# **Nigericin**

# NLRP3 inflammasome Inducer

Catalog code: tlrl-nig, tlrl-nig-5 https://www.invivogen.com/nigericin

# For research use only

Version 21D28-MM

# PRODUCT INFORMATION

#### Contents

- Nigericin sodium salt is available in two quantities:
  - 10 mg: tlrl-nig
  - 50 mg: tlrl-nig-5

#### Storage and stability

- Nigericin is shipped at room temperature. Upon receipt, store at -20  $^{\circ}\text{C}.$
- Upon resuspension, Nigericin can be stored at 4°C or at -20°C. Resuspended product is stable for 1 month at 4°C and for 1 year at -20°C when properly stored. Avoid repeated freeze-thaw cycles.

#### Quality control

- The biological activity has been confirmed using cellular assays.
- The absence of bacterial contamination (e.g. lipoproteins and endotoxins) has been confirmed using HEK-Blue<sup>™</sup> TLR2 and HEK-Blue<sup>™</sup> TLR4 cells.

### DESCRIPTION

Nigericin is a microbial toxin derived from the Gram-positive bacteria <code>Streptomyces hygroscopicus</code>. It induces the release of IL-1 $\beta$  in response to the activation of the NLRP3 (also known as NALP3 or cryopyrin) inflammasome<sup>1</sup>. The NLRP3 inflammasome is a caspase-1-activating complex comprising the NLR protein NLRP3 and the adaptor ASC required for the maturation and secretion of IL-1 $\beta$ <sup>2</sup>.

Nigericin acts as a potassium ionophore. It probably activates NLRP3 through its ionophore function which allows the intracellular K<sup>+</sup> efflux across the membrane<sup>3</sup>. Whether the formation of non-selective pannexin-1 pores also contributes to NLRP3 activation upon Nigericin treatment remains unclear<sup>4</sup>.

1. Mariathasan S. et al., 2006. Cryopyrin activates the inflammasome and ATP. Nature 440;228-32. 2. Martinon F. & Tschopp J., 2004. Inflammatory caspases: linking an intracellular innate immune system to autoinflammatory diseases. Cell. 117(5):561-74. 3. Muñoz-Planillo R. et al., 2013. K<sup>+</sup> efflux is the common trigger of NLRP3 inflammasome activation by bacterial toxins and particulate matter. Immunity. 38(6):1142-53. 4. Pelegrin P, & Surprenant A., 2007. Pannexin-1 couples to maitotoxin- and nigericin-induced interleukin-1beta release through a dye uptake-independent pathway. J Biol Chem. 282(4):2386-94.

### CHEMICAL PROPERTIES

**CAS Number:** 28643-80-3

Synonym: Antibiotic K178, Polyetherin A

Linear formula: C<sub>40</sub>H<sub>67</sub>NaO<sub>11</sub> Molecular weight: 746.94 g/mol Solubility: 5 mg/ml in ethanol

# **METHODS**

# Solubilization of Nigercin (5 mg/ml)

- Prepare a 5 mg/ml (6.7 mM) Nigericin stock solution in 100% ethanol.
- Prepare further dilutions by adding the appropriate amount of endotoxin-free water.

# Detection of NLRP3 inflammasome induction

Secretion of IL-1 $\beta$  is an indicator of the NLRP3 inflammasome induction. The activation and release of IL-1 $\beta$  requires two distinct signals: the first signal leads to the transcriptional upregulation and synthesis of pro-IL-1 $\beta$ ; the second signal leads to IL-1 $\beta$  maturation and secretion through the activation of NLRP3 inflammasome.

The synthesis of pro-IL-1 $\beta$  can be induced by priming human monocytic THP-1 cells for 3 h with PMA (phorbol 12-myristate 13-acetate; 20-50 ng/ml) or LPS (lipopolysaccharide, 1 µg/ml). Subsequent stimulation with 1-10 µM of Nigericin leads to the formation of NLRP3 inflammasome resulting in IL-1 $\beta$  maturation and secretion.

Secreted IL-1 $\beta$  can be detected by Western blot or by ELISA. Alternatively, InvivoGen recommends the use of HEK-Blue<sup>™</sup> IL-1 $\beta$  cells, a reporter cell line that specifically detects bioactive IL-1 $\beta$ . These cells express an NF- $\kappa$ B and AP-1-inducible SEAP (secreted alkaline phosphatase) reporter gene. The presence of IL-1 $\beta$  leads to NF- $\kappa$ B and AP-1 activation and the subsequent secretion of SEAP. Levels of SEAP can be easily determined with QUANTI-Blue<sup>™</sup> Solution, a detection reagent that turn purple/ blue in the presence of alkaline phosphatase.

# RELATED PRODUCTS

Product	Description	Cat. Code
Alum Hydroxide ATP CPPD crystals HEK-Blue" IL-1β Cells Hemozoin LPS-EK MSU crystals QUANTI-Blue™ Solution THP1-Null2 cells	Inflammasome inducer Inflammasome inducer Inflammasome inducer IL-1β reporter cells Inflammasome inducer LPS from <i>E. coli</i> K12 Inflammasome inducer SEAP detection reagent THP-1-derived monocytes	tlrl-aloh tlrl-atp tlrl-cppd hkb-il1bv2 tlrl-hz tlrl-eklps tlrl-msu rep-qbs thp-nullz



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