abcam

Product datasheet

Human VCAM1 knockout A549 cell lysate ab275504

画像数 3

製品の概要

製品名	Human VCAM1 knockout A549 cell lysate
製品の概要	Knockout cell lysate achieved by CRISPR/Cas9. **Treatments:** Human VCAM1 knockout A549 cell lysate - Untreated Human wild-type A549 cell lysate - Untreated Human VCAM1 knockout A549 cell lysate - TNF-a (10 ng/ml, 16h) Human wild-type A549 cell lysate - TNF-a (10 ng/ml, 16h)
Parental Cell Line	A549
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, Homozygous: 4 bp 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. **Usage of SDS sample buffer is not recommended with these lyophilized lysates.*
特記事項	**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.

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アプリケーション **適用あり:** WB

製品の特性

保存方法 Store at -80°C. Please refer to protocols.

内容	1 kit
ab277498 - Human VCAM1 knockout A549 cell lysate - TNF-alpha treated	1 x 100µg
ab277312 - Human VCAM1 knockout A549 cell lysate	1 x 100µg
ab277499 - Human wild-type A549 cell lysate - TNF-alpha treated	1 x 100µg
ab277305 - Human wild-type A549 cell lysate	1 x 100µg

Cell type epithelial

Disease Carcinoma

Gender Male

ターゲット情報

機能 Important in cell-cell recognition. Appears to function in leukocyte-endothelial cell adhesion. Interacts with the beta-1 integrin VLA4 on leukocytes, and mediates both adhesion and signal transduction. The VCAM1/VLA4 interaction may play a pathophysiologic role both in immune responses and in leukocyte emigration to sites of inflammation.

組織特異性 Expressed on inflammed vascular endothelium, as well as on macrophage-like and dendritic cell types in both normal and inflammed tissue.

配列類似性 Contains 7 Ig-like C2-type (immunoglobulin-like) domains.

ドメイン Either the first or the fourth Ig-like C2-type domain is required for VLA4-dependent cell adhesion.

翻訳後修飾 Sialoglycoprotein.

細胞内局在 Membrane.

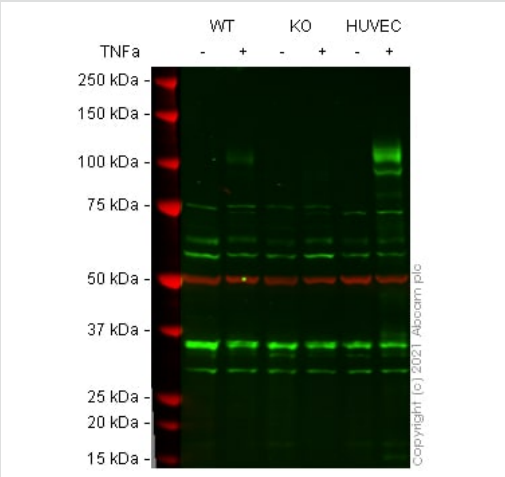
アプリケーション

The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab275504の使用に適用されます

アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

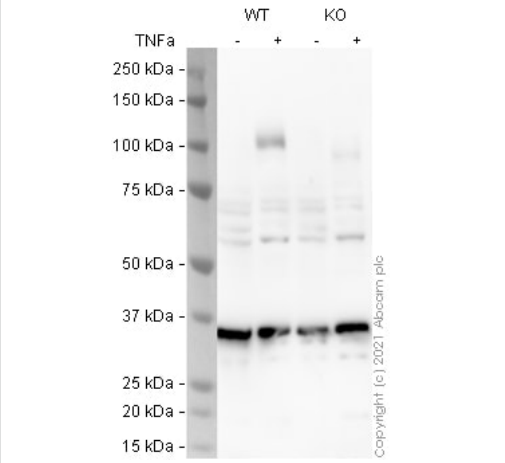
アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration.

画像



Western blot - Human VCAM1 knockout A549 cell lysate (ab275504)

Lane 1: Wild-type A549 cell lysate 30 ug
Lane 2: Wild-type A549 TNF-a treated (10 ng/mL, 16h) cell lysate 30 ug
Lane 3: VCAM1 knockout A549 cell lysate 30 ug
Lane 4: VCAM1 knockout A549 TNF-a treated (10 ng/mL, 16h) cell lysate 30 ug
Lane 5: HUVEC cell lysate 30 ug
Lane 6: HUVEC TNF-a treated (16 ng/mL, 16h) cell lysate 30 ug
Lanes 1 - 6: Merged signal (red and green). Green - **ab134047** observed at 105 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.
ab134047 was shown to react with VCAM1 in wild-type A549 cells in Western blot with loss of signal observed in VCAM1 knockout cell line **ab273758** (knockout cell lysate ab275504). Wild-type A549 and VCAM1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab134047** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 2000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with HRP conjugated Goat anti-Rabbit (H+L) 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.



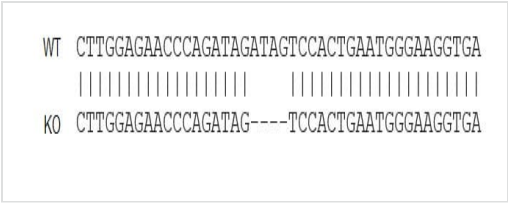
Western blot - Human VCAM1 knockout A549 cell lysate (ab275504)

Lane 1: Wild-type A549 cell lysate 30 ug
Lane 2: Wild-type A549 TNF-a treated (10 ng/mL, 16h) cell lysate 30 ug
Lane 3: VCAM1 knockout A549 cell lysate 30 ug
Lane 4: VCAM1 knockout A549 TNF-a treated (10 ng/mL, 16h) cell lysate 30 ug

Lanes 1: Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate, 20 ug
Lanes 2: GPX4 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate, 20 ug
Lanes 3: Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate, 20 ug
Lanes 3: Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate, 20 ug

ab174279 was shown to react with VCAM1 in treated wild-type A549 cells in western blot. Loss of signal was observed when treated VCAM1 knockout cell line **ab273758** (knockout cell lysate ab275504) was used. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab174279** overnight

at 4 °C at a 1 in 1000 dilution. Blots were incubated with HRP conjugated Goat anti-Rabbit (H+L) secondary antibody at 1 in 5000 for 1 hour at room temperature before development with Optiblot ECL reagent (**ab133456**) and imaging.



Sanger Sequencing - Human VCAM1 knockout A549 cell lysate (ab275504)

Allele-1: 4 bp deletion in exon 2

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