

Human HTT (Huntingtin) knockout HeLa cell lysate ab256946

画像数 3

製品の概要

製品名	Human HTT (Huntingtin) knockout HeLa 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	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon21.
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.](#)

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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
ab262819 - Human HTT 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 May play a role in microtubule-mediated transport or vesicle function.

組織特異性 Expressed in the brain cortex (at protein level). Widely expressed with the highest level of expression in the brain (nerve fibers, varicosities, and nerve endings). In the brain, the regions where it can be mainly found are the cerebellar cortex, the neocortex, the striatum, and the hippocampal formation.

関連疾患 Defects in HTT are the cause of Huntington disease (HD) [MIM:143100]. HD is an autosomal dominant neurodegenerative disorder characterized by involuntary movements (chorea), general motor impairment, psychiatric disorders and dementia. Onset of the disease occurs usually in the third or fourth decade of life and symptoms progressively worsen leading to death in 10 to 20 years. Onset and clinical course depend on the degree of poly-Gln repeat expansion, longer expansions resulting in earlier onset and more severe clinical manifestations. HD affects 1 in 10,000 individuals of European origin. Neuropathology of Huntington disease displays a distinctive pattern with loss of neurons, especially in the caudate and putamen (striatum).

配列類似性 Belongs to the huntingtin family.
Contains 10 HEAT repeats.

ドメイン The N-terminal Gln-rich and Pro-rich domain has great conformational flexibility and is likely to exist in a fluctuating equilibrium of alpha-helical, random coil, and extended conformations.

翻訳後修飾 Cleaved by apopain downstream of the polyglutamine stretch. The resulting N-terminal fragment is cytotoxic and provokes apoptosis.
Forms with expanded polyglutamine expansion are specifically ubiquitinated by SYVN1, which promotes their proteasomal degradation.

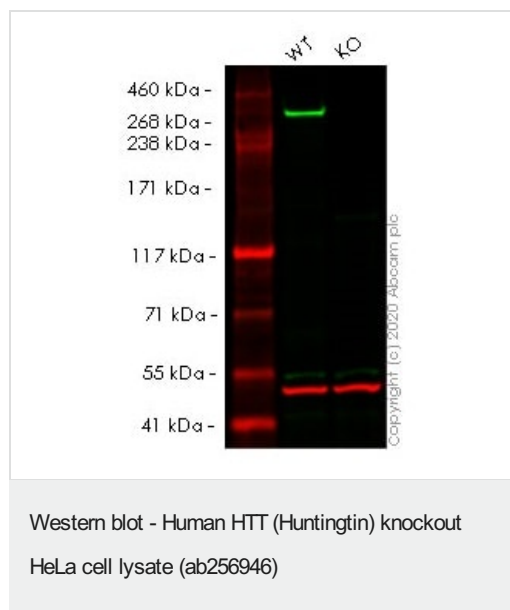
細胞内局在 Cytoplasm. Nucleus. The mutant Huntingtin protein colocalizes with AKAP8L in the nuclear matrix of Huntington's disease neurons.

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ab256946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. 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

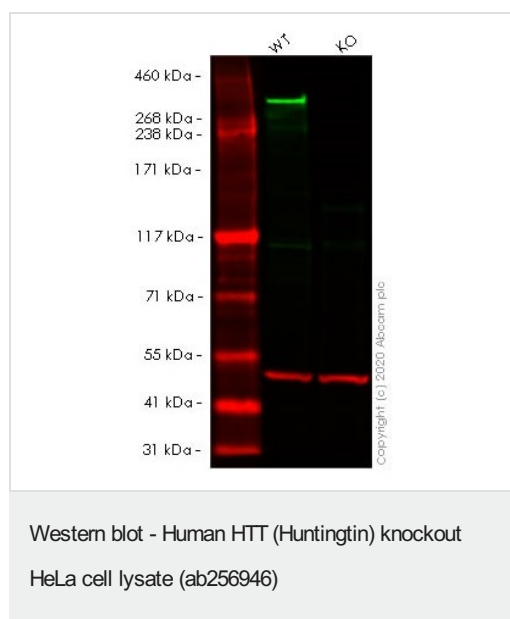


Lane 1: Wild-type HeLa cell lysate (20µg)

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

Lanes 1- 2: Merged signal (red and green). Green - **ab109115** observed at 348 kDa. Red - loading control **ab7291** observed at 50 kDa.

ab109115 Anti-Huntingtin antibody [EPR5526] was shown to specifically react with Huntingtin in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab265976** (knockout cell lysate ab256946) was used. Wild-type and Huntingtin 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. **ab109115** and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) were incubated overnight at 4 °C at 1 in 10000 dilution 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.



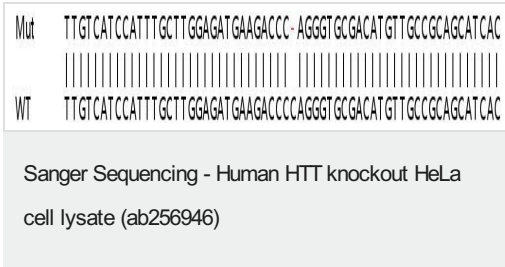
Lane 1: Wild-type HeLa cell lysate (20µg)

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

Lanes 1- 2: Merged signal (red and green). Green - **ab45169** observed at 348 kDa. Red - loading control **ab7291** observed at 50 kDa.

ab45169 Anti-Huntingtin antibody [EP867Y] was shown to specifically react with Huntingtin in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab265976** (knockout cell lysate ab256946) was used. Wild-type and Huntingtin 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. **ab45169** and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) were incubated overnight at 4 °C at 1 in 10000 dilution 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.



Homozygous: 1 bp deletion in exon21

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