abcam

Product datasheet

Anti-Huntingtin antibody [EP867Y] ab45169

KO 評価済 リコンビナント RabMAb

5 References 画像数 7

製品の概要

製品名	Anti-Huntingtin antibody [EP867Y]
製品の詳細	Rabbit monoclonal [EP867Y] to Huntingtin
由来種	Rabbit
アプリケーション	**適用あり:** Flow Cyt (Intra), WB, IHC-P, ICC/IF **適用なし:** IP
種交差性	**交差種:** Mouse, Human **交差が予測される動物種:** Rat
免疫原	Synthetic peptide within Human Huntingtin aa 550-650. The exact sequence is proprietary. Corresponding to residues specific to the apopain cleavage site.
ポジティブ・コントロール	ICC/IF: SK-N-SH cells. WB: HAP1, SH-SY5Y and HeLa whole cell lysates. IHC-P: Human brain tissue. Flow Cyt (intra): SH-SY5Y cells.
特記事項	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
バッファー	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture supernatant
精製度	Protein A purified

ポリ/モノ	モノクローナル
クローン名	EP867Y
アイソタイプ	IgG

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ab45169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
Flow Cyt (Intra)		1/100. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
WB		1/10000. Predicted molecular weight: 348 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		1/50 - 1/100.

追加情報 Is unsuitable for IP.

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).

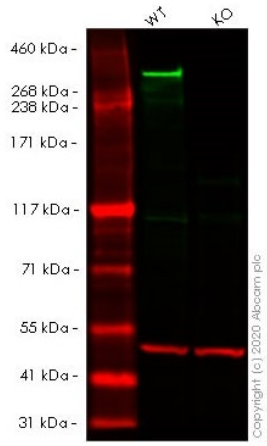
配列類似性 Belongs to the huntingtin family.
Contains 10 HEAT repeats.

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.

画像



Western blot - Anti-Huntingtin antibody [EP867Y] (ab45169)

All lanes : Anti-Huntingtin antibody [EP867Y] (ab45169) at 1/10000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : HTT knockout HeLa cell lysate

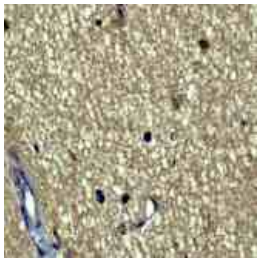
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 348 kDa
Observed band size: 348 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab45169 observed at 348 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) observed at 50 kDa.

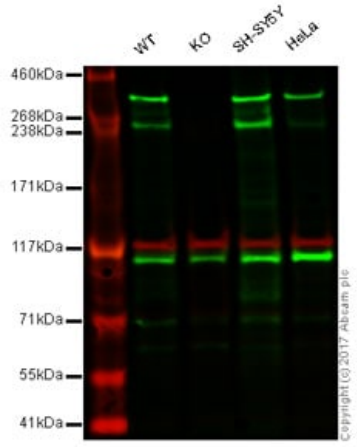
ab45169 was shown to react with Huntingtin in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab265976** (knockout cell lysate **ab256946**) was used. Wild-type HeLa and HTT knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab45169 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (**ab7291**) overnight at 4°C at a 1 in 10000 dilution and a 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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Huntingtin antibody [EP867Y] (ab45169)

Ab45169 staining human Huntingtin in human brain tissue by immunohistochemistry using paraffin embedded tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-Huntingtin antibody [EP867Y] (ab45169)

All lanes : Anti-Huntingtin antibody [EP867Y] (ab45169) at 1/10000 dilution

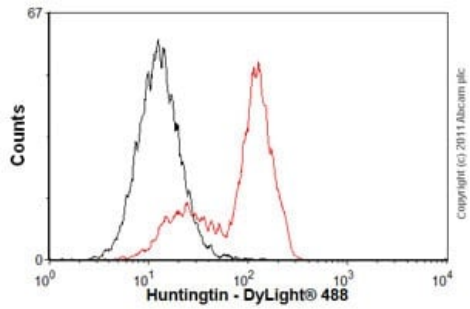
Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : Huntingtin knockout HAP1 whole cell lysate
Lane 3 : SH-SY5Y whole cell lysate
Lane 4 : HeLa whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 348 kDa

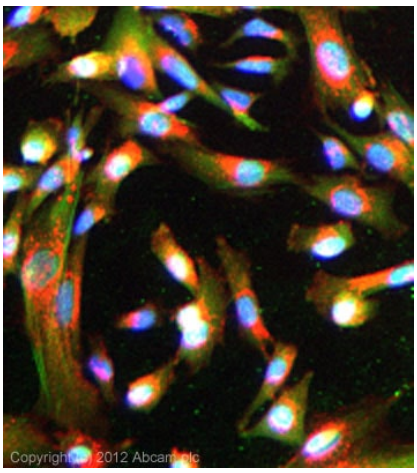
Lanes 1 - 4: Merged signal (red and green). Green - ab45169 observed at 348 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

ab45169 was shown to specifically recognize Huntingtin in wild-type HAP1 cells along with additional cross reactive bands. No band was observed when Huntingtin knockout samples were examined. Wild-type and Huntingtin knockout samples were subjected to SDS-PAGE. ab45169 and **ab18058** (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/10,000 dilution and 1/10,000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed **ab216777** secondary antibodies at 1/10,000 dilution for 1 hour at room temperature before imaging.



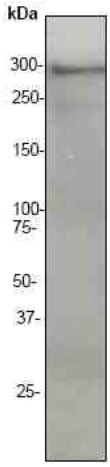
Flow Cytometry (Intracellular) - Anti-Huntingtin antibody [EP867Y] (ab45169)

Overlay histogram showing SH-SY5Y cells stained with ab45169 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab45169, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (**ab96899**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in SH-SY5Y cells fixed with 4% paraformaldehyde/permeabilized in 0.1% PBS-Tween used under the same conditions.



Immunocytochemistry/ Immunofluorescence - Anti-Huntingtin antibody [EP867Y] (ab45169)

ICC/IF image of ab45169 stained SKNSH cells. The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab45169, 1/200 dilution) overnight at +4°C. The secondary antibody (green) was **ab96899**, DyLight® 488 goat anti-rabbit IgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Western blot - Anti-Huntingtin antibody [EP867Y] (ab45169)

Anti-Huntingtin antibody [EP867Y] (ab45169) at 1/10000 dilution + SH-SY-5Y cell lysate

Predicted band size: 348 kDa

Observed band size: 300 kDa

Anti-Huntingtin antibody [EP867Y] (ab45169)

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery

- Response to your inquiry within 24 hours
- 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