abcam

Product datasheet

# Human NLRP3 knockout THP-1 cell lysate ab280122

3 Images

## Overview

| | |
|---|---|
| **Product name** | Human NLRP3 knockout THP-1 cell lysate |
| **Product overview** | Knockout cell lysate achieved by CRISPR/Cas9. |
| **Parental Cell Line** | THP-1 |
| **Organism** | Human |
| **Mutation description** | Knockout achieved by using CRISPR/Cas9, Homozygous: 58bp deletion in exon 2 |
| **Passage number** | <20 |
| **Knockout validation** | Sanger Sequencing, Western Blot (WB) |
| **Reconstitution notes** | To use as WB control, resuspend the lyophilizate in 50 µL of LDS* Sample Buffer to have a final concentration of 2 mg/ml. For reducing conditions, we recommend a final concentration of 0.1 M DTT.<br>**Usage of SDS sample buffer is not recommended with these lyophilized lysates.* |
| **Notes** | **Lysate preparation:** Our lysates are made using RIPA buffer to which we add a protease inhibitor cocktail and phosphatase inhibitor cocktail (ratio: 300:100:10). *This means that the protein of interest is denatured.* If you require a native form of the protein please use the live cell version - found **here**. Please refer to our lysis protocol for further details on how our lysates are prepared.<br><br>**User storage instructions:** Lyophilizate may be stored at 4°C. After reconstitution, store at -20°C for short-term storage or -80°C for long-term storage.<br><br>Access thousands of knockout cell lysates, generated from commonly used cancer cell lines.<br>**See here for more information on knockout cell lysates.**<br><br>Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of products that contain European Authorisation list (Annex XIV) substances.<br>It is the responsibility of our customers to check the necessity of application of REACH Authorisation, and any other relevant authorisations, for their intended uses.<br><br>This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our **limited use license** and **patent pages**. |
| **Tested applications** | **Suitable for:** WB, Sanger Sequencing |

## Properties

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**Storage instructions** Store at -80°C. Please refer to protocols.

| Components | 1 kit |
|---|---|
| ab280165 - Human NLRP3 knockout THP-1 cell lysate | 1 x 100µg |
| ab282895 - Human wild-type THP-1 cell lysate | 1 x 100µg |

**Cell type** acute monocytic leukemia

**Disease** Acute Monocytic Leukemia

**Gender** Male

## Target

**Function** May function as an inducer of apoptosis. Interacts selectively with ASC and this complex may function as an upstream activator of NF-kappa-B signaling. Inhibits TNF-alpha induced activation and nuclear translocation of RELA/NF-KB p65. Also inhibits transcriptional activity of RELA. Activates caspase-1 in response to a number of triggers including bacterial or viral infection which leads to processing and release of IL1B and IL18.

**Tissue specificity** Expressed in blood leukocytes. Strongly expressed in polymorphonuclear cells and osteoblasts. Undetectable or expressed at a lower magnitude in B- and T-lymphoblasts, respectively. High level of expression detected in chondrocytes. Detected in non-keratinizing epithelia of oropharynx, esophagus and ectocervix and in the urothelial layer of the bladder.

**Involvement in disease** Defects in NLRP3 are the cause of familial cold autoinflammatory syndrome type 1 (FCAS1) [MIM:120100]; also known as familial cold urticaria. FCAS are rare autosomal dominant systemic inflammatory diseases characterized by episodes of rash, arthralgia, fever and conjunctivitis after generalized exposure to cold.
Defects in NLRP3 are a cause of Muckle-Wells syndrome (MWS) [MIM:191900]; also known as urticaria-deafness-amyloidosis syndrome. MWS is a hereditary periodic fever syndrome characterized by fever, chronic recurrent urticaria, arthralgias, progressive sensorineural deafness, and reactive renal amyloidosis. The disease may be severe if generalized amyloidosis occurs.
Defects in NLRP3 are the cause of chronic infantile neurologic cutaneous and articular syndrome (CINCA) [MIM:607115]; also known as neonatal onset multisystem inflammatory disease (NOMID). CINCA is a rare congenital inflammatory disorder characterized by a triad of neonatal onset of cutaneous symptoms, chronic meningitis and joint manifestations with recurrent fever and inflammation.

**Sequence similarities** Belongs to the NLRP family.
Contains 1 DAPIN domain.
Contains 9 LRR (leucine-rich) repeats.
Contains 1 NACHT domain.

**Cellular localization** Cytoplasm.

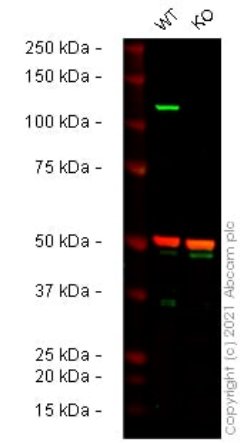
## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab280122 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

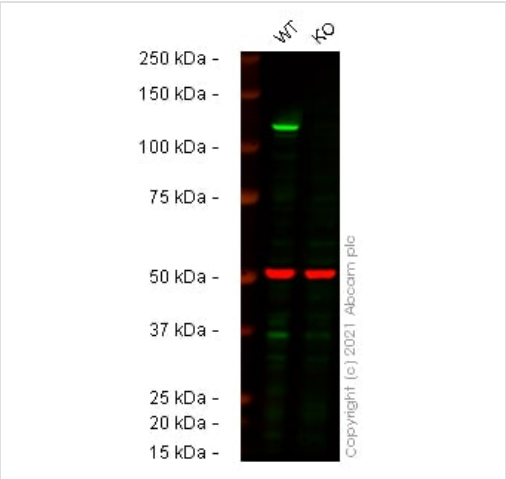
| Application | Abreviews | Notes |
|---|---|---|
| **WB** | | Use at an assay dependent concentration. Predicted molecular weight: 118 kDa. |
| **Sanger Sequencing** | | Use at an assay dependent concentration. |

## Images



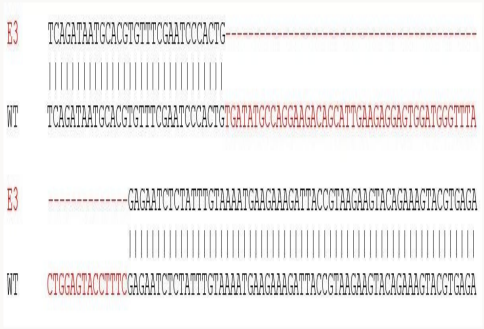

Western blot - Human NLRP3 knockout THP-1 cell lysate (ab280122)

**Lane 1:** Wild-type THP-1 cell lysate 20 µg
**Lane 2:** NLRP3 knockout THP-1 cell lysate 20 µg
False colour image of Western blot: Anti-NLRP3 antibody staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (**ab7291**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab4207** was shown to bind specifically to NLRP3. A band was observed at 118 kDa in wild-type THP-1 cell lysates with no signal observed at this size in NLRP3 knockout cell line **ab280063** (knockout cell lysate ab280122). To generate this image, wild-type and NLRP3 knockout THP-1 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Donkey anti-Goat IgG H&L (IRDye® 800CW) preabsorbed (**ab216775**) and Donkey anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216778**) at 1/20000 dilution.




Western blot - Human NLRP3 knockout THP-1 cell lysate (ab280122)

**Lane 1:** Wild-type THP-1 cell lysate 20 µg
**Lane 2:** NLRP3 knockout THP-1 cell lysate 20 µg
False colour image of Western blot: Anti-NLRP3 antibody [EPR23094-1] staining at 1/500 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (**ab7291**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab263899** was shown to bind specifically to NLRP3. A band was observed at 118 kDa in wild-type THP-1 cell lysates with no signal observed at this size in NLRP3 knockout cell line **ab280063** (knockout cell lysate ab280122). To generate this image, wild-type and NLRP3 knockout THP-1 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (**ab216772**) at 1/20000 dilution.




Human NLRP3 knockout THP-1 cell lysate

58bp deletion in exon 2

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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