abcam

Product datasheet

Human USP9X knockout HeLa cell lysate ab257790

画像数 4

製品の概要

製品名 Human USP9X knockout HeLa cell lysate

製品の概要

Knockout cell lysate achieved by CRISPR/Cas9.

Parental Cell Line HeLa

Organism Human

Mutation description Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon9 and 1 bp insertion in exon9.

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.

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アプリケーション 適用あり: WB

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製品の特性

保存方法

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

内容	1 kit
ab262215 - Human USP9X 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

ターゲット情報

機能

Deubiquitinase involved both in the processing of ubiquitin precursors and of ubiquitinated proteins. May therefore play an important role regulatory role at the level of protein turnover by preventing degradation of proteins through the removal of conjugated ubiquitin. Essential component of TGF-beta/BMP signaling cascade. Regulates chromosome alignment and segregation in mitosis by regulating the localization of BIRC5/survivin to mitotic centromeres. Specifically hydrolyzes both 'Lys-29'- and 'Lys-33'-linked polyubiquitins chains. Specifically deubiquitinates monoubiquitinated SMAD4, opposing the activity of E3 ubiquitin-protein ligase

TRIM33.

組織特異性 Widely expressed in embryonic and adult tissues.

配列類似性 Belongs to the peptidase C19 family.

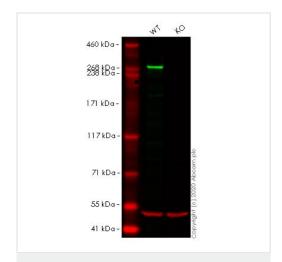
細胞内局在 Cytoplasm.

アプリケーション

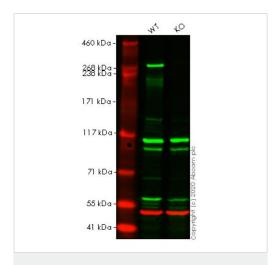
The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab257790の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration. Predicted molecular weight: 292 kDa.

画像



Western blot - Human USP9X knockout HeLa cell lysate (ab257790)



Western blot - Human USP9X knockout HeLa cell lysate (ab257790)

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

Lane 2: USP9X knockout HeLa cell lysate (20µg)

Lanes 1-2: Merged signal (red and green). Green - <u>ab180191</u> observed at 290 kDa. Red - loading control <u>ab7291</u> observed at 50 kDa.

ab180191 Anti-USP9x antibody [EPR13809(B)] - N-terminal was shown to specifically react with USP9x in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265665 (knockout cell lysate ab257790) was used. Wild-type and USP9x knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab180191 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) were incubated overnight at 4°C at 1 in 1000 and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

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

Lane 2: USP9X knockout HeLa cell lysate (20µg)

Lanes 1-2: Merged signal (red and green). Green - <u>ab19879</u> observed at 290 kDa. Red - loading control <u>ab7291</u> observed at 50 kDa.

ab19879 Anti-USP9x antibody was shown to specifically react with USP9x in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab265665 (knockout cell lysate ab257790) was used. Wild-type and USP9x knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab19879 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) were incubated overnight at 4°C at 1 and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-

Mouse IgG H&L (IRDye[®] 680RD) preadsorbed (<u>ab216776</u>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Mut ACTCATCGACATGGTAATCCTGAGGAGGAA-AGTGGCTCACAGCTGAACGAATGGCAGTG

Allele-1: 1 bp deletion in exon9

Sanger Sequencing - Human USP9X knockout HeLa cell lysate (ab257790)

Mut ACTCATCGACATGGTAATCCTGAGGAGGAAAGAGTGGCTCACAGCTGAACGAATGGCAGT
WT ACTCATCGACATGGTAATCCTGAGGAGGAA GAGTGGCTCACAGCTGAACGAATGGCAGT

Allele-2: 1 bp insertion in exon9

Sanger Sequencing - Human USP9X knockout HeLa cell lysate (ab257790)

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