abcam

Product datasheet

Human CD44 knockout HeLa cell line ab262515

画像数 14

製品の概要

製品名	Human CD44 knockout HeLa cell line
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by CRISPR/Cas9; X = 1 bp insertion, 1 bp deletion; Frameshift = 98.54%
Passage number	<20
Knockout validation	Immunocytochemistry (ICC), Next Generation Sequencing (NGS), Western Blot (WB)
アプリケーション	**適用あり:** Next Generation Sequencing, ICC/IF, WB
Biosafety level	2
特記事項	**Recommended control:** Human wild-type HeLa cell line (**ab271142**). Please note a wild-type cell line is not automatically included with a knockout cell line order, if required please add recommended wild-type cell line at no additional cost using the code WILDTYPE-TMTK1. **Cryopreservation cell medium:** Cell Freezing Medium-DMSO Serum free media, contains 8.7% DMSO in MEM supplemented with methyl cellulose. **Culture medium:** DMEM (High Glucose) + 10% FBS **Initial handling guidelines:** Upon arrival, the vial should be stored in liquid nitrogen vapor phase and not at -80°C. Storage at -80°C may result in loss of viability. 1. Thaw the vial in 37°C water bath for approximately 1-2 minutes. 2. Transfer the cell suspension (0.8 mL) to a 15 mL/50 mL conical sterile polypropylene centrifuge tube containing 8.4 mL pre-warmed culture medium, wash vial with an additional 0.8 mL culture medium (total volume 10 mL) to collect remaining cells, and centrifuge at 201 x g (rcf) for 5 minutes at room temperature. 10 mL represents minimum recommended dilution. 20 mL represents maximum recommended dilution. 3. Resuspend the cell pellet in 5 mL pre-warmed culture medium and count using a haemocytometer or alternative cell counting method. Based on cell count, seed cells in an appropriate cell culture flask at a density of $2x10^4$ cells/cm^2. Seeding density is given as a guide only and should be scaled to align with individual lab schedules. 4. Incubate the culture at 37°C incubator with 5% CO_2. Cultures should be monitored daily. **Subculture guidelines:** All seeding densities should be based on cell counts gained by established methods. A guide seeding density of $2x10^4$ cells/cm^2 is recommended. A partial media change 24 hours prior to subculture may be helpful to encourage growth, if required.

Cells should be passaged when they have achieved 80-90% confluence.
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**.

We will provide viable cells that proliferate on revival.

製品の特性

Number of cells	1 x 10^6 cells/vial, 1 mL
Adherent /Suspension	Adherent
Tissue	Cervix
Cell type	epithelial
Disease	Adenocarcinoma
Gender	Female
Mycoplasma free	Yes
保存方法	Shipped on Dry Ice. Store in liquid nitrogen.
バッファー	Constituents: 8.7% Dimethylsulfoxide, 2% Cellulose, methyl ether

ターゲット情報

機能	Receptor for hyaluronic acid (HA). Mediates cell-cell and cell-matrix interactions through its affinity for HA, and possibly also through its affinity for other ligands such as osteopontin, collagens, and matrix metalloproteinases (MMPs). Adhesion with HA plays an important role in cell migration, tumor growth and progression. Also involved in lymphocyte activation, recirculation and homing, and in hematopoiesis. Altered expression or dysfunction causes numerous pathogenic phenotypes. Great protein heterogeneity due to numerous alternative splicing and post-translational modification events.
組織特異性	Isoform 10 (epithelial isoform) is expressed by cells of epithelium and highly expressed by carcinomas. Expression is repressed in neuroblastoma cells.
配列類似性	Contains 1 Link domain.
ドメイン	The lectin-like LINK domain is responsible for hyaluronan binding.
翻訳後修飾	Proteolytically cleaved in the extracellular matrix by specific proteinases (possibly MMPs) in several cell lines and tumors. N-glycosylated. O-glycosylated; contains more-or-less-sulfated chondroitin sulfate glycans, whose number may affect the accessibility of specific proteinases to their cleavage site(s). Phosphorylated; activation of PKC results in the dephosphorylation of Ser-706 (constitutive phosphorylation site), and the phosphorylation of Ser-672.
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ab262515の使用に適用されます**

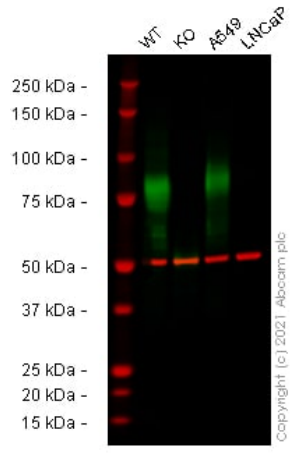
アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Next Generation Sequencing		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration.

画像

Next Generation Sequencing - Human CD44 knockout HeLa cell line (ab262515)

1 bp deletion after Arg149 (allele 1) and 1 bp deletion after Asn148 (allele 2) of the WT protein



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [EPR18668] (**ab189524**) at 1/1000 dilution

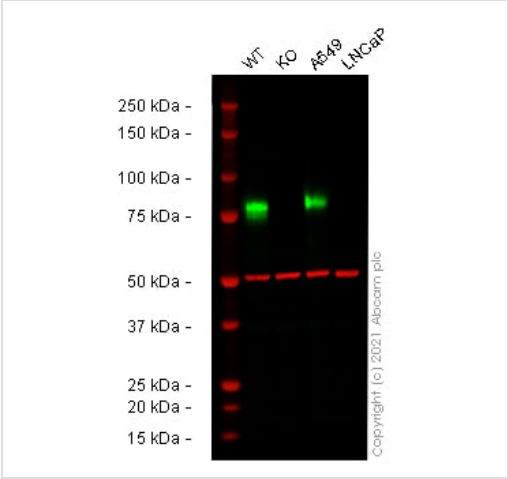
Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD44 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : LNCaP (Human prostate cancer cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 70-85 kDa

False colour image of Western blot: Anti-CD44 antibody [EPR18668] 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, **ab189524** was shown to bind specifically to CD44. A band was observed at 70-85 kDa in wild-type HeLa cell lysates with no signal observed at this size in CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**). To generate this image, wild-type and CD44 knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) 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® 680RD) preabsorbed (**ab216776**) at 1/20000 dilution.



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [SP37] (**ab101531**) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : CD44 knockout HeLa cell lysate
Lane 3 : A549 cell lysate
Lane 4 : LNCaP cell lysate

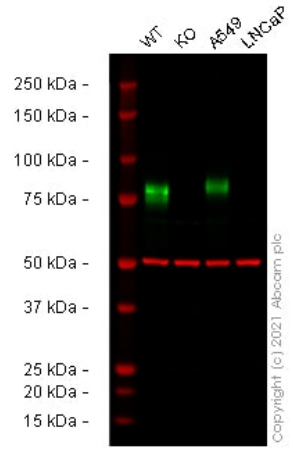
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 75-80 kDa

False colour image of Western blot: Anti-CD44 antibody [SP37] 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, **ab101531** was shown to bind specifically to CD44. A band was observed at 75-80 kDa in wild-type HeLa cell lysates with no signal observed at this size in CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**). To generate this image, wild-type and CD44 knockout HeLa cell

lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) 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® 680RD) preabsorbed (**ab216776**) at 1/20000 dilution.



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [EPR1013Y] (**ab51037**) at 1/5000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : CD44 knockout HeLa cell lysate
Lane 3 : A549 cell lysate
Lane 4 : LNCaP cell lysate

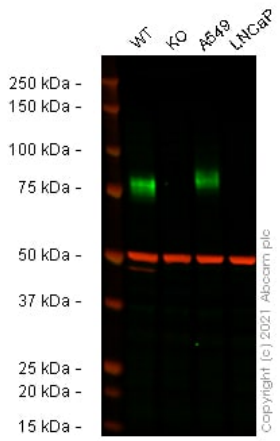
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 75-80 kDa

False colour image of Western blot: Anti-CD44 antibody [EPR1013Y] staining at 1/5000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (**ab7291**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab51037** was shown to bind specifically to CD44. A band was observed at 75-80 kDa in wild-type HeLa cell lysates with no signal observed at this size in CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**). To generate this image, wild-type and CD44 knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) 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® 680RD) preabsorbed (**ab216776**) at

1/20000 dilution.



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [C44Mab-5] (**ab264539**)

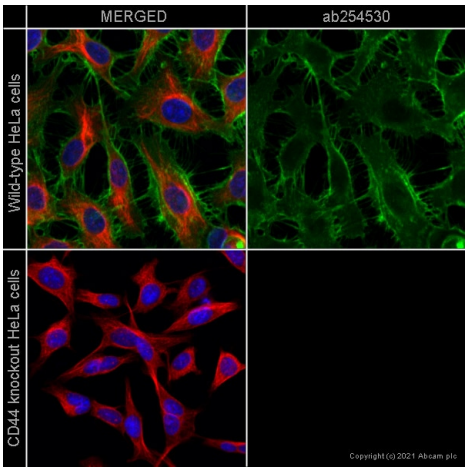
Lane 1 : Wild-type HeLa cell lysate
Lane 2 : CD44 knockout HeLa cell lysate
Lane 3 : A549 cell lysate
Lane 4 : LNCaP cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

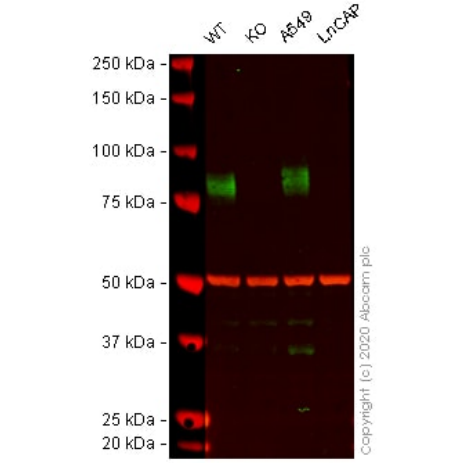
Observed band size: 75-80 kDa

False colour image of Western blot: Anti-CD44 antibody [C44Mab-5] staining at 1.226 µg/ml, shown in green; Rabbit anti-alpha Tubulin antibody [EP1332Y] (**ab52866**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab264539** was shown to bind specifically to CD44. A band was observed at 75-80 kDa in wild-type HeLa cell lysates with no signal observed at this size in CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**). To generate this image, wild-type and CD44 knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) 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-Mouse IgG H&L (IRDye® 800CW) preabsorbed (**ab216772**) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (**ab216777**) at 1/20000 dilution.



Immunocytochemistry/ Immunofluorescence - Human CD44 knockout HeLa cell line (ab262515)

ab254530 staining CD44 in wild-type HeLa cells (top panel) and CD44 knockout HeLa cells (ab262515) (bottom panel). The cells were fixed with 100% methanol (5 min) then permeabilized with 0.1% Tween for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with **ab254530** at 0.4µg/ml concentration and **ab6046** (Rabbit polyclonal to beta Tubulin) at 1/1000 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat secondary antibody to mouse IgG (Alexa Fluor® 488) (**ab150117**) at 2 µg/ml (shown in green) and a goat secondary antibody to rabbit IgG (Alexa Fluor® 594) (**ab150080**) at 2 µg/ml (shown in red). Nuclear DNA was labelled in blue with DAPI.
Image was taken with a confocal microscope (Leica-Microsystems TCS SP8).



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [MEM-263] (**ab9524**) at 2 µg/ml

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD44 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : LNCaP (Human prostate cancer cell line) whole cell lysate

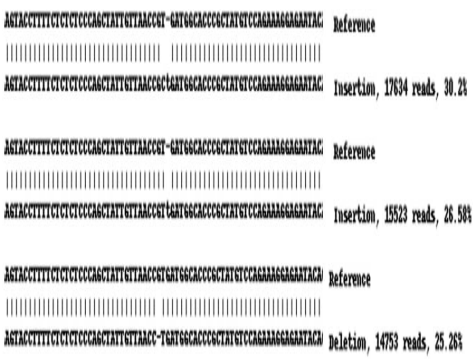
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 80 kDa

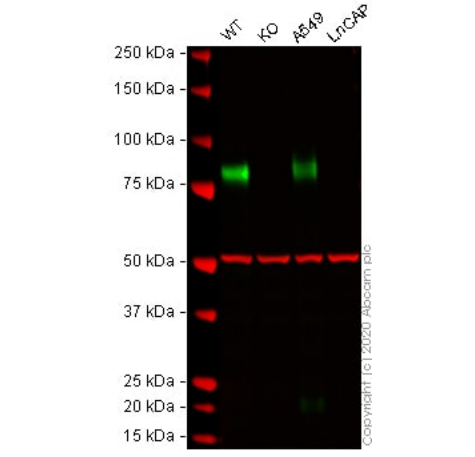
Lanes 1 - 4: Merged signal (red and green). Green - **ab9524** observed at 80 kDa. Red - loading control, **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) observed at 55kDa.

ab9524 was shown to react with CD44 in wild-type HeLa cells in western blot Loss of signal was observed when CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**) was used. Wild-type and CD44 knockout HeLa cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab9524** and **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4°C at 2 µg/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 hour at room temperature before imaging.



Next Generation Sequencing - Human CD44 knockout HeLa cell line (ab262515)

Knockout achieved by CRISPR/Cas9; X = 1 bp insertion, 1 bp deletion; Frameshift = 98.54%



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [BLR038F] (**ab243894**) at 1/1000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD44 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : LNCaP (Human prostate cancer cell line) whole cell lysate

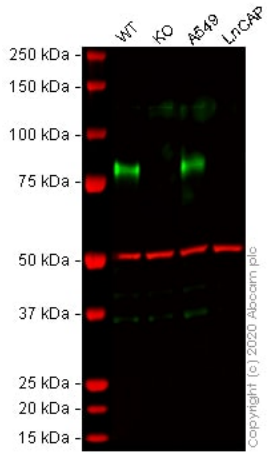
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 80 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab243894** observed at 80 kDa. Red - loading control, **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

ab243894 was shown to react with CD44 in wild-type HeLa cells in western blot Loss of signal was observed when CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**) was used. Wild-type HeLa and CD44 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab243894** 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 hour at room temperature before imaging.



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : HRP Anti-CD44 antibody [EPR1013Y] (**ab194989**) at 1/2500 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD44 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : LNCaP (Human prostate cancer cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

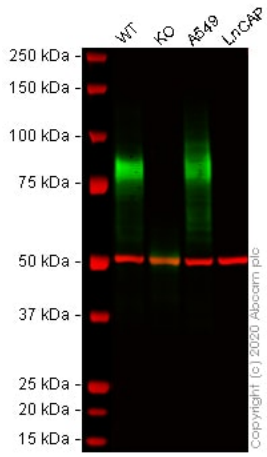
Performed under reducing conditions.

Observed band size: 80 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab194989** observed at 80 kDa. Red - loading control, **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

ab194989 was shown to react with CD44 (HRP) in wild-type HeLa cells in western blot Loss of signal was observed when CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**) was used. Wild-type HeLa and CD44 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in

TBS-T (0.1% Tween®) before incubation with **ab194989** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4°C at a 1 in 2500 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 CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [EPR18668] (**ab189524**) at 1/1000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD44 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : LNCaP (Human prostate cancer cell line) whole cell lysate

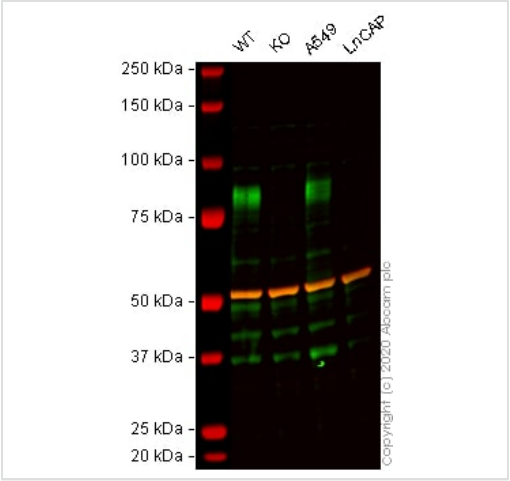
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 80 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab189524** observed at 80 kDa. Red - loading control, **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

ab189524 was shown to react with CD44 in wild-type HeLa cells in western blot Loss of signal was observed when CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**) was used. Wild-type HeLa and CD44 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab189524** 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 hour at room temperature before imaging.



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44v6 antibody [VFF-7] (**ab30436**) at 1/1000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD44 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : LNCaP (Human prostate cancer cell line) whole cell lysate

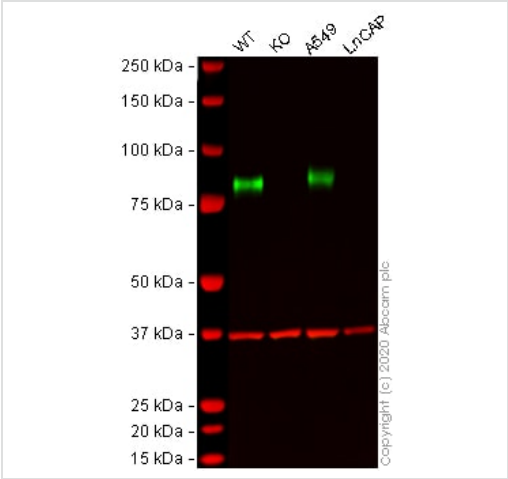
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 80 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab30436** observed at 80 kDa. Red - loading control, **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) observed at 55kDa.

ab30436 was shown to react with CD44v6 in wild-type HeLa cells in western blot Loss of signal was observed when CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**) was used. Wild-type HeLa and CD44 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab30436** and **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4°C at a 1 in 1000 dilution 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 hour at room temperature before imaging.



Western blot - Human CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [SP37] (**ab101531**) at 1/1000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD44 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : LNCaP (Human prostate cancer cell line) whole cell lysate

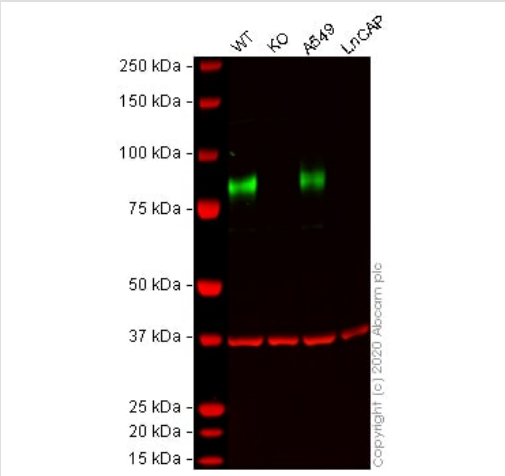
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 80 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab101531** observed at 80 kDa. Red - loading control, **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab101531 was shown to react with CD44 in wild-type HeLa cells in western blot Loss of signal was observed when CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**) was used. Wild-type HeLa and CD44 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab101531** 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 CD44 knockout HeLa cell line (ab262515)

All lanes : Anti-CD44 antibody [EPR1013Y] (**ab51037**) at 1/5000 dilution

Lane 1 : Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : CD44 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 3 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 4 : LNCaP (Human prostate cancer cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Observed band size: 80 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab51037** observed at 80 kDa. Red - loading control, **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab51037 was shown to react with CD44 in wild-type HeLa cells in western blot Loss of signal was observed when CD44 knockout cell line ab262515 (knockout cell lysate **ab263938**) was used. Wild-type HeLa and CD44 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with **ab51037** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 5000 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.

- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.co.jp/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors