abcam

Product datasheet

# Human ANXA6 (Annexin-6) knockout HeLa cell lysate ab257351

5 Images

## Overview

| | |
|---|---|
| **Product name** | Human ANXA6 (Annexin-6) knockout HeLa cell lysate |
| **Product overview** | Knockout cell lysate achieved by CRISPR/Cas9. |
| **Parental Cell Line** | HeLa |
| **Organism** | Human |
| **Mutation description** | Knockout achieved by using CRISPR/Cas9, 1 bp insertion in exon7 and 5 bp deletion in exon7. |
| **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, ERS Genomics Limited and Sigma-Aldrich Co. LLC, and is developed with patented technology. For full details of the licenses and patents please refer to our **limited use license** and **patent pages**. |
| **Tested applications** | **Suitable for:** WB |

## Properties

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

| Components | 1 kit |
|---|---|
| ab260969 - Human ANXA6 knockout HeLa cell lysate | 1 x 100µg |
| ab255929 - Human wild-type HeLa cell lysate | 1 x 100µg |

**Cell type** epithelial

**Disease** Adenocarcinoma

**Gender** Female

**STR Analysis** Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18 TH01: 7 TPOX: 8,12 CSF1PO: 9, 10

## Target

**Function** May associate with CD21. May regulate the release of Ca(2+) from intracellular stores.

**Sequence similarities** Belongs to the annexin family.
Contains 8 annexin repeats.

**Domain** A pair of annexin repeats may form one binding site for calcium and phospholipid.

**Post-translational modifications** Phosphorylated in response to growth factor stimulation.

**Cellular localization** Cytoplasm. Melanosome. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

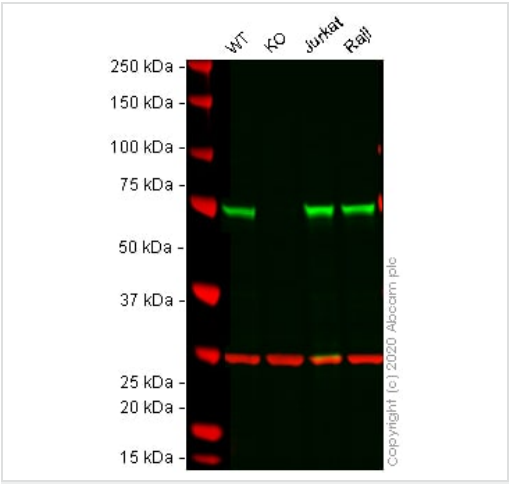
## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab257351 in the following tested applications.

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

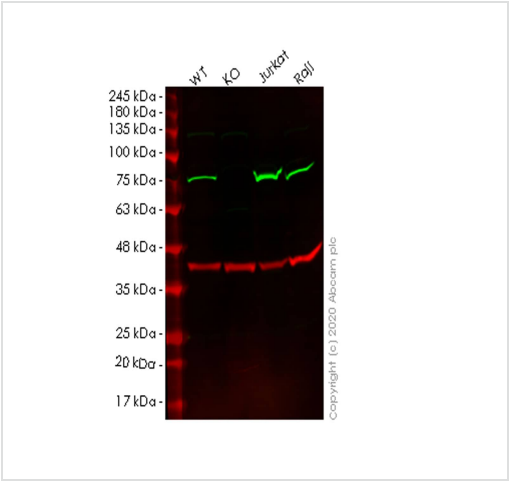
| Application | Abreviews | Notes |
|---|---|---|
| **WB** | | Use at an assay dependent concentration. Predicted molecular weight: 76 kDa. |

## Images




Western blot - Human ANXA6 (Annexin-6) knockout HeLa cell lysate (ab257351)

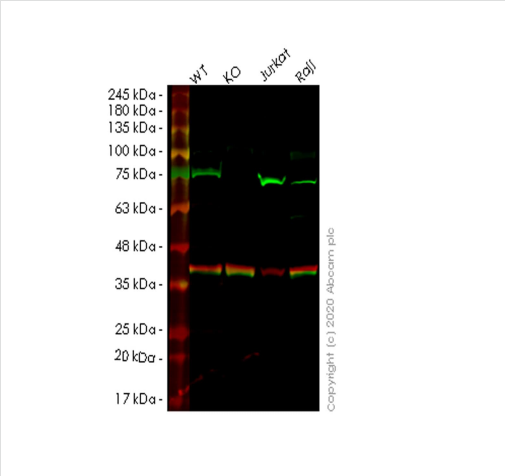
**Lane 1:** Wild-type HeLa cell lysate 20 ug
**Lane 2:** ANXA6 knockout HeLa cell lysate 20 ug
**Lane 3:** Jurkat cell lysate 20 ug
**Lane 4:** Raji cell lysate 20 ug
**Lanes 1 - 4:**Merged signal (red and green). Green - **ab199422** observed at 75 kDa. Red - loading control, **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.
**ab199422** was shown to react with Annexin-6/ANXA6 in wild-type HeLa cells in western blot with loss of signal observed in ANXA6 knockout cell line **ab265677** (ANXA6 knockout cell lysate ab257351). Wild-type and ANXA6 knockout HeLa cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab199422** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.




Western blot - Human ANXA6 knockout HeLa cell lysate (ab257351)

**Lane 1:**Wild-type HeLa cell lysate (20 ug)
**Lane 2:**ANXA6 knockout HeLa cell lysate (20 ug)
**Lane 3:**Jurkat cell lysate (20 ug)
**Lane 4:**Raji cell lysate (20 ug)

**ab201023** was shown to specifically react with Annexin-6/ANXA6 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265677** (knockout cell lysate ab257351) was used. Wild-type and Annexin-6/ANXA6 knockout samples were subjected to SDS-PAGE. **ab201023** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated at room temperature for 2.5 hours at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.




Western blot - Human ANXA6 knockout HeLa cell lysate (ab257351)

**Lane 1:** Wild-type HeLa cell lysate (20 ug)
**Lane 2:** ANXA6 knockout HeLa cell lysate (20 ug)
**Lane 3:** Jurkat cell lysate (20 ug)
**Lane 4:** Raji cell lysate (20 ug)

**ab201024** was shown to specifically react with Annexin-6/ANXA6 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265677** (knockout cell lysate ab257351) was used. Wild-type and Annexin-6/ANXA6 knockout samples were subjected to SDS-PAGE. **ab201024** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated at room temperature for 2.5 hours at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.


Sanger Sequencing - Human ANXA6 knockout HeLa cell lysate (ab257351)

Allele-1: 5 bp deletion in exon7


Sanger Sequencing - Human ANXA6 knockout HeLa cell lysate (ab257351)

Allele-2: 1 bp insertion in exon7

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

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