

# Alum Hydroxide

Aluminium hydroxide suspension; NLRP3 inflammasome inducer

Catalog code: tlr1-aloh

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

For research use only

Version 19L06-MM

## PRODUCT INFORMATION

### Contents

• 500 µl Alum Hydroxide (aluminium hydroxide suspension). This product is provided as a white suspension at 30 mg/ml.

### Storage and stability

- Alum Hydroxide is shipped at room temperature.  
- Upon receipt, store Alum Hydroxide at room temperature (15-25°C).  
**DO NOT FREEZE.**

### Quality control

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

## PRODUCT DESCRIPTION

Alum Hydroxide (also referred to as alum) is a ready-to-use suspension of aluminium hydroxide. Aluminium salts, such as aluminium hydroxide, aluminium phosphate or potassium aluminium sulfate, are commonly used vaccine adjuvants, although their mode of action *in vivo* remains unclear. Comprehensive research *in vitro* clearly demonstrates that alum induces an NLRP3 inflammasome-dependent induction of interleukin-1β (IL-1β) and IL-18<sup>1</sup>. Of note, alum has been shown to trigger NLRP3 activation through lysosomal destabilization<sup>2</sup>.

The NLRP3 inflammasome is an intracellular multi-protein complex that plays a central role in innate immunity<sup>3,4</sup>. It is activated by a two-step process; a first signal ('priming') is provided by microbial molecules such as lipopolysaccharide (LPS), while the second signal is provided by a wide array of stimuli including bacterial toxins, endogenous molecules or crystalline substances such as alum. This triggers inflammasome multimerization and caspase-1 activation with the subsequent maturation and secretion of IL-1β and IL-18.

Interestingly, *in vivo* studies indicate a minimal role for the NLRP3 inflammasome in alum's adjuvanticity<sup>5</sup>. The reason for this divergence between the *in vitro* and *in vivo* action of alum needs to be further investigated.

InvivoGen's Alum Hydroxide is designed for *in vitro* assays. Its ability to induce the NLRP3 inflammasome has been validated using InvivoGen's THP1-Null cells (see Methods section).

1. Li H. *et al.*, 2008. Cutting Edge: Inflammasome activation by Alum and Alum's adjuvant effect are mediated by NLRP3. *J Immunol.* 181:17-21. 2. Hornung V. *et al.*, 2008. Silica crystals and aluminium salts activate the NALP3 inflammasome through phagosomal destabilization. *Nature Immunol.* 9:847-856. 3. Schroder K. & Tschopp J., 2010. The inflammasomes. *Cell* 140(6):821-32. 4. Franchi L. *et al.*, 2012. Sensing and reacting to microbes through the inflammasomes. *Nat Immunol* 13(4):325-32. 5. Wen Y. & Shi Y., 2016. Alum: an old dog with new tricks. *Emerg Microbes Infect.* 5:e25.

## CHEMICAL PROPERTIES

CAS number: 21645-51-2

Chemical formula: Al(OH)<sub>3</sub>

Molecular weight: 78 g/mol

## METHODS

### Preparation of serial dilutions

- Prepare dilutions of Alum Hydroxide with sterile water. Do **not** store Alum Hydroxide dilutions for more than 1 day.

Working concentrations: 50-500 µg/ml

## NLRP3 INFLAMMASOME INDUCTION

Alum Hydroxide can be used to induce the NLRP3 inflammasome in cellular assays, such as InvivoGen's THP-1/HEK-Blue™ IL-1β assay. This assay uses the secretion of IL-1β by THP1-Null cells as an indicator of NLRP3 inflammasome induction. The production IL-1β by these cells is measured using HEK-Blue™ IL-1β cells. For more information about this assay please visit <https://www.invivogen.com/thp1-null>.

### Production of IL-1β by THP-1 cells

1. Prepare a THP-1 cell suspension at 2 x 10<sup>6</sup> cells/ml and add 180 µl of this cell suspension per well of a 96-well plate (3 x 10<sup>6</sup> cells/well).
2. Prime THP-1 cells with 20 µl of LPS (final concentration 1 µg/ml) for 3 hours at 37 °C in 5% CO<sub>2</sub>.
3. Remove gently medium and add 180 µl supplemented RPMI.
4. Add 20 µl of Alum Hydroxide (50-500 µg/ml final concentration).
5. Incubate overnight at 37 °C in 5% CO<sub>2</sub>.

### Detection of IL-1β

Secreted IL-1β from the supernatant of the treated THP-1 cells can be detected using InvivoGen's HEK-Blue™ IL-1β cells. For more information, visit <https://www.invivogen.com/hek-blue-il1b>.

## RELATED PRODUCTS

Product	Description	Cat. Code
ATP	Inflammasome inducer	tlr1-atpl
CPPD Crystals	Inflammasome inducer	tlr1-cppd
HEK-Blue™ IL-1β Cells	IL-1β reporter cells	hkb-il1b
Hemozoin	Inflammasome inducer	tlr1-hz
LPS-EK	LPS from <i>E. coli</i> K12	tlr1-eklps
MSU crystals	Inflammasome inducer	tlr1-msu
Nigericin	Inflammasome inducer	tlr1-nig
THP1-Null Cells	Human monocytic cells	thp-null

## TECHNICAL SUPPORT

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