

# Alum Crystals

## NLRP3 inflammasome Inducer

Catalog code: tlrl-alk

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

For research use only

Version 19E23-MM

## PRODUCT INFORMATION

### Contents

- 1 g Alum crystals (aluminium potassium sulfate)

### Storage and stability

- Alum crystals are shipped at room temperature. Store at room temperature (15-25°C).
- Upon resuspension, Alum crystals should be stored at 4°C for short term storage or -20°C for long storage. Resuspended product is stable 6 months at 4°C and 1 year at -20°C when properly stored.

### Quality control

- Purity: ≥98.0%
- 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.

## DESCRIPTION

Aluminum hydroxide and potassium salts (alum) are commonly used vaccine adjuvants. Adjuvants are vaccine additives that stimulate the immune system without having any specific antigenic effect. Alum has been demonstrated to activate caspase-1 and triggers IL-1 $\beta$  and IL-18 secretion<sup>1</sup>. All alum preparations contain crystals. The alum-induced release of IL-1 $\beta$  in macrophages is dependent on NALP3 and ASC, indicating that alum triggers inflammation through activation of the NALP3 inflammasome<sup>2</sup>. Alum has been shown to trigger NALP3 activation through lysosomal destabilization<sup>2</sup>.

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.

## CHEMICAL PROPERTIES

CAS Number: 7784-24-9

Linear formula:  $\text{AlK}(\text{SO}_4)_2 \cdot 12\text{H}_2\text{O}$

Molecular weight: 474.39 g/mol

Solubility: Clear to slightly hazy colorless solution at 0.5 M in water

## METHODS

### Solubilization of alum crystals

- Prepare a 40 mM (20 mg/ml) alum stock solution by using endotoxin-free water.
- Prepare further dilutions by adding the appropriate amount of endotoxin-free water or PBS.

### 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; 300 ng/ml) or LPS (lipopolysaccharide; 1  $\mu\text{g}/\text{ml}$ ). Subsequent stimulation with 10-200 mg/ml alum 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 ELISA. Alternatively, InvivoGen recommends the use of HEK-Blue™ IL-1 $\beta$  cells, a reporter cell line that specifically detects bioactive IL-1 $\beta$ . These cells express an NF- $\kappa\text{B}$  and AP-1-inducible SEAP (secreted alkaline phosphatase) reporter gene. The presence of IL-1 $\beta$  leads to NF- $\kappa\text{B}$  and AP-1 activation and the subsequent secretion of SEAP. Levels of SEAP can be easily determined with HEK-Blue™ Detection or QUANTI-Blue™ Solution, detection media that turn purple/blue in the presence of alkaline phosphatase.

## RELATED PRODUCTS

Product	Catalog Code
ATP	tlrl-atp
CPPD crystals	tlrl-cppd
HEK-Blue™ Detection	hb-det2
HEK-Blue™ IL-1 $\beta$ Cells	hkb-il1b
Hemozoin	tlrl-hz
LPS-EK (LPS from <i>E. coli</i> K12)	tlrl-eklps
MSU crystals	tlrl-msu
Nigericin	tlrl-nig
Poly(dA:dT)	tlrl-pat
QUANTI-Blue™ Solution	rep-qbs

### TECHNICAL SUPPORT

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