# abcam

# Product datasheet

# Human RIPK2 (RIP2) knockout HeLa cell lysate ab258636

3 Images

Overview

Product name Human RIPK2 (RIP2) 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 exon 1 and 2 bp deletion in exon 1.

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.

\*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.

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.

Access thousands of knockout cell lysates, generated from commonly used cancer cell lines.

See here for more information on knockout cell lysates.

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.

It is the responsibility of our customers to check the necessity of application of REACH

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licenses and patents please refer to our <u>limited use license</u> and <u>patent pages</u>.

Tested applications Suitable for: WB

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#### **Properties**

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

Components	1 kit
ab260606 - Human RIPK2 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 Activates pro-caspase-1 and pro-caspase-8. Potentiates CASP8-mediated apoptosis. Activates

NF-kappa-B.

**Tissue specificity** Detected in heart, brain, placenta, lung, peripheral blood leukocytes, spleen, kidney, testis,

prostate, pancreas and lymph node.

**Sequence similarities**Belongs to the protein kinase superfamily. TKL Ser/Thr protein kinase family.

Contains 1 CARD domain.

Contains 1 protein kinase domain.

**Post-translational** Autophosphorylated. Phosphorylated upon DNA damage, probably by ATM or ATR.

modifications Ubiquitinated; undergoes 'Lys-63'-linked polyubiquitination catalyzed by ITCH.

Cellular localization Cytoplasm.

#### **Applications**

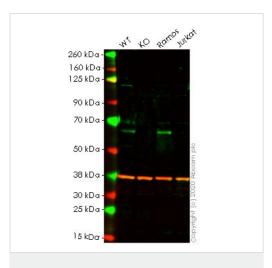
#### The Abpromise guarantee

Our Abpromise guarantee covers the use of ab258636 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: 61 kDa.

#### **Images**



Western blot - Human RIPK2 knockout HeLa cell lysate (ab258636)

Lane 1: Wild-type HeLa cell lysate (20 µg)

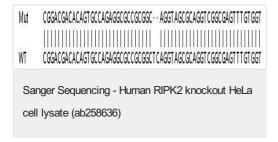
Lane 2: RIPK2 knockout HeLa cell lysate (20 µg)

Lane 3: Ramos cell lysate (20 µg)

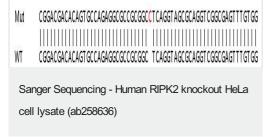
Lane 4: Jurkat cell lysate (20 µg)

**Lanes 1-4:** Merged signal (red and green). Green - <u>ab75257</u> observed at 65 kDa. Red - loading control <u>ab181602</u> observed at 37 kDa.

ab75257 Anti-RIP2 antibody [AF28D3] was shown to specifically react with RIP2 in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab264688 (knockout cell lysate ab258636) was used. Wild-type and RIP2 knockout samples were subjected to SDS-PAGE. ab75257 and Anti-GAPDH antibody[EPR16891] - Loading Control (ab181602) were incubated overnight at 4°C at 1 in 2000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) and Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Allele-1: 2 bp deletion in exon 1



Allele-2: 1 bp insertion in exon 1

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