

# Poly(I:C) LMW

## Low Molecular Weight

Synthetic analog of dsRNA; TLR3 ligand

Catalog code: tlr1-picw, tlr1-picw-250

<https://www.invivogen.com/polyic-lmw>

For research use only

Version 19D27-MM

## PRODUCT INFORMATION

### Contents

- Poly(I:C) LMW is provided lyophilized and is available in two sizes:
  - tlr1-picw: 25 mg
  - tlr1-picw-250: 250 mg
- Sterile endotoxin-free physiological water (NaCl 0.9%)
  - 10 ml with catalog code tlr1-picw
  - 2 x 25 ml with catalog code tlr1-picw-250

### Storage and stability

- Product is shipped at room temperature. Upon receipt, store at 4°C.
- Lyophilized product is stable for 1 year at 4°C when properly stored.
- 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.

## DESCRIPTION

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)<sup>1,2</sup>. Upon poly(I:C) recognition, TLR3 activates the transcription factor interferon regulatory factor 3 (IRF3), through the adapter protein Toll-IL-1 receptor (TIR) domain-containing adapter inducing IFN- $\beta$  (TRIF, also known as TICAM-1)<sup>3</sup>. Activation of IRF3 leads to the production of type I IFNs, especially IFN- $\beta$ . A second pathway involves the recruitment of TNF receptor-associated factor 6 (TRAF6) or receptor interacting protein 1 (RIP1), with the subsequent activation of the transcription factors NF- $\kappa$ B and AP-1<sup>4</sup>. Activation of this pathway triggers the production of inflammatory cytokines and chemokines such as TNF- $\alpha$ , IL-6 and CXCL10. Poly(I:C) is also recognized by the cytosolic RNA helicases retinoic acid-inducible protein 1 (RIG-I) and melanoma differentiation-associate gene 5 (MDA-5)<sup>5</sup>.

1. Alexopoulou L. et al., 2001. Recognition of double-stranded RNA and activation of NF- $\kappa$ B 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. Toll-like receptor 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.

## METHODS

### Preparation of sterile stock solution (20 mg/ml)

Stimulation of TLR3 can be achieved with 30 ng-10  $\mu$ g/ml Poly(I:C) LMW.  
- Add 1.25 ml of the endotoxin-free physiological water provided 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- $\kappa$ B. Stimulation with a TLR3 ligand activates NF- $\kappa$ B and AP-1 which induces the production of SEAP. Levels of SEAP can be easily determined with QUANTI-Blue™ (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  $\mu$ l of the cell suspension per well of a 96-well plate.
3. Stimulate cells with 30 ng-10  $\mu$ g/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™ 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.*

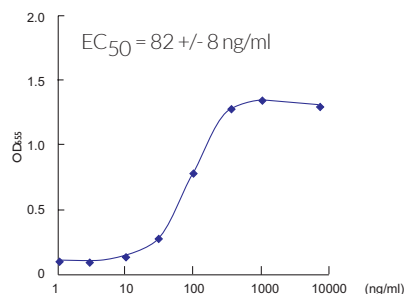


Figure 1. A typical stimulation curve. HEK-Blue™ hTLR3 cells were stimulated with increasing concentrations of Poly(I:C) LMW. After 18h incubation, NF- $\kappa$ B-induced SEAP activity was assessed using QUANTI-Blue™.

## RELATED PRODUCTS

Product	Catalog Code
HEK-Blue hTLR3	hkb-htlr3
QUANTI-Blue™	rep-qb1
Poly(I:C) HMW	tlr1-pic
Poly(A:U)	tlr1-pau

### TECHNICAL SUPPORT

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