abcam

Product datasheet

Human CD74 knockout Raji cell lysate ab275529

6 Images

Overview

Product name	Human CD74 knockout Raji cell lysate
Product overview	Western blot data indicates that the CRISPR gene edit may have resulted in a truncation of the protein of interest. Please see data images.
Parental Cell Line	Raji
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 13 bp deletion in exon 2
Passage number	<20
Knockout validation	Sanger Sequencing
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 Authorisation, and any other relevant authorisations, for their intended uses. 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

Properties

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

Components	1 kit
ab277364 - Human CD74 knockout Raji cell lysate	1 x 100µg
ab277365 - Human wild-type Raji cell lysate	1 x 100µg

Cell type Burkitt's lymphoma

Disease Lymphoma

Gender Male

Target

Function Plays a critical role in MHC class II antigen processing by stabilizing peptide-free class II alpha/beta heterodimers in a complex soon after their synthesis and directing transport of the complex from the endoplasmic reticulum to the endosomal/lysosomal system where the antigen processing and binding of antigenic peptides to MHC class II takes place. Serves as cell surface receptor for the cytokine MIF.

Sequence similarities Contains 1 thyroglobulin type-1 domain.

Cellular localization Cell membrane. Endoplasmic reticulum membrane. Golgi apparatus > trans-Golgi network. Endosome. Lysosome. Transits through a number of intracellular compartments in the endocytic pathway. It can either undergo proteolysis or reach the cell membrane.

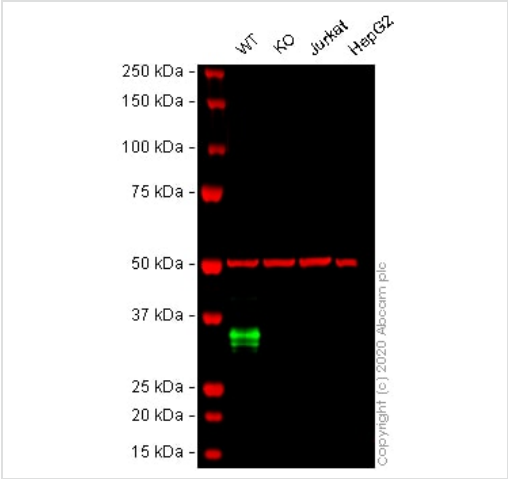
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab275529 in the following tested applications.

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

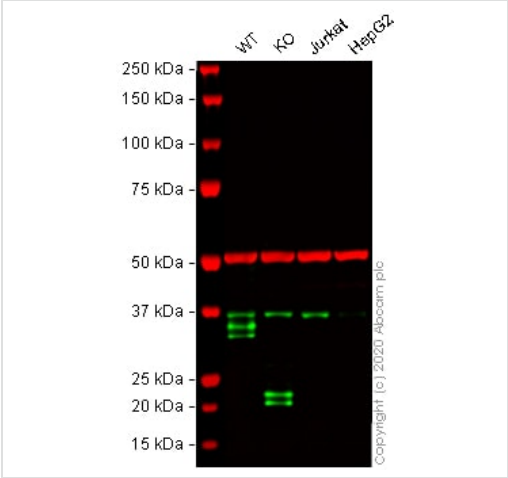
Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 34 kDa. Western blot data indicates that the CRISPR gene edit may have resulted in a truncation of the protein of interest. Please see data images.

Images



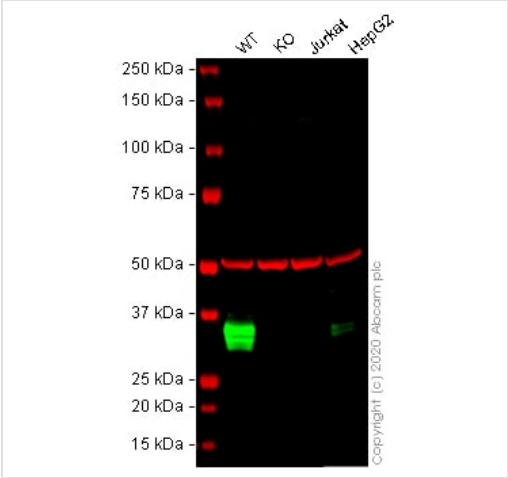
Western blot - Human CD74 knockout Raji cell lysate (ab275529)

Lane 1: Wild-type Raji cell lysate 30 ug
Lane 2: CD74 knockout Raji cell lysate 30 ug
Lane 3: Jurkat cell lysate 30 ug
Lane 4: HepG2 cell lysate 30 ug
Lanes 1 - 4: Merged signal (red and green). Green - **ab270265** observed at 35 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.
ab270265 was shown to react with CD74 in western blot. The band observed in the knockout lysate lane below 35kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab270265** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at 1 ug/ml 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 h at room temperature before imaging.



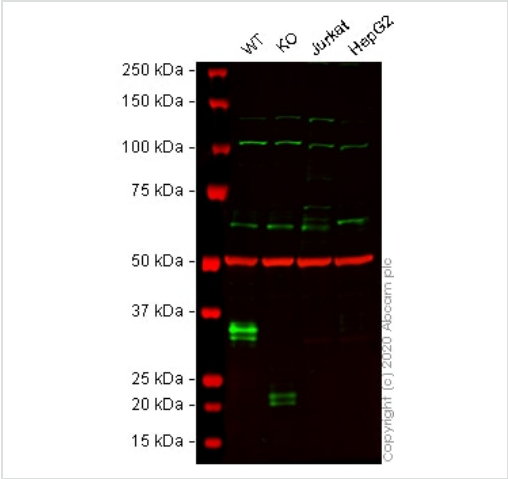
Western blot - Human CD74 knockout Raji cell lysate (ab275529)

Lane 1: Wild-type Raji cell lysate 30 ug
Lane 2: CD74 knockout Raji cell lysate 30 ug
Lane 3: Jurkat cell lysate 30 ug
Lane 4: HepG2 cell lysate 30 ug
Lanes 1 - 4: Merged signal (red and green). Green - **ab108393** observed at 35 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.
ab108393 was shown to react with CD74 in western blot. The band observed in the knockout lysate lane below 35kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab108393** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) 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 h at room temperature before imaging.



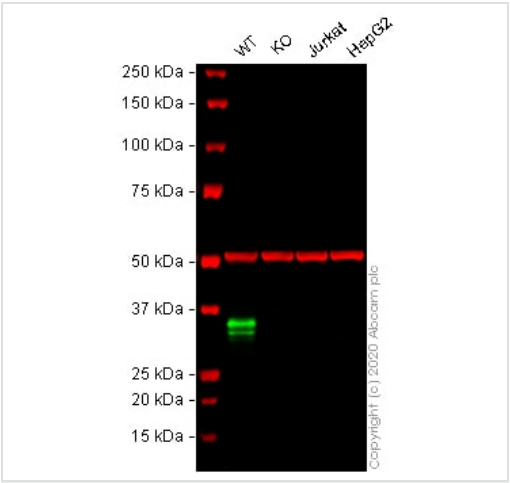
Western blot - Human CD74 knockout Raji cell lysate (ab275529)

Lane 1: Wild-type Raji cell lysate 30 ug
Lane 2: CD74 knockout Raji cell lysate 30 ug
Lane 3: Jurkat cell lysate 30 ug
Lane 4: HepG2 cell lysate 30 ug
Lanes 1 - 4: Merged signal (red and green). Green - **ab64772** observed at 35 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.
ab64772 was shown to react with CD74 in western blot. The band observed in the knockout lysate lane below 35kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab64772** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at 1 ug/ml 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 h at room temperature before imaging.



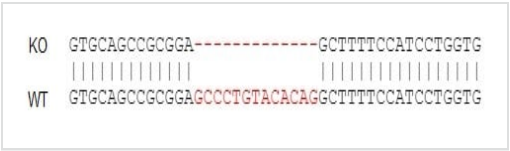
Western blot - Human CD74 knockout Raji cell lysate (ab275529)

Lane 1: Wild-type Raji cell lysate 30 ug
Lane 2: CD74 knockout Raji cell lysate 30 ug
Lane 3: Jurkat cell lysate 30 ug
Lane 4: HepG2 cell lysate 30 ug
Lanes 1 - 4: Merged signal (red and green). Green - **ab22603** observed at 35 kDa. Red - loading control **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) observed at 55 kDa.
ab22603 was shown to react with CD74 in western blot. The band observed in the knockout lysate lane below 35kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab22603** and **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4 °C at 1 ug/ml and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (**ab216772**) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (**ab216777**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Western blot - Human CD74 knockout Raji cell lysate (ab275529)

Lane 1: Wild-type Raji cell lysate 30 ug
Lane 2: CD74 knockout Raji cell lysate 30 ug
Lane 3: Jurkat cell lysate 30 ug
Lane 4: HepG2 cell lysate 30 ug
Lanes 1 - 4: Merged signal (red and green). Green - **ab9514** observed at 35 kDa. Red - loading control **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) observed at 55 kDa.
ab9514 was shown to react with CD74 in western blot. The band observed in the knockout lysate lane below 35kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution before incubation with **ab9514** and **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4 °C at 5 ug/ml and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (**ab216772**) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (**ab216777**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Sanger Sequencing - Human CD74 knockout Raji cell lysate

Homozygous: 13 bp deletion in exon 2

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

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