TLR3 ligand activates NF- κ B and AP-1 which induces the production of SEAP. Levels of SEAP can be easily determined with QUANTI-Blue Solution, a detection medium that turns purple/blue in the presence of alkaline phosphatase.

- 1. Prepare a HEK-Blue™ hTLR3 cell suspension (250,000 cells/ml).
- 2. Add 180 µl of the cell suspension per well of a 96-well plate.
- 3. Stimulate cells with 30 ng-10 µg/ml Poly(I:C) LMW for 6 to 24 hours.
- 4. Determine poly(I:C) stimulation on TLR3 by assessing reporter gene expression using QUANTI-Blue[™] Solution or HEK-Blue[™] detection. Note: InvivoGen provides also a high molecular weight poly(I:C) HMW (see "Related Products"), with an average size of 1.5-8 kb that may activate the immune system differently.

RELATED PRODUCTS

| Product | Catalog Code |
|--|--|
| HEK-Blue [®] hTLR3 QUANTI-Blue [®] Solution Poly(I:C) HMW Poly(A:U) | hkb-htlr3 rep-qbs tlrl-pic tlrl-pau |
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