abcam

Product datasheet

Anti-TDP43 antibody [EPR5810] ab109535



ייבער RabMAb

★★★★★ 1 Abreviews 12 References 画像数 14

製品の概要

製品名 Anti-TDP43 antibody [EPR5810]

製品の詳細 Rabbit monoclonal [EPR5810] to TDP43

由来種 Rabbit

アプリケーション 適用あり: ICC/IF, IHC-Fr, WB, IHC-P, Flow Cyt (Intra)

適用なし: IP

種交差性 交差種: Mouse, Rat, Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: HAP1, HeLa, Jurkat, 293T, K562, and A431 cell lysates, Mouse and Rat brain lysates; IHC-

> Fr: Mouse cerebrum tissue, Human prostate carcinoma IHC-P: Human papillary carcinoma and glioma tissue, Mouse and Rat cerebrum tissues; ICC/IF: HAP1-TARDBP, Hek293 and HeLa

cells; Flow Cyt (intra): K562 cells.

特記事項 TARDBP is a protein encoded by the TARDBP gene. A hyper-phosphorylated, ubiquitinated and

cleaved formof TARDBP, known as TDP-43 is the significant protein in several diseases,

including amyotrophic lateral sclerosis (ALS).

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. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

精製度 Protein A purified

ポリ/モノ モノクローナル

クローン名 EPR5810

アイソタイプ lgG

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ab109535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
ICC/IF		Use a concentration of 1 μ g/ml. This antibody is suitable to detect TDP43 using MeOH fixation in ICC. We have compared methanol and paraformaldehyde (PFA) fixation methods with this product and recommend to use methanol only.
IHC-Fr		1/50. Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20)
WB		1/1000 - 1/10000. Predicted molecular weight: 45 kDa.
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
Flow Cyt (Intra)		Use at an assay dependent concentration.

追加情報 Is unsuitable for IP.

ターゲット情報

機能 DNA and RNA-binding protein which regulates transcription and splicing. Involved in the

regulation of CFTR splicing. It promotes CFTR exon 9 skipping by binding to the UG repeated motifs in the polymorphic region near the 3'-splice site of this exon. The resulting aberrant splicing is associated with pathological features typical of cystic fibrosis. May also be involved in microRNA biogenesis, apoptosis and cell division. Can repress HIV-1 transcription by binding to

the HIV-1 long terminal repeat. Stabilizes the low molecular weight neurofilament (NFL) mRNA

through a direct interaction with the 3' UTR.

組織特異性 Ubiquitously expressed. In particular, expression is high in pancreas, placenta, lung, genital tract

and spleen.

関連疾患 Defects in TARDBP are the cause of amyotrophic lateral sclerosis type 10 (ALS10)

[MIM:612069]. ALS is a neurodegenerative disorder affecting upper and lower motor neurons and resulting in fatal paralysis. Sensory abnormalities are absent. Death usually occurs within 2 to 5 years. The etiology of ALS is likely to be multifactorial, involving both genetic and environmental

factors. The disease is inherited in 5-10% of the cases.

配列類似性 Contains 2 RRM (RNA recognition motif) domains.

ドメイン The RRM domains can bind to both DNA and RNA.

翻訳後修飾 Hyperphosphorylated in hippocampus, neocortex, and spinal cord from individuals affected with

ALS and FTLDU.

 $Ubiquitinated\ in\ hippocampus,\ neocortex,\ and\ spinal\ cord\ from\ individuals\ affected\ with\ ALS\ and$

FTLDU.

Cleaved to generate C-terminal fragments in hippocampus, neocortex, and spinal cord from

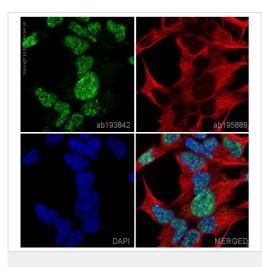
individuals affected with ALS and FTLDU.

細胞内局在 Nucleus. In patients with frontotemporal lobar degeneration and amyotrophic lateral sclerosis, it is

absent from the nucleus of affected neurons but it is the primary component of cytoplasmic

ubiquitin-positive inclusion bodies.

画像

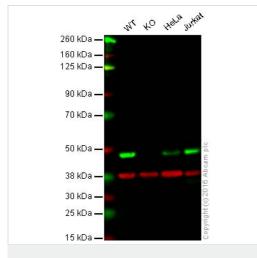


Immunocytochemistry/ Immunofluorescence - Anti-TDP43 antibody [EPR5810] (ab109535)

ab193842 staining TDP43 in Hek293 cells. The cells were fixed with 100% methanol (5 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated overnight at +4°C with **ab193842** at a 1/250 dilution (shown in green) and **ab195889**, Mouse monoclonal to alpha Tubulin (Alexa Fluor® 594), at a 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

This antibody is not suitable to detect TDP43 using PFA fixation in ICC.



Western blot - Anti-TDP43 antibody [EPR5810] (ab109535)

Lane 1: Wild-type HAP1 cell lysate (40 μg)

Lane 2: TDP43 knockout HAP1 cell lysate (40 μg)

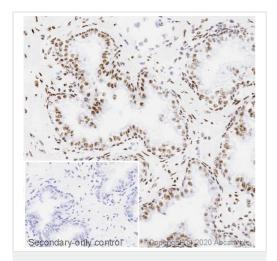
Lane 3: HeLa cell lysate (40 µg)

Lane 4: Jurkat cell lysate (40 µg)

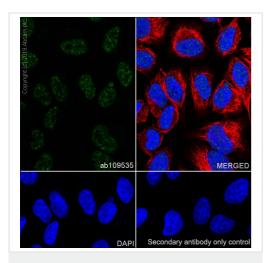
Lanes 1 - 4: Merged signal (red and green). Green - ab109535 observed at 48 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

Unpurified ab 109535 was shown to specifically react with TDP43 when TDP43 knockout samples were used. Wild-type and TDP43 knockout samples were subjected to SDS-PAGE. Ab 109535 and ab8245 (loading control to GAPDH) were diluted at 1/1000 and 1/10,000 dilution respectively and incubated overnight at 4C. Blots were developed with IRDye® 800CW Goat anti-Rabbit IgG (H + L) and IRDye® 680 Goat anti-Mouse IgG (H + L) secondary antibodies

at 1/10,000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Frozen sections) - Anti-TDP43 antibody [EPR5810] (ab109535)



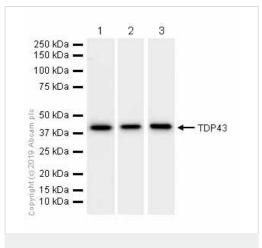
Immunocytochemistry/ Immunofluorescence - Anti-TDP43 antibody [EPR5810] (ab109535)

IHC image of TDP43 staining in a section of frozen human prostate carcinoma performed on a Leica BONDTM system using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab109535, 1/100 dilution, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset secondary-only control image is taken from an identical assay without primary antibody.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling TDP43 with purified ab109535 at 1/50 dilution (6.2 μ g/ml). Cells were fixed in 100% Methanol and permeabilized with None. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 μ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 μ g/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

This antibody is not suitable to detect TDP43 using PFA fixation in ICC.



Western blot - Anti-TDP43 antibody [EPR5810] (ab109535)

All lanes : Anti-TDP43 antibody [EPR5810] (ab109535) at 1/5000 dilution (Purified)

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates

Lane 2 : Mouse brain lysates

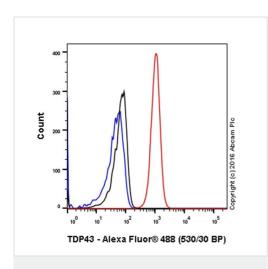
Lane 3 : Rat brain lysates

Lysates/proteins at 15 µg per lane.

Secondary

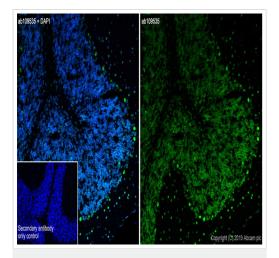
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 45 kDa **Observed band size:** 45 kDa



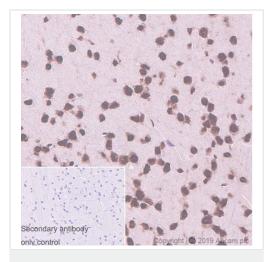
Flow Cytometry (Intracellular) - Anti-TDP43 antibody [EPR5810] (ab109535)

Intracellular Flow Cytometry analysis of K562 (human chronic myelogenous leukemia) cells labeling TDP43 with purified ab109535 at 1/20 dilution (10ug/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit lgG (Alexa Fluorr[®] 488) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal lgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



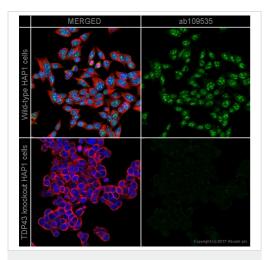
Immunohistochemistry (Frozen sections) - Anti-TDP43 antibody [EPR5810] (ab109535)

Immunohistochemistry (Frozen sections) analysis of mouse cerebrum tissue sections labeling TDP43 with Purified unpurified ab109535 at 1/50 (0.5 µg/ml). Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20). Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. DAPI was used as a counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TDP43 antibody
[EPR5810] (ab109535)

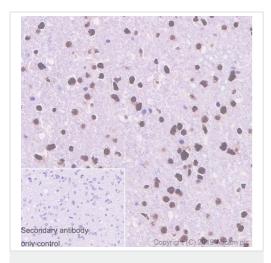
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse cerebrum tissue sections labeling TDP43 with purified ab109535 at 1/100 dilution (0.3 µg/ml). Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0). Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunocytochemistry/ Immunofluorescence - Anti-TDP43 antibody [EPR5810] (ab109535)

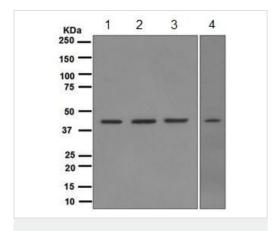
Unpurified ab109535 staining TDP43 in wild-type HAP1 cells (top panel) and TDP43 knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab109535 at 1 μ g/ml concentration and <u>ab195889</u> at 1/250 dilution (shown in pseudo colour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit lgG (Alexa Fluor® 488) (<u>ab150081</u>) at 2 μ g/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

This antibody is not suitable to detect TDP43 using PFA fixation in ICC.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TDP43 antibody
[EPR5810] (ab109535)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human glioma tissue sections labeling TDP43 with purified ab109535 at 1/100 dilution (0.3 µg/ml). Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0). Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Western blot - Anti-TDP43 antibody [EPR5810] (ab109535)

All lanes : Anti-TDP43 antibody [EPR5810] (ab109535) at 1/1000 dilution ((unpurified))

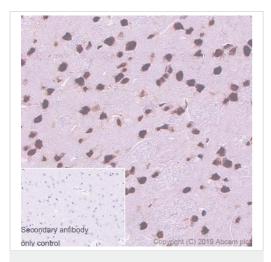
Lane 1: HeLa cell lysate
Lane 2: 293T cell lysate
Lane 3: K562 cell lysate
Lane 4: A431 cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP-labelled goat anti-rabbit at 1/2000 dilution

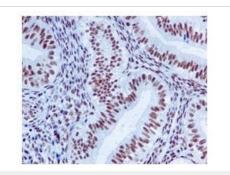
Predicted band size: 45 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TDP43 antibody
[EPR5810] (ab109535)

Immunohistochemical analysis of paraffin-embedded Rat cerebrum tissue labeling TDP43 with unpurified ab109535, followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Nuclear staining on rat cerebrum. The section was incubated with ab229902 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2)

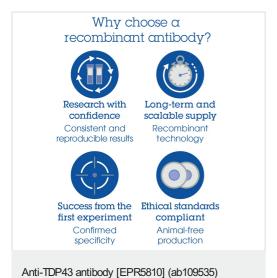
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TDP43 antibody
[EPR5810] (ab109535)

Unpurified ab109535 at 1/100 dilution staining TARDBP in paraffinembedded Human papillary carcinoma tissue by Immunohistochemistry.

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



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