abcam

Product datasheet

Human USP7 (HAUSP) knockout HEK-293T cell lysate ab257284

画像数 4

製品の概要

製品名	Human USP7 (HAUSP) knockout HEK-293T cell lysate
製品の概要	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	HEK293T
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp insertion in exon2 and Insertion of the selection cassette in exon2.
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.*
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. 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**.

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
ab260944 - Human USP7 knockout HEK293T cell lysate	1 x 100µg
ab255553 - Human wild-type HEK293T cell lysate	1 x 100µg

Cell type epithelial

STR Analysis Amelogenin X D5S818: 8, 9 D13S317: 12, 14 D7S820: 11 D16S539: 9, 13 vWA: 16, 19 TH01: 7, 9.3 TPOX: 11 CSF1PO: 11, 12

ターゲット情報

機能 Hydrolase that deubiquitinates target proteins such as FOXO4, p53/TP53, MDM2, ERCC6, DNMT1, UHRF1, PTEN and DAXX (PubMed:11923872, PubMed:15053880, PubMed:16964248, PubMed:18716620, PubMed:25283148). Together with DAXX, prevents MDM2 self-ubiquitination and enhances the E3 ligase activity of MDM2 towards p53/TP53, thereby promoting p53/TP53 ubiquitination and proteasomal degradation. Deubiquitinates p53/TP53, preventing degradation of p53/TP53, and enhances p53/TP53-dependent transcription regulation, cell growth repression and apoptosis (PubMed:25283148). Deubiquitinates p53/TP53 and MDM2 and strongly stabilizes p53/TP53 even in the presence of excess MDM2, and also induces p53/TP53-dependent cell growth repression and apoptosis. Deubiquitination of FOXO4 in presence of hydrogen peroxide is not dependent on p53/TP53 and inhibits FOXO4-induced transcriptional activity. In association with DAXX, is involved in the deubiquitination and translocation of PTEN from the nucleus to the cytoplasm, both processes that are counteracted by PML. Involved in cell proliferation during early embryonic development. Involved in transcription-coupled nucleotide excision repair (TC-NER) in response to UV damage: recruited to DNA damage sites following interaction with KIAA1530/UVSSA and promotes deubiquitination of ERCC6, preventing UV-induced degradation of ERCC6. Contributes to the overall stabilization and trans-activation capability of the herpesvirus 1 trans-acting transcriptional protein ICP0/VMW110 during HSV-1 infection. Involved in maintenance of DNA methylation via its interaction with UHRF1 and DNMT1: acts by mediating deubiquitination of UHRF1 and DNMT1, preventing their degradation and promoting DNA methylation by DNMT1 (PubMed:21745816). Exhibits a preference towards 'Lys-48'-linked ubiquitin chains. Increases regulatory T-cells (Treg) suppressive capacity by deubiquitinating and stabilizing the transcription factor FOXP3 which is crucial for Treg cell function (PubMed:23973222).

組織特異性 Widely expressed. Overexpressed in prostate cancer.

配列類似性 Belongs to the peptidase C19 family.
Contains 1 MATH domain.
Contains 1 USP domain.

ドメイン The C-terminus plays a role in its oligomerization.

翻訳後修飾 Isoform 1: Phosphorylated. Isoform 1 is phosphorylated at positions Ser-18 and Ser-963. Isoform 2: Not phosphorylated.

	Isoform 1: Polyneddylated. Isoform 2: Not Polyneddylated. Isoform 1 and isoform 2: Not sumoylated. Isoform 1 and isoform 2: Polyubiquitinated by herpesvirus 1 trans-acting transcriptional protein ICP0/VMW110; leading to its subsequent proteasomal degradation. Isoform 1: Ubiquitinated at Lys-869.
細胞内局在	Nucleus. Cytoplasm. Nucleus, PML body. Present in a minority of ND10 nuclear bodies. Association with ICP0/VMW110 at early times of infection leads to an increased proportion of USP7-containing ND10. Colocalizes with ATXN1 in the nucleus. Colocalized with DAXX in speckled structures. Colocalized with PML and PTEN in promyelocytic leukemia protein (PML) nuclear bodies.

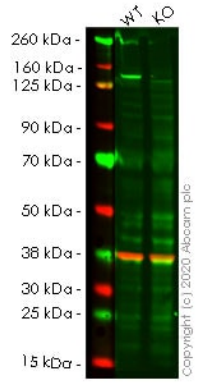
アプリケーション

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ab257284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: 128 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.

画像



Western blot - Human USP7 (HAUSP) knockout HEK293T cell lysate (ab257284)

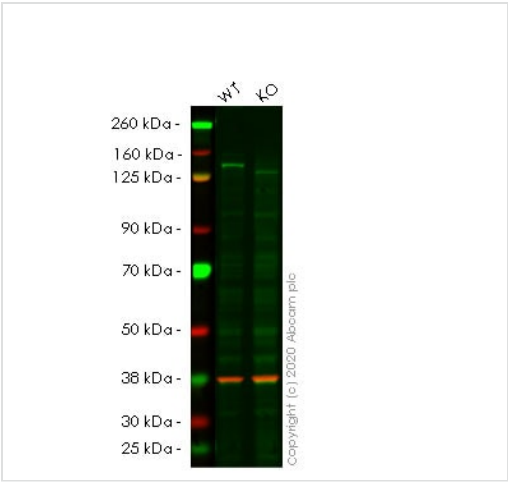
Lane 1: Wild-type HEK-293T cell lysate (20µg)

Lane 2: USP7 knockout HEK-293T cell lysate (20µg)

Lanes 1- 2: Merged signal (red and green). Green - **ab109109** observed at 128 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab109109 Anti-HAUSP / USP7 antibody [EPR4254] was shown to specifically react with HAUSP / USP7 in wild-type HEK-293T cells in western blot. The band observed in the knockout cell line **ab266535** (knockout cell lysate ab257284) lane below 128kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type and HAUSP / USP7 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. **ab109109** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 and 1 in 20000 dilution respectively. Blots were developed with Goat anti-

Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human USP7 (HAUSP) knockout HEK293T cell lysate (ab257284)

Lane 1: Wild-type HEK-293T cell lysate (20µg)

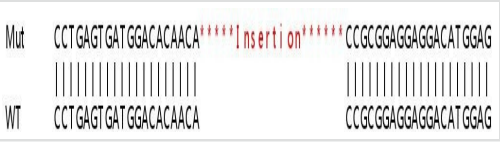
Lane 2: USP7 knockout HEK-293T cell lysate (20µg)

Lanes 1- 2: Merged signal (red and green). Green - **ab108931** observed at 128 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab108931 Anti-HAUSP / USP7 antibody [EPR4253] was shown to specifically react with HAUSP / USP7 in wild-type HEK-293T cells in western blot. The band observed in the knockout cell line **ab266535** (knockout cell lysate ab257284) lane below 128kDa may represent truncated forms and cleaved fragments. This has not been investigated further. Wild-type and HAUSP / USP7 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. **ab108931** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Sanger Sequencing - Human USP7 knockout HEK293T cell lysate (ab257284)

Allele-1: 1 bp insertion in exon2



Sanger Sequencing - Human USP7 knockout HEK293T cell lysate (ab257284)

Allele-2: Insertion of the selection cassette in exon2

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