

Human S100A4 knockout HeLa cell lysate ab257046

7 图像

概述

产品名称	人S100A4 knockout HeLa cell裂解物
产品概述	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 exon2 and 5 bp deletion in exon2.
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.*

说明

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.](#)

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经测试应用

适用于: Sanger Sequencing, WB

性能

存放说明 Store at -80°C. Please refer to protocols.

组件	1 kit
ab260907 - Human S100A4 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

靶标

组织特异性	Ubiquitously expressed.
序列相似性	Belongs to the S-100 family. Contains 2 EF-hand domains.

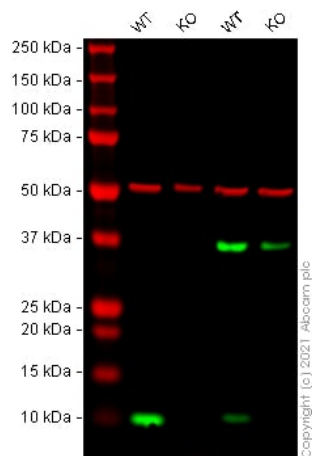
应用

The Abpromise guarantee **Abpromise™**承诺保证使用ab257046于以下的经测试应用

“应用说明”部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Sanger Sequencing		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 12 kDa.

图片



Western blot - Human S100A4 knockout HeLa cell lysate (ab257046)

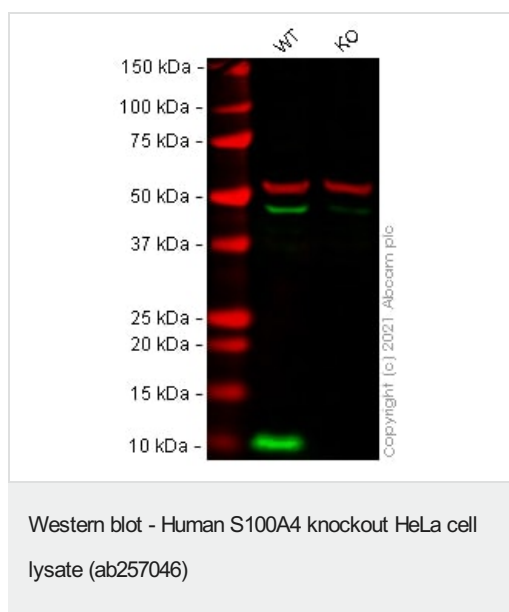
Lane 1: Wild-type HeLa cell lysate 20 µg

Lane 2: S100A4 knockout HeLa cell lysate 20 µg

Lane 3: Wild-type A549 cell lysate 20 µg

Lane 4: S100A4 knockout A549 cell lysate 20 µg

False colour image of Western blot: Anti-S100A4 antibody [EPR14639(2)] staining at 1/1000 dilution, shown in green; loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) staining at 1/20000 dilution, shown in red. In Western blot, **ab197896** was shown to bind specifically to S100A4. A band was observed at 11 kDa in wild-type and A549 cell lysates with no signal observed at this size in S100A4 knockout HeLa cell line **ab265709** (knockout cell lysate ab257046) and S100A4 knockout A549 cell line **ab261865** (knockout cell lysate **ab261674**). To generate this image, wild-type and S100A4 knockout HeLa and S100A4 knockout A549 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.

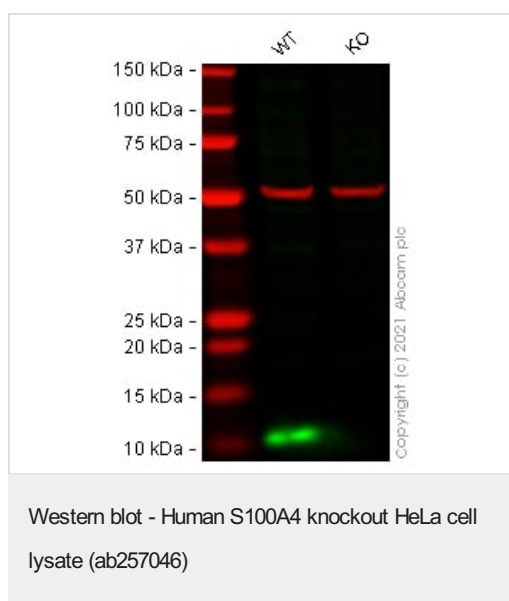


Lane 1: Wild-type HeLa cell lysate 20 µg

Lane 2: S100A4 knockout HeLa cell lysate 20 µg

Lanes 1 - 2: Merged signal (red and green). Green - **ab124805** observed at 11 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

ab124805 was shown to react with S100A4 in wild-type HeLa cells in Western blot with loss of signal observed in S100A4 knockout cell line **ab265709** (S100A4 knockout cell lysate ab257046). Wild-type HeLa and S100A4 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab124805** 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.



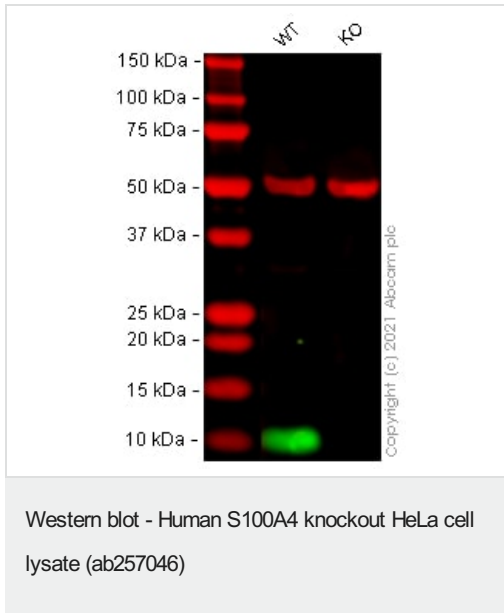
Lane 1: Wild-type HeLa cell lysate µg

Lane 2: S100A4 knockout HeLa cell lysate 20 µg

Lanes 1 - 2: Merged signal (red and green). Green - **ab197896** observed at 11 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

ab197896 was shown to react with S100A4 in wild-type HeLa cells in Western blot with loss of signal observed in S100A4 knockout cell line **ab265709** (S100A4 knockout cell lysate ab257046). Wild-type HeLa and S100A4 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab197896** 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.

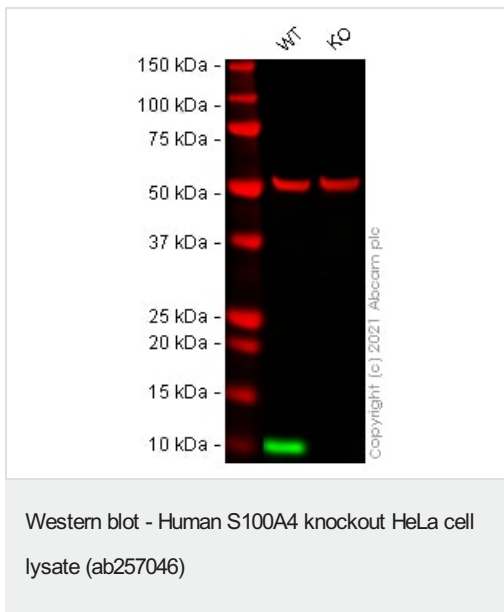


Lane 1: Wild-type HeLa cell lysate 20 µg

Lane 2: S100A4 knockout HeLa cell lysate 20 µg

Lanes 1 - 2: Merged signal (red and green). Green - **ab218511** observed at 11 kDa. Red - loading control **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) observed at 55kDa.

ab218511 was shown to react with S100A4 in wild-type HeLa cells in Western blot with loss of signal observed in S100A4 knockout cell line **ab265709** (S100A4 knockout cell lysate ab257046). Wild-type HeLa and S100A4 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab218511** and **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4 °C at 0.5 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 h at room temperature before imaging.



Lane 1: Wild-type HeLa cell lysate 20 µg

Lane 2: S100A4 knockout HeLa cell lysate 20 µg

Lanes 1 - 2: Merged signal (red and green). Green - **ab218512** observed at 11 kDa. Red - loading control **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) observed at 55kDa.

ab218512 was shown to react with S100A4 in wild-type HeLa cells in Western blot with loss of signal observed in S100A4 knockout cell line **ab265709** (S100A4 knockout cell lysate ab257046). Wild-type HeLa and S100A4 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab218512** and **ab52866** (Rabbit anti-alpha Tubulin antibody [EP1332Y]) overnight at 4 °C at 0.5 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 h at room temperature before imaging.

Mut	CTTGTGAGCTTGAACCTGTCACC-----TGCCCAGTACTTGTGGAAGGTGGACACCAT
WT	CTTGTGAGCTTGAACCTGTCACCCCTTTGCCCGAGTACTTGTGGAAGGTGGACACCAT
Sanger Sequencing - Human S100A4 knockout	
HeLa cell lysate (ab257046)	

Allele-1: 5 bp deletion in exon2

Mut	CTTGTGAGCTTGAACCTGTCACCTCTTTGCCCGAGTACTTGTGGAAGGTGGACACCA
WT	CTTGTGAGCTTGAACCTGTCACCCTTTGCCCGAGTACTTGTGGAAGGTGGACACCA
Sanger Sequencing - Human S100A4 knockout	
HeLa cell lysate (ab257046)	

Allele-2: 1 bp insertion in exon2

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