# Loxoribine

## Guanosine analog; a TLR7 ligand

Catalog # tlrl-lox

#### For research use only

Version # 16E12-MM

#### PRODUCT INFORMATION

- 50 mg (147 µmol) Loxoribine
- 25 ml sterile endotoxin-free water

#### Storage:

- Loxoribine is provided as a white to off-white solid and shipped at room temperature. Store at 4°C. Loxoribine is stable for 1 year when properly stored.
- Upon resuspension, prepare aliquots of loxoribine and store at -20 °C. Resuspended product is stable for 6 months when properly stored. Avoid repeated freeze-thaw cycles.

#### **QUALITY CONTROL**

- Purity: >94% (UHPLC)
- The TLR7 activity has been confirmed using HEK-Blue™ TLR7
- The absence of TLR8 activity has been confirmed using HEK-Blue™ hTLR8 cells.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue™ TLR2 and HEK-Blue™ TLR4 cells.

#### DESCRIPTION

Loxoribine is a guanosine analog derivatized at position N<sup>7</sup> and C<sup>8</sup>. This nucleoside is a very powerful stimulator of the immune system but until recently the mechanism responsible for this immunostimulatory activity was unknown<sup>1</sup>. It appears that similarly to imiquimod, a small synthetic antiviral molecule, loxoribine activates the innate immune system through Toll-like receptor 7 and that this activation requires endosomal maturation<sup>2</sup>. Loxoribine recognition is restricted to TLR7.

1. Gorden KB. et al., 2005. Synthetic TLR agonists reveal functional differences between human TLR7 and TLR8. J Immunol. 174(3):1259-68. 2. Heil F. et al., 2003. The Toll-like receptor 7 (TLR7)-specific stimulus loxoribine uncovers a strong relationship within the TLR7, 8 and 9 subfamily. Eur J Immunol. 33(11):2987-97.

#### CHEMICAL PROPERTIES

**Synonym:** 7-allyl-7,8-dihydro-8-oxo-guanosine

Formula: C<sub>13</sub>H<sub>17</sub>N<sub>5</sub>O<sub>6</sub> Molecular weight: 339.3 **CAS number:** 121288-39-9 **Structure:** 

#### **METHODS**

### Preparation of stock solution (100 mM)

Stimulation of TLR7 can be achieved with Loxoribine at a concentration of 1 mM.

- 1. Resuspend Loxoribine in 1.47 ml DMSO to 50 mg of Loxoribine.
- 2. Further dilutions can be prepared using the endotoxin-free water (provided) or aqueous buffers.

#### TLR7 stimulation using Loxoribine

Loxoribine can be used to stimulate TLR7 in HEK-Blue™ TLR7 cells. HEK-Blue™ TLR7 cells stably express an NF-κB-inducible secreted embryonic alkaline phosphatase (SEAP) and overexpress the TLR7 gene. For more information visit: www.invivogen.com

- 1. Prepare a HEK-Blue™ TLR7 cell suspension according to the data
- 2. Add 20 µl of Loxoribine at a final concentration of 1 mM per well of a 96-well plate.
- 3. Add 180 µl of cell suspension per well.
- 2. Incubate cells and Loxoribine for 6-24 h at 37 °C, 5% CO2.
- 3. Determine TLR7 stimulation with Loxoribine by assessing cytokine expression using an ELISA, or SEAP expression using a SEAP detection medium, such as QUANTI-Blue™ or HEK-Blue™ Detection.

#### RELATED PRODUCTS

Product	Catalog Code
CL264 (TLR7 ligand) HEK-Blue™ hTLR7 cells HEK-Blue™ mTLR7 cells HEK-Blue™ Detection (SEAP detection medium) pUNO1-hTLR7 (human gene) pUNO1-mTLR7 (mouse gene)	tlrl-c264e hkb-htlr7 hkb-mtlr7 hb-det2 puno1-htlr7 puno1-mtlr7
QUANTI-Blue <sup>™</sup> (SEAP detection medium)	rep-qb1



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