Revision 4

Store at -20C

#12456

GSK-3β (D5C5Z) XP® Rabbit mAb

Cell Signaling TECHNOLOGY®

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Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
WB, IP, IHC-P, IF-IC, F	H M R Mk	Endogenous	46	Rabbit IgG	P49841	2932

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:50
Immunohistochemistry (Paraffin)	1:400 - 1:1600
Immunofluorescence (Immunocytochemistry)	1:200 - 1:800
Flow Cytometry	1:100 - 1:400

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.

Specificity / Sensitivity

GSK-3β (D5C5Z) XP® Rabbit mAb recognizes endogenous levels of total GSK-3β protein. This antibody does not cross-react with GSK-3α protein.
Species Reactivity:
Human, Mouse, Rat, Monkey

Source / Purification

Monoclonal antibody is produced by immunizing animals with recombinant protein specific to the carboxy terminus of human GSK-3β protein.

Background

Glycogen synthase kinase-3 (GSK-3) was initially identified as an enzyme that regulates glycogen synthesis in response to insulin (1). GSK-3 is a ubiquitously expressed serine/threonine protein kinase that phosphorylates and inactivates glycogen synthase. GSK-3 is a critical downstream element of the PI3K/Akt cell survival pathway whose activity can be inhibited by Akt-mediated phosphorylation at Ser21 of GSK-3α and Ser9 of GSK-3β (2,3). GSK-3 has been implicated in the regulation of cell fate in *Dictyostelium* and is a component of the Wnt signaling pathway required for *Drosophila, Xenopus,* and mammalian development (4). GSK-3 has been shown to regulate cyclin D1 proteolysis and subcellular localization (5).

1. Welsh, G.I. et al. (1996) *Trends Cell Biol* 6, 274-9.
2. Srivastava, A.K. and Pandey, S.K. (1998) *Mol Cell Biochem* 182, 135-41.
3. Cross, D.A. et al. (1995) *Nature* 378, 785-9.
4. Nusse, R. (1997) *Cell* 89, 321-3.
5. Diehl, J.A. et al. (1998) *Genes Dev* 12, 3499-511.

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

APPLICATIONS KEY **WB:** Western Blot **IP:** Immunoprecipitation **IHC:** Immunohistochemistry **ChIP:** Chromatin Immunoprecipitation **IF:** Immunofluorescence **F:** Flow Cytometry **E-P:** ELISA-Peptide

CROSS-REACTIVITY KEY **H:** human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **Mi:** mink **C:** chicken **Dm:** D. melanogaster **X:** Xenopus **Z:** zebrafish **B:** bovine **Dg:** dog **Pg:** pig **Sc:** S. cerevisiae **Ce:** C. elegans **Hr:** horse **All:** all species expected

Revision 4

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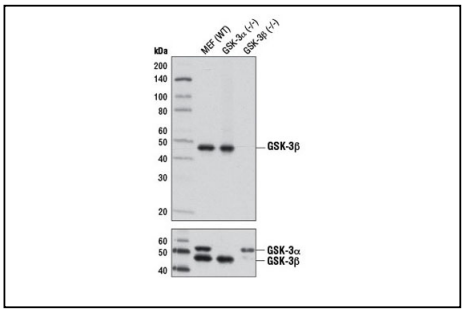
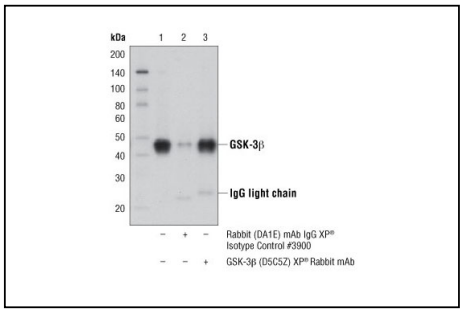
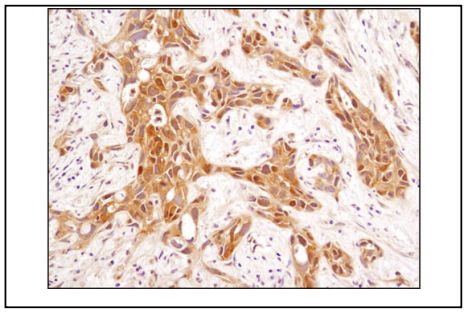
GSK-3β (D5C5Z) XP® Rabbit mAb



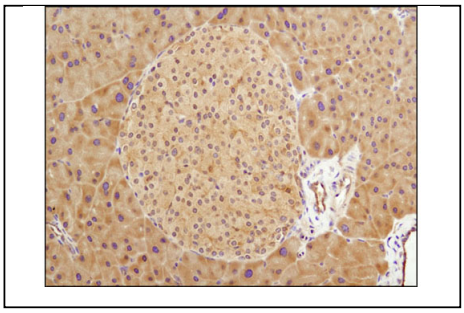
Western blot analysis of extracts from wild-type, GSK-3α (-/-), and GSK3β (-/-) mouse embryonic fibroblasts (MEFs) using GSK-3β (D5C5Z) XP® Rabbit mAb (upper) and GSK-3α/β (D75D3) XP® Rabbit mAb #5676 (lower). (MEF wild type, GSK-3α (-/-), and GSK-3β (-/-) cells were kindly provided by Dr. Jim Woodgett, University of Toronto, Canada).



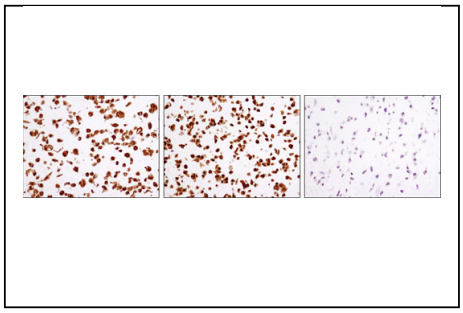
Immunoprecipitation of GSK-3β from PC-12 cell extracts, using Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (lane 2) or GSK-3β (D5C5Z) XP® Rabbit mAb (lane 3). Lane 1 is 10% input. Western blot analysis was performed using GSK-3β (D5C5Z) XP® Rabbit mAb.



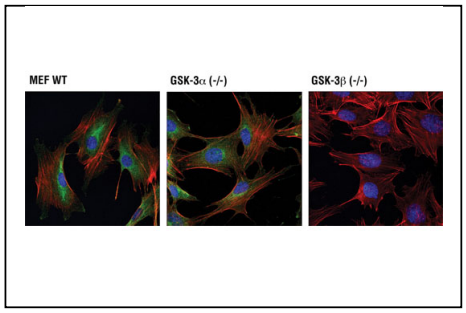
Immunohistochemical analysis of paraffin-embedded human breast carcinoma using GSK-3β (D5C5Z) XP® Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded mouse pancreas using GSK-3β (D5C5Z) XP® Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded MEF cell pellets, wild type (left), GSK-3α (-/-) (middle) and GSK-3β (-/-) (right) using GSK-3β (D5C5Z) XP® Rabbit mAb. (MEF wild type, GSK-3β (-/-), and GSK-3α (-/-) cells were kindly provided by Dr. Jim Woodgett, University of Toronto, Canada).



Confocal immunofluorescent analysis of wild-type mouse embryonic fibroblasts (MEFs) (left), GSK-3α (-/-) MEFs (center)and GSK-3β (-/-) MEFs (right) using GSK-3β (D5C5Z) XP® Rabbit mAb (green). Actin filaments were labeled with DY-554 phalloidin (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye). (MEF wild type, GSK-3α (-/-), and GSK-3β (-/-) cells were kindly provided by Dr. Jim Woodgett, University of Toronto, Canada).

Revision 4

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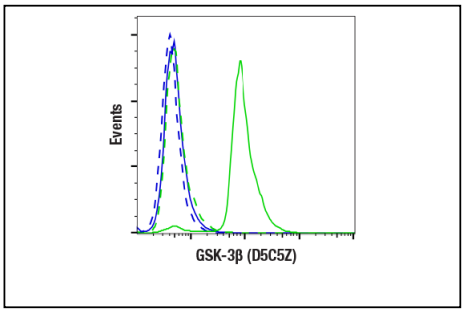
GSK-3β (D5C5Z) XP® Rabbit mAb



Flow cytometric analysis of GSK-3β (-/-) MEFs (blue, negative) and wild type mouse embryonic fibroblasts (MEFs) (green, positive) using GSK-3β (D5C5Z) XP® Rabbit mAb or a concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (dashed lines). Anti-rabbit IgG (H+L), F(ab')2 Fragment (Alexa Fluor® 488 Conjugate) #4412 was used as a secondary antibody. (MEF wild type and GSK-3β (-/-) cells were kindly provided by Dr. Jim Woodgett, University of Toronto, Canada).

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GSK-3β (D5C5Z) XP® Rabbit mAb

限制使用

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Revision 1

Store at -20C

#5558

Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb

Cell Signaling TECHNOLOGY®

Orders: 877-616-CELL (2355)
orders@cellsignal.com

Support: 877-678-TECH (8324)

Web: info@cellsignal.com
www.cellsignal.com

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For Research Use Only. Not For Use In Diagnostic Procedures.

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
WB, IP, IF-IC, F	H M R Hm	Endogenous	46	Rabbit IgG	P49841	2932

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:50
Immunofluorescence (Immunocytochemistry)	1:400
Flow Cytometry	1:200

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.

Specificity / Sensitivity

Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb detects endogenous levels of GSK-3β only when phosphorylated at Ser9. This antibody reacts with denatured components of bovine serum, including BSA.
Species Reactivity:
Human, Mouse, Rat, Hamster

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser9 of human GSK-3β.

Background

Glycogen synthase kinase-3 (GSK-3) was initially identified as an enzyme that regulates glycogen synthesis in response to insulin (1). GSK-3 is a ubiquitously expressed serine/threonine protein kinase that phosphorylates and inactivates glycogen synthase. GSK-3 is a critical downstream element of the PI3K/Akt cell survival pathway whose activity can be inhibited by Akt-mediated phosphorylation at Ser21 of GSK-3α and Ser9 of GSK-3β (2,3). GSK-3 has been implicated in the regulation of cell fate in *Dictyostelium* and is a component of the Wnt signaling pathway required for *Drosophila, Xenopus,* and mammalian development (4). GSK-3 has been shown to regulate cyclin D1 proteolysis and subcellular localization (5).

1. Welsh, G.I. et al. (1996) *Trends Cell Biol* 6, 274-9.
2. Srivastava, A.K. and Pandey, S.K. (1998) *Mol Cell Biochem* 182, 135-41.
3. Cross, D.A. et al. (1995) *Nature* 378, 785-9.
4. Nusse, R. (1997) *Cell* 89, 321-3.
5. Diehl, J.A. et al. (1998) *Genes Dev* 12, 3499-511.

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

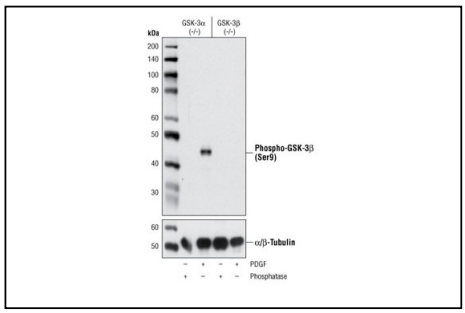
APPLICATIONS KEY **WB:** Western Blot **IP:** Immunoprecipitation **IHC:** Immunohistochemistry **ChIP:** Chromatin Immunoprecipitation **IF:** Immunofluorescence **F:** Flow Cytometry **E-P:** ELISA-Peptide

CROSS-REACTIVITY KEY **H:** human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **Mi:** mink **C:** chicken **Dm:** D. melanogaster **X:** Xenopus **Z:** zebrafish **B:** bovine **Dg:** dog **Pg:** pig **Sc:** S. cerevisiae **Ce:** C. elegans **Hr:** horse **All:** all species expected

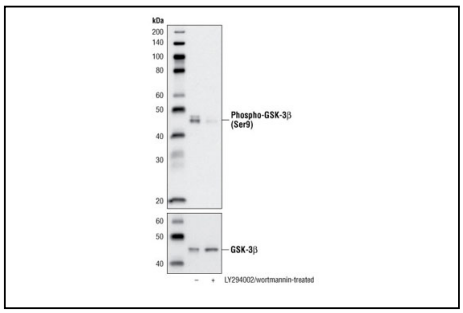
Revision 1

#5558

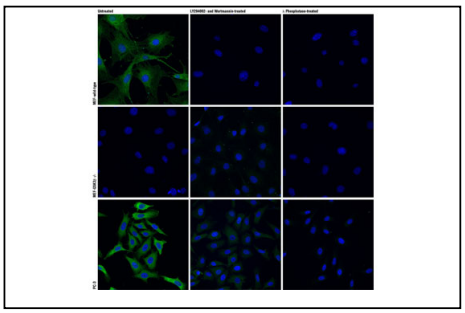
Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb



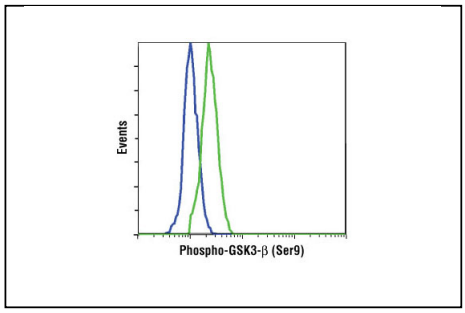
Western blot analysis of extracts from GSK-3α (-/-) (lanes 1,2) and GSK-3β (-/-) (lanes 3,4) mouse embryonic fibroblast (MEF) cells, λ phosphatase or PDGF-treated, using Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb (upper) and α/β-Tubulin Antibody #2148 (lower). (MEF wild type, GSK-3α (-/-) and GSK-3β (-/-) cells were kindly provided by Dr. Jim Woodgett, University of Toronto, Canada).



Western blot analysis of extracts from PC-3 cells, untreated or LY294002/wortmannin-treated, using Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb (upper) or GSK-3β (27C10) Rabbit mAb #9315 (lower).



Confocal immunofluorescent analysis of wild type mouse embryonic fibroblasts (MEFs) (top row), GSK-3β (-/-) MEFs (middle row) , or PC-3 cells (bottom row), untreated (left), LY294002- and Wortmannin-treated (#9901 and #9951 respectively; center) or lambda phosphatase-treated (right), using Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb (green). Actin filaments were labeled with DY-554 phalloidin (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye). (MEF wild type and GSK-3β (-/-) cells were kindly provided by Dr. Jim Woodgett, University of Toronto, Canada).



Flow cytometric analysis of NIH/3T3 cells, untreated (blue) or PDGF-treated (green), using Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb.

Revision 1

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Phospho-GSK-3β (Ser9) (D85E12) XP® Rabbit mAb

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Certificate of Analysis

Anti-phospho-GSK-3 (Y279/Y216), clone 5G-2F

(mouse monoclonal $IgG_{1\kappa}$)
Catalog # 05-413
Lot # 21442

Immunogen: Synthetic peptide (C-KQLLHGEPNVS[pY]ICSRY) corresponding to amino acids 203-219 derived from a region of the catalytic domain (between subdomain VII and VIII) of the Drosophila GSK-3/shaggy enzyme coupled to ovalbumin.

Specificity: GSK-3 is constitutively phosphorylated at this tyrosine residue in resting cells. This immunogen is identical in 15 of 17 residues in human and rat GSK-3α (Y279) and GSK-3β (Y216). The phosphotyrosine residue can be removed from GSK-3 by treatment with PTP1B.

Species Cross-reactivity: Mouse, rat, human, *S. pombe* and *Dictyostelium*. Clone 5G-2F should cross-react with nearly all species because of the extremely high conservation of this region among the known enzymes of many different species.

Storage and Stability: Stable for 2 years at -20°C from date of shipment. Aliquot to avoid repeated freezing and thawing. For maximum recovery of product, centrifuge the original vial after thawing and prior to removing the cap.

Formulation: **200µg** of protein G purified mouse $IgG_{1\kappa}$ in **200µl** of 0.1M Tris-glycine, pH 7.4, 0.15M NaCl and 0.05% sodium azide. Frozen solution.

FOR IN VITRO RESEARCH USE ONLY
NOT FOR USE IN HUMANS OR IN ANIMALS

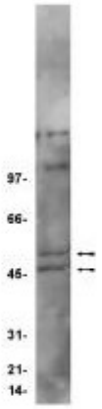
Quality Control Testing

Immunoblot Analysis: 0.5-2µg/ml of this lot detected phospho-GSK-3 in RIPA lysates from human Jurkat cells. Previous lots detected phospho-GSK-3 in mouse 3T3, human A431 and rat L6 cell lysates.

Included Positive Antigen Control: Catalog # 12-303, Jurkat lysate. **Add 2.5µl of 2-mercaptoethanol/100µl of lysate and boil for 5 minutes to reduce the preparation.** Load 20µg of reduced lysate per lane for minigels.

Immunoprecipitation: Not recommended.

Immunocytochemistry: 10µg/ml of this lot shows positive immunostaining for phospho-GSK-3 in L6 cells fixed with acetone.



Immunoblot Analysis
Representative blot from a previous lot. Jurkat cell lysate was resolved by electrophoresis, transferred to nitrocellulose and probed with **anti-phospho-GSK-3 (Y279/Y216), clone 5G-2F,** (0.5µg/ml). Proteins were visualized using a goat anti-mouse secondary antibody conjugated to HRP and a chemiluminescence detection system. Arrows indicate phospho-GSK-3 alpha and beta isoforms (51 and 46kDa).

General References:
Hughes, K., *et al.*, Eur. J. Biochem. **203:** 305-311, 1992.
Siddle, K., *et al.*, Biochem. J. **305:** 25-28, 1995.
Woodgett, J.R., *et al.*, EMBO **12:** 803-808, 1993.

Application Reference:
Harwood, A.J., *et al.,* Cell **80:** 139-148. 1995.

Immunoblot Protocol

1. Perform SDS-polyacrylamide gel electrophoresis (SDS-PAGE) on a cell lysate sample (cell lysis buffer: 50mM Tris-HCl, pH7.4; 1% NP-40; 0.25% sodium deoxycholate; 150mM NaCl; 1mM EGTA; 1mM PMSF; 1μg/ml each aprotinin, leupeptin, pepstatin; 1mM Na_3VO_4; 1mM NaF) and transfer the proteins to nitrocellulose. Wash the blotted nitrocellulose twice with water.
2. Block the blotted nitrocellulose in freshly prepared TBS containing 0.05% Tween 20 (TBS-T) for 1 hour at 20-25°C with constant agitation. Block again using TBS containing 3% nonfat dry skim milk and 0.05% Tween 20 (TBS-MLK/Tw) for 1-2 hours at room temperature or overnight a 4 ℃ with constant agitation.
3. Incubate the nitrocellulose with **0.5-2μg/ml of anti-phospho-GSK-3 (Y279/Y216), clone 5G-2F,** diluted in freshly prepared TBS-MLK/Tw for 1-2 hours at room temperature or overnight at with agitation at 4°C.
4. Wash the nitrocellulose twice with water.
5. Incubate the nitrocellulose in the secondary reagent of choice (a goat anti-mouse HRP conjugated IgG, Catalog # 12-349, 1:3000 dilution was used) in TBS-MLK/Tw for 1.5 hours at room temperature with agitation.
6. Wash the nitrocellulose with water twice.
7. Wash the nitrocellulose in TBS-0.05% Tween 20 for 15 minutes.
8. Rinse the nitrocellulose in 4-5 changes of water.
9. Use detection method of choice (enhanced chemiluminescence was used).

Immunocytochemistry Protocol

1. Plate approximately 200μl of cell suspension into each well of a slide. Incubate 24 hours in a 37°C CO_2 incubator.
2. Wash the cells three times for 5 minutes with TBS. Do not shake cells.
3. Add fix (100% acetone) for 10 minutes at -20 ℃.
4. Wash the cells with TBS, twice, for 15 minutes. Do not shake.
5. Add 400μl of 1% BSA in TBS and incubate for 1 hour at room temperature.
6. Wash the cells with TBS for 15 minutes.
7. Incubate the cells with **10μg/ml anti-phospho-GSK-3 (Y279/Y216), clone 5G-2F,**in 1% BSA in TBS and incubate for 1 hour at room temperature.
8. Wash the cells twice with TBS for 5 minutes.
9. Incubate the cells with a 1:200 dilution of goat anti mouse IgG fluorescein conjugated secondary antibody in 1% BSA in TBS for 1 hour at room temperature.
10. Wash the cells three times with TBS for 5 minutes.
11. Examine the cells under a fluorescent microscope.

abcam

Product datasheet

Anti-Tau antibody [TAU-5] - BSA and Azide free ab80579

5 Abreviews 104 References 3 图像

概述

产品名称	Anti-Tau抗体[TAU-5] - BSA and Azide free
描述	小鼠单克隆抗体[TAU-5] to Tau - BSA and Azide free
宿主	Mouse
经测试应用	**适用于:** WB, IP, ICC/IF
种属反应性	**与反应:** Mouse, Rat, Human **预测可用于:** Sheep, Cow
免疫原	Full length native protein (purified) corresponding to Cow Tau. Purified bovine microtubule-associated proteins.
阳性对照	WB: Human Alzheimer's brain whole tissue lysate. Human, mouse and rat brain whole tissue lysate. ICC: Human iPSC-Derived Glutamatergic Neurons. SKNSH cells treated with prostaglandin J2.
常规说明	This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or conjugation for your experiments, please contact orders@abcam.com. The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
存储溶液	Constituent: 100% PBS
无载体	是
纯度	Protein G purified
克隆	单克隆

克隆编号	TAU-5
同种型	IgG1
轻链类型	kappa

应用

The Abpromise guarantee Abpromise™承诺保证使用ab80579于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★★ (4)	Use a concentration of 1 µg/ml. Predicted molecular weight: 79 kDa.
IP		Use at 2 µg/mg of lysate.
ICC/IF		Use a concentration of 5 µg/ml.

靶标

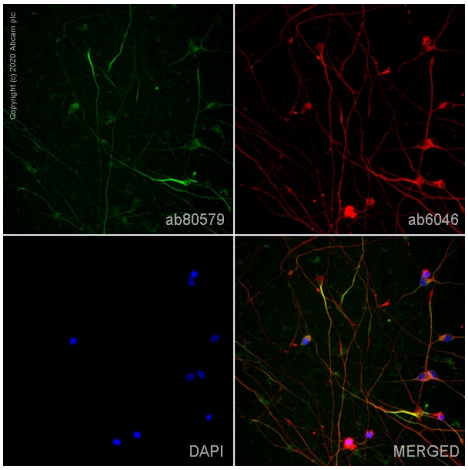
功能 Promotes microtubule assembly and stability, and might be involved in the establishment and maintenance of neuronal polarity. The C-terminus binds axonal microtubules while the N-terminus binds neural plasma membrane components, suggesting that tau functions as a linker protein between both. Axonal polarity is predetermined by tau localization (in the neuronal cell) in the domain of the cell body defined by the centrosome. The short isoforms allow plasticity of the cytoskeleton whereas the longer isoforms may preferentially play a role in its stabilization.

组织特异性 Expressed in neurons. Isoform PNS-tau is expressed in the peripheral nervous system while the others are expressed in the central nervous system.

疾病相关 Note=In Alzheimer disease, the neuronal cytoskeleton in the brain is progressively disrupted and replaced by tangles of paired helical filaments (PHF) and straight filaments, mainly composed of hyperphosphorylated forms of TAU (PHF-TAU or AD P-TAU).
Defects in MAPT are a cause of frontotemporal dementia (FTD) [MIM:600274]; also called frontotemporal dementia (FTD), pallido-ponto-nigral degeneration (PPND) or historically termed Pick complex. This form of frontotemporal dementia is characterized by presenile dementia with behavioral changes, deterioration of cognitive capacities and loss of memory. In some cases, parkinsonian symptoms are prominent. Neuropathological changes include frontotemporal atrophy often associated with atrophy of the basal ganglia, substantia nigra, amygdala. In most cases, protein tau deposits are found in glial cells and/or neurons.
Defects in MAPT are a cause of Pick disease of the brain (PIDB) [MIM:172700]. It is a rare form of dementia pathologically defined by severe atrophy, neuronal loss and gliosis. It is characterized by the occurrence of tau-positive inclusions, swollen neurons (Pick cells) and argentophilic neuronal inclusions known as Pick bodies that disproportionally affect the frontal and temporal cortical regions. Clinical features include aphasia, apraxia, confusion, anomia, memory loss and personality deterioration.
Note=Defects in MAPT are a cause of corticobasal degeneration (CBD). It is marked by extrapyramidal signs and apraxia and can be associated with memory loss. Neuropathologic features may overlap Alzheimer disease, progressive supranuclear palsy, and Parkinson disease.

	Defects in MAPT are a cause of progressive supranuclear palsy type 1 (PSNP1) [MIM:601104, 260540]; also abbreviated as PSP and also known as Steele-Richardson-Olszewski syndrome. PSNP1 is characterized by akinetic-rigid syndrome, supranuclear gaze palsy, pyramidal tract dysfunction, pseudobulbar signs and cognitive capacities deterioration. Neurofibrillary tangles and gliosis but no amyloid plaques are found in diseased brains. Most cases appear to be sporadic, with a significant association with a common haplotype including the MAPT gene and the flanking regions. Familial cases show an autosomal dominant pattern of transmission with incomplete penetrance; genetic analysis of a few cases showed the occurrence of tau mutations, including a deletion of Asn-613.
序列相似性	Contains 4 Tau/MAP repeats.
发展阶段	Four-repeat (type II) tau is expressed in an adult-specific manner and is not found in fetal brain, whereas three-repeat (type I) tau is found in both adult and fetal brain.
结构域	The tau/MAP repeat binds to tubulin. Type I isoforms contain 3 repeats while type II isoforms contain 4 repeats.
翻译后修饰	Phosphorylation at serine and threonine residues in S-P or T-P motifs by proline-directed protein kinases (PDPK: CDK1, CDK5, GSK-3, MAPK) (only 2-3 sites per protein in interphase, seven-fold increase in mitosis, and in PHF-tau), and at serine residues in K-X-G-S motifs by MAP/microtubule affinity-regulating kinase (MARK) in Alzheimer diseased brains. Phosphorylation decreases with age. Phosphorylation within tau's repeat domain or in flanking regions seems to reduce tau's interaction with, respectively, microtubules or plasma membrane components. Phosphorylation on Ser-610, Ser-622, Ser-641 and Ser-673 in several isoforms during mitosis. Polyubiquitinated. Requires functional TRAF6 and may provoke SQSTM1-dependent degradation by the proteasome (By similarity). PHF-tau can be modified by three different forms of polyubiquitination. 'Lys-48'-linked polyubiquitination is the major form, 'Lys-6'-linked and 'Lys-11'-linked polyubiquitination also occur. Glycation of PHF-tau, but not normal brain tau. Glycation is a non-enzymatic post-translational modification that involves a covalent linkage between a sugar and an amino group of a protein molecule forming ketoamine. Subsequent oxidation, fragmentation and/or cross-linking of ketoamine leads to the production of advanced glycation endproducts (AGES). Glycation may play a role in stabilizing PHF aggregation leading to tangle formation in AD.
细胞定位	Cytoplasm > cytosol. Cell membrane. Cytoplasm > cytoskeleton. Cell projection > axon. Mostly found in the axons of neurons, in the cytosol and in association with plasma membrane components.
形式	There are 9 isoforms produced by alternative splicing.

图片

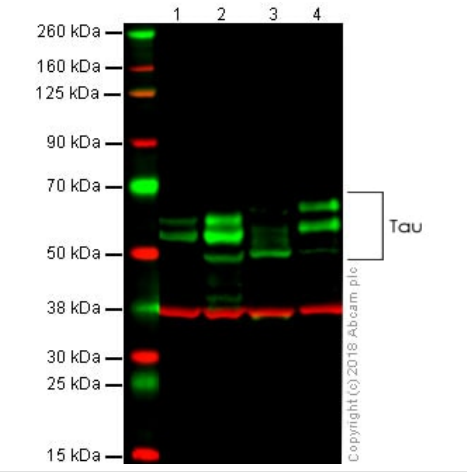


Immunocytochemistry/ Immunofluorescence - Anti-Tau antibody [TAU-5] - BSA and Azide free (ab80579)

Ab80579 staining Tau in ab259259 ioNEURONS/glut cells (Human iPSC-Derived Glutamatergic Neurons).

The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% PBS-Tween for 5 mins 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 ab80579 at 5 µg/ml and ab6046, Rabbit polyclonal to beta Tubulin - Loading Control, at 1/1000 dilution. Cells were then incubated with ab150117, Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preabsorbed at 1/1000 dilution (shown in green) and ab150088, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 594) preabsorbed at 1/1000 dilution (shown in pseudocolor red). Nuclear DNA was labelled with DAPI (shown in blue).

Images were acquired with the Perkin Elmer Operetta HCA and a single confocal plane is shown.



Western blot - Anti-Tau antibody [TAU-5] - BSA and Azide free (ab80579)

All lanes :

Lane 1 : Human Alzheimer's brain whole tissue lysate
Lane 2 : Human brain whole tissue lysate
Lane 3 : Mouse brain whole tissue lysate
Lane 4 : Rat brain whole tissue lysate

Lysates/proteins at 40 µg per lane.

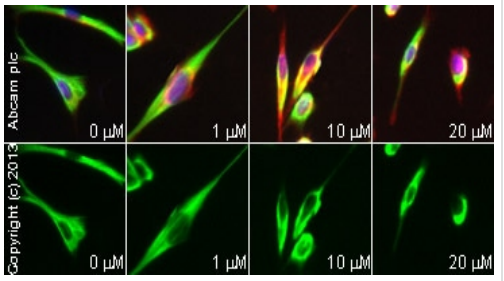
Performed under reducing conditions.

Predicted band size: 79 kDa

Tau cleavage products are shown between 70kDa and 50kDa.

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 55 minutes before being

transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 3% milk before ab80579 and ab181602 (Rabbit anti GAPDH loading control) were incubated overnight at 4°C at a 1ug/ml concentration and 1/10000 dilution respectively. Antibody binding was detected using Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-Tau antibody [TAU-5] - BSA and Azide free (ab80579)

ab80579 staining tau in SKNSH cells treated with prostaglandin J2 (ab120913), by ICC/IF. Expression of tau expression is restringed to the perinuclear zone with increased concentration of prostaglandin J2, as described in literature.
The cells were incubated at 37°C for 6 hours in media containing different concentrations of ab120913 (prostaglandin J2) in DMSO, fixed with 100% methanol for 5 minutes at -20°C and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with ab80579 (10 µg/ml) was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A DyLight® 488 anti-mouse polyclonal antibody (ab96879) at 1/250 dilution was used as the secondary antibody. Nuclei (blue) were counterstained with DAPI and membrane is was stained using WGA (red).

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Product datasheet

Anti-beta Actin antibody [AC-15] ab6276

敲除验证

★★★★★ 94 Abreviews 1949 References 6 图像

概述

产品名称	Anti-beta Actin抗体[AC-15]
描述	小鼠单克隆抗体[AC-15] to beta Actin
宿主	Mouse
经测试应用	**适用于:** ICC/IF, WB
种属反应性	**与反应:** Mouse, Rat, Cow, Dog, Human, African green monkey, Chinese hamster **不与反应:** Drosophila melanogaster, Dictyostelium discoideum
免疫原	Synthetic peptide corresponding to beta Actin aa 1-100 (N terminal) conjugated to keyhole limpet haemocyanin. Slightly modified ß-cytoplasmic actin N-terminal peptide, Ac-Asp-Asp-Asp-Ile-Ala-Al?a-Leu-Val-Ile-Asp-Asn-Gl y?-Ser-Gly-Lys, conjugated to KLH. Run BLAST with ExPASy Run BLAST with NCBI
表位	N-terminal of the beta isoform of actin.
阳性对照	ICC/IF: SV40LT-SMC cells WB: HAP1, HeLa, Jurkat, A431, HEK-293, COS-7, NIH/3T3, PC-12, Rat2, CHO, MDBK and MDCK cell lysates.
常规说明	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
存储溶液	pH: 7.40 Preservative: 0.097% Sodium azide Constituent: PBS
纯度	Affinity purified

纯化说明	Purified from hybridoma cell culture.
克隆	单克隆
克隆编号	AC-15
同种型	IgG1

应用

The Abpromise guarantee Abpromise™承诺保证使用ab6276于以下的经测试应用

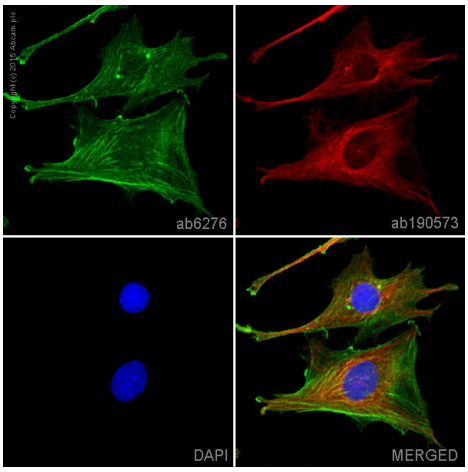
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
ICC/IF	★★★★★ (9)	Use a concentration of 5 µg/ml.
WB	★★★★★ (76)	1/5000 - 1/16000. Predicted molecular weight: 42 kDa.

靶标

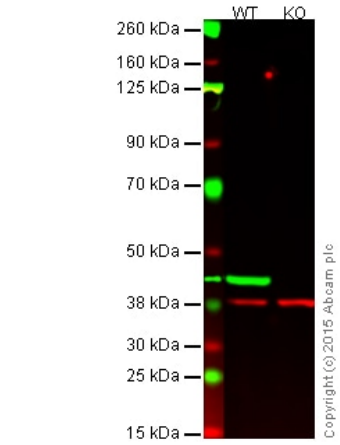
功能	Actins are highly conserved proteins that are involved in various types of cell motility and are ubiquitously expressed in all eukaryotic cells.
疾病相关	Defects in ACTB are a cause of dystonia juvenile-onset (DYTJ) [MIM:607371]. DYTJ is a form of dystonia with juvenile onset. Dystonia is defined by the presence of sustained involuntary muscle contraction, often leading to abnormal postures. DYTJ patients manifest progressive, generalized, dopa-unresponsive dystonia, developmental malformations and sensory hearing loss.
序列相似性	Belongs to the actin family.
翻译后修饰	ISGylated.
细胞定位	Cytoplasm > cytoskeleton. Localized in cytoplasmic mRNP granules containing untranslated mRNAs.

图片



Immunocytochemistry/ Immunofluorescence - Anti-beta Actin antibody [AC-15] (ab6276)

ab6276 staining beta Actin in SV40LT-SMC cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized in 0.1% Triton X-100 for 5 minutes and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab6276 at a working concentration of 5µg/ml and ab190573, Rabbit monoclonal [EP1332Y] to alpha Tubulin (Alexa Fluor® 647, shown in red) at 1/250 overnight at +4°C, followed by a further incubation at room temperature for 1h with an anti-mouse AlexaFluor® 488 (ab150117) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

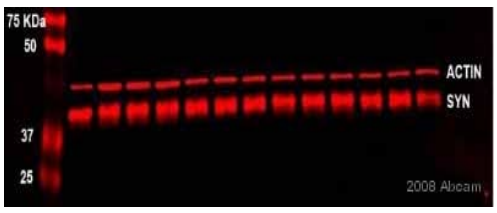


Western blot - Anti-beta Actin antibody [AC-15] (ab6276)

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: Beta actin knockout HAP1 cell lysate (20 µg)

Lanes 1 and 2: Merged signal (red and green). Green - beta actin, ab6276 observed at 42 kDa. Red - loading control, ab181602 observed at 37 kDa.

Ab6276 was shown to specifically react with beta actin in wild-type HAP1 cells. No band was observed when beta actin knockout samples were used. Wild-type and beta actin knockout samples were subjected to SDS-PAGE. ab6276 (beta actin) and ab181602 (loading control to GAPDH) were diluted 1/5000 and 1/10 000 and incubated overnight at 4°C. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (ab216772) and Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (ab216777) secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.

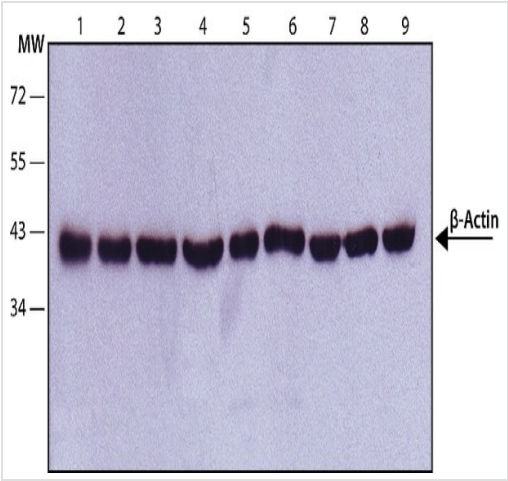


Western blot - Anti-beta Actin antibody [AC-15] (ab6276)

This image is courtesy of an Abreview submitted by Dr Mark Elliott

Western Blot of ab6276 (used as loading control) with whole tissue lysate of human grey matter from BA20 (temporal cortex). Ab6276 was diluted 1/50000 and incubated with the sample for 16 hours at 4°C. 5 µg of lysate was loaded onto the gel, which was blocked with 5% milk for 1 hour at 20°C. An Alexa Fluor® 680 conjugated goat anti-mouse antibody, diluted 1/5000, was used as the secondary.

Bands below actin in image are synaptophysin (SYN).



Western blot - Anti-beta Actin antibody [AC-15] (ab6276)

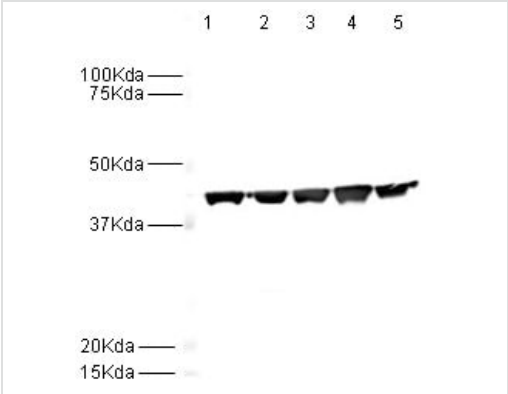
All lanes : Anti-beta Actin antibody [AC-15] (ab6276) at 1 µg/ml

Lane 1 : HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate
Lane 2 : Jurkat (human T cell leukemia cell line from peripheral blood) cell lysate
Lane 3 : COS-7 (african green monkey kidney fibroblast-like cell line) cell lysate
Lane 4 : NIH/3T3 (mouse embryonic fibroblast cell line) cell lysate
Lane 5 : PC-12 (rat adrenal gland pheochromocytoma cell line) cell lysate
Lane 6 : Rat2 (rat fibroblast cell line) cell lysate
Lane 7 : CHO (chinese hamster ovary cell line) cell lysate
Lane 8 : MDBK (bovine kidney cell line) cell lysate
Lane 9 : MDCK (canine kidney cell line) cell lysate

Secondary
All lanes : Goat Anti-Mouse IgG-Peroxidase

Developed using the ECL technique.

Predicted band size: 42 kDa



Western blot - Anti-beta Actin antibody [AC-15] (ab6276)

All lanes : Anti-beta Actin antibody [AC-15] (ab6276) at 1/5000 dilution

Lane 1 : HeLa nuclear
Lane 2 : HeLa whole cell lysate
Lane 3 : A431 cell lysate
Lane 4 : Jurkat cell lysate
Lane 5 : HEK293 cell lysate

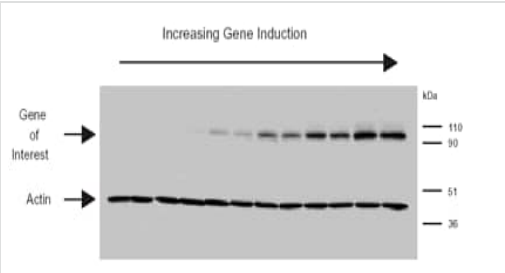
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Alexa Fluor anti mouse at 1/5000 dilution

Performed under reducing conditions.

Predicted band size: 42 kDa

Observed band size: 42 kDa



Western blot - Anti-beta Actin antibody [AC-15] (ab6276)

This image is courtesy of David Grimm, Yale University, USA.

MDCK cells induced with increasing amounts of doxycycline to control expression of the gene of interest. All cells were normalized for loading with an albumin protein standard assay. Anti-beta actin (ab6276) was used at a concentration of 1:5000 in a milk blocking solution. B-actin blotting confirms the albumin assay in showing that an equal amount of lysate was loaded in each lane.

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Revision 4

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#9441

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Applications:	Reactivity:	Sensitivity:	Source:
WB, IP, IHC-P, IF-IC, ChIP, E-P	All	Endogenous	Rabbit

Product Usage Information

For optimal ChIP results, use 10 µl of antibody and 10 µg of chromatin (approximately 4 x 10^6 cells) per IP. This antibody has been validated using SimpleChIP® Enzymatic Chromatin IP Kits.

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:100
Immunohistochemistry (Paraffin)	1:400 - 1:1600
Immunofluorescence (Immunocytochemistry)	1:800
Chromatin IP	1:50
Peptide ELISA (DELFIA)	1:2000

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA and 50% glycerol. Store at –20°C. Do not aliquot the antibody.

Specificity / Sensitivity

Acetylated-Lysine Antibody detects proteins posttranslationally modified by acetylation on the epsilon-amine groups of lysine residues. The antibody recognizes acetylated lysine in a wide range of sequence contexts. It has been demonstrated to recognize acetylated histones, p53, CBP, PCAF and chemically acetylated BSA. The antibody has been shown to react with as little as 0.04 ng of chemically acetylated BSA while not recognizing up to 25 µg of nonacetylated BSA. (U.S. Patent No's.: 6,441,140; 6,982,318; 7,259,022; 7,344,714; U.S.S.N. 11,484,485; and all foreign equivalents.)

Species Reactivity:
All Species Expected

Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic acetylated lysine-containing peptide. Antibodies are purified by protein A and peptide affinity chromatography.

Background

Acetylation of lysine, like phosphorylation of serine, threonine or tyrosine, is an important reversible modification controlling protein activity. The conserved amino-terminal domains of the four core histones (H2A, H2B, H3, and H4) contain lysines that are acetylated by histone acetyltransferases (HATs) and deacetylated by histone deacetylases (HDACs) (1). Signaling resulting in acetylation/deacetylation of histones, transcription factors, and other proteins affects a diverse array of cellular processes including chromatin structure and gene activity, cell growth, differentiation, and apoptosis (2-6). Recent proteomic surveys suggest that acetylation of lysine residues may be a widespread and important form of posttranslational protein modification that affects thousands of proteins involved in control of cell cycle and metabolism, longevity, actin polymerization, and nuclear transport (7,8). The regulation of protein acetylation status is impaired in cancer and polyglutamine diseases (9), and HDACs have become promising targets for anti-cancer drugs currently in development (10).

1. Hassig, C.A. and Schreiber, S.L. (1997) *Curr Opin Chem Biol* 1, 300-8.
2. Allfrey, V.G. et al. (1964) *Proc Natl Acad Sci USA* 51, 786-94.
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7. Kim, S.C. et al. (2006) *Mol Cell* 23, 607-18.
8. Choudhary, C. et al. (2009) *Science* 325, 834-40.
9. Hughes, R.E. (2002) *Curr Biol* 12, R141-3.
10. Vigushin, D.M. and Coombes, R.C. (2004) *Curr Cancer Drug Targets* 4, 205-18.

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

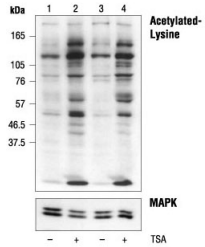
APPLICATIONS KEY **WB:** Western Blot **IP:** Immunoprecipitation **IHC:** Immunohistochemistry **ChIP:** Chromatin Immunoprecipitation **IF:** Immunofluorescence **F:** Flow Cytometry **E-P:** ELISA-Peptide

CROSS-REACTIVITY KEY **H:** human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **Mi:** mink **C:** chicken **Dm:** D. melanogaster **X:** Xenopus **Z:** zebrafish **B:** bovine **Dg:** dog **Pg:** pig **Sc:** S. cerevisiae **Ce:** C. elegans **Hr:** horse **All:** all species expected

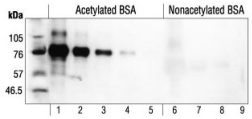
Revision 4

#9441

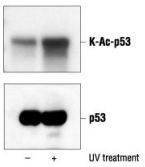
Acetylated-Lysine Antibody



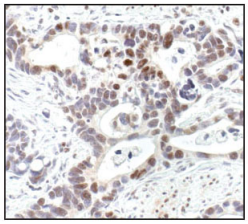
Western blot analysis of extracts from COS cells, untreated or TSA-treated, grown in 10% FBS (lanes 1 and 2) or serum starved for 18 hours (lanes 3 and 4), using Acetylated-Lysine Antibody (upper) or p44/42 MAP Kinase Antibody #9102 (lower).



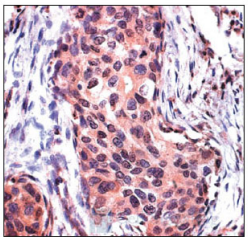
Specificity and sensitivity of Acetylated-Lysine Antibody assayed on acetylated BSA (4; 1; 0.2; 0.04 or 0.008 ng in lanes 1-5) or nonacetylated BSA (25,000; 5,000; 1,000 or 200 ng in lanes 6-9).



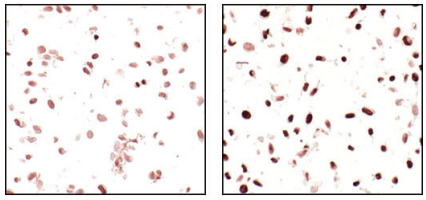
Western blot analysis of immunoprecipitated p53 showing an increase in p53 acetylation using Acetylated-Lysine Antibody (upper) or p53 antibody (lower). p53 was immunoprecipitated from lysates from 293 cells, untreated or UV-treated, using p53 Antibody #9282.



Immunohistochemical analysis of paraffin-embedded human colon carcinoma using Acetylated-Lysine Antibody.



Immunohistochemical staining of a paraffin-embedded human breast tumor section showing nuclear and cytoplasmic localization of proteins with acetylated lysine residues using Acetylated-Lysine Antibody.

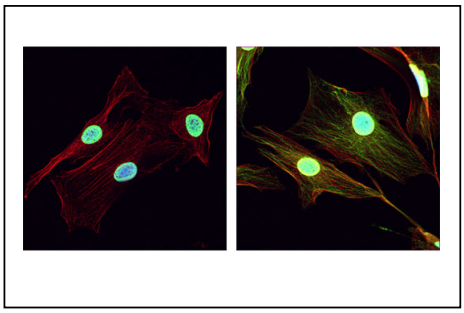


Immunohistochemical analysis of paraffin-embedded NIH/3T3 untreated (left) or TSA-treated (right) using Acetylated-Lysine Antibody.

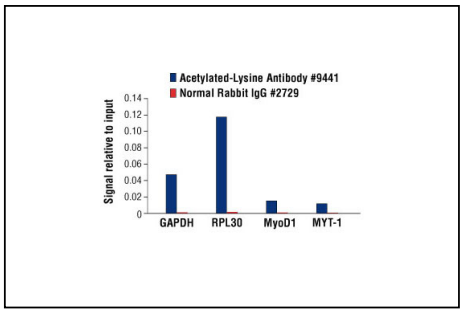
Revision 4

#9441

Acetylated-Lysine Antibody



Confocal immunofluorescent analysis of NIH/3T3 cells, untreated (left) or SAHA-treated (right), labeled with Acetylated-Lysine Antibody (green). Actin filaments have been labeled with Alexa Fluor R 555 phalloidin (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).



Chromatin immunoprecipitations were performed with cross-linked chromatin from HeLa cells and either Acetylated-Lysine Antibody or Normal Rabbit IgG #2729, using SimpleChIP® Enzymatic Chromatin IP Kit (Agarose Beads) #9002. The enriched DNA was quantified by real-time PCR, using SimpleChIP® Human GAPDH Exon 1 Primers #5516, SimpleChIP® Human RPL30 Exon 3 Primers #7014, SimpleChIP® Human MyoD1 Exon 1 Primers #4490, and SimpleChIP® Human MYT-1 Exon 1 Primers #4493. The amount of immunoprecipitated DNA in each sample is represented as signal relative to the total amount of input chromatin, which is equivalent to one.

#9441

Acetylated-Lysine Antibody

限制使用

除非 CST 的合法授书代表以书面形式书行明确同意，否书以下条款适用于 CST、其关书方或分书商提供的书品。 任何书充本条款或与本条款不同的客书条款和条件，除非书 CST 的合法授书代表以书面形式书独接受， 否书均被拒书，并且无效。

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abcam

Product datasheet

Anti-Ubiquitin antibody [Ubi-1] ab7254

6 Abreviews 76 References 5 图像

概述

产品名称	Anti-Ubiquitin抗体[Ubi-1]
描述	小鼠单克隆抗体[Ubi-1] to Ubiquitin
宿主	Mouse
经测试应用	**适用于:** IHC-P, WB **不适用于:** IP
种属反应性	**与反应:** Mouse, Human
免疫原	Full length protein corresponding to Cow Ubiquitin conjugated to keyhole limpet haemocyanin (Glutaraldehyde).
阳性对照	H9C2 cells IHC-P:FFPE mouse brain alzheimer
常规说明	This product was changed from ascites to tissue culture supernatant on 17th May 2016. The following lots are from ascites and are still in stock as of 17th May 2016: GR159581-2, GR159581-12, GR159581-13. Lot numbers higher than GR159581-13 will be from tissue culture supernatant. Please note that the dilutions may need to be adjusted accordingly. The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
存储溶液	Preservative: 0.1% Sodium azide
纯度	Affinity purified
克隆	单克隆
克隆编号	Ubi-1

同种型	IgG1
轻链类型	kappa

应用

The Abpromise guarantee Abpromise™承诺保证使用ab7254于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
IHC-P		Use a concentration of 1 - 5 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB	★★★★☆ (4)	1/100 - 1/5000.

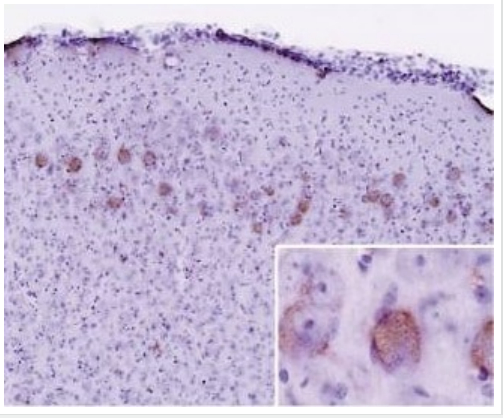
应用说明 Is unsuitable for IP.

靶标

相关性 Function: Ubiquitin exists either covalently attached to another protein, or free (unanchored). When covalently bound, it is conjugated to target proteins via an isopeptide bond either as a monomer (monoubiquitin), a polymer linked via different Lys residues of the ubiquitin (polyubiquitin chains) or a linear polymer linked via the initiator Met of the ubiquitin (linear polyubiquitin chains). Polyubiquitin chains, when attached to a target protein, have different functions depending on the Lys residue of the ubiquitin that is linked: Lys-6-linked may be involved in DNA repair; Lys-11-linked is involved in ERAD (endoplasmic reticulum-associated degradation) and in cell-cycle regulation; Lys-29-linked is involved in lysosomal degradation; Lys-33-linked is involved in kinase modification; Lys-48-linked is involved in protein degradation via the proteasome; Lys-63-linked is involved in endocytosis, DNA-damage responses as well as in signaling processes leading to activation of the transcription factor NF-kappa-B. Linear polymer chains formed via attachment by the initiator Met lead to cell signaling. Ubiquitin is usually conjugated to Lys residues of target proteins, however, in rare cases, conjugation to Cys or Ser residues has been observed. When polyubiquitin is free (unanchored-polyubiquitin), it also has distinct roles, such as in activation of protein kinases, and in signaling. Similarity: Belongs to the ubiquitin family. Contains 3 ubiquitin-like domains.

细胞定位 Cell Membrane, Cytoplasmic and Nuclear

图片

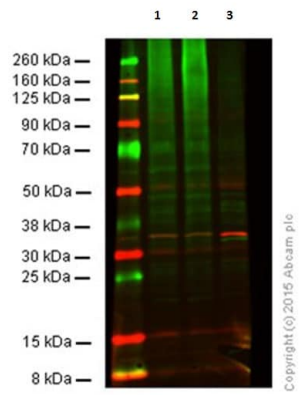


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [Ubi-1] (ab7254)

Immunohistochemical staining of ubiquitin in a 7 months-old MPS IIIA mouse brain (PFA fixed) using ab7254.

Sections, 40 µm thick, were cut sagittally on a vibratome. Slices were permeabilized in 1% triton X-100/PBS for 1 hour at room temperature. The samples were placed on vectabond-coated glass slides and dried overnight. They were then rehydrated in PBS and washed in ice-cold methanol for 10 min.

The slices were washed in 0.3% H_2O_2 in methanol at room temperature and incubated sequentially with 0.5% normal donkey serum, ab7254 and biotin-labeled F(ab')$_2$ donkey secondary antibody in Tris-buffered saline (pH 7.5) containing 2% BSA and 0.02% Tween 20. The signal was detected with an ABC kit, visualized with diaminobenzidine and counterstained with hematoxylin.



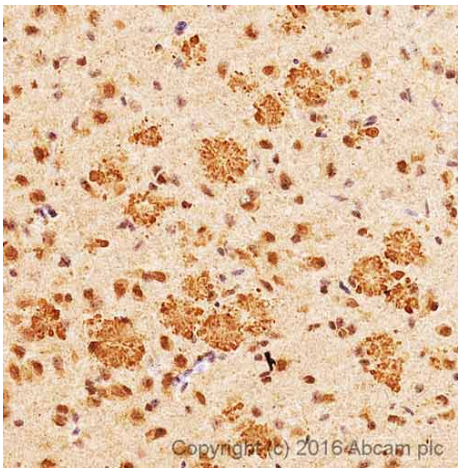
Western blot - Anti-Ubiquitin antibody [Ubi-1] (ab7254)

All lanes : Anti-Ubiquitin antibody [Ubi-1] (ab7254) at 1/100 dilution

Lane 1 : HeLa cell lysate
Lane 2 : HeLa cell lysate with MG132
Lane 3 : Mouse brain tissue lysate

Lysates/proteins at 20 µg per lane.

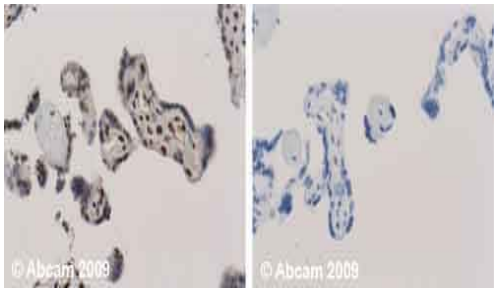
Secondary
All lanes : Anti mouse IgG at 1/10000 dilution



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [Ubi-1] (ab7254)

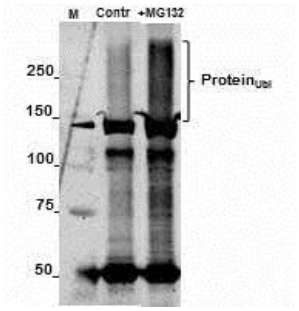
IHC image of ab7254 staining in mouse alzheimer brain formalin fixed paraffin embedded tissue section, using MOM detection kit, ab127055. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins. The section was incubated with ab7254, 5µg/ml, for 15 mins at room temperature. DAB was used as the chromogen (ab103723). The section was counterstained with haematoxylin and mounted with DPX.

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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ubiquitin antibody [Ubi-1] (ab7254)

Human normal placenta (ab29745). Staining is localised in the cytoplasm and in the nuclei. Left panel: with primary antibody at 1 ug/ml. Right panel: isotype control. Sections were stained using an automated system DAKO Autostainer Plus, at room temperature: sections were rehydrated and antigen retrieved with the Dako 3 in 1 AR buffers citrate pH6.1 in a DAKO PT Link. Slides were peroxidase blocked in 3% H2O2 in methanol for 10 mins. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 min and detected with Dako envision flex amplification kit for mouse for 30 minutes. Colorimetric detection was completed with Diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin and coverslipped under DePeX. Please note that for manual staining we recommend to optimize the primary antibody concentration and incubation time (overnight incubation), and amplification may be required.



Western blot - Anti-Ubiquitin antibody [Ubi-1] (ab7254)

Image courtesy of an anonymous Abreview.

HeLa cells were co-transfected with a plasmid expressing a target protein together with Ubi expressing vector for 24 hours and either left untreated (Contr) or were treated with 10 µM MG-132 for 6 hours (+MG132). Then the protein of interest was pulled down using Flag agarose beads and and probed with ab7254 at a 1/2000 dilution. The secondary used was an Alexa-Fluor 680 conjugated goat anti-mouse polyclonal used at a 1/10000 dilution. The protein is known to be degraded through proteasome.

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- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
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For Research Use Only

GST TAG Polyclonal ANTIBODY

proteintech®
Antibodies | ELISA kits | Proteins
www.ptglab.com

Catalog Number:10000-0-AP 103 Publications

Basic Information

Catalog Number:
10000-0-AP
Size:
513 μg/ml
Source:
Rabbit
Isotype:
IgG
Purification Method:
Antigen affinity purification

GenBank Accession Number:
FN315687
GeneID (NCBI):
Full Name:
Calculated MW:
26 kDa

Recommended Dilutions:
WB: 1:1000-1:4000

Applications

Tested Applications:
FC, WB,ELISA
Cited Applications:
ChIP, IF, IP, WB
Species Specificity:
recombinant protein
Cited Species:
human, mouse, plant, rat

Positive Controls:
WB : Recombinant protein,

Background Information

GST(Glutathione S-Transferase) is a widely used protein tag encoded by the Schistosoma japonicum. GST provides both an easily detectable tag and a simple purification process with little effect on the biological function of the protein of interest. This GST-fusion protein can then be purified from cells via its high affinity for glutathione. Antibodies to GST are useful for checking protein expression both in plaques and on western blots as well as for immunoaffinity purification of proteins expressed in cells. This antibody can be used to detect the GST tagged protein. This antibody can be used as Rabbit IgG control in FC.

Notable Publications

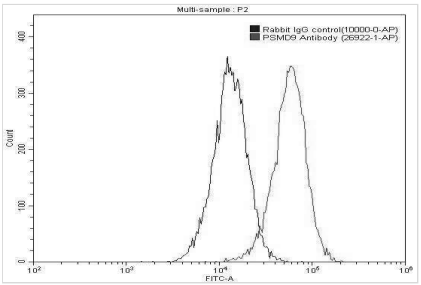
Author	Pubmed ID	Journal	Application
Tianyu Han	30231667	Autophagy	
Xiangying Luo	34536269	Aging (Albany NY)	WB
Yunchao Liu	32931829	Int J Biol Macromol	WB

Storage

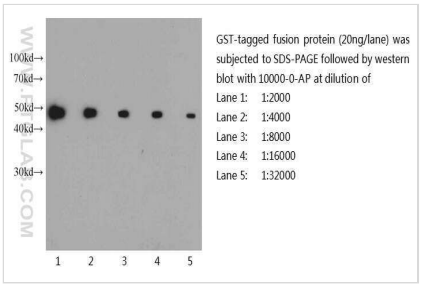
Storage:
Store at -20°C. Stable for one year after shipment.
Storage Buffer:
PBS with 0.02% sodium azide and 50% glycerol pH 7.3.
Aliquoting is unnecessary for -20°C storage

For technical support and original validation data for this product please contact:
T: 1 (888) 4PTGLAB (1-888-478-4522) (toll free in USA), or 1(312) 455-8498 (outside USA)
E: proteintech@ptglab.com
W: ptglab.com

Selected Validation Data



1X10^6 HeLa cells were stained with 0.20 ug PSMD9 antibody (26922-1-AP, red) and 0.20 ug Rabbit IgG control (10000-0-AP, blue). Fixed with 90% MeOH.



Western blot of GST-tagged fusion protein with anti-GST-tag (10000-0-AP) at various dilutions.

abcam

Product datasheet

Anti-Histone H4 antibody [EPR16599] - ChIP Grade ab177840

重组 RabMAb

3 Abreviews 11 References 10 图像

概述

产品名称	Anti-Histone H4抗体[EPR16599] - ChIP Grade
描述	兔单克隆抗体[EPR16599] to Histone H4 - ChIP Grade
宿主	Rabbit
经测试应用	**适用于:** PepArr, ChIP, IHC-P, WB, ICC/IF
种属反应性	**与反应:** Mouse, Rat, Human, Drosophila melanogaster, Recombinant fragment **预测可用于:** a wide range of other species
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: HeLa and NIH/3T3 whole cell lysates; Drosophila whole lysate. ICC/IF: HeLa cells. IHC-P: Human colon, mouse pancreas and rat cerebral cortex tissues. ChIP: Chromatin from HeLa cells.
常规说明	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.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
存储溶液	Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR16599
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab177840于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
PepArr		Use at an assay dependent concentration.
ChIP		Use 2 µg for 25 µg of chromatin.
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB	★★★★☆ (3)	1/1000. Detects a band of approximately 11 kDa (predicted molecular weight: 11 kDa). We recommend using 3% milk as the blocking agent for Western blot.
ICC/IF		1/100.

靶标

功能

Core component of nucleosome. Nucleosomes wrap and compact DNA into chromatin, limiting DNA accessibility to the cellular machineries which require DNA as a template. Histones thereby play a central role in transcription regulation, DNA repair, DNA replication and chromosomal stability. DNA accessibility is regulated via a complex set of post-translational modifications of histones, also called histone code, and nucleosome remodeling.

序列相似性

Belongs to the histone H4 family.

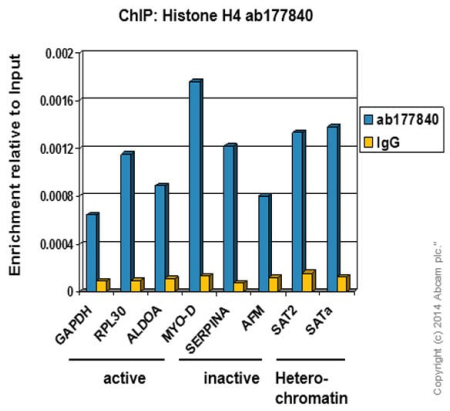
翻译后修饰

Acetylation at Lys-6 (H4K5ac), Lys-9 (H4K8ac), Lys-13 (H4K12ac) and Lys-17 (H4K16ac) occurs in coding regions of the genome but not in heterochromatin.
Citrullination at Arg-4 (H4R3ci) by PADI4 impairs methylation.
Monomethylation and asymmetric dimethylation at Arg-4 (H4R3me1 and H4R3me2a, respectively) by PRMT1 favors acetylation at Lys-9 (H4K8ac) and Lys-13 (H4K12ac).
Demethylation is performed by JMJD6. Symmetric dimethylation on Arg-4 (H4R3me2s) by the PRDM1/PRMT5 complex may play a crucial role in the germ-cell lineage.
Monomethylated, dimethylated or trimethylated at Lys-21 (H4K20me1, H4K20me2, H4K20me3). Monomethylation is performed by SET8. Trimethylation is performed by SUV420H1 and SUV420H2 and induces gene silencing.
Ubiquitinated by the CUL4-DDB-RBX1 complex in response to ultraviolet irradiation. This may weaken the interaction between histones and DNA and facilitate DNA accessibility to repair proteins. Monoubiquitinated at Lys-92 of histone H4 (H4K91ub1) in response to DNA damage. The exact role of H4K91ub1 in DNA damage response is still unclear but it may function as a licensing signal for additional histone H4 post-translational modifications such as H4 Lys-21 methylation (H4K20me).
Sumoylated, which is associated with transcriptional repression.

细胞定位

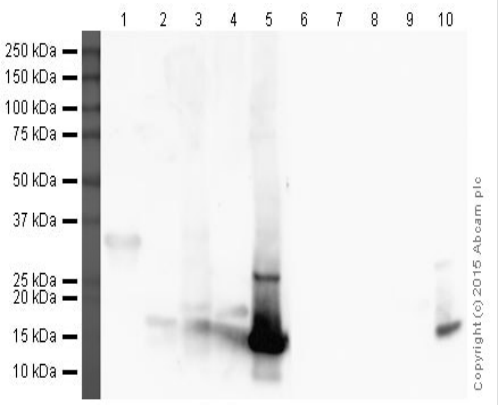
Nucleus. Chromosome.

图片



ChIP - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

Chromatin was prepared from Hela cells according to the Abcam X-ChIP protocol. Cells were fixed with 0.75% formaldehyde for 10min. The ChIP was performed with 25µg of chromatin, 2µg of ab177840 (blue), and 20µl of Anti rabbit IgG sepharose beads. 2µg of rabbit normal IgG was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (SYBR approach). Primers are located in the first kb of the transcribed region.



Western blot - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

All lanes : Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840) at 1/1000 dilution

Lanes 1 & 6 : Histone H1 Recombinant Protein
Lanes 2 & 7 : Histone H2A Recombinant Protein
Lanes 3 & 8 : Histone H2B Recombinant Protein
Lanes 4 & 9 : Histone H3.1 Recombinant Protein
Lanes 5 & 10 : Histone H4 Recombinant Protein

Lysates/proteins at 0.1 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG (H+L), Peroxidase Conjugated at 1/50000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

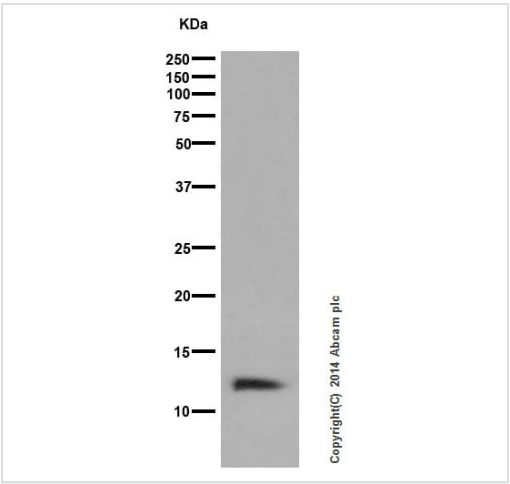
Predicted band size: 11 kDa
Observed band size: 15 kDa

Exposure time: 4 minutes

Lanes 1-5: 1% BSA blocking buffer

Lanes 6-10: 3% Milk blocking buffer

We recommend using 3% milk as the blocking agent for Western blot.



Western blot - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840) at 1/1000 dilution + Drosophila whole lysates at 10 µg

Secondary
Goat Anti-Rabbit IgG, (H+L),Peroxidase conjugated at 1/1000 dilution

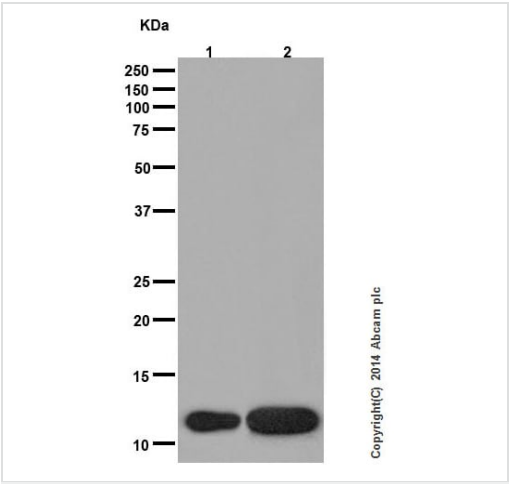
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 11 kDa
Observed band size: 11 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

All lanes : Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840) at 1/5000 dilution

Lane 1 : HeLa whole cell lysates
Lane 2 : NIH/3T3 whole cell lysates

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L),Peroxidase conjugated at 1/1000 dilution

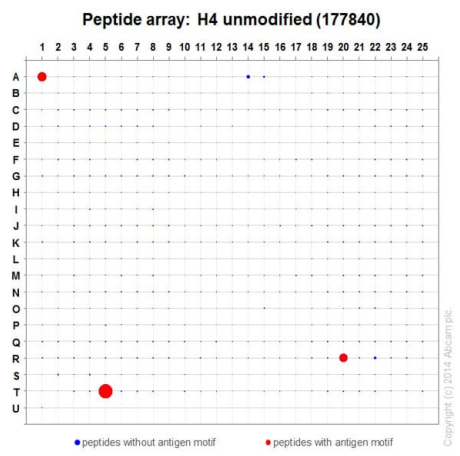
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 11 kDa
Observed band size: 11 kDa

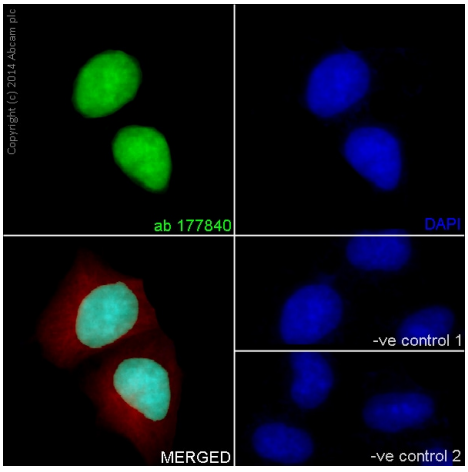
Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



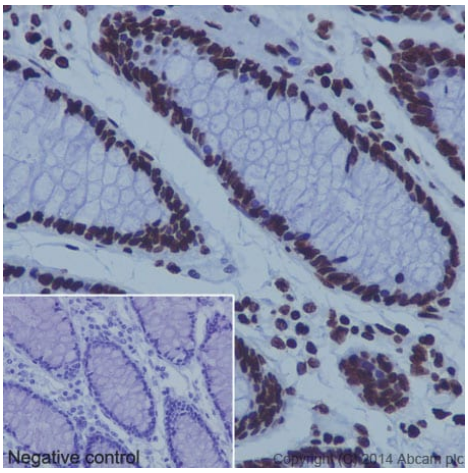
Peptide Array - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

ab177840 was tested in Peptide Array against 501 different modified and unmodified histone peptides; each peptide is printed on the array at six concentrations (each in triplicate).
Circle area represents affinity between the antibody and a peptide: all antigen-containing peptides are displayed as red circles, all other peptides as blue circles. The affinity is calculated as area under curve when antibody binding values are plotted against the corresponding peptide concentration. Each circle area is normalized to the peptide with the strongest affinity.
The complete dataset, including full list of all peptides and information on the position of each peptide in the diagram, can be downloaded here.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa cells labeling Histone H4 with ab177840 at 1/100 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/400 dilution (green). Nuclear staining on HeLa cell line is observed. The nuclear counter stain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/500 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows:
1. ab177840 at 1/100 dilution followed by ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
2. ab7291 (anti-Tubulin mouse mAb) at 1/500 dilution followed by ab150077 (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/400 dilution.

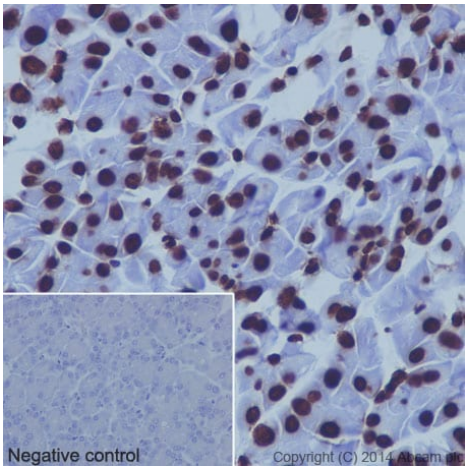


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling Histone H4 with ab177840 at 1/2000 dilution, followed by prediluted Goat Anti-Rabbit IgG H&L (HRP). Nucleus staining on glandular epithelium of Human colon tissue is observed. Counter stained with Hematoxylin.

Negative control: PBS instead of primary antibody; secondary antibody is prediluted Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

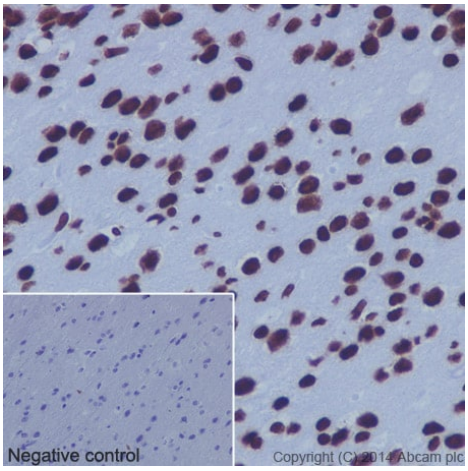


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

Immunohistochemical analysis of paraffin-embedded Mouse pancreas tissue labeling Histone H4 with ab177840 at 1/2000 dilution, followed by prediluted Goat Anti-Rabbit IgG H&L (HRP). Nucleus staining on glandular epithelium of mouse pancreas tissue is observed. Counter stained with Hematoxylin.

Negative control: PBS instead of primary antibody; secondary antibody is prediluted Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

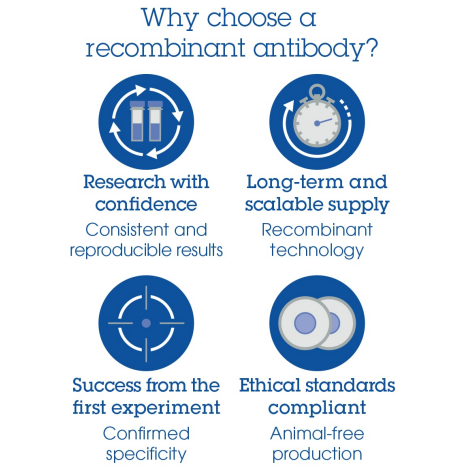


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

Immunohistochemical analysis of paraffin-embedded Rat cerebral cortex tissue labeling Histone H4 with ab177840 at 1/2000 dilution, followed by prediluted Goat Anti-Rabbit IgG H&L (HRP). Nuclear staining on neuron cells of cerebral cortex tissue is observed. Counter stained with Hematoxylin.

Negative control: PBS instead of primary antibody; secondary antibody is prediluted Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Anti-Histone H4 antibody [EPR16599] - ChIP Grade (ab177840)

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Terms and conditions

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Anti-acetyl-Histone H4

Polyclonal Antibody

Cat. # 06-866

Lot # 2384725

pack size: 200 µL

Store at -20ºC

FOR RESEARCH USE ONLY

Certificate of Analysis

Applications	Species Cross-Reactivity	Antibody Isotype	Epitope/ Region	Host Species	Molecular Weight	Accession #
WB, ChIP, IC	H, T, Eu	IgG	a.a. 2-19	Rb	10 kDa	NM_175054

Background

Histone H4 is one of the 5 main histone proteins involved in the structure of chromatin in eukaryotic cells. Featuring a main globular domain and a long N terminal tail H4 is involved with the structure of the nucleosomes of the 'beads on a string' structure. Acetylation of histone H4 occurs at several different lysine positions in the histone tail and is performed by a family of enzymes known as Histone Acetyl Transferases (HATs).

Presentation

Whole antiserum containing 0.05% sodium azide.

Specificity

Acetylated histone H4, acetylated histone H2B from *Tetrahymena* and weakly cross-reacts with acetylated histone H2B from HeLa cells, may cross-react with other acetylated proteins.

Species Cross-reactivity

Human and *Tetrahymena.* Other species not tested, but expected to cross-react since Histone H4 is well conserved.

Immunogen

KLH-conjugated peptide corresponding to amino acids 2-19 of *Tetrahymena* histone H4 [AGGAcKGGAcKGMGAcKVGAAcKRHS-C], acetylated on lysines 5, 8, 12 and 16.

Molecular Weight

10 kDa

Storage and Handling

Stable for 1 year at -20°C from date of receipt.

Aliquot to avoid repeated freezing and thawing. For maximum recovery of product, centrifuge the original vial after thawing and prior to removing the cap.

Control

TSA-treated Jurkat cells. For a negative control, perform a no-antibody immunoprecipitation by incubating the supernatant fraction with 60µl of Salmon Sperm DNA/Protein A Agarose- 50% Slurry. Transcriptionally unactivated DNA samples should be prepared as controls for PCR.

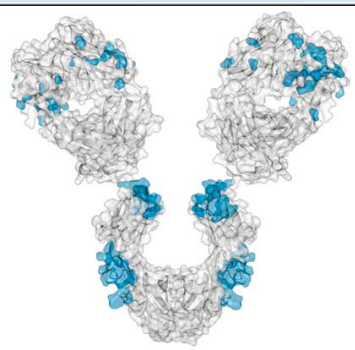
Quality Control Testing

Routinely evaluated by western blot on *Tetrahymena* macronuclei or acid extracted proteins from HeLa cells treated with 5 mM sodium butyrate.

Western Blot Analysis: 1:2000 dilution of this lot detected acetylated histone H4 in acid extracted proteins from HeLa cells treated with 5 mM sodium butyrate. Sodium butyrate, an inhibitor of deacetylases, was used to enhance detection of acetylated histone H4.

Western Blot Analysis
Representative lot data. Acid-extracted proteins from normal HeLa cells (Lane 1) and HeLa cells treated with 5 mM sodium butyrate for 24 hours (Lane 2) were resolved by electrophoresis, transferred to nitrocellulose and probed with anti-acetyl Histone H4 (1:2000). Proteins were visualized using a goat-anti rabbit secondary antibody conjugated to HRP and a chemi-luminescence detection system.

Arrow indicates acetylated histone H4 (~10 kDa).



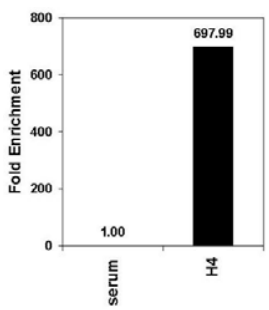
References

1. Dammer, Eric B., *et al.* (2007). *Mol Endocrinol.* 21: 415-38.
2. Sakamoto, A., *et al.* (2004). *Hum Mol Genet.* 13: 819-28.
3. Caretti, G., *et al.* (2003). *J Biol Chem.* 278: 30435-40.
4. Park, S. W. and Wei, L. N. (2003). *J Biol Chem.* 278: 29776-29782.
5. Siegel, P. M., *et al.* (2003). *J Biol Chem.* 278: 35444-35450.
6. Toyota, M., *et al.* (2003). *Proc Natl Acad Sci USA. 100: 7818-23.*

Additional Research Applications

Chromatin Immunoprecipitation:

Representative lot data. 5-10 µL immunoprecipitated transcriptionally active chromatin containing acetylated histone H4 from 2 X 10^6 serum stimulated HeLa cells.



APPLICATION LEGEND: **WB** Western Blotting **ChIP** Chromatin Immunoprecipitation **IP** Immunoprecipitation **IC** Immunocytochemistry **IH** Immunohistochemistry (Tissue)

SPECIES LEGEND: **Eu** Eukaryote **H** Human **M** Mouse **R** Rat **Rb** Rabbit **T** Tetrahymena

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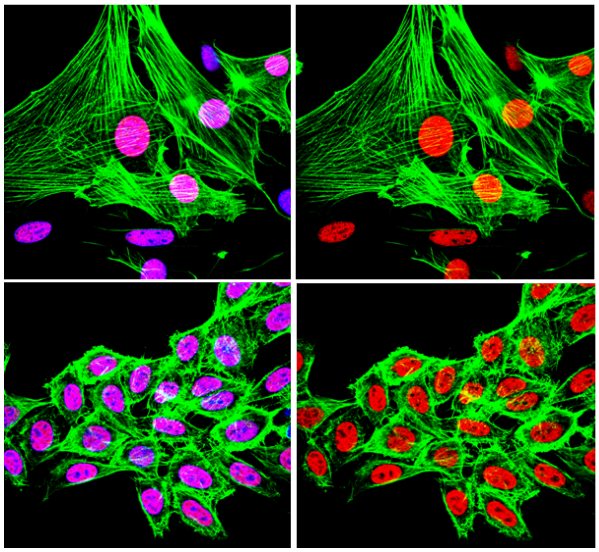
Rev.B/2009-08-26/06-866CA/JM

Additional Research Applications

Immunocytochemistry:

Representative lot data.

Confocal fluorescent analysis of HeLa and NIH/3T3 cells using 06-866 (Red). Actin filaments have been labeled with AlexaFluor®488-Phalloidin (Green). Nucleus is stained with DAPI (Blue). This antibody positively stains the nucleus.



PROTOCOL

Western Blot

1. Perform SDS-polyacrylamide gel electrophoresis (SDS-PAGE) on acid-extracted protein from cells treated with or without sodium butyrate (see the protocol below) and transfer the proteins to nitrocellulose. Wash the blotted nitrocellulose twice with water.
2. Block the blotted nitrocellulose in freshly prepared PBS containing 3% nonfat dry milk (Catalog # 20-200), (PBS-MLK) for one hour at room temperature with constant agitation.
3. Incubate the nitrocellulose with **1:2000 dilution of anti-acetyl Histone H4** in freshly prepared PBS-MLK, at for 3 hours with agitation at room temperature.
4. Wash the nitrocellulose three times with water.
5. Incubate the nitrocellulose in the secondary reagent of choice (a goat anti-rabbit IgG conjugated to HRP, Catalog # 12-348, 1:5000 dilution was used) in PBS-MLK for 1.5 hours at room temperature with agitation.
6. Wash the nitrocellulose with water three times.
7. Wash the nitrocellulose in PBS-0.05% Tween 20 for 5 minutes.
8. Rinse the nitrocellulose with 4-5 changes of water.
9. Use detection method of choice (enhanced chemiluminescence was used).

Acid Extraction of Proteins from Sodium Butyrate Treated HeLa Cells

1. Grow cells to 70% confluency in DMEM supplemented with 10% FBS.
2. Add sodium butyrate (100 mM sterile stock solution), which inhibits histone deacetylases, to a final concentration of 5 mM and continue to grow the cells for 24 hours.
3. Scrape the cells from the plate.
4. Pellet the cells by centrifugation at 200 x g for 10 minutes.
5. Decant the supernatant fraction.
6. Suspend the cells with 10-15 volumes of PBS and centrifuge at 200 x g for 10 minutes.
7. Decant supernatant fraction (PBS wash).
8. Suspend the cell pellet in 5-10 volumes of **lysis buffer**.
9. Add sulfuric acid to a final concentration of 0.2 M (0.4N). **Use polypropylene tubes.**
10. Incubate on ice for 30 minutes.
11. Centrifuge at 11,000 x g for 10 minutes at 4°C.
12. Keep the supernatant fraction, which contains the acid soluble proteins, and discard the acid-insoluble pellet.
13. Dialyze the supernatant against 200 mL 0.1 M (0.1N) acetic acid, twice for 1-2 hours each.
14. Dialyze three times against 200 mL H20 for 1 hour, 3 hours, and overnight, respectively. The protein can be quantified and lyophilized or stored at -70°C.

Lysis buffer:

10 mM HEPES, pH 7.9
1.5 mM MgCl2
10 mM KCl
*0.5 mM DTT

*1.5 mM PMSF

*Add PMSF and DTT just prior to use of the buffer.

Chromatin Immunoprecipitation

Part A. Optimization of DNA Shearing

Establish optimal conditions required for shearing cross-linked DNA to 200-1000 base pairs in length by following steps 1- 9 below. Vary the power setting and/or the number of 10-second pulses during sonication of the samples. Be sure to keep the sample on ice at all times (the sonication generates heat which will denature the DNA). Check the size of sonicated DNA by gel electrophoresis after reversion of cross-links. Our experience shows DNA is sheared to the appropriate length with 3-4 sets of 10-second pulses using a Cole Parmer, High Intensity Ultrasonic Processor/Sonicator, 50-watt model equipped with a 2 mm tip and set to 30% of maximum power.

Once sonication conditions have been optimized, keep cell number consistent for subsequent experiments. The protocol below for the optimization of DNA Shearing is for one Chip assay (~1 x 10^6 cells per condition).

Note: Steps 3-7 should be done on ice.

1. Stimulate or treat 1 x 10^6 cells on a 10 cm dish as appropriate. (Cells should be treated under conditions for which transcriptional activation of the gene of interest has been demonstrated). Include one extra dish (1 X 10^6 cells) to be used solely for estimation of cell number.
2. Cross link histones to DNA by adding formaldehyde directly to culture medium to a final concentration of 1% and incubate for 10 minutes at 37°C. (For example, add 270 µL 37% formaldehyde into 10 mL of growth medium on plate).
3. Aspirate medium, removing as much medium as possible. Wash cells twice using ice cold PBS containing protease inhibitors (1 mM phenylmethylsulfonyl fluoride (PMSF), 1 µg/mL aprotinin and 1 µg/mL pepstatin A). Note: Add protease inhibitors to PBS just prior to use. PMSF has a half-life of approximately 30 minutes in aqueous solutions.
4. Scrape cells into conical tube.
5. Pellet cells for 4 minutes at 2000 rpm at 4°C. Warm **SDS Lysis Buffer (Catalog # 20-163)** to room temperature to dissolve precipitated SDS and add protease inhibitors (inhibitors: 1 mM PMSF, 1 µg/mL aprotinin and 1 µg/mL pepstatin A).
6. Resuspend cell pellet in 200 µL of **SDS Lysis Buffer (Catalog # 20-163)** and incubate for 10 minutes on ice. **Note:** The 200 µL of SDS Lysis Buffer is per 1 X 10^6 cells; if more cells are used, the resuspended cell pellet should be divided into 200 µL aliquots so that each 200 µL aliquot contains ~1 X 10^6 cells.
7. Sonicate lysate to shear DNA to lengths between 200 and 1000 basepairs being sure to keep samples ice cold (***Note:*** Once sonication conditions have been optimized following steps 1 to 9, proceed to Part B, step 1 below).
8. Add 8 µL **5 M NaCl (Catalog # 20-159)** and reverse crosslinks at 65°C for 4 hours.
9. Recover DNA by phenol/chloroform extraction and run sample (example 5 µL,10 µL and 20 µL samples) in an agarose gel to visualize shearing efficiency.

Part B. Experimental protocol.

If sonication conditions have been optimized (Part A), complete steps 1 through 7 and continue with the protocol below. For a negative/background control, prepare a sample to use as a no-antibody immunoprecipitation control in step 5 below. Additionally, transcriptionally unactivated DNA samples should be prepared as controls for PCR in section II.

1. Centrifuge samples (part A, step 7) for 10 minutes at 13,000 rpm at 4°C, and add 200 µL of the sonicated cell pellet suspension to a new 2 mL-microcentrifuge tube.
2. Dilute the sonicated cell pellet suspension 10 fold in **ChIP Dilution Buffer (Catalog # 20-153)**, adding protease inhibitors as above. This is done by adding 1800 µL ChIP Dilution Buffer to the 200 µL sonicated cell pellet suspension for a final volume of 2 mL in each immunoprecipitation condition. **Note**: *If proceeding to PCR a portion of the diluted cell pellet suspension 1% (~20 µL) can be kept to quantitate the amount of DNA present in different samples at the PCR protocol, Part B, section II, step 6. This sample is considered to be your input/starting material and needs to have the Histone-DNA crosslinks reversed by heating at 65°C for 4 hours (see section II, step 3.)*
3. To reduce nonspecific background, pre-clear the 2 mL diluted cell pellet suspension with 80 µL of **Salmon Sperm DNA/Protein A Agarose-50% Slurry (Catalog # 16-157)** for 30 minutes at 4°C with agitation.
4. Pellet agarose by brief centrifugation and collect the supernatant fraction.
5. Add the immunoprecipitating antibody (the amount will vary per antibody) to the 2 mL supernatant fraction and incubate overnight at 4°C with rotation. *For a negative control, perform a no-antibody immunoprecipitation by incubating the supernatant fraction with 60 µL of **Salmon Sperm DNA/Protein A Agarose- 50% Slurry (Catalog # 16-157)** for one hour at 4°C with rotation and proceed to step 7.*
6. Add 60 µL of **Salmon Sperm DNA/Protein A Agarose Slurry (Catalog # 16-157)** for one hour at 4°C with rotation to collect the antibody/histone complex.
7. Pellet agarose by gentle centrifugation (700 to 1000 rpm at 4°C, ~1 min). Carefully remove the supernatant that contains unbound, non-specific DNA. Wash the protein A agarose/antibody/histone complex for 3-5 minutes on a rotating platform with 1 mL of each of the buffers listed in the order as given below:
   - **a.** Low Salt Immune Complex Wash Buffer (Catalog # 20-154), **one wash**
   - **b.** High Salt Immune Complex Wash Buffer (Catalog # 20-155), **one wash**
   - **c.** LiCl Immune Complex Wash Buffer (Catalog # 20-156), **one wash**
   - **d.** TE Buffer (Catalog # 20-157), **two washes.**

After step 7 above, the sample is now a protein A/antibody/histone/DNA complex ready for either an Immunoprecipitation/Western Blot assay (Section I) or Polymerase Chain Reaction (PCR) assay (Section II).

Section I. Immunoprecipitation/Western Blot protocol to detect histone.

1. Following washing of the beads in part B, step 7, immunoprecipitated histones can be analyzed by Western Blot analysis. Add 25 µL of 1X Laemmli buffer per sample and boil for 10 minutes. Load 20 µL per lane and perform western blot procedure as described per appropriate antibody.

Section II. PCR protocol to amplify DNA that is bound to the immunoprecipitated histone.

1. Freshly prepare elution buffer (1%SDS, 0.1 M NaHCO3).
2. Elute the histone complex from the antibody by adding 250 µL elution buffer to the pelleted protein A agarose/antibody/histone complex from step 7d above. Vortex briefly to mix and incubate at room temperature for 15 minutes with rotation. Spin down agarose, and carefully transfer the supernatant fraction (eluate) to another tube and repeat elution. Combine eluates (total volume = ~500 µL).
3. Add 20 µL 5 M NaCl (Catalog # 20-159) to the combined eluates (500 µL) and reverse histone-DNA crosslinks by heating at 65°C for 4 hours. At this step the sample can be stored and -20°C and the protocol continued the next day.

Note: Include the input/starting material *(the sample saved from Part B, step 2, which has had the Histone-DNA crosslinks reversed) as well as a transcriptionally - unactivated DNA sample as negative and background controls for the PCR reaction. Previously, a 5µl sample has been used in a nested PCR reaction. However, the amount of sample used per reaction must be determined empirically (e.g., titrate the sample at this step by using 1, 2, 5, or 10µl per PCR reaction)*. If PCR results are poor, complete steps 4, 5 and 6 below to purify the DNA sample. NOTE: Handle the samples carefully, some DNA may be lost during the purification steps.

4. Add 10 µL of 0.5 M EDTA (Catalog # 20-158), 20 µL 1 M Tris-HCl, pH 6.5 (Catalog # 20-160) and 2 µL of 10 mg/mL Proteinase K to the combined eluates and incubate for one hour at 45°C.
5. Recover DNA by phenol/chloroform extraction and ethanol precipitation. Addition of an inert carrier, such as 20 µg glycogen or yeast tRNA, helps visualize the DNA pellet. Wash pellets with 70% ethanol and air dry.
6. Resuspend pellets in an appropriate buffer for PCR or slot-blot reactions. PCR or slot-blot conditions must be determined empirically.

RELATED PRODUCTS (specific)

cat #	description
06-866	Anti-acetyl-Histone H4
06-761	Anti-acetyl-Histone H4 (Lys12)
07-329	Anti-acetyl-Histone H4 (Lys16)
06-759	Anti-acetyl-Histone H4 (Lys5)
04-118	Anti-acetyl-Histone H4 (Lys5), rabbit monoclonal
06-760	Anti-acetyl-Histone H4 (Lys8)
04-557	Anti-acetyl-Histone H4, pan (Lys 5,8,12)
07-213	Anti-dimethyl-Histone H4 (Arg3)
05-672	Anti-dimethyl-Histone H4 (Lys20), clone 6G7/H4
05-734	Anti-dimethyl-Histone H4 (Lys79), clone ER133
05-754	Anti-di-tri-methyl-Histone H4 (Lys20), clone AW317
07-596	Anti-Histone H4 (citrulline 3)
05-858	Anti-Histone H4, pan
06-946	Anti-hyperacetylated Histone H4 (Penta)
05-735	Anti-monomethyl-Histone H4 (Lys20), clone NL314
04-079	Anti-trimethyl-Histone H4 (Lys20), rabbit monoclonal
17-211	Acetyl-Histone H4 Antibody Set
17-229	Acetyl-Histone H4 Immunoprecipitation (ChIP) Assay Kit
17-212	Acetyl-Histone H4 Peptide Pack
17-217	Acetyl-Histone H4 Site Specificity Pack
12-348	Goat Anti-Rabbit IgG
17-500	Catch and Release Reversible Immunoprecipitation System
16-266	Protein G Agarose Fast Flow 10 mL
16-125	Protein A-Agarose 10 mL

RELATED PRODUCTS (non-specific)

cat #	description
IPVH00010	Immobilon-P 26.5 cm x 3.75 m Roll PVDF 0.45 um IPVH07850
IPFL00010	Immobilon-FL 26.5 cm x 3.75 m Roll PVDF 0.45 um
IPVH07850	Immobilon-P 7 x 8.4 cm PVDF 0.45 mm (sheet) 50/pk
ISEQ00010	Immobilon-P SQ 26.5 cm x 3.75 m 1 roll PVDF 0.2 um
ISEQ07850	Immobilon-P 7 x 8.4 cm PVDF 0.2 mm (sheet) 50/pk
IPFL07810	Immobilon-FL 7 x 8.4 cm PVDF 0.45 mm (sheet) 10/pk
WBKLS0050	IMMOBILON WESTERN CHEMILUM HRP SUBSTRATE 50 mL
17-373SP	Spray & Glow™ ECL Western Blotting 40 mL
2060	Re-Blot Western Blot Recycling Kit
2500	Re-Blot Plus Western Blot Recycling Kit
B2080-175GM	Blot Quick Blocker Membrane Blocking Agent 175G
2170	CHEMIBLOCKER-1LT
20-200	IMMUNOBLOT BLOCKING REAGENT 20G
12-302	EGF-Stimulated A431 Cell Lysate
12-349	Goat Anti-Mouse IgG, HRP conjugate
12-110	Phosphotyrosine control (EGF-stim A431 cell lysate)

Tau Monoclonal Antibody (HT7)

Product Details	
Size	100 µg
Species Reactivity	Bovine, Human
Published Species	Pig, Rat, Non-human primate, Human, Mouse
Host/Isotype	Mouse / IgG1, kappa
Class	Monoclonal
Type	Antibody
Clone	HT7
Conjugate	Unconjugated
Immunogen	Purified human Tau, epitope human Tau between residue 159 and 163 (numbering according to human Tau40), corresponding to the amino acid sequence PPGQK.
Form	Liquid
Concentration	0.2 mg/mL
Purification	Protein A
Storage buffer	PBS
Contains	no preservative
Storage conditions	-20° C, Avoid Freeze/Thaw Cycles
RRID	AB_2314654

Applications	Tested Dilution	Publications
Western Blot (WB)	1-2 µg/mL	78 Publications
Immunohistochemistry (IHC)	-	35 Publications
Immunohistochemistry (Paraffin) (IHC (P))	5-10 µg/mL	6 Publications
Immunohistochemistry (PFA fixed) (IHC (PFA))	-	3 Publications
Immunohistochemistry - Free Floating (IHC (Free))	Assay-dependent	2 Publications
Immunocytochemistry (ICC/IF)	-	13 Publications
ELISA (ELISA)	2-10 µg/mL	9 Publications
Immunoprecipitation (IP)	-	7 Publications
Neutralization (Neu)	-	1 Publication
Dot blot (DB)	-	2 Publications
Miscellaneous PubMed (Misc)	-	1 Publication

Product Specific Information

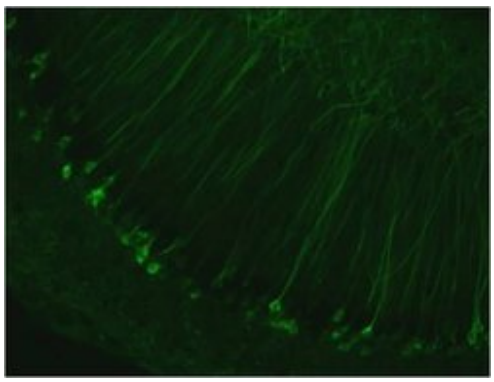
MN1000 targets Tau in ELISA, IF, ICC, IHC (P), and WB applications and shows reactivity with Bovine, and Human samples.

The MN1000 immunogen is purified human Tau, epitope human Tau between residue 159 and 163 (numbering according to

human Tau40), corresponding to the amino acid sequence PPGQK.

MN1000 detects Tau which has a predicted molecular weight of approximately 79 kDa.

Product Images For Tau Monoclonal Antibody (HT7)



Tau Antibody (MN1000) in IHC (Free)
Immunohistochemistry was performed on paraformaldehyde-fixed free-floating hippocampus tissue sections from transgenic (3xTg-AD) mice that express human Tau. Tissues were blocked in 5% normal horse serum and 0.4% Triton X-100 for 60 minutes at room temperature and probed with a Tau monoclonal antibody (Product # MN1000) at a dilution of 1:1000 at 4°C overnight. Tissues were washed extensively with PBS. Detection was performed using a fluorophore-conjugated anti-mouse IgG secondary antibody at a dilution of 1:1000. Tissues were visualized by fluorescence microscopy. Data courtesy of the Innovators Program.

Tau Antibody (MN1000) in WB
Western blot analysis of human Tau was performed by loading 15 µg of sarkosyl-soluble cortex lysate from transgenic (3xTg-AD) mice that express human Tau per well onto an SDS-PAGE gel. Proteins were transferred to a PVDF membrane and blocked with 5% milk + 0.5% BSA in PBST buffer for 1 hour at room temperature. The membrane was probed with a Tau monoclonal antibody (Product # MN1000) at a dilution of 1:1000 at 4°C overnight, washed in PBST, and probed with an HRP-conjugated anti-mouse IgG secondary antibody at a dilution of 1:60,000. Detection was performed using a chemiluminescent substrate. Data courtesy of the Innovators Program.

157 References

Western Blot (78)

Molecular psychiatry

Loss of function of the mitochondrial peptidase PITRM1 induces proteotoxic stress and Alzheimer's disease-like pathology in human cerebral organoids.

"MN1000 was used in Western Blotting to support a mechanistic link between mitochondrial function and common neurodegenerative proteinopathies."

Authors: Pérez MJ,Ivanyuk D,Panagiotakopoulou V,Di Napoli G,Kalb S,Brunetti D,Al-Shaana R,Kaeser SA,Fraschka SA,Jucker M,Zeviani M,Viscomi C,Deleidi M

Species
Human

Dilution
1:1000

Year
2021

Journal of Alzheimer's disease : JAD

Partial Inhibition of Mitochondrial Complex I Reduces Tau Pathology and Improves Energy Homeostasis and Synaptic Function in 3xTg-AD Mice.

"MN1000 was used in Western Blotting to investigate the effect of specific MCI inhibitor tricyclic pyrone compound CP2 on levels of human pTau, memory function, long term potentiation (LTP), and energy homeostasis in 18-month-old 3xTg-AD mice and explore the potential mechanisms."

Authors: Stojakovic A,Chang SY,Nesbitt J,Pichurin NP,Ostroot MA,Aikawa T,Kanekiyo T,Trushina E

Species
Mouse

Dilution
1:1000

Year
2021

View more WB references on thermofisher.cn

Immunohistochemistry (35)

eNeuro

Increased Tau Expression Correlates with Neuronal Maturation in the Developing Human Cerebral Cortex.

"MN1000 was used in Immunohistochemistry to demonstrate that tau increases with neuronal maturation in both the developing fetal brain and iPSC-derived organoids."

Authors: Fiock KL,Smalley ME,Crary JF,Pasca AM,Hefti MM

Species
Human

Dilution
Not Cited

Year
2021

The Journal of biological chemistry

Differential compartmental processing and phosphorylation of pathogenic human tau and native mouse tau in the line 66 model of frontotemporal dementia.

"MN1000 was used in Western Blot, Immunohistochemistry to examine the cellular distribution of tau protein species in human tau overexpressing line 66 mice, a transgenic mouse model akin to genetic variants of frontotemporal dementia."

Authors: Lemke N,Melis V,Lauer D,Magbagbeolu M,Neumann B,Harrington CR,Riedel G,Wischik CM,Theuring F,Schwab K

Species
Human

Dilution
1:2000

Year
2020

View more IHC references on thermofisher.cn

More applications with references on thermofisher.cn

IHC (P) (6) | IHC (PFA) (3) | IHC (Free) (2) | ICC/IF (13) | ELISA (9) | IP (7) | Neu (1) | DB (2) | Misc (1)

Revision 1

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#3724

HA-Tag (C29F4) Rabbit mAb

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Applications:	Reactivity:	Sensitivity:	Source/Isotype:
WB, IP, IHC-P, IF-IC, F, ChIP	All	Transfected Only	Rabbit IgG

Product Usage Information

For optimal ChIP results, use 10 µl of antibody and 10 µg of chromatin (approximately 4 x 10^6 cells) per IP. This antibody has been validated using SimpleChIP® Enzymatic Chromatin IP Kits.

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:50
Immunohistochemistry (Paraffin)	1:800 - 1:3200
Immunofluorescence (Immunocytochemistry)	1:800 - 1:1600
Flow Cytometry	1:800 - 1:1600
Chromatin IP	1:50

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.

Specificity / Sensitivity

HA-Tag (C29F4) Rabbit mAb detects exogenously expressed proteins containing the HA epitope tag. The antibody may cross-react with a protein of unknown origin ~100kDa.

Species Reactivity:
All Species Expected

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic peptide containing the influenza hemagglutinin epitope (YPYDVPDYA).

Background

Epitope tags are useful for the labeling and detection of proteins using immunoblotting, immunoprecipitation, and immunostaining techniques. Because of their small size, they are unlikely to affect the tagged protein's biochemical properties.

The HA tag is derived from an epitope of the influenza hemagglutinin protein which has been used extensively as a general epitope tag in expression vectors (1).

1. Field, J. et al. (1988) *Mol Cell Biol* 8, 2159-65.

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

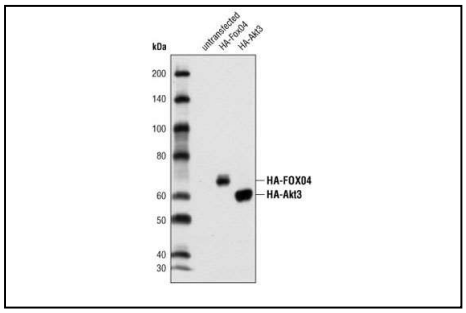
APPLICATIONS KEY **WB:** Western Blot **IP:** Immunoprecipitation **IHC:** Immunohistochemistry **ChIP:** Chromatin Immunoprecipitation **IF:** Immunofluorescence **F:** Flow Cytometry **E-P:** ELISA-Peptide

CROSS-REACTIVITY KEY **H:** human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **Mi:** mink **C:** chicken **Dm:** D. melanogaster **X:** Xenopus **Z:** zebrafish **B:** bovine **Dg:** dog **Pg:** pig **Sc:** S. cerevisiae **Ce:** C. elegans **Hr:** horse **All:** all species expected

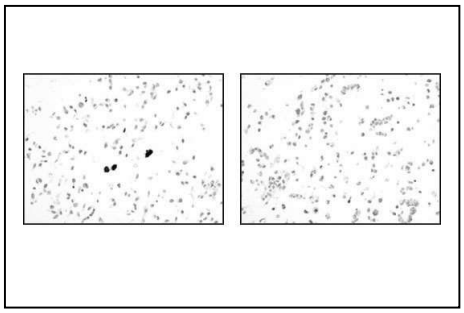
Revision 1

#3724

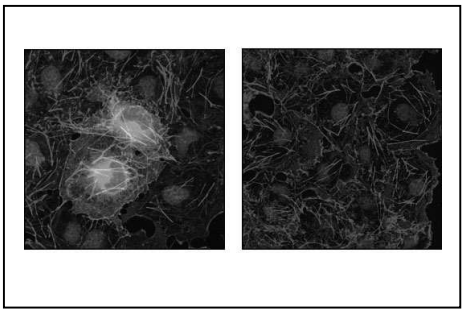
HA-Tag (C29F4) Rabbit mAb



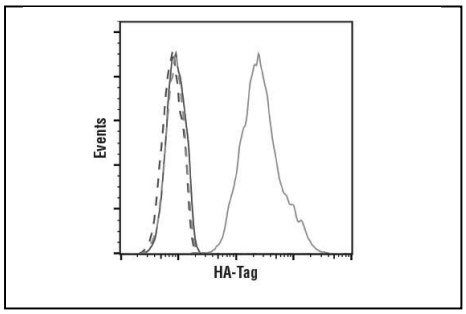
Western blot analysis of extracts from HeLa cells, untransfected or transfected with either HA-FoxO4 or HA-Akt3, using HA-Tag (C29F4) Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded COS cells, untransfected (right) or HA-Tag transfected (left), using HA-Tag (C29F4) Rabbit mAb.

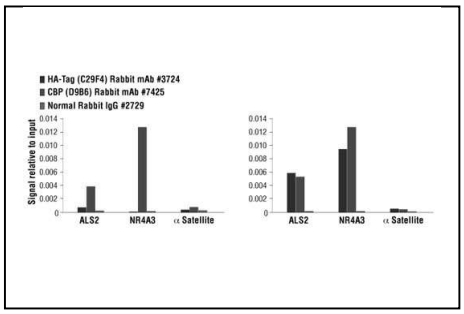


Confocal immunofluorescent analysis of COS cells, transfected with an HA-tagged protein (left) or mock-transfected (right), using HA-Tag (C29F4) Rabbit mAb (green). Actin filaments have been labeled using DY-554 phalloidin (red). Blue pseudocolor = DRAQ5® (fluorescent DNA dye).



Flow cytometric analysis of 293T cells untransfected (blue) or transfected with HA-tagged Akt (green), using HA-Tag (C29F4) Rabbit mAb (solid lines) or concentration matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (dashed lines).

Anti-rabbit IgG (H+L), F(ab')2 Fragment (Alexa Fluor® 488 Conjugate) #4412 was used as a secondary antibody.



293T cells were either untransfected (left panel) or transfected with an HA-tagged human CBP construct (right panel), then treated with Forskolin #3828 (30 µM) . Chromatin immunoprecipitations were performed with cross-linked chromatin from cells and HA-Tag (C29F4) Rabbit mAb, CBP (D9B6) Rabbit mAb, or Normal Rabbit IgG #2729 using SimpleChIP® Enzymatic Chromatin IP Kit (Magnetic Beads) #9003. The enriched DNA was quantified by real-time PCR using human ALS2 exon 1 primers, SimpleChIP® Human NR4A3 Promoter Primers #4829, and SimpleChIP® Human α Satellite Repeat Primers #4486. The amount of immunoprecipitated DNA in each sample is represented as signal relative to the total amount of input chromatin, which is equivalent to one.

#3724

HA-Tag (C29F4) Rabbit mAb

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abcam

Product datasheet

Anti-GFP antibody ab290

★★★★★ 178 Abreviews 2676 References 11 图像

概述

产品名称	Anti-GFP抗体
描述	**兔多克隆抗体**to GFP
宿主	Rabbit
特异性	Anti-GFP antibody (ab290) is a highly versatile antibody that gives a stronger signal than other anti-GFP antibodies available. On Western blot the antibody detects the GFP fraction from cell extracts expressing recombinant GFP fusion proteins and has also been shown to be useful on mouse sections fixed with formalin. In Immunocytochemistry, the antibody gives a very good signal on recombinant YES-GFP chimeras expressed in COS cells (McCabe et al. 1999 and figure below). It is routinely used in Immunoprecipitation (IP) and IP-Western protocols and has been used successfully in HRP Immunohistochemistry at 1:200 on whole-mount mouse embryos. **GFP antibody is reactive against all variants of *Aequorea victoria* GFP such as S65T-GFP, RS-GFP, YFP, CFP, RFP and EGFP.**
经测试应用	**适用于:** ELISA, IHC-Fr, ICC, IHC-P, IP, WB, IHC-FoFr, IHC-FrFl, Electron Microscopy
种属反应性	**与反应:** Species independent
免疫原	Recombinant full length protein corresponding to GFP. Green fluorescent protein (GFP) from Aequorea victoria. Database link: P42212
阳性对照	The Recombinant A. victoria GFP protein (ab84191), any other purified recombinant GFP, any cell line confirmed to overexpress GFP. ICC: NIH3T3, U2OS and glandular stomach cells. IHC: Mouse brain and dog heart tissue. WB: Sample: COS7 and LNCaP whole cell lysate - transfected with GFP-Eml4.
常规说明	The total IgG concentration has been determined to be 5 mg/mL. The specific IgG concentration is unknown. This product should be kept refrigerated at all times whilst in short term storage. Using sterilised equipment will reduce the risk of bacterial contamination. Anti-GFP antibody (ab6556) is the purified version of this antibody (see Related Products). The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or

contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
存储溶液	Preservative: 0.05% Sodium azide Constituent: 1.25% Sodium chloride
纯度	Whole antiserum
纯化说明	This antibody is provided as whole antiserum. It is not possible to determine the exact antibody concentration, since whole serum contains many other host serum proteins besides the antibody of interest.
克隆	**多克隆**
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab290于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
ELISA		Use at an assay dependent concentration.
IHC-Fr	★★★★★ (8)	Use at an assay dependent concentration. Reported to work at dilutions up to 1/3000. Use secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab15077).
ICC	★★★★★ (2)	1/200 - 1/1000. We recommend Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150081) secondary antibody.
IHC-P	★★★★★ (24)	1/500 - 1/1000. Perform heat mediated antigen retrieval via the microwave method before commencing with IHC staining protocol.
IP	★★★★★ (24)	Use at an assay dependent concentration. Use at 1µl per 10cm tissue culture dish (use 10µl protein A agarose CL4B to precipitate the immune complex).

应用	Ab评论	说明
WB	★★★★★ (69)	1/1000 - 1/2500. It is recommended to use 12.5% SDS-PAGE and to transfer to PVDF membrane. Use 1x Blotto (or 3% BSA in PBS) for diluting and blocking. Use PBS in 3x 5min washing steps throughout the immunolabelling. Probe with ab290 at 1:1000 - 1:5000 dilution and use Goat Anti-Rabbit IgG H&L (HRP) (ab205718) at 1:5000 dilution with ECL detection method. ab290 has been reported to
IHC-FoFr	★★★★★ (5)	1/200 - 1/500.
IHC-FrFl	★★★★★ (2)	Use at an assay dependent concentration.
Electron Microscopy		1/1000 - 1/4000.

靶标

相关性

Function: Energy-transfer acceptor. Its role is to transduce the blue chemiluminescence of the protein aequorin into green fluorescent light by energy transfer. Fluoresces in vivo upon receiving energy from the Ca^{2+} -activated photoprotein aequorin.

Subunit structure: Monomer.

Tissue specificity: Photocytes.

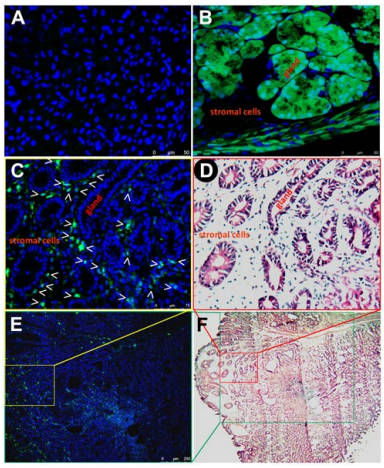
Post-translational modification: Contains a chromophore consisting of modified amino acid residues. The chromophore is formed by autocatalytic backbone condensation between Ser-65 and Gly-67, and oxidation of Tyr-66 to didehydrotyrosine. Maturation of the chromophore requires nothing other than molecular oxygen.

Biotechnological use: Green fluorescent protein has been engineered to produce a vast number of variously colored mutants, fusion proteins, and biosensors. Fluorescent proteins and its mutated allelic forms, blue, cyan and yellow have become a useful and ubiquitous tool for making chimeric proteins, where they function as a fluorescent protein tag. Typically they tolerate N- and C-terminal fusion to a broad variety of proteins. They have been expressed in most known cell types and are used as a noninvasive fluorescent marker in living cells and organisms. They enable a wide range of applications where they have functioned as a cell lineage tracer, reporter of gene expression, or as a measure of protein-protein interactions. Can also be used as a molecular thermometer, allowing accurate temperature measurements in fluids. The measurement process relies on the detection of the blinking of GFP using fluorescence correlation spectroscopy.

Sequence similarities: Belongs to the GFP family.

Biophysicochemical properties: Absorption: Abs(max)=395 nm
Exhibits a smaller absorbance peak at 470 nm. The fluorescence emission spectrum peaks at 509 nm with a shoulder at 540 nm.

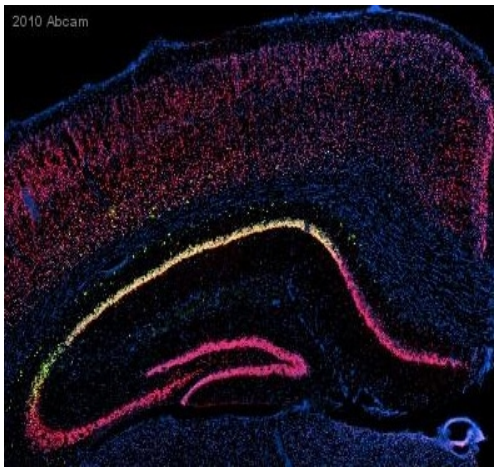
图片



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GFP antibody (ab290)

Bone marrow-derived infiltrating cells in the stromal tissue of gastric intraepithelial tumor traced by GFP direct fluorescence.

(**A**) Normal tissues of the glandular stomach of a regular GFP(−) control mouse. (**B**) Normal tissues of the glandular stomach of a GFP(+) transgenic control mouse; (**C, E, D, F**) An induced gastric intraepithelial neoplasia (GIN) in a bone marrow transplanted mouse. GFP(+) BMDCs tracked with direct fluorescence localized in the GIN stromal tissue are shown in **C** and **E**. The same GIN lesion slide stained by H&E after the fluorescence observation are shown in **D** and **F**. DAPI (**A–C** and **E**) and hematoxylin (**D** and **F**) are used to visualize nuclei, respectively. Locations of the images **C** and **D** in the images **E** and **F**, and the image **E** in the image **F** are marked in the corresponding color. The gastric glands and stromal cells are also labeled.

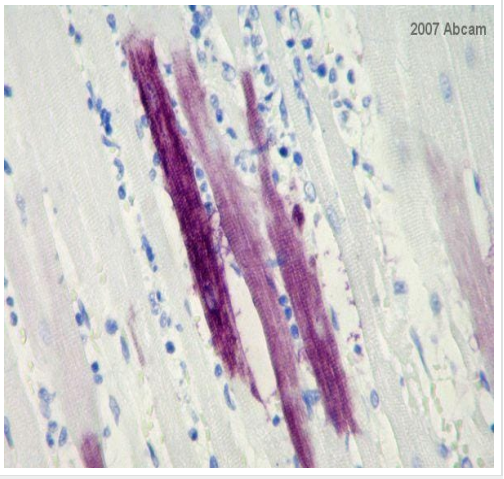


Immunohistochemistry - Free Floating - Anti-GFP antibody (ab290)

This image is courtesy of an Abreview submitted by Judith Kranz

Immunohistochemistry (Free Floating) analysis of mouse brain tissue sections labelling GFP with ab290. Tissue was fixed with 4% PFA, frozen 30 µm sections were blocked for 1 hour at room temperature with 10% normal goat serum + donkey anti-mouse IgG Fab fragments (0.1 mg/ml). Sections were incubated with the primary antibody at a dilution of 1/1000 in TBS + 0.25% Triton-X for 16 hours at 4°C. A Cy2®-conjugated donkey anti-rabbit IgG (H+L) at a dilution of 1/200 was used as the secondary antibody.

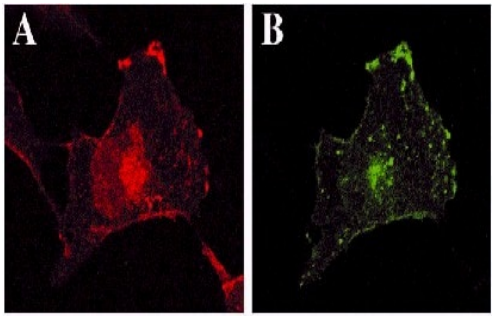
Image shows anti-NeuN (red), DAPI (blue), and anti-GFP staining of GFP-cre (green, yellow with NeuN colocalization).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-GFP antibody (ab290)

This image is courtesy of an anonymous Abreview

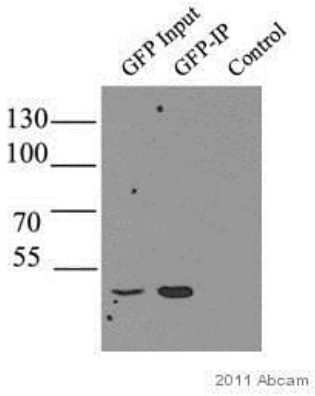
ab290 staining dog hearts (Adv-GFP injection) tissue sections by IHC-P. Sections were PFA fixed and subjected to heat mediated antigen retrieval in citric acid (Ph6.0, 0.05% Tween20) prior to blocking with 10% serum for 30 mins at 37°C. The primary antibody was diluted 1/1000 in PBS and incubated with the sample for 1 hour at 25°C. A HRP conjugated secondary like Goat Anti-Rabbit IgG H&L (HRP) (ab205718) was used.



Immunocytochemistry - Anti-GFP antibody (ab290)

Immunofluorescence images showing similar localization of Yes-GFP (first 10 aa's of Yes PTK fused to the N-terminus of GFP) to full length Yes PTK. A: Distribution of Yes detected using mouse anti-Yes Ab followed by Texas Red-conjugated anti-mouse Ab. B: Chimeric GFP's detected using rabbit anti-GFP Ab (Abcam ab290) followed by FITC-conjugated anti-rabbit Ab.

Image kindly provided by L.G. Berthiaume. Taken from J. McCabe and L.G. Berthiaume, Functional Roles for Fatty Acylated Amino-terminal Domains in Subcellular Localization, *Molecular Biology of the Cell* **10**:3771-3786, 1999

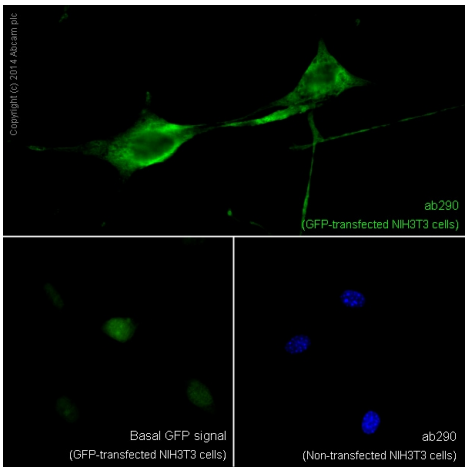


Immunoprecipitation - Anti-GFP antibody (ab290)

This image is courtesy of an Abreview submitted by William Hung

ab290 immunoprecipitating GFP in HEK293 nuclear lysate expressing GFP. 20µg of lysate was incubated with primary antibody (1 µg/mg lysate) and matrix (Protein G) for 16 hours at 4°C in AFC low salt buffer. For western blotting ab290 (1/5000) was used to confirm successful immunoprecipation.

Lane 1: HEK293 nuclear lysate expressing GFP input.
Lane 2: IP of HEK293 nuclear lysate expressing GFP.
Lane 3: Cells with no GFP.

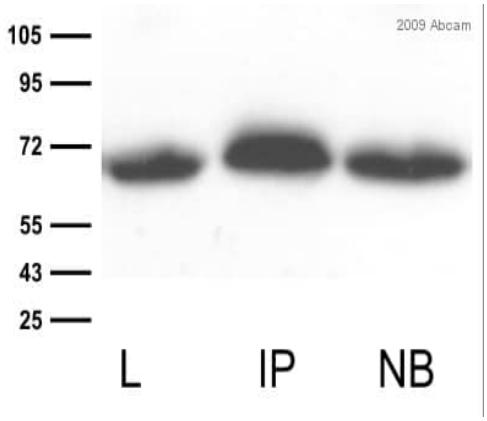


Immunocytochemistry - Anti-GFP antibody (ab290)

ab290 staining GFP in GFP-transfected NIH3T3 cells. The cells were fixed with 4% formaldehyde (10min) and then blocked in 1% BSA / 0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated with ab290 at 1/200 dilution overnight at +4°C followed by incubation with Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150081), for 1 hour, at 1µg/ml.

Under identical experimental conditions, when compared to the basal level of GFP expression in transfected NIH3T3 cells, the cells upon which ab290 was applied gave a stronger signal in the 488 channel, indicating that ab290 is binding to GFP and therefore eliciting signal amplification.

ab290 was also applied to non-GFP-transfected NIH3T3 cells, which produced no positive staining, indicating specificity for GFP. Nuclear DNA was labelled with 1.43µM DAPI (blue).

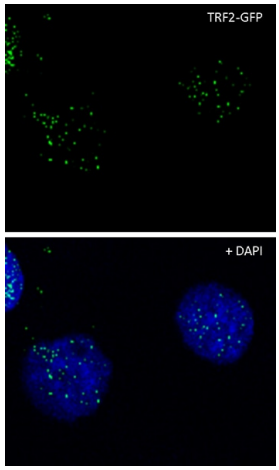


Immunoprecipitation - Anti-GFP antibody (ab290)

This image is courtesy of an Abreview submitted by Vladimir Milenkovic

ab290 immunoprecipitate in human HEK293 cells transfected with Annexin1-GFP. 25µg of cell lysate was incubated with the primary antibody and matrix (Protein G) in 1% TX-100, 10% glycerol, 1X PBS for 16 hours at 4°C. For Western blotting anti-rabbit HRP conjugated secondary antibody was used at a dilution at 1/5000.

Lane 1: Lysate of HEK293 cells expressing Annexin1-GFP fusion protein.
Lane 2: IP with anti-GFP.
Lane 3: Not bound fraction.

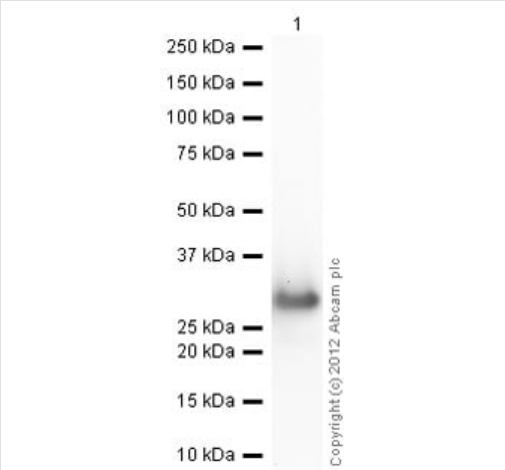


Immunocytochemistry - Anti-GFP antibody (ab290)

This image is courtesy of an anonymous Abreview

ab290 staining GFP in U2OS cells expressing TRF2-GFP fusion protein by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with formaldehyde, permeabilized with NP40 and blocked with 3% BSA for 1 hour at 21°C. Samples were incubated with the primary antibody (1/1000 in PBS + 3% BSA) for 12 hours at 4°C. An Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) at a dilution of 1/500 was used as the secondary antibody.

Green - GFP.
Blue - DAPI.



Western blot - Anti-GFP antibody (ab290)

Anti-GFP antibody (ab290) at 1/2500 dilution + Recombinant *A. victoria* GFP protein (ab84191) at 0.01 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

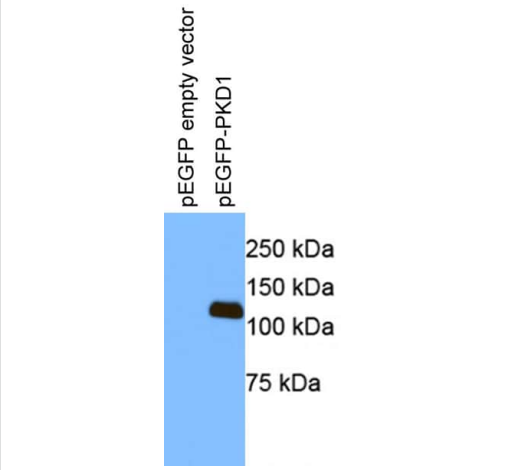
Developed using the ECL technique.

Performed under reducing conditions.

Observed band size: 27 kDa

Exposure time: 30 seconds

Secondary antibody - goat anti-rabbit HRP preadsorbed (ab97080)



Western blot - Anti-GFP antibody (ab290)

This image is courtesy of an anonymous Abreview

All lanes : Anti-GFP antibody (ab290) at 1/5000 dilution

Lane 1 : LNCaP whole cell lysate - pEGFP empty vector

Lane 2 : LNCaP whole cell lysate - pEGFP-PKD1 transfected

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : HRP-conjugated goat anti-rabbit IgG at 1/10000 dilution

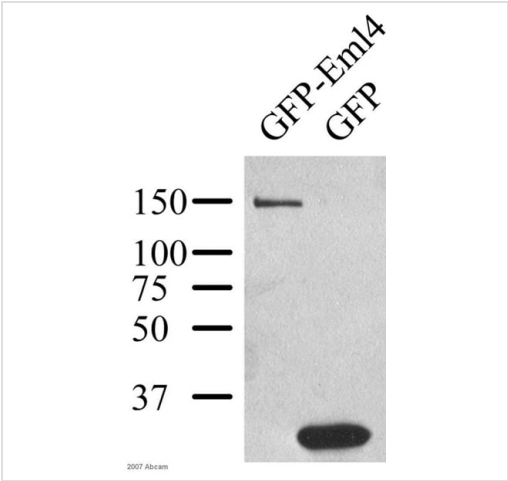
Developed using the ECL technique.

Performed under reducing conditions.

Exposure time: 10 seconds

Blocked with 5% milk for 1 hour at 23°C.

Incubated with the primary antibody for 16 hours at 4°C.



Western blot - Anti-GFP antibody (ab290)

This image is courtesy of an Abreview submitted by S Houtman

All lanes : Anti-GFP antibody (ab290) at 1/5000 dilution

Lane 1 : COS7 whole cell lysate - transfected with GFP-Eml4

Lane 2 : COS7 whole cell lysate - transfected with GFP

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : HRP-conjugated pig anti-rabbit IgG at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Observed band size: 30 kDa

Exposure time: 10 seconds

Blocked with 5% milk for 1 hour at 20°C.

Incubated with the primary antibody for 18 hours at 4°C in TBS containing 2% milk and 1% Tween.

Predicted MW of Eml4 ~ 120 kDa.

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Product datasheet

Anti-NMDAR1 antibody [EPR2481(2)] ab109182

重组 RabMAb

4 Abreviews 47 References 6 图像

概述

产品名称	Anti-NMDAR1抗体[EPR2481(2)]
描述	兔单克隆抗体[EPR2481(2)] to NMDAR1
宿主	Rabbit
经测试应用	**适用于:** WB, ICC/IF **不适用于:** IHC-P
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	Fetal brain cell lysate. ICC/IF: Mouse primary neuron cells
常规说明	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. **We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.**

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
存储溶液	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 40% Glycerol, 59% PBS, 0.05% BSA
纯度	Protein A purified

克隆	单克隆
克隆编号	EPR2481(2)
同种型	IgG

应用

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"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

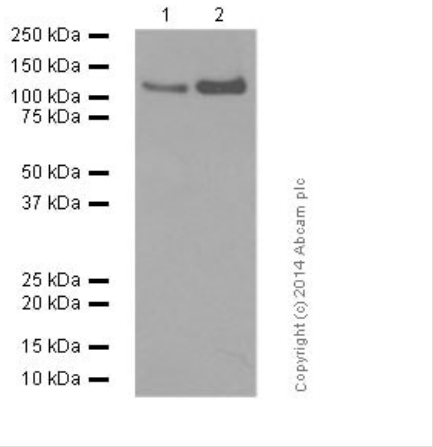
应用	Ab评论	说明
WB	★★★★☆ (4)	1/1000 - 1/10000. Detects a band of approximately 120 kDa (predicted molecular weight: 105 kDa).
ICC/IF		1/50.

应用说明 Is unsuitable for IHC-P.

靶标

功能	NMDA receptor subtype of glutamate-gated ion channels with high calcium permeability and voltage-dependent sensitivity to magnesium. Mediated by glycine. This protein plays a key role in synaptic plasticity, synaptogenesis, excitotoxicity, memory acquisition and learning. It mediates neuronal functions in glutamate neurotransmission. Is involved in the cell surface targeting of NMDA receptors.
序列相似性	Belongs to the glutamate-gated ion channel (TC 1.A.10.1) family. NR1/GRIN1 subfamily.
翻译后修饰	NMDA is probably regulated by C-terminal phosphorylation of an isoform of NR1 by PKC. Dephosphorylated on Ser-897 probably by protein phosphatase 2A (PPP2CB). Its phosphorylated state is influenced by the formation of the NMDAR-PPP2CB complex and the NMDAR channel activity.
细胞定位	Cell membrane. Cell junction > synapse > postsynaptic cell membrane. Cell junction > synapse > postsynaptic cell membrane > postsynaptic density. Enriched in post-synaptic plasma membrane and post-synaptic densities.

图片



Western blot - Anti-NMDAR1 antibody [EPR2481(2)] (ab109182)

All lanes : Anti-NMDAR1 antibody [EPR2481(2)] (ab109182) at 1/5000 dilution (purified)

Lane 1 : Mouse brain tissue lysate
Lane 2 : Rat brain tissue lysate

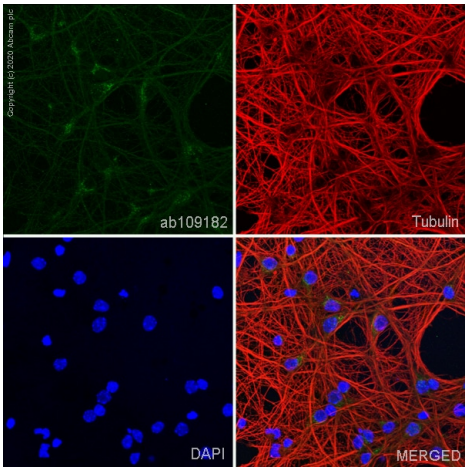
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 105 kDa
Observed band size: 120 kDa

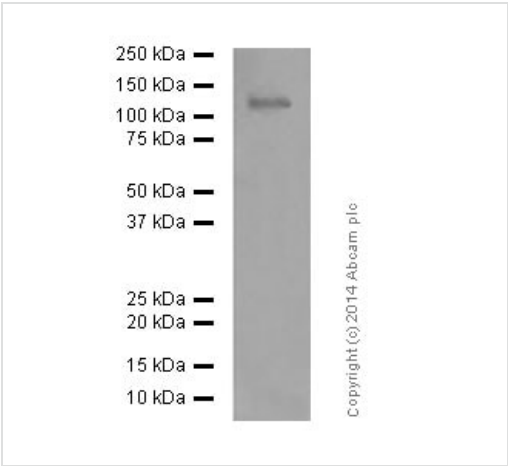
Blocking buffer: 5% NFDM/TBST

Dilution buffer: 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Anti-NMDAR1 antibody [EPR2481(2)] (ab109182)

Immunocytochemistry/ Immunofluorescence analysis of mouse primary neuron cells labeling NMDAR1 with purified ab109182 at 1/50 (9.5µg/mL). Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 µg/mL). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1/1000 (2 µg/mL) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.
Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection.



Western blot - Anti-NMDAR1 antibody [EPR2481(2)] (ab109182)

Anti-NMDAR1 antibody [EPR2481(2)] (ab109182) at 1/1000 dilution (purified) + Human cerebellum tissue lysate at 20 µg

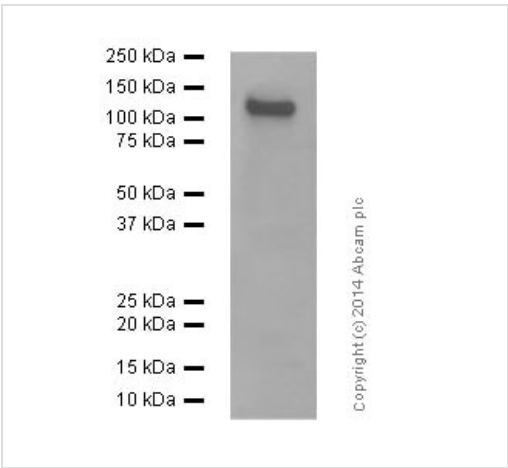
Secondary

HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 105 kDa
Observed band size: 120 kDa

Blocking buffer: 5% NFDM/TBST

Dilution buffer: 5% NFDM/TBST



Western blot - Anti-NMDAR1 antibody [EPR2481(2)] (ab109182)

Anti-NMDAR1 antibody [EPR2481(2)] (ab109182) at 1/5000 dilution (purified) + Human fetal brain tissue lysate at 20 µg

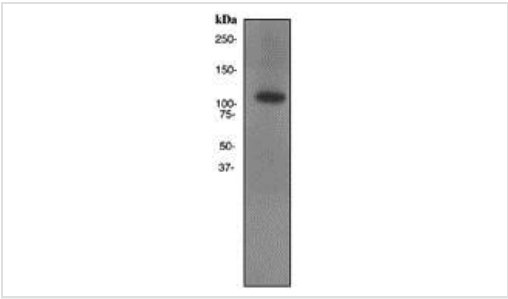
Secondary

HRP goat anti-rabbit (H+L) at 1/1000 dilution

Predicted band size: 105 kDa
Observed band size: 120 kDa

Blocking buffer: 5% NFDM/TBST

Dilution buffer: 5% NFDM/TBST

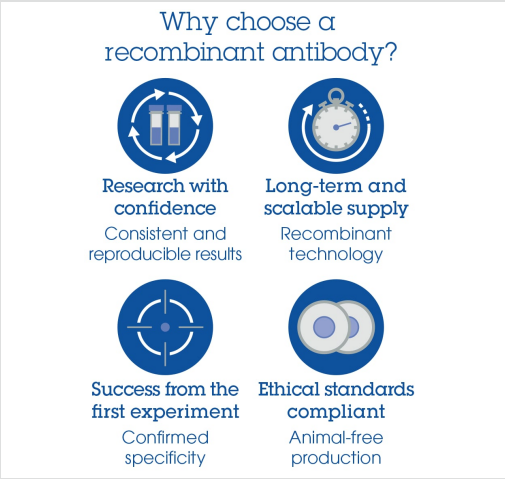


Western blot - Anti-NMDAR1 antibody [EPR2481(2)] (ab109182)

Anti-NMDAR1 antibody [EPR2481(2)] (ab109182) at 1/1000 dilution (unpurified) + Human fetal brain cell lysate at 10 µg

Predicted band size: 105 kDa
Observed band size: 120 kDa

Secondary antibody - anti-rabbit HRP (ab6721)



Anti-NMDAR1 antibody [EPR2481(2)] (ab109182)

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Product datasheet

Anti-NMDAR2A antibody ab14596

2 Abreviews 33 References

概述

产品名称	Anti-NMDAR2A抗体
描述	兔多克隆抗体to NMDAR2A
宿主	Rabbit
种属反应性	**与反应:** Mouse, Monkey
免疫原	Fusion protein (Mouse) (C terminal last 200 amino acids).

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.40 Preservative: 0.05% Sodium azide Constituents: 0.184% Tris glycine, 0.87% Sodium chloride
纯度	Protein A purified
克隆	多克隆
同种型	IgG

靶标

功能	NMDA receptor subtype of glutamate-gated ion channels possesses high calcium permeability and voltage-dependent sensitivity to magnesium. Activation requires binding of agonist to both types of subunits.
序列相似性	Belongs to the glutamate-gated ion channel (TC 1.A.10.1) family. NR2A/GRIN2A subfamily.
细胞定位	Cell membrane. Cell junction > synapse > postsynaptic cell membrane.

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Product datasheet

Anti-NMDAR2B antibody ab65783

5 Abreviews 77 References 5 图像

概述

产品名称	Anti-NMDAR2B抗体
描述	兔多克隆抗体to NMDAR2B
宿主	Rabbit
经测试应用	**适用于:** WB, ICC/IF, IP, IHC-FoFr
种属反应性	**与反应:** Mouse, Rat, Chicken, Human, Xenopus laevis **预测可用于:** Dog, Zebrafish
免疫原	Synthetic peptide conjugated to KLH derived from within residues 1450 to the C-terminus of Rat NMDAR2B.参阅Abcam的专有抗源政策(Peptide available as ab71176.)
常规说明	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.40 Preservative: 0.02% Sodium azide Constituent: PBS Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising agent. If you would like information about the formulation of a specific lot, please contact our scientific support team who will be happy to help.
纯度	Immunogen affinity purified
克隆	多克隆
同种型	IgG

应用

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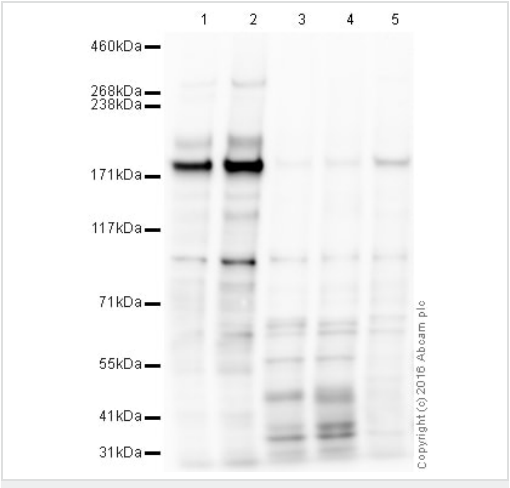
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★☆ (3)	Use a concentration of 1 µg/ml. Detects a band of approximately 180 kDa (predicted molecular weight: 166 kDa).
ICC/IF		Use a concentration of 5 µg/ml.
IP		Use a concentration of 5 µg/ml.
IHC-FoFr	★★★★☆ (1)	Use at an assay dependent concentration.

靶标

功能	NMDA receptor subtype of glutamate-gated ion channels with high calcium permeability and voltage-dependent sensitivity to magnesium. Mediated by glycine.
组织特异性	Primarily found in the fronto-parieto-temporal cortex and hippocampus pyramidal cells, lower expression in the basal ganglia.
序列相似性	Belongs to the glutamate-gated ion channel (TC 1.A.10.1) family. NR2B/GRIN2B subfamily.
细胞定位	Cell membrane. Cell junction > synapse > postsynaptic cell membrane.

图片



Western blot - Anti-NMDAR2B antibody (ab65783)

All lanes : Anti-NMDAR2B antibody (ab65783) at 1 µg/ml

Lane 1 : Rat Hippocampus Tissue Lysate at 10 µg

Lane 2 : Mouse Hippocampus Tissue Lysate (ab48631) at 10 µg

Lane 3 : Human brain tissue lysate - total protein (ab29466) at 20 µg

Lane 4 : Human brain hippocampus tissue lysate - total protein (ab30180) at 20 µg

Lane 5 : Human brain amygdala tissue lysate - total protein at 10 µg

Secondary

All lanes : Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/50000 dilution

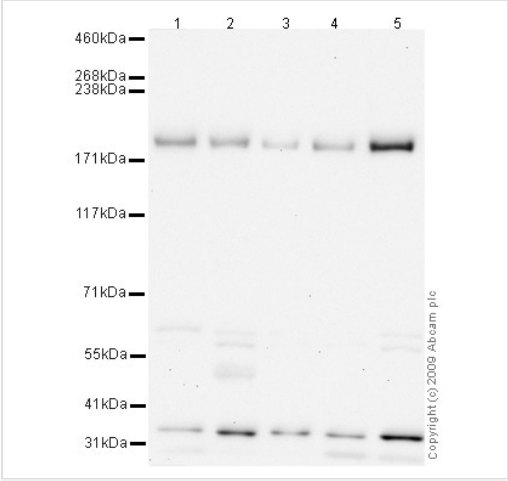
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 166 kDa
Observed band size: 180 kDa
Additional bands at: 100 kDa, 200 kDa, 35 kDa, 45 kDa, 56 kDa, 65 kDa. We are unsure as to the identity of these extra bands.

Exposure time: 8 minutes

This blot was produced using a 3-8% Tris Acetate gel under the TA buffer system. The gel was run at 150V for 60 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with ab65783 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution ab133406.



Western blot - Anti-NMDAR2B antibody (ab65783)

All lanes : Anti-NMDAR2B antibody (ab65783) at 1 µg/ml

Lane 1 : Human brain tissue lysate - total protein (ab29466)
Lane 2 : Brain (Mouse) Tissue Lysate
Lane 3 : Brain (Rat) Tissue Lysate
Lane 4 : Hippocampus (Mouse) Tissue Lysate
Lane 5 : Rat Hippocampus Tissue Lysate

Lysates/proteins at 10 µg per lane.

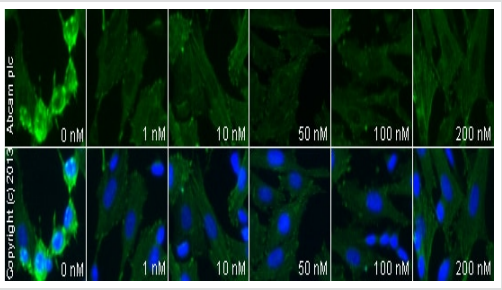
Secondary
All lanes : Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Performed under reducing conditions.

Predicted band size: 166 kDa
Observed band size: 180 kDa
Additional bands at: 35 kDa. We are unsure as to the identity of these extra bands.

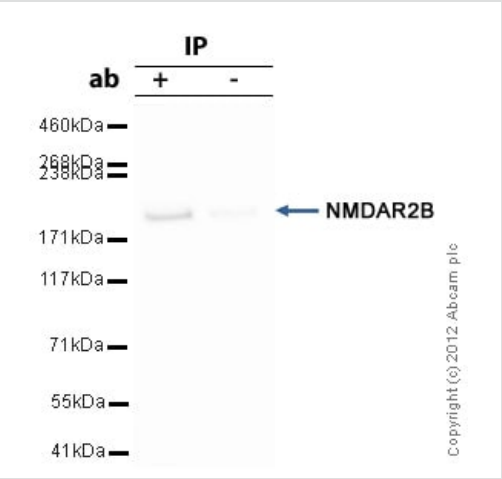
Exposure time: 1 minute

NMDAR2B contains a number of potential phosphorylation and glycosylation sites (SwissProt) which may explain its migration at a higher molecular weight than predicted.



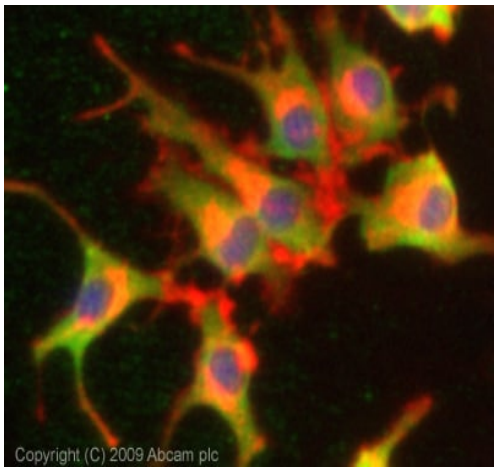
Immunocytochemistry/ Immunofluorescence - Anti-NMDAR2B antibody (ab65783)

ab65783 staining NR2B in SKNSH cells treated with GBR 12909 dihydrochloride (ab120607), by ICC/IF. Decrease in NR2B expression correlates with increased concentration of GBR 12909 dihydrochloride, as described in literature.
The cells were incubated at 37°C for 10 minutes in media containing different concentrations of ab120607 (GBR 12909 dihydrochloride) in DMSO, fixed with 100% methanol for 5 minutes at -20°C and blocked with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% tween for 2h at room temperature. Staining of the treated cells with ab65783 (5 µg/ml) was performed overnight at 4°C in PBS containing 1% BSA and 0.1% tween. A DyLight 488 goat anti-rabbit polyclonal antibody (ab96899) at 1/250 dilution was used as the secondary antibody. Nuclei were counterstained with DAPI and are shown in blue.



Immunoprecipitation - Anti-NMDAR2B antibody (ab65783)

NMDAR2B was immunoprecipitated using 0.5mg Mouse Brain tissue lysate, 5µg of Rabbit polyclonal to NMDAR2B and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).
The antibody was incubated under agitation with Protein G beads for 10min, Mouse Brain tissue lysate lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.
Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70^{o}C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab65783.
Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697).
Band: 180kDa; NMDAR2B



Immunocytochemistry/ Immunofluorescence - Anti-NMDAR2B antibody (ab65783)

ICC/IF image of ab65783 stained PC12 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 (ab65783, 5µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) used at a 1/1000 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. This antibody also gave a positive result in 4% PFA fixed (10 min) PC12 cells at 5µg/ml.

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Revision 3

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Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source:	UniProt ID:	Entrez-Gene Id:
WB, IP	H M R	Endogenous	95	Rabbit	P78352	1742

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:50

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA and 50% glycerol. Store at –20°C. Do not aliquot the antibody.

Specificity / Sensitivity

PSD95 Antibody detects endogenous levels of total PSD95 protein.
Species Reactivity:
Human, Mouse, Rat

Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic peptide corresponding to residues of human PSD95. Antibodies are purified by protein A and peptide affinity chromatography.

Background

Postsynaptic Density protein 95 (PSD95) is a member of the membrane-associated guanylate kinase (MAGUK) family of proteins. These family members consist of an amino-terminal variable segment followed by three PDZ domains, an SH3 domain, and an inactive guanylate kinase (GK) domain. PSD95 is a scaffolding protein involved in the assembly and function of the postsynaptic density complex (1-2). PSD95 participates in synaptic targeting of AMPA receptors through an indirect manner involving Stargazin and related transmembrane AMPA receptor regulatory proteins (TARPs) (3). It is implicated in experience-dependent plasticity and plays an indispensable role in learning (4). Mutations in PSD95 are associated with autism (5).

1. Cao, J. et al. (2005) *J. Cell Biol* 168, 117-26.
2. Chetkovich, D.M. et al. (2002) *J. Neurosci.* 22, 6415-25.
3. Cai, C. et al. (2006) *J. Biol. Chem.* 281, 4267-73.
4. Yao, W.D. et al. (2004) *Neuron* 41, 625-38.
5. Cline, H. (2005) *Curr. Biol.* 15, R203-5.

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

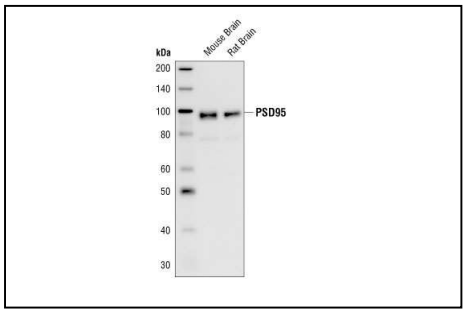
APPLICATIONS KEY **WB:** Western Blot **IP:** Immunoprecipitation **IHC:** Immunohistochemistry **ChIP:** Chromatin Immunoprecipitation **IF:** Immunofluorescence **F:** Flow Cytometry **E-P:** ELISA-Peptide

CROSS-REACTIVITY KEY **H:** human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **Mi:** mink **C:** chicken **Dm:** D. melanogaster **X:** Xenopus **Z:** zebrafish **B:** bovine **Dg:** dog **Pg:** pig **Sc:** S. cerevisiae **Ce:** C. elegans **Hr:** horse **All:** all species expected

Revision 3

#2507

PSD95 Antibody



Western blot analysis of extracts from mouse and rat brain, using PSD95 Antibody.

#2507

PSD95 Antibody

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Product datasheet

Anti-Synaptotagmin antibody [ASV30] ab13259

★★★★★ 1 Abreviews 21 References 3 图像

概述

产品名称	Anti-Synaptotagmin抗体[ASV30]
描述	小鼠单克隆抗体[ASV30] to Synaptotagmin
宿主	Mouse
经测试应用	**适用于:** WB, ICC/IF, IP
种属反应性	**与反应:** Mouse, Rat
免疫原	Full length protein corresponding to Rat Synaptotagmin. Rat brain synaptic junction protein complexes Database link: P21707
阳性对照	Mouse or Rat brain tissue extract.
常规说明	This product was changed from ascites to tissue culture supernatant on 5th July 2019. Lot numbers higher than GR3258922 are from tissue culture supernatant. Please note that the dilutions may need to be adjusted accordingly. If you have any questions, please do not hesitate to contact our scientific support team. The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
存储溶液	Preservative: 0.09% Sodium azide Constituents: PBS, 50% Glycerol (glycerin, glycerine)
纯度	Protein G purified
纯化说明	Purified from TCS.
克隆	单克隆

克隆编号	ASV30
同种型	IgG2a

应用

The Abpromise guarantee Abpromise™承诺保证使用ab13259于以下的经测试应用

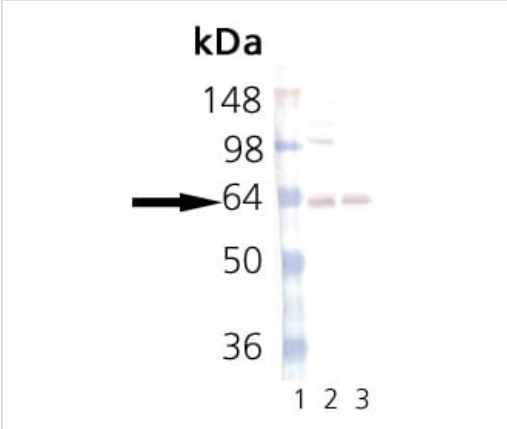
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★★ (1)	1/1000.
ICC/IF		Use a concentration of 5 µg/ml.
IP		Use a concentration of 5 µg/ml.

靶标

功能	May have a regulatory role in the membrane interactions during trafficking of synaptic vesicles at the active zone of the synapse. It binds acidic phospholipids with a specificity that requires the presence of both an acidic head group and a diacyl backbone.
序列相似性	Belongs to the synaptotagmin family. Contains 2 C2 domains.
结构域	The first C2 domain mediates Ca(2+)-dependent phospholipid binding. The second C2 domain mediates interaction with Stonin 2.
细胞定位	Cytoplasmic vesicle, secretory vesicle, synaptic vesicle membrane. Cytoplasmic vesicle, secretory vesicle, chromaffin granule membrane. Synaptic vesicles and chromaffin granules.

图片



Western blot - Anti-Synaptotagmin antibody [ASV30] (ab13259)

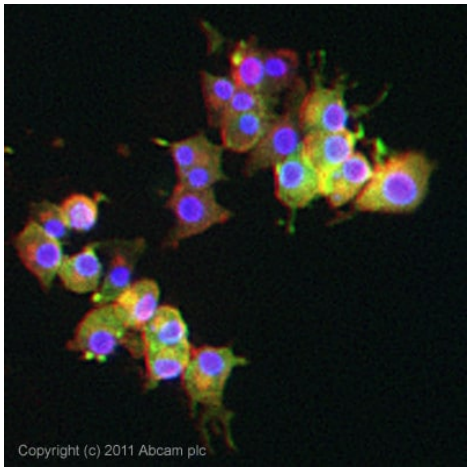
All lanes : Anti-Synaptotagmin antibody [ASV30] (ab13259) at 1/1000 dilution

Lane 1 : Molecular weight ladder

Lane 2 : Lysates prepared from mouse brain

Lane 3 : Lysates prepared from rat brain

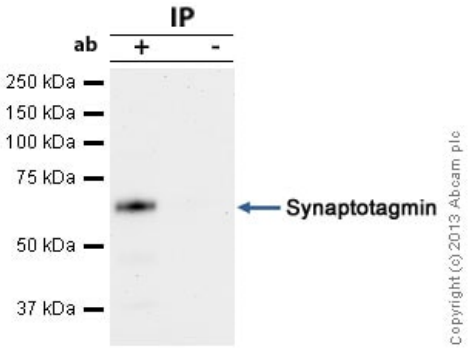
This image was generated using the ascites version of the product.



Immunocytochemistry/ Immunofluorescence - Anti-Synaptotagmin antibody [ASV30] (ab13259)

ICC/IF image of ab13259 stained PC12 cells. The cells were 4% formaldehyde fixed (10 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 (ab13259, 5µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-mouse IgG (H+L) used at a 1/1000 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.

This image was generated using the ascites version of the product.



Immunoprecipitation - Anti-Synaptotagmin antibody [ASV30] (ab13259)

Synaptotagmin was immunoprecipitated using 0.5mg Mouse Brain tissue lysate, 5µg of Mouse monoclonal to Synaptotagmin and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).
The antibody was incubated under agitation with Protein G beads for 10min, Mouse Brain tissue lysate lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.
Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70^{o}C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab13259.
Secondary: Goat polyclonal to mouse IgG light chain specific (HRP) at 1/20,000 dilution.
Band: 64kDa; Synaptotagmin

This image was generated using the ascites version of the product.

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Product datasheet

Anti-Synapsin I antibody - Synaptic Marker ab64581

20 Abreviews 72 References 3 图像

概述

产品名称	Anti-Synapsin I抗体- Synaptic Marker
描述	兔多克隆抗体to Synapsin I - Synaptic Marker
宿主	Rabbit
经测试应用	**适用于:** WB, IP
种属反应性	**与反应:** Mouse, Rat, Goat **预测可用于:** Sheep, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
常规说明	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.40 Preservative: 0.02% Sodium azide Constituent: PBS Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising agent. If you would like information about the formulation of a specific lot, please contact our scientific support team who will be happy to help.
纯度	Immunogen affinity purified
克隆	**多克隆**
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab64581于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★★ (3)	Use a concentration of 1 µg/ml. Detects a band of approximately 74, 70 kDa (predicted molecular weight: 74 kDa). Abcam recommends using 3-5% milk as the blocking agent.
IP		Use at an assay dependent concentration.

靶标

功能 Neuronal phosphoprotein that coats synaptic vesicles, binds to the cytoskeleton, and is believed to function in the regulation of neurotransmitter release. The complex formed with NOS1 and CAPON proteins is necessary for specific nitric-oxid functions at a presynaptic level.

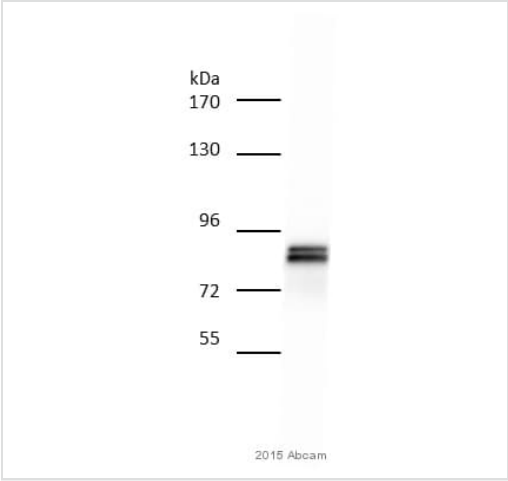
疾病相关 Defects in SYN1 are a cause of epilepsy X-linked with variable learning disabilities and behavior disorders [MIM:300491]. XELBD is characterized by variable combinations of epilepsy, learning difficulties, macrocephaly, and aggressive behavior.

序列相似性 Belongs to the synapsin family.

翻译后修饰 Substrate of at least four different protein kinases. It is probable that phosphorylation plays a role in the regulation of synapsin-1 in the nerve terminal. Phosphorylated upon DNA damage, probably by ATM or ATR.

细胞定位 Cell junction > synapse. Golgi apparatus.

图片



Western blot - Anti-Synapsin I antibody - Synaptic Marker (ab64581)

This image is courtesy of an anonymous Abreview

Anti-Synapsin I antibody - Synaptic Marker (ab64581) at 1/5000 dilution + Rat whole brain tissue lysate at 15 µg

Secondary

Donkey anti-Rabbit IgG at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

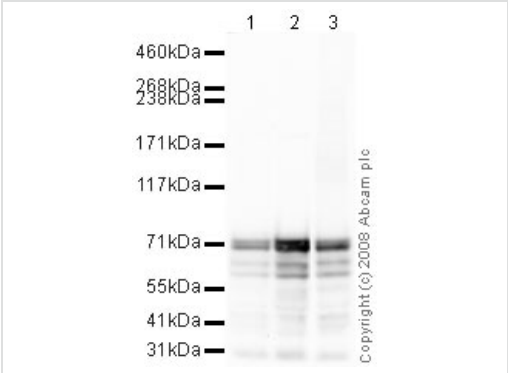
Predicted band size: 74 kDa

Observed band size: 80,85 kDa

Exposure time: 15 seconds

Blocked with 5% milk for 1 hour at 25°C.

Sample was incubated with primary antibody for 18 hours at 4°C in TBS + 0.1% Tween.



Western blot - Anti-Synapsin I antibody - Synaptic Marker (ab64581)

All lanes : Anti-Synapsin I antibody - Synaptic Marker (ab64581) at 1 µg/ml

Lane 1 : Spinal Cord (Mouse) Tissue Lysate
Lane 2 : Brain (Mouse) Tissue Lysate
Lane 3 : Brain (Rat) Tissue Lysate - normal tissue

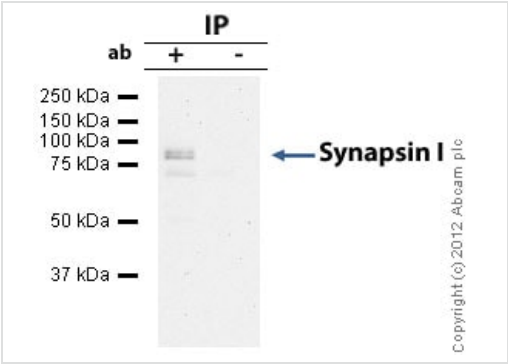
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Performed under reducing conditions.

Predicted band size: 74 kDa
Observed band size: 70,74 kDa

The immunogen for ab64581 is predicted to recognise both the 70 kDa and 74 kDa isoforms of Synapsin I. Synapsin I is also known to contain a number of potential phosphorylation and glycosylation sites (SwissProt data).



Immunoprecipitation - Anti-Synapsin I antibody - Synaptic Marker (ab64581)

Synapsin I - Synaptic Marker was immunoprecipitated using 0.5mg Mouse Brain whole tissue lysate, 5µg of Rabbit polyclonal to Synapsin I - Synaptic Marker and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).
The antibody was incubated under agitation with Protein G beads for 10min, Mouse Brain whole tissue lysate lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.
Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70^{o}C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab64581.
Secondary: Goat polyclonal to mouse IgG light chain specific (HRP)

at 1/5000 dilution.
Band: 80kDa: Synapsin I - Synaptic Marker contains a number of potential glycosylation sites (SwissProt) which may explain the higher migrating band.

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Product datasheet

Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] ab183797

重组 RabMAb

11 References 11 图像

概述

产品名称	Anti-Glutamate Receptor 1 (AMPA subtype)抗体[EPR19522]
描述	兔单克隆抗体[EPR19522] to Glutamate Receptor 1 (AMPA subtype)
宿主	Rabbit
经测试应用	**适用于:** WB, IHC-P, IP, IHC-Fr
种属反应性	**与反应:** Mouse, Rat, Human, Recombinant fragment
免疫原	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: Mouse Glutamate Receptor 1 fragment recombinant protein; Mouse brain, hippocampus and cerebellum lysates; Rat brain, cerebellum and hippocampus lysates; Human cerebellum and fetal brain lysates. IHC-P: Mouse and rat hippocampus tissues. IHC-Fr: Mouse hippocampus tissue. IP: Mouse brain whole cell lysate.
常规说明	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.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
存储溶液	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
纯度	Protein A purified

克隆	单克隆
克隆编号	EPR19522
同种型	IgG

应用

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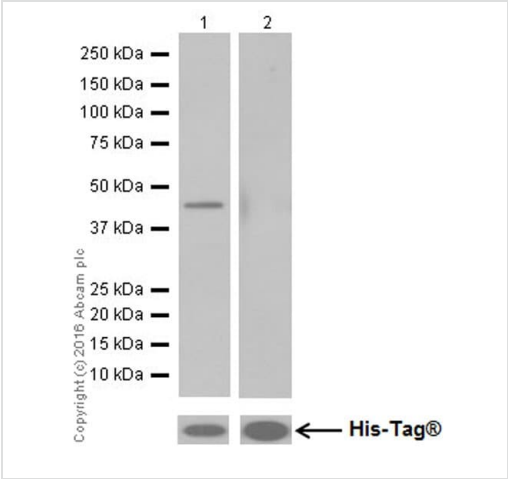
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		1/1000. Detects a band of approximately 102 kDa (predicted molecular weight: 102 kDa).
IHC-P		1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. IHC is recommended for mouse and rat only.
IP		1/30.
IHC-Fr		1/100. Antigen retrieval: Heated citrate solution (10mM citrate PH 6.0 + 0.05% Tween-20) IHC is recommended for mouse and rat only

靶标

功能	Ionotropic glutamate receptor. L-glutamate acts as an excitatory neurotransmitter at many synapses in the central nervous system. Binding of the excitatory neurotransmitter L-glutamate induces a conformation change, leading to the opening of the cation channel, and thereby converts the chemical signal to an electrical impulse. The receptor then desensitizes rapidly and enters a transient inactive state, characterized by the presence of bound agonist.
组织特异性	Widely expressed in brain.
序列相似性	Belongs to the glutamate-gated ion channel (TC 1.A.10.1) family. GRIA1 subfamily.
翻译后修饰	Palmitoylated. Depalmitoylated upon glutamate stimulation. Cys-603 palmitoylation leads to Golgi retention and decreased cell surface expression. In contrast, Cys-829 palmitoylation does not affect cell surface expression but regulates stimulation-dependent endocytosis.
细胞定位	Cell membrane. Endoplasmic reticulum membrane. Cell junction > synapse > postsynaptic cell membrane. Interaction with CACNG2 promotes cell surface expression.

图片



Western blot - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

All lanes : Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797) at 1/5000 dilution

Lane 1 : Mouse Glutamate Receptor 1 fragment recombinant protein
Lane 2 : Mouse Glutamate Receptor 2 fragment recombinant protein

Lysates/proteins at 0.01 µg per lane.

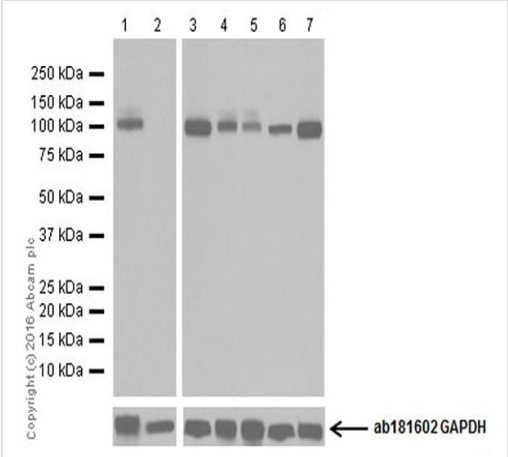
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 102 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure times: Lane 1: 2 seconds; Lane 2: 3 minutes.

Mouse Glutamate Receptor 1 fragment recombinant protein contains aa19-184 with a GST/His-Tag®. Mouse Glutamate Receptor 2 fragment recombinant protein contains aa25-288 with a His-Tag®.



Western blot - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

All lanes : Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797) at 1/1000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Rat liver lysate
Lane 3 : Mouse hippocampus lysate
Lane 4 : Mouse cerebellum lysate
Lane 5 : Rat brain lysate
Lane 6 : Rat cerebellum lysate
Lane 7 : Rat hippocampus lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at

1/100000 dilution

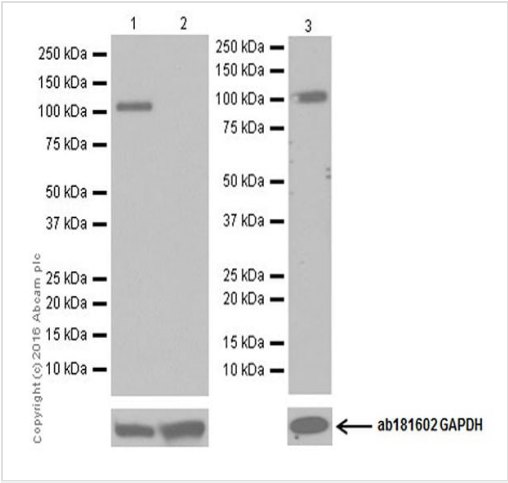
Predicted band size: 102 kDa
Observed band size: 102 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure times: Lane 1 and 2: 30 seconds; Lane 3, 4, 5, 6 and 7: 15 seconds.

Rodent Glutamate Receptor 1 is widely expressed in brain and represents the predominant excitatory neurotransmitter system but not in liver.

Negative control: Rat liver (PMID: 2480522).



Western blot - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

All lanes : Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797) at 1/1000 dilution

Lane 1 : Human cerebellum lysate
Lane 2 : Human muscle lysate
Lane 3 : Human fetal brain lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

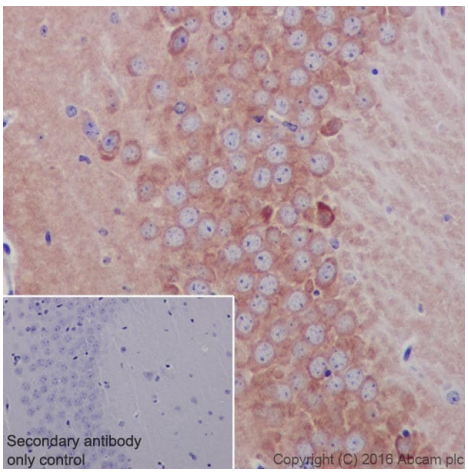
Predicted band size: 102 kDa
Observed band size: 102 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

Human Glutamate Receptor 1 is widely expressed in brain and represents the predominant excitatory neurotransmitter system but not in muscle.

Negative control: human muscle (PMID: 1652753).

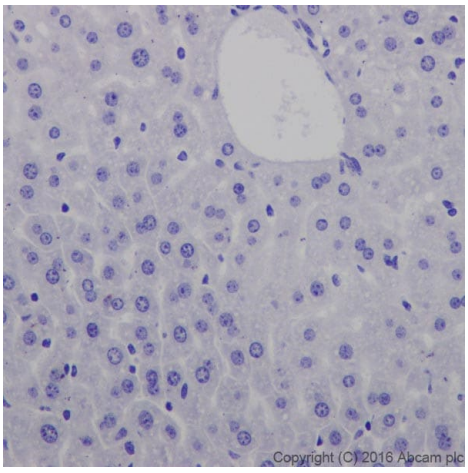


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

Immunohistochemical analysis of paraffin-embedded Mouse hippocampus tissue labeling Glutamate Receptor 1 (AMPA subtype) with ab183797 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasm staining on mouse hippocampus was observed [PMID: 15723058]. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

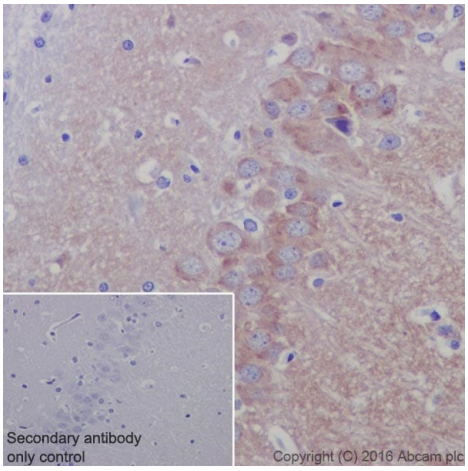
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

Immunohistochemical analysis of paraffin-embedded Mouse liver tissue labeling Glutamate Receptor 1 (AMPA subtype) with ab183797 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. No staining on mouse liver is observed. Counter stained with Hematoxylin.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

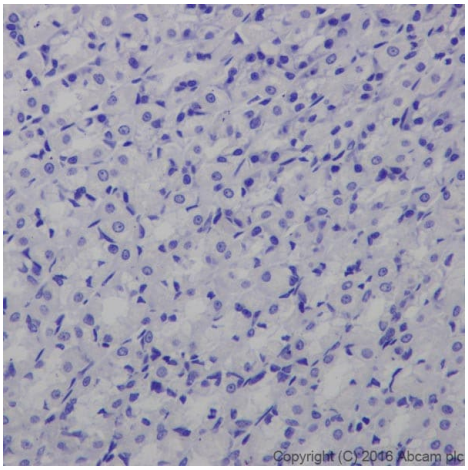


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

Immunohistochemical analysis of paraffin-embedded Rat hippocampus tissue labeling Glutamate Receptor 1 (AMPA subtype) with ab183797 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Cytoplasm staining on rat hippocampus is observed [PMID: 15723058]. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

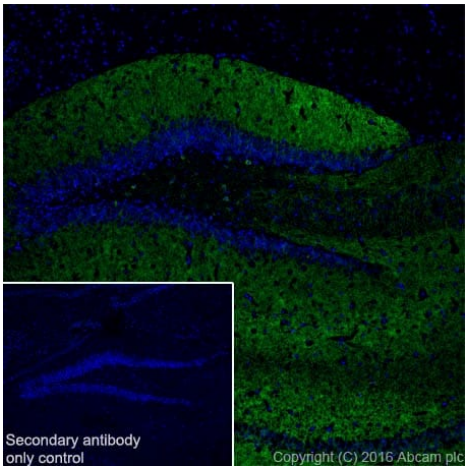
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

Immunohistochemical analysis of paraffin-embedded Rat stomach tissue labeling Glutamate Receptor 1 (AMPA subtype) with ab183797 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. No staining on rat stomach is observed. Counter stained with Hematoxylin.

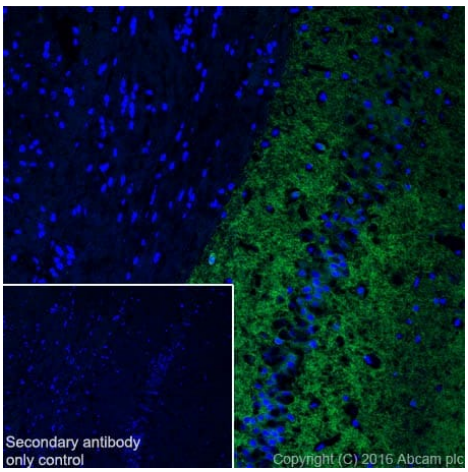
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Frozen sections) - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

Immunohistochemical analysis of 4% paraformaldehyde-fixed, 0.2% Triton X-100 permeabilized frozen Mouse hippocampus tissue labeling Glutamate Receptor 1 (AMPA subtype) with ab183797 at 1/100 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Cytoplasm staining on mouse hippocampus was observed. The nuclear counterstain is DAPI (blue).

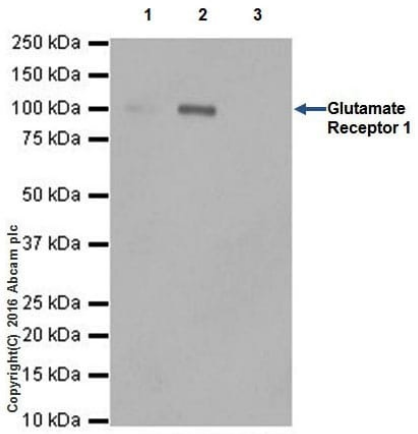
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab150077 secondary antibody at 1/1000 dilution.



Immunohistochemistry (Frozen sections) - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

Immunohistochemical analysis of 4% paraformaldehyde-fixed, 0.2% Triton X-100 permeabilized frozen Mouse hippocampus tissue labeling Glutamate Receptor 1 (AMPA subtype) with ab183797 at 1/100 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Cytoplasm staining on mouse hippocampus was observed. The nuclear counterstain is DAPI (blue).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab150077 at 1/1000 dilution.



Immunoprecipitation - Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

Glutamate Receptor 1 (AMPA subtype) was immunoprecipitated from 0.35 mg of Mouse brain whole cell lysate with ab183797 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab183797 at 1/1000 dilution. VeriBlot for IP Detection Reaction (HRP) (ab131366), was used for detection at 1/10000 dilution.

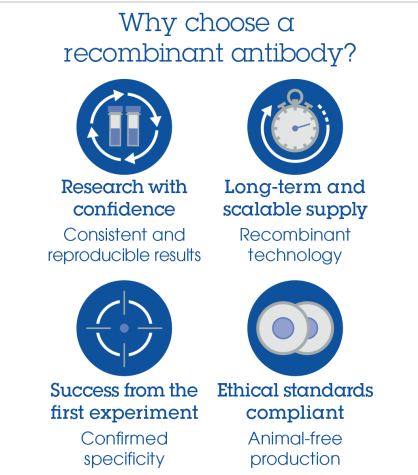
Lane 1: Mouse brain whole cell lysate, 10ug (Input).

Lane 2: ab183797 IP in Mouse brain whole cell lysate.

Lane 3: Rabbit IgG,monoclonal [EPR25A]-Isotype Control (ab172730) instead of ab183797 in Mouse brain whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 3 minutes.



Anti-Glutamate Receptor 1 (AMPA subtype) antibody [EPR19522] (ab183797)

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Product datasheet

Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] ab206293

重组 RabMAb

3 Abreviews 15 References 6 图像

概述

产品名称	Anti-Ionotropic Glutamate receptor 2抗体[EPR18115]
描述	兔单克隆抗体[EPR18115] to Ionotropic Glutamate receptor 2
宿主	Rabbit
经测试应用	**适用于:** ICC/IF, IP, IHC-P, WB
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: Mouse, rat and Human brain lysates; Human fetal brain lysate. IHC-P: Human solitary fibrous tumor tissue. ICC/IF: HT1080 cells. IP: C6 whole cell lysates.
常规说明	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.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
存储溶液	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR18115

同种型	IgG

应用

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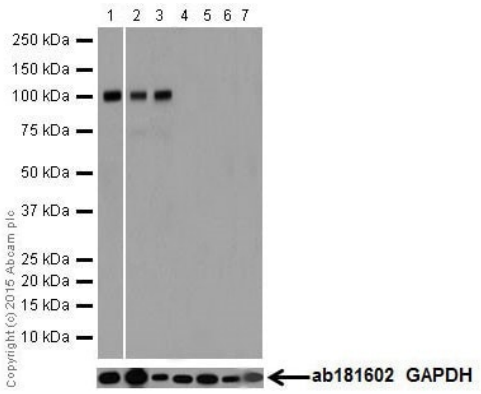
应用	Ab评论	说明
ICC/IF		1/700.
IP		1/40.
IHC-P		1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. IHC is recommended for human only.
WB	★★★★★ (2)	1/2000. Detects a band of approximately 99 kDa (predicted molecular weight: 99 kDa).

靶标

功能	Ionotropic glutamate receptor. L-glutamate acts as an excitatory neurotransmitter at many synapses in the central nervous system. Binding of the excitatory neurotransmitter L-glutamate induces a conformation change, leading to the opening of the cation channel, and thereby converts the chemical signal to an electrical impulse. The receptor then desensitizes rapidly and enters a transient inactive state, characterized by the presence of bound agonist. In the presence of CACNG4 or CACNG7 or CACNG8, shows resensitization which is characterized by a delayed accumulation of current flux upon continued application of glutamate.
序列相似性	Belongs to the glutamate-gated ion channel (TC 1.A.10.1) family. GRIA2 subfamily.
翻译后修饰	Palmitoylated. Depalmitoylated upon glutamate stimulation. Cys-610 palmitoylation leads to Golgi retention and decreased cell surface expression. In contrast, Cys-836 palmitoylation does not affect cell surface expression but regulates stimulation-dependent endocytosis.
细胞定位	Cell membrane. Endoplasmic reticulum membrane. Cell junction > synapse > postsynaptic cell membrane. Interaction with CACNG2, CNIH2 and CNIH3 promotes cell surface expression.

图片



Western blot - Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] (ab206293)

All lanes : Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] (ab206293) at 1/5000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Rat brain lysate
Lane 3 : Human fetal brain lysate
Lane 4 : Rat spleen lysate
Lane 5 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 6 : Human fetal kidney lysate
Lane 7 : HT1080 (Human fibrosarcoma cell line) whole cell lysate

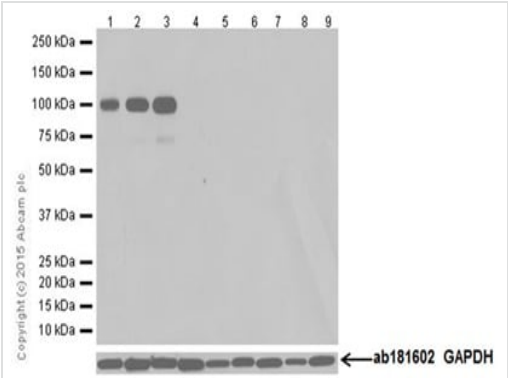
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 99 kDa
Observed band size: 99 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1: 1 second; Lane 2,3,4,5,6 and 7: 10 seconds.



Western blot - Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] (ab206293)

All lanes : Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] (ab206293) at 1/2000 dilution

Lane 1 : Human brain lysate
Lane 2 : Mouse brain lysate
Lane 3 : Rat brain lysate
Lane 4 : C6 (Rat glial tumor cell line) whole cell lysate
Lane 5 : Human fetal kidney lysate
Lane 6 : HT1080 (Human fibrosarcoma cell line) whole cell lysate
Lane 7 : Mouse heart lysate
Lane 8 : Rat spleen lysate
Lane 9 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

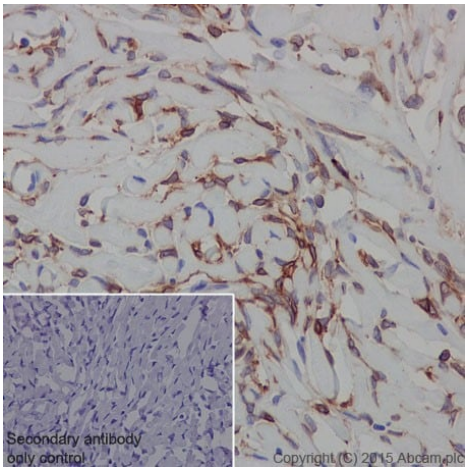
Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 99 kDa
Observed band size: 99 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] (ab206293)

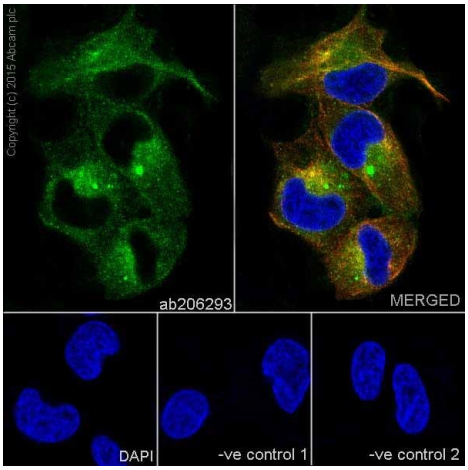
Immunohistochemical analysis of paraffin-embedded Human solitary fibrous tumor tissue labeling Ionotropic Glutamate receptor 2 with ab206293 at 1/500 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Membrane staining on Human solitary fibrous tumor is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] (ab206293)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HT1080 (Human fibrosarcoma cell line) cells labeling Ionotropic Glutamate receptor 2 with ab206293 at 1/700 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing cytoplasmic staining on HT1080 cell line.

The nuclear counterstain is DAPI (blue).

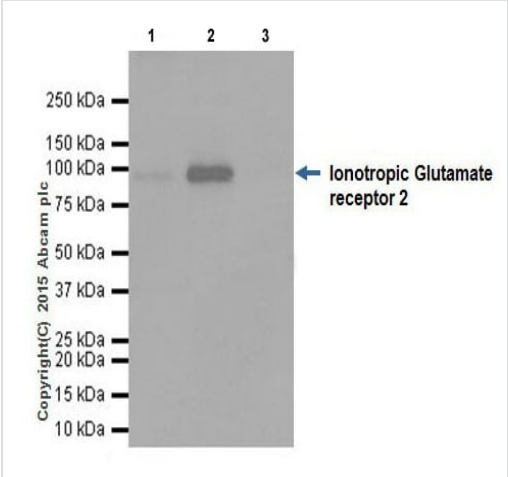
Tubulin is detected with Anti-alpha Tubulin antibody [EPR18115] - Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab206293 at 1/700 dilution followed by ab150120 at 1/1000 dilution.

-ve control 2: ab7291 at 1/1000 dilution followed by ab150077 at

1/1000 dilution.



Immunoprecipitation - Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] (ab206293)

Ionotropic Glutamate receptor 2 was immunoprecipitated from 1mg of C6 (Rat glial tumor cell line) whole cell lysate with ab206293 at 1/40 dilution.

Western blot was performed from the immunoprecipitate using ab206293 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

Lane 1: C6 whole cell lysate 10µg (Input).

Lane 2: ab206293 IP in C6 whole cell lysate.

Lane 3:Rabbit IgG, monoclonal [EPR18115] - Isotype Control (ab172730) instead of ab206293 in C6 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 10 seconds.

Anti-Ionotropic Glutamate receptor 2 antibody [EPR18115] (ab206293)

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Product datasheet

Anti-Tau (phospho S202) antibody [EPR2402] ab108387

重组 RabMAb

2 Abreviews 9 References 4 图像

概述

产品名称	Anti-Tau (phospho S202)抗体[EPR2402]
描述	兔单克隆抗体[EPR2402] to Tau (phospho S202)
宿主	Rabbit
特异性	*Stimulation may be required to allowdetection of the phosphorylated protein. Please see images belowfor recommended treatment conditions and positive controls.*
经测试应用	**适用于:** WB **不适用于:** Flow Cyt,ICC or IHC-P
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: Human brain lysate, mouse hippocampus, rat hippocampus and cerebral cortex lysates.
常规说明	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.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
存储溶液	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 0.05% BSA, 40% Glycerol (glycerin, glycerine), 59% PBS
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR2402

同种型 IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab108387于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		1/5000 - 1/10000. Predicted molecular weight: 79 kDa.

应用说明 Is unsuitable for Flow Cyt,ICC or IHC-P.

靶标

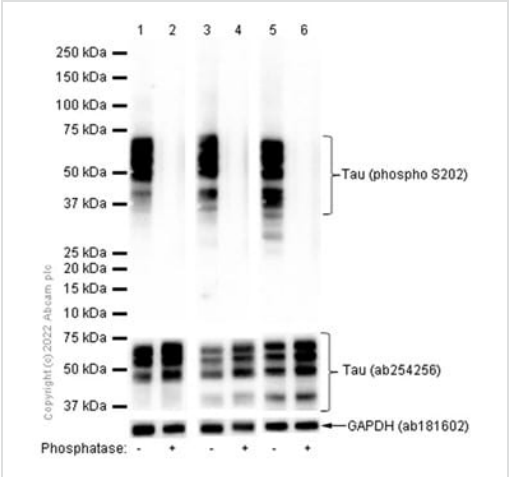
功能 Promotes microtubule assembly and stability, and might be involved in the establishment and maintenance of neuronal polarity. The C-terminus binds axonal microtubules while the N-terminus binds neural plasma membrane components, suggesting that tau functions as a linker protein between both. Axonal polarity is predetermined by tau localization (in the neuronal cell) in the domain of the cell body defined by the centrosome. The short isoforms allow plasticity of the cytoskeleton whereas the longer isoforms may preferentially play a role in its stabilization.

组织特异性 Expressed in neurons. Isoform PNS-tau is expressed in the peripheral nervous system while the others are expressed in the central nervous system.

疾病相关 Note=In Alzheimer disease, the neuronal cytoskeleton in the brain is progressively disrupted and replaced by tangles of paired helical filaments (PHF) and straight filaments, mainly composed of hyperphosphorylated forms of TAU (PHF-TAU or AD P-TAU).
Defects in MAPT are a cause of frontotemporal dementia (FTD) [MIM:600274]; also called frontotemporal dementia (FTD), pallido-ponto-nigral degeneration (PPND) or historically termed Pick complex. This form of frontotemporal dementia is characterized by presenile dementia with behavioral changes, deterioration of cognitive capacities and loss of memory. In some cases, parkinsonian symptoms are prominent. Neuropathological changes include frontotemporal atrophy often associated with atrophy of the basal ganglia, substantia nigra, amygdala. In most cases, protein tau deposits are found in glial cells and/or neurons.
Defects in MAPT are a cause of Pick disease of the brain (PIDB) [MIM:172700]. It is a rare form of dementia pathologically defined by severe atrophy, neuronal loss and gliosis. It is characterized by the occurrence of tau-positive inclusions, swollen neurons (Pick cells) and argentophilic neuronal inclusions known as Pick bodies that disproportionally affect the frontal and temporal cortical regions. Clinical features include aphasia, apraxia, confusion, anomia, memory loss and personality deterioration.
Note=Defects in MAPT are a cause of corticobasal degeneration (CBD). It is marked by extrapyramidal signs and apraxia and can be associated with memory loss. Neuropathologic features may overlap Alzheimer disease, progressive supranuclear palsy, and Parkinson disease.
Defects in MAPT are a cause of progressive supranuclear palsy type 1 (PSNP1) [MIM:601104, 260540]; also abbreviated as PSP and also known as Steele-Richardson-Olszewski syndrome. PSNP1 is characterized by akinetic-rigid syndrome, supranuclear gaze palsy, pyramidal tract dysfunction, pseudobulbar signs and cognitive capacities deterioration. Neurofibrillary tangles and gliosis but no amyloid plaques are found in diseased brains. Most cases appear to be sporadic, with a significant association with a common haplotype including the MAPT gene and

	the flanking regions. Familial cases show an autosomal dominant pattern of transmission with incomplete penetrance; genetic analysis of a few cases showed the occurrence of tau mutations, including a deletion of Asn-613.
序列相似性	Contains 4 Tau/MAP repeats.
发展阶段	Four-repeat (type II) tau is expressed in an adult-specific manner and is not found in fetal brain, whereas three-repeat (type I) tau is found in both adult and fetal brain.
结构域	The tau/MAP repeat binds to tubulin. Type I isoforms contain 3 repeats while type II isoforms contain 4 repeats.
翻译后修饰	Phosphorylation at serine and threonine residues in S-P or T-P motifs by proline-directed protein kinases (PDPK: CDK1, CDK5, GSK-3, MAPK) (only 2-3 sites per protein in interphase, seven-fold increase in mitosis, and in PHF-tau), and at serine residues in K-X-G-S motifs by MAP/microtubule affinity-regulating kinase (MARK) in Alzheimer diseased brains. Phosphorylation decreases with age. Phosphorylation within tau's repeat domain or in flanking regions seems to reduce tau's interaction with, respectively, microtubules or plasma membrane components. Phosphorylation on Ser-610, Ser-622, Ser-641 and Ser-673 in several isoforms during mitosis. Polyubiquitinated. Requires functional TRAF6 and may provoke SQSTM1-dependent degradation by the proteasome (By similarity). PHF-tau can be modified by three different forms of polyubiquitination. 'Lys-48'-linked polyubiquitination is the major form, 'Lys-6'-linked and 'Lys-11'-linked polyubiquitination also occur. Glycation of PHF-tau, but not normal brain tau. Glycation is a non-enzymatic post-translational modification that involves a covalent linkage between a sugar and an amino group of a protein molecule forming ketoamine. Subsequent oxidation, fragmentation and/or cross-linking of ketoamine leads to the production of advanced glycation endproducts (AGES). Glycation may play a role in stabilizing PHF aggregation leading to tangle formation in AD.
细胞定位	Cytoplasm > cytosol. Cell membrane. Cytoplasm > cytoskeleton. Cell projection > axon. Mostly found in the axons of neurons, in the cytosol and in association with plasma membrane components.
形式	There are 9 isoforms produced by alternative splicing.

图片



Western blot - Anti-Tau (phospho S202) antibody [EPR2402] (ab108387)

All lanes : Anti-Tau (phospho S202) antibody [EPR2402] (ab108387) at 1/1000 dilution

Lane 1 : Rat hippocampus lysate

Lane 2 : Rat hippocampus lysate then the membrane treated with Alkaline Phosphatase for 1 hour

Lane 3 : Rat cerebral cortex lysate

Lane 4 : Rat cerebral cortex lysate then the membrane treated with Alkaline Phosphatase for 1 hour

Lane 5 : Mouse hippocampus lysate

Lane 6 : Mouse hippocampus lysate then the membrane treated with Alkaline Phosphatase for 1 hour

Lysates/proteins at 15 µg per lane.

Secondary

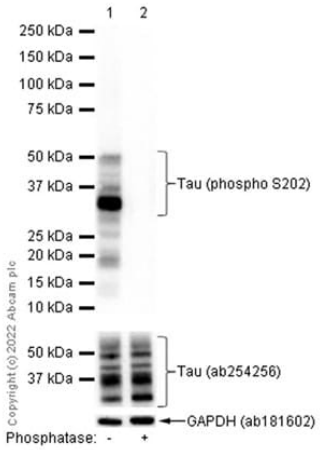
All lanes : Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 79 kDa
Observed band size: 32-72 kDa

The molecular weight observed is consistent with what has been described in the literature (PMID: 28382304, 32692785 and 30120733).

Exposure time: 3 seconds

Blocking and diluting buffer: 5% NFDM/TBST



Western blot - Anti-Tau (phospho S202) antibody [EPR2402] (ab108387)

All lanes : Anti-Tau (phospho S202) antibody [EPR2402] (ab108387) at 1/1000 dilution

Lane 1 : Human brain lysate
Lane 2 : Human brain lysate then the membrane treated with Alkaline Phosphatase for 1 hour

Lysates/proteins at 15 µg per lane.

Secondary

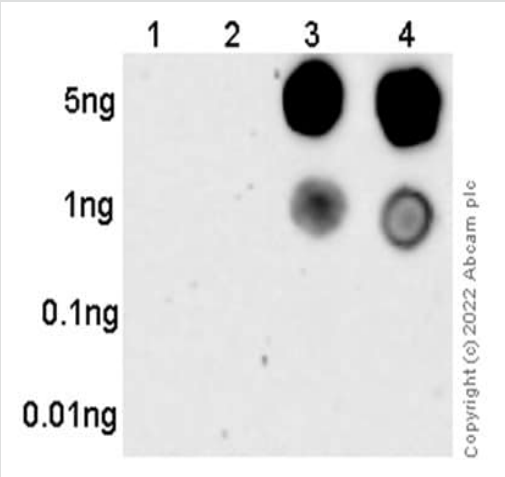
All lanes : Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 79 kDa
Observed band size: 32-72 kDa

The molecular weight observed is consistent with what has been described in the literature (PMID: 28382304, 32692785 and 30120733).

Exposure time: 5 seconds

Blocking and diluting buffer: 5% NFDM/TBST



Dot Blot - Anti-Tau (phospho S202) antibody [EPR2402] (ab108387)

Dot blot analysis using 1/1000 dilution ab108387 and Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) secondary at 1/100000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST

Lane 1: Tau non-phospho peptide

Lane 2: Tau S199 phospho peptide

Lane 3: Tau S202 phospho peptide

Lane 4: Tau S199+S202 phospho peptide

Exposure time: 3 minutes

Anti-Tau (phospho S202) antibody [EPR2402] (ab108387)

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Product Datasheet

Tau(Phospho-Thr205) Antibody

Catalog No: #11108

Package Size: #11108-1 50ul #11108-2 100ul

Orders: order@signalwayantibody.com
Support: tech@signalwayantibody.com

Description

Product Name	Tau(Phospho-Thr205) Antibody
Host Species	Rabbit
Clonality	Polyclonal
Purification	Antibodies were produced by immunizing rabbits with synthetic phosphopeptide and KLH conjugates. Antibodies were purified by affinity-chromatography using epitope-specific phosphopeptide. Non-phospho specific antibodies were removed by chromatogramphy using non-phosphopeptide.
Applications	WB IHC
Species Reactivity	Hu Ms Rt
Specificity	The antibody detects endogenous level of Tau only when phosphorylated at threonine 205.
Immunogen Type	Peptide-KLH
Immunogen Description	Peptide sequence around phosphorylation site of threonine 205 (P-G-T(p)-P-G) derived from Human Tau.
Target Name	Tau
Modification	Phospho
Other Names	MAPT; MTAPT; MTBT1; Neurofibrillary tangle protein; PHF-tau
Accession No.	Swiss-Prot: P10636NCBI Protein: NP _001116538.1
Uniprot	P10636
GeneID	4137;
Concentration	1.0mg/ml
Formulation	Supplied at 1.0mg/mL in phosphate buffered saline (without Mg2+ and Ca2+), pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol.
Storage	Store at -20°C for long term preservation (recommended). Store at 4°C for short term use.

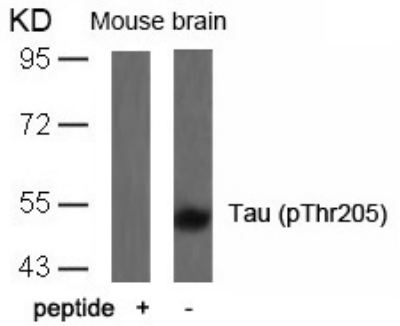
Application Details

Predicted MW: 48 62 78 kd

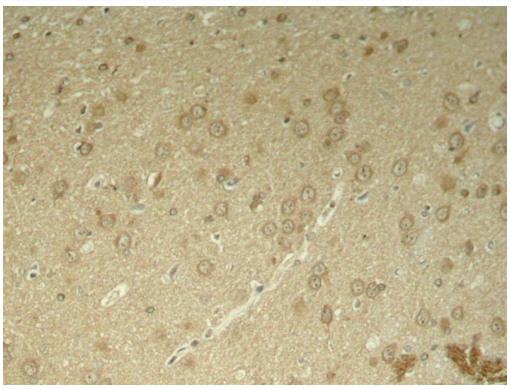
Western blotting: 1:500~1:1000

Immunohistochemistry: 1:50~1:100

Images



Western blot analysis of extracts from mouse brain tissue using Tau(Phospho-Thr205) Antibody #11108 and the same antibody preincubated with blocking peptide.



Immunohistochemical analysis of paraffin-embedded rat hippocampal region tissue from a model with Alzheimer

Background

Promotes microtubule assembly and stability, and might be involved in the establishment and maintenance of neuronal polarity. The C-terminus binds axonal microtubules while the N-terminus binds neural plasma membrane components, suggesting that tau functions as a linker protein between both. Axonal polarity is predetermined by tau localization (in the neuronal cell) in the domain of the cell body defined by the centrosome. The short isoforms allow plasticity of the cytoskeleton whereas the longer isoforms may preferentially play a role in its stabilization.

del C Alonso A, et al. (2004) J Biol Chem; 279(33): 34873-81

Li G, Yin H, Kuret J (2004) J Biol Chem; 279(16): 15938-45

Giasson BI, et al. (2002) Biochemistry; 41(51): 15376-87

Liu F, et al. (2002) FEBS Lett; 530(1-3): 209

Taniguchi T, et al. (2001) J Biol Chem; 276(13): 10025-31

Note: This product is for in vitro research use only

Tau(Phospho-Ser396) Antibody

Catalog No: #11102

Package Size: #11102-1 50ul #11102-2 100ul

Orders: order@signalwayantibody.com
Support: tech@signalwayantibody.com

Description

Product Name	Tau(Phospho-Ser396) Antibody
Host Species	Rabbit
Clonality	Polyclonal
Purification	Antibodies were produced by immunizing rabbits with synthetic phosphopeptide and KLH conjugates. Antibodies were purified by affinity-chromatography using epitope-specific phosphopeptide. Non-phospho specific antibodies were removed by chromatogramphy using non-phosphopeptide.
Applications	WB IHC
Species Reactivity	Hu Ms Rt
Specificity	The antibody detects endogenous level of Tau only when phosphorylated at serine396.
Immunogen Type	Peptide-KLH
Immunogen Description	Peptide sequence around phosphorylation site of serine 396 (Y-K-S(p)-P-V) derived from Human Tau.
Target Name	Tau
Modification	Phospho
Other Names	MAPT; MTAPT; MTBT1; Neurofibrillary tangle protein; PHF-tau
Accession No.	Swiss-Prot: P10636NCBI Protein: NP _001116538.1
Uniprot	P10636
GeneID	4137;
Concentration	1.0mg/ml
Formulation	Supplied at 1.0mg/mL in phosphate buffered saline (without Mg2+ and Ca2+), pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol.
Storage	Store at -20°C for long term preservation (recommended). Store at 4°C for short term use.

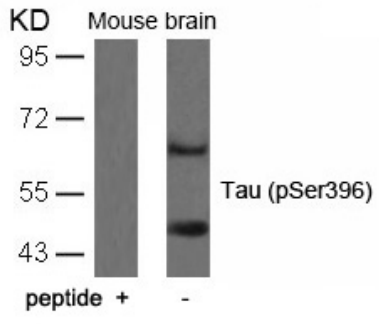
Application Details

Predicted MW: 48 62 78 kd

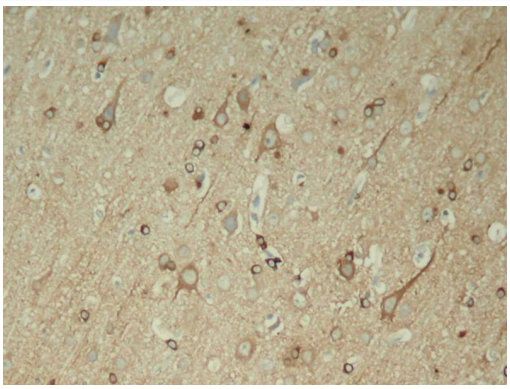
Western blotting: 1:500~1:1000

Immunohistochemistry: 1:50~1:100

Images



Western blot analysis of extracts from mouse brain tissue using Tau(Phospho-Ser396) Antibody #11102 and the same antibody preincubated with blocking peptide.



Immunohistochemical analysis of paraffin-embedded rat hippocampal region tissue from a model with Alzheimer

Background

Promotes microtubule assembly and stability, and might be involved in the establishment and maintenance of neuronal polarity. The C-terminus binds axonal microtubules while the N-terminus binds neural plasma membrane components, suggesting that tau functions as a linker protein between both. Axonal polarity is predetermined by tau localization (in the neuronal cell) in the domain of the cell body defined by the centrosome. The short isoforms allow plasticity of the cytoskeleton whereas the longer isoforms may preferentially play a role in its stabilization.

Puig B, et al. (2005) Acta Neuropathol (Berl). 110(3):261-268.

DeGiorgis JA, et al. (2005) J Proteome Res. 4(2): 306-315.

Alonso Adel C, et al. (2004) J Biol Chem. 279(33): 34873-34881.

Note: This product is for in vitro research use only

Tau(Phospho-Ser404) Antibody

Catalog No: #11112

Package Size: #11112-1 50ul #11112-2 100ul

Orders: order@signalwayantibody.com
Support: tech@signalwayantibody.com

Description

Product Name	Tau(Phospho-Ser404) Antibody
Host Species	Rabbit
Clonality	Polyclonal
Purification	Antibodies were produced by immunizing rabbits with synthetic phosphopeptide and KLH conjugates. Antibodies were purified by affinity-chromatography using epitope-specific phosphopeptide. Non-phospho specific antibodies were removed by chromatogramphy using non-phosphopeptide.
Applications	WB IHC IF
Species Reactivity	Hu Ms Rt
Specificity	The antibody detects endogenous level of Tau only when phosphorylated at serine 404.
Immunogen Type	Peptide-KLH
Immunogen Description	Peptide sequence around phosphorylation site of serine 404 (D-T-S(p)-P-R) derived from Human Tau.
Target Name	Tau
Modification	Phospho
Other Names	MAPT; MTAPT; MTBT1; Neurofibrillary tangle protein; PHF-tau
Accession No.	Swiss-Prot: P10636NCBI Protein: NP_001116538.1
Uniprot	P10636
GeneID	4137;
Concentration	1.0mg/ml
Formulation	Supplied at 1.0mg/mL in phosphate buffered saline (without Mg2+ and Ca2+), pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol.
Storage	Store at -20°C for long term preservation (recommended). Store at 4°C for short term use.

Application Details

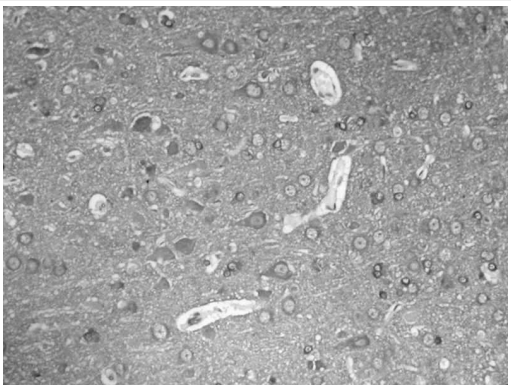
Predicted MW: 48 62 78 kd

Western blotting: 1:500~1:1000

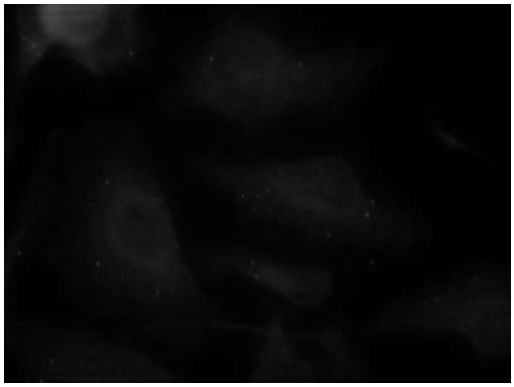
Immunohistochemistry: 1:50~1:100

Immunofluorescence: 1:100~1:200

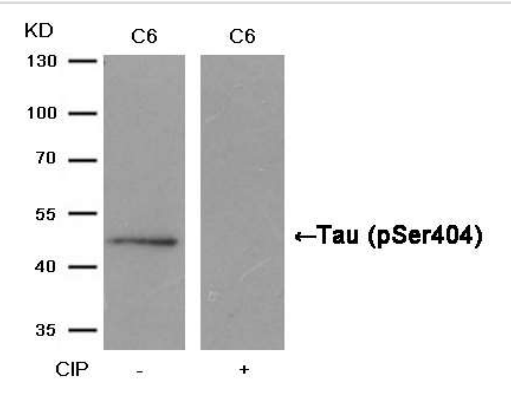
Images



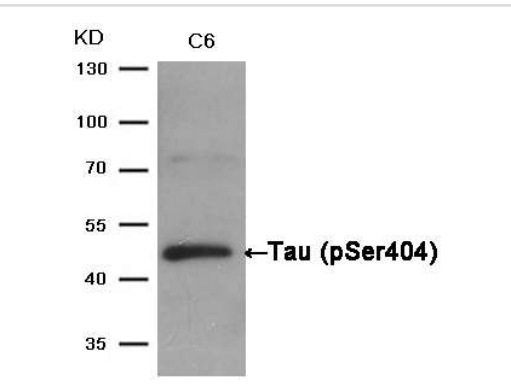
Immunohistochemical analysis of paraffin-embedded rat hippocampal region tissue from a model with Alzheimer



Immunofluorescence staining of methanol-fixed Hela cells using Tau(Phospho-Ser404) Antibody #11112.



Western blot analysis of extracts from C6 cells, treated with calf intestinal phosphatase (CIP), using Tau (Phospho-Ser404) Antibody #11112.



Western blot analysis of extracts from C6 cells using Tau(Phospho-Ser404) Antibody #11112.

Background

Promotes microtubule assembly and stability, and might be involved in the establishment and maintenance of neuronal polarity. The C-terminus binds axonal microtubules while the N-terminus binds neural plasma membrane components, suggesting that tau functions as a linker protein between both. Axonal polarity is predetermined by tau localization (in the neuronal cell) in the domain of the cell body defined by the centrosome. The short isoforms allow plasticity of the cytoskeleton whereas the longer isoforms may preferentially play a role in its stabilization.

Li G, Yin H, et.al .(2004) J Biol Chem ; 279(16): 15938-45.

Noble W, et al. (2003) Neuron ; 38(4): 555-65.

Giasson BI, et al.(2002). Biochemistry; 41(51): 15376-87.

Lee G., et.al. (1989). Neuron 2:1615-1624.
Andreadis A.et.al. (1992) Biochemistry 31:10626-10633.

Note: This product is for in vitro research use only

abcam

Product datasheet

Anti-Tau (phospho S199) antibody ab4749

★★★★★ 1 Abreviews 4 References 3 图像

概述

产品名称	Anti-Tau (phospho S199)**抗体**
描述	**兔多克隆抗体**to Tau (phospho S199)
宿主	Rabbit
经测试应用	**适用于:** IHC-P, WB
种属反应性	**与反应:** Mouse, Human, African green monkey **预测可用于:** Rat
免疫原	Synthetic peptide corresponding to Human Tau (phospho S199). Database link: P10636
阳性对照	IHC-P: Mouse brain tissue, human brain tissue. WB: African green monkey kidney.
常规说明	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
存储溶液	pH: 7.30 Preservative: 0.05% Sodium azide Constituents: PBS, 50% Glycerol (glycerin, glycerine), 0.1% BSA
纯度	Immunogen affinity purified
纯化说明	Purified from rabbit serum by sequential epitope-specific chromatography. The antibody has been negatively preadsorbed using a non-phosphopeptide corresponding to the site of phosphorylation to remove antibody that is reactive with non-phosphorylated tau. The final product is generated by affinity chromatography using a tau-derived peptide that is phosphorylated at serine 199.
克隆	**多克隆**

同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab4749于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

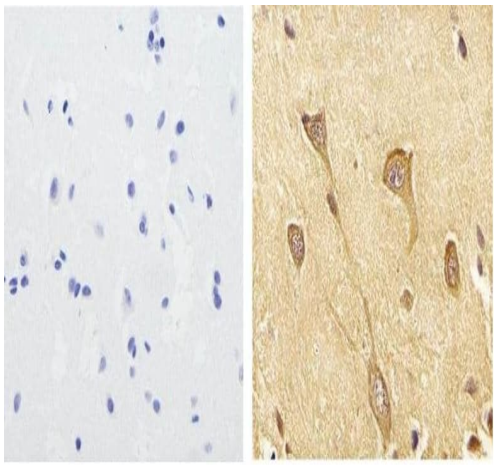
应用	Ab评论	说明
IHC-P		1/20 - 1/200.
WB	★★★★★ (1)	1/1000. Detects a band of approximately 60 kDa.

靶标

功能	Promotes microtubule assembly and stability, and might be involved in the establishment and maintenance of neuronal polarity. The C-terminus binds axonal microtubules while the N-terminus binds neural plasma membrane components, suggesting that tau functions as a linker protein between both. Axonal polarity is predetermined by tau localization (in the neuronal cell) in the domain of the cell body defined by the centrosome. The short isoforms allow plasticity of the cytoskeleton whereas the longer isoforms may preferentially play a role in its stabilization.
组织特异性	Expressed in neurons. Isoform PNS-tau is expressed in the peripheral nervous system while the others are expressed in the central nervous system.
疾病相关	Note=In Alzheimer disease, the neuronal cytoskeleton in the brain is progressively disrupted and replaced by tangles of paired helical filaments (PHF) and straight filaments, mainly composed of hyperphosphorylated forms of TAU (PHF-TAU or AD P-TAU). Defects in MAPT are a cause of frontotemporal dementia (FTD) [MIM:600274]; also called frontotemporal dementia (FTD), pallido-ponto-nigral degeneration (PPND) or historically termed Pick complex. This form of frontotemporal dementia is characterized by presenile dementia with behavioral changes, deterioration of cognitive capacities and loss of memory. In some cases, parkinsonian symptoms are prominent. Neuropathological changes include frontotemporal atrophy often associated with atrophy of the basal ganglia, substantia nigra, amygdala. In most cases, protein tau deposits are found in glial cells and/or neurons. Defects in MAPT are a cause of Pick disease of the brain (PIDB) [MIM:172700]. It is a rare form of dementia pathologically defined by severe atrophy, neuronal loss and gliosis. It is characterized by the occurrence of tau-positive inclusions, swollen neurons (Pick cells) and argentophilic neuronal inclusions known as Pick bodies that disproportionally affect the frontal and temporal cortical regions. Clinical features include aphasia, apraxia, confusion, anomia, memory loss and personality deterioration. Note=Defects in MAPT are a cause of corticobasal degeneration (CBD). It is marked by extrapyramidal signs and apraxia and can be associated with memory loss. Neuropathologic features may overlap Alzheimer disease, progressive supranuclear palsy, and Parkinson disease. Defects in MAPT are a cause of progressive supranuclear palsy type 1 (PSNP1) [MIM:601104, 260540]; also abbreviated as PSP and also known as Steele-Richardson-Olszewski syndrome. PSNP1 is characterized by akinetic-rigid syndrome, supranuclear gaze palsy, pyramidal tract dysfunction, pseudobulbar signs and cognitive capacities deterioration. Neurofibrillary tangles and gliosis but no amyloid plaques are found in diseased brains. Most cases appear to be

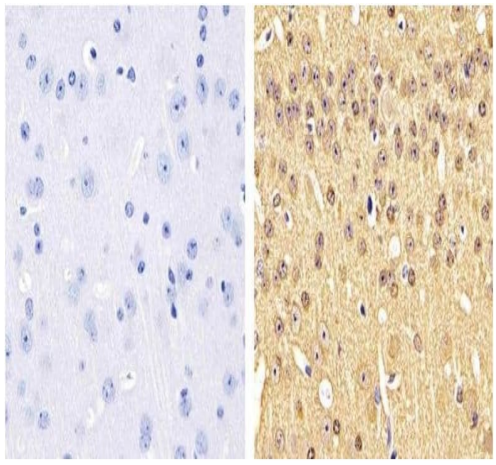
	sporadic, with a significant association with a common haplotype including the MAPT gene and the flanking regions. Familial cases show an autosomal dominant pattern of transmission with incomplete penetrance; genetic analysis of a few cases showed the occurrence of tau mutations, including a deletion of Asn-613.
序列相似性	Contains 4 Tau/MAP repeats.
发展阶段	Four-repeat (type II) tau is expressed in an adult-specific manner and is not found in fetal brain, whereas three-repeat (type I) tau is found in both adult and fetal brain.
结构域	The tau/MAP repeat binds to tubulin. Type I isoforms contain 3 repeats while type II isoforms contain 4 repeats.
翻译后修饰	Phosphorylation at serine and threonine residues in S-P or T-P motifs by proline-directed protein kinases (PDPK: CDK1, CDK5, GSK-3, MAPK) (only 2-3 sites per protein in interphase, seven-fold increase in mitosis, and in PHF-tau), and at serine residues in K-X-G-S motifs by MAP/microtubule affinity-regulating kinase (MARK) in Alzheimer diseased brains. Phosphorylation decreases with age. Phosphorylation within tau's repeat domain or in flanking regions seems to reduce tau's interaction with, respectively, microtubules or plasma membrane components. Phosphorylation on Ser-610, Ser-622, Ser-641 and Ser-673 in several isoforms during mitosis. Polyubiquitinated. Requires functional TRAF6 and may provoke SQSTM1-dependent degradation by the proteasome (By similarity). PHF-tau can be modified by three different forms of polyubiquitination. 'Lys-48'-linked polyubiquitination is the major form, 'Lys-6'-linked and 'Lys-11'-linked polyubiquitination also occur. Glycation of PHF-tau, but not normal brain tau. Glycation is a non-enzymatic post-translational modification that involves a covalent linkage between a sugar and an amino group of a protein molecule forming ketoamine. Subsequent oxidation, fragmentation and/or cross-linking of ketoamine leads to the production of advanced glycation endproducts (AGES). Glycation may play a role in stabilizing PHF aggregation leading to tangle formation in AD.
细胞定位	Cytoplasm > cytosol. Cell membrane. Cytoplasm > cytoskeleton. Cell projection > axon. Mostly found in the axons of neurons, in the cytosol and in association with plasma membrane components.
形式	There are 9 isoforms produced by alternative splicing.

图片



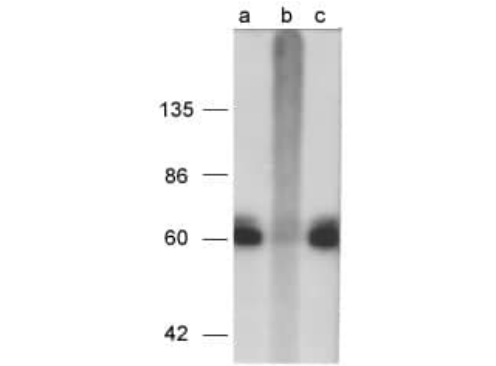
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Tau (phospho S199) antibody (ab4749)

Paraffin embedded human brain tissue (right) stained for Tau using ab4749 at 1/100 dilution in immunohistochemical analysis. Negative control without primary antibody (left).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Tau (phospho S199) antibody (ab4749)

Paraffin embedded mouse brain tissue (right) stained for Tau using ab4749 at 1/100 dilution in immunohistochemical analysis. Negative control without primary antibody (left).



Western blot - Anti-Tau (phospho S199) antibody (ab4749)

Cell extracts from African green monkey kidney (CV-1) cells, stably expressing human four repeat tau and a protein phosphatase inhibitor, were resolved by SDS-PAGE on a 10% Tris-glycine gel. The proteins were transferred to nitrocellulose. Membranes were incubated with 0.50 µg/mL anti-phospho tau [pS199] (ab4749), following prior incubation in the absence (a) or presence of the peptide immunogen (b), or the nonphosphopeptide corresponding to the tau phosphopeptide (c). Cell extracts from African green monkey kidney (CV-1) cells, stably expressing human four repeat tau and a protein phosphatase inhibitor, were resolved by SDS-PAGE on a 10% Tris-glycine gel. The proteins were transferred to nitrocellulose. Membranes were incubated with 0.50 µg/mL anti-phospho tau [pS199] (ab4749), following prior incubation in the

absence (a) or presence of the peptide immunogen (b), or the nonphosphopeptide corresponding to the tau phosphopeptide (c).

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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- 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

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Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors

Phospho-Tau (Ser202, Thr205) Monoclonal Antibody (AT8)

Product Details	
Size	100 µg
Species Reactivity	Human, Mouse, Rat
Published Species	Dog, Rabbit, Rat, Fruit fly, Non-human primate, Hamster, Cat, Human, Mouse
Host/Isotype	Mouse / IgG1, kappa
Class	Monoclonal
Type	Antibody
Clone	AT8
Conjugate	Unconjugated
Immunogen	Partially purified human PHF-Tau
Form	Liquid
Concentration	0.2 mg/mL
Purification	Protein A
Storage buffer	PBS
Contains	no preservative
Storage conditions	-20° C, Avoid Freeze/Thaw Cycles
RRID	AB_223647

Applications	Tested Dilution	Publications
Western Blot (WB)	1:250-1:2,000	195 Publications
Immunohistochemistry (IHC)	-	319 Publications
Immunohistochemistry (Paraffin) (IHC (P))	5-10 µg/mL	40 Publications
Immunohistochemistry (Frozen) (IHC (F))	-	9 Publications
Immunohistochemistry - Free Floating (IHC (Free))	-	18 Publications
Immunocytochemistry (ICC/IF)	1:50-1:1,000	35 Publications
Flow Cytometry (Flow)	-	1 Publication
ELISA (ELISA)	-	5 Publications
Dot blot (DB)	-	1 Publication
Miscellaneous PubMed (Misc)	-	21 Publications

Product Specific Information

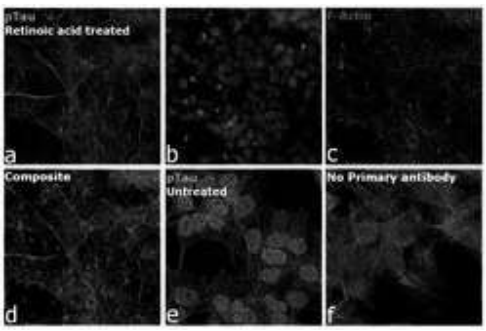
MN1020 targets PHF-tau (Ser202/Thr205)a in ELISA, IF, IHC(P), and WB applications and shows reactivity with Human samples.

The MN1020 immunogen is partially purified human PHF-Tau.

MN1020 detects PHF-tau (Ser202/Thr205)a which has a predicted molecular weight of approximately 79 kDa.

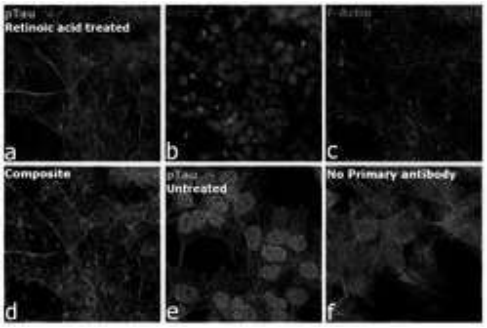
This product is a Low Endotoxin formulation.

Advanced Verification Data

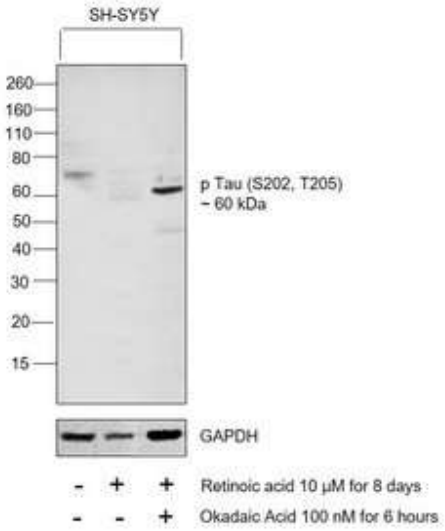


Phospho-Tau (Ser202, Thr205) Antibody (MN1020)
Detection of altered subcellular localization of the target protein by cell treatment demonstrates antibody specificity. Immunofluorescence analysis using Phospho-Tau (Ser202, Thr205) Monoclonal Antibody (AT8) (Product # MN1020), shows plasma membrane and cytoskeleton localization in SH-SY5Y cells treated with retinoic acid as compared to untreated SH-SY5Y cells which is reported to be low to negative for PhosphoTau expression. Cell treatment validation info.

Product Images For Phospho-Tau (Ser202, Thr205) Monoclonal Antibody (AT8)



Phospho-Tau (Ser202, Thr205) Antibody (MN1020) in ICC/IF
Immunofluorescence analysis of Phospho-Tau (Ser202, Thr205) was performed using 70% confluent log phase SH-SY5Y Retinoic acid 100 nM, 8 days. The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 15 minutes, and blocked with 2% BSA for 45 minutes at room temperature. The cells were labeled with Phospho-Tau (Ser202, Thr205) Monoclonal Antibody (AT8) (Product # MN1020) at 1:100 in 0.1% BSA, incubated at 4 degree celsius overnight and then labeled with Donkey anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor Plus 488 (Product # A32766), (1:2000), for 45 minutes at room temperature (Panel a: Green). Nuclei (Panel b:Blue) were stained with ProLong™ Diamond Antifade Mountant with DAPI (Product # P36962). F-actin (Panel c: Red) was stained with Rhodamine Phalloidin (Product # R415, 1: 300). Panel d represents the merged image showing Plasma membrane and cytoskeleton localization. Panel e represents untreated cells showing faint signal Panel f represents control cells with no primary antibody to assess background. The images were captured at 60X magnification.



Phospho-Tau (Ser202, Thr205) Antibody (MN1020) in WB
Western blot was performed using Anti-Phospho-Tau (Ser202, Thr205) Monoclonal Antibody (AT8) (Product # MN1020) and a 60kDa band corresponding to Phospho-Tau (Ser202, Thr205) was observed across cell line tested and increased upon Retinoic acid and the successive Okadaic acid treatments. Whole cell extracts (30 µg lysate) of SH-SY5Y (Lane 1), SH-SY5Y treated with Retinoic acid (10 uM for 8 days) (Lane 2), SH-SY5Y differentiated with retinoic acid and treated with Okadaic acid (100 nM for 6 hours) (Lane 3) were electrophoresed using NuPAGE™ 10% Bis-Tris Protein Gel (Product # NP0301BOX). Resolved proteins were then transferred onto a Nitrocellulose membrane (Product # IB23001) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with the primary antibody (1:1000) and detected by chemiluminescence with Goat anti-Mouse IgG (H+L) Superclonal™ Recombinant Secondary Antibody, HRP (Product # A28177, 1:5000 dilution) using the iBright FL 1000 (Product # A32752). Chemiluminescent detection was performed using SuperSignal™ West Dura Extended Duration Substrate (Product # 34076).

644 References

Western Blot (195)

Journal of Alzheimer's disease : JAD

Partial Inhibition of Mitochondrial Complex I Reduces Tau Pathology and Improves Energy Homeostasis and Synaptic Function in 3xTg-AD Mice.

"MN1020 was used in Western Blotting to investigate the effect of specific MCI inhibitor tricyclic pyrone compound CP2 on levels of human pTau, memory function, long term potentiation (LTP), and energy homeostasis in 18-month-old 3xTg-AD mice and explore the potential mechanisms."

Authors: Stojakovic A,Chang SY,Nesbitt J,Pichurin NP,Ostroot MA,Aikawa T,Kanekiyo T,Trushina E

Species Mouse

Dilution 1:1000

Year 2021

Communications biology

The anesthetic sevoflurane induces tau trafficking from neurons to microglia.

"MN1020 was used in Western Blotting to assess mice and neurons treated with anesthetics sevoflurane and desflurane, and apply nanobeam-sensor technology, an ultrasensitive method, to measure tau/p-tau amounts."

Authors: Dong Y,Liang F,Huang L,Fang F,Yang G,Tanzi RE,Zhang Y,Quan Q,Xie Z

Species Mouse

Dilution 1:200

Year 2021

View more WB references on thermofisher.cn

Immunohistochemistry (319)

Molecular psychiatry

Loss of function of the mitochondrial peptidase PITRM1 induces proteotoxic stress and Alzheimer's disease-like pathology in human cerebral organoids.

"MN1020 was used in Immunohistochemistry to support a mechanistic link between mitochondrial function and common neurodegenerative proteinopathies."

Authors: Pérez MJ,Ivanyuk D,Panagiotakopoulou V,Di Napoli G,Kalb S,Brunetti D,Al-Shaana R,Kaeser SA,Fraschka SA,Jucker M,Zeviani M,Viscomi C,Deleidi M

Species Human

Dilution 1:1000

Year 2021

Frontiers in neuroscience

Reduction of the RNA Binding Protein TIA1 Exacerbates Neuroinflammation in Tauopathy.

"MN1020 was used in Immunohistochemistry to demonstrate that TIA1 plays an important role in the neuroimmune response to chronic stress in the form of tauopathy."

Authors: LeBlang CJ,Medalla M,Nicoletti NW,Hays EC,Zhao J,Shattuck J,Cruz AL,Wolozin B,Luebke JI

Species Mouse

Dilution 1:100

Year 2021

View more IHC references on thermofisher.cn

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IHC (P) (40) | IHC (F) (9) | IHC (Free) (18) | ICC/IF (35) | Flow (1) | ELISA (5) | DB (1) | Misc (21)

abcam

Product datasheet

Anti-Iba1 antibody [EPR16588] ab178846

重组 RabMAb

★★★★★ 40 Abreviews 160 References 13 图像

概述

产品名称	Anti-Iba1抗体[EPR16588]
描述	兔单克隆抗体[EPR16588] to Iba1
宿主	Rabbit
特异性	For ab178846 Abcam recommends blocking in 3% milk for cleanest results in WB. Blocking with BSA gives slightly higher background.
经测试应用	**适用于:** Flow Cyt (Intra), IHC-P, WB, ICC/IF
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: HL-60, THP-1, U937, RAW 264.7 and NR8383 whole cell lysates; Human, mouse and rat spleen lysates; Mouse testis and liver lysates. IHC-P: Human Cerebral cortex, human hippocampus; Rat and mouse normal brain tissues. Flow Cyt (intra): U937 cells. IHC-Fr: Mouse Cerebral cortex tissue.
常规说明	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.

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
存储溶液	Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
纯度	Protein A purified
克隆	单克隆

克隆编号	EPR16588
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab178846于以下的经测试应用

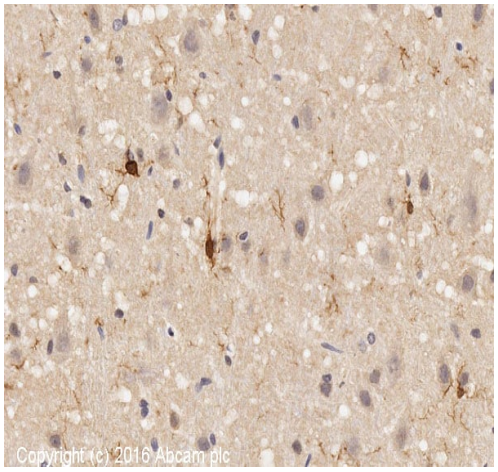
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		1/160.
IHC-P	★★★★★ (14)	1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB	★★★★★ (2)	1/500 - 1/2000. Detects a band of approximately 10, 15 kDa (predicted molecular weight: 17 kDa). Abcam recommends blocking in 3% milk for cleanest results in WB. Blocking with BSA gives slightly higher background.
ICC/IF	★★★★★ (5)	1/500.

靶标

功能	Actin-binding protein that enhances membrane ruffling and RAC activation. Enhances the actin-bundling activity of LCP1. Binds calcium. Plays a role in RAC signaling and in phagocytosis. May play a role in macrophage activation and function. Promotes the proliferation of vascular smooth muscle cells and of T-lymphocytes. Enhances lymphocyte migration. Plays a role in vascular inflammation.
组织特异性	Detected in T-lymphocytes and peripheral blood mononuclear cells.
序列相似性	Contains 2 EF-hand domains.
翻译后修饰	Phosphorylated on serine residues.
细胞定位	Cytoplasm > cytoskeleton. Cell projection > ruffle membrane. Associated with the actin cytoskeleton at membrane ruffles and at sites of phagocytosis.

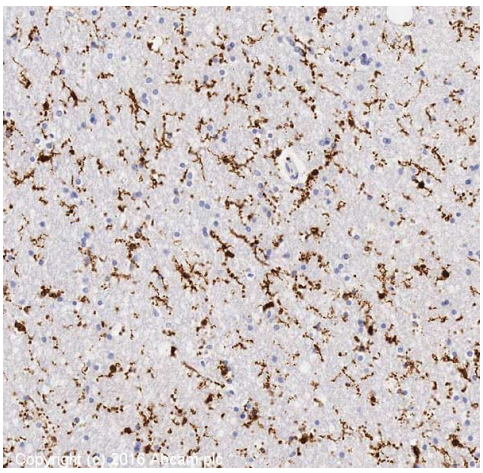
图片



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody [EPR16588] (ab178846)

IHC image of Iba1 staining in rat normal brain formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab178846, 1/2000 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.

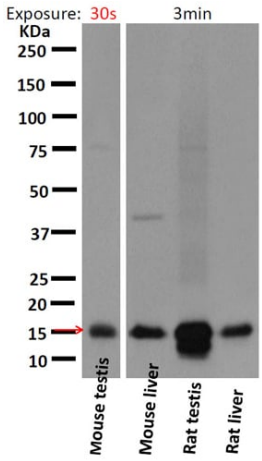
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody [EPR16588] (ab178846)

IHC image of Iba1 staining in human normal hippocampus formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab178846, 1/2000 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.

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.



Western blot - Anti-Iba1 antibody [EPR16588] (ab178846)

All lanes : Anti-Iba1 antibody [EPR16588] (ab178846)

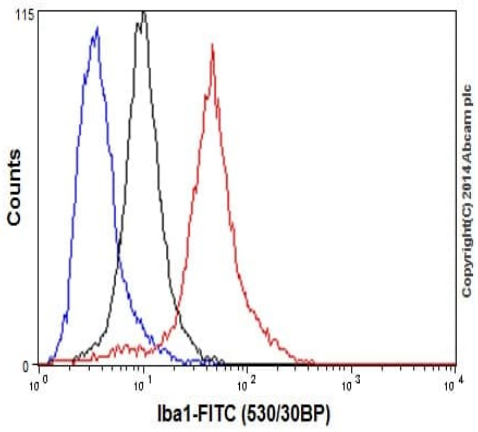
Lane 1 : Mouse testis

Lane 2 : Mouse liver

Lane 3 : Rat testis

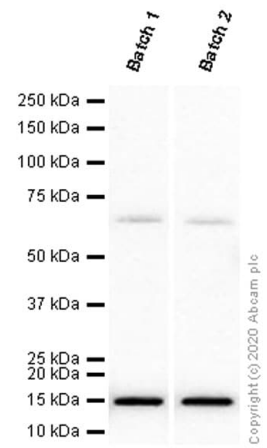
Lane 4 : Rat liver

Predicted band size: 17 kDa



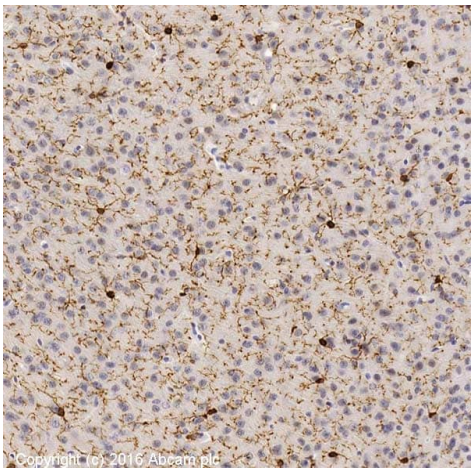
Flow Cytometry (Intracellular) - Anti-Iba1 antibody [EPR16588] (ab178846)

Intracellular Flow Cytometry analysis of2% paraformaldehyde fixed U937 (human histiocytic lymphoma cell line) cells labeling Iba1with ab178846 at 1/160 dilution (red line). Secondary antibody used is a goat anti rabbit IgG (FITC) at 1/150 dilution. The isotype control is rabbit monoclonal IgG (black line). The unlabeled control is cells without incubation with primary and secondary antibodies (blue line).



Western blot - Anti-Iba1 antibody [EPR16588] (ab178846)

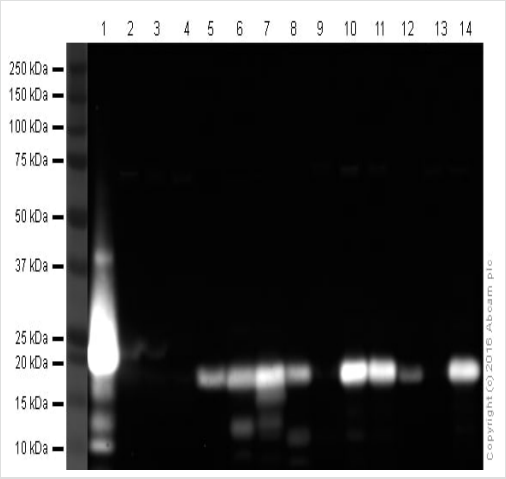
Different batches of ab178846 were tested on THP-1 (Human monocytic leukemia monocyte) lysate at 0.02 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 15 kDa.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody [EPR16588] (ab178846)

IHC image of Iba1 staining in mouse normal brain formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol B. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab178846, 1/2000 dilution, for 15 mins at room temperature. A goat anti-rabbit biotinylated secondary antibody was used to detect the primary, and visualized using an HRP conjugated ABC system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

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.



Western blot - Anti-Iba1 antibody [EPR16588] (ab178846)

All lanes : Anti-Iba1 antibody [EPR16588] (ab178846) at 1/500 dilution

Lane 1 : Human Iba1 recombinant protein at 0.1 µg
Lane 2 : HEK-293 (human epithelial cell line from embryonic kidney) whole cell lysate at 20 µg
Lane 3 : A431 (human epidermoid carcinoma cell line) whole cell lysate at 20 µg
Lane 4 : NIH/3T3 (mouse embyro fibroblast cell line) whole cell lysate at 30 µg
Lane 5 : Human spleen tissue lysate at 20 µg
Lane 6 : Mouse spleen tissue lysate at 30 µg
Lane 7 : Rat spleen tissue lysate at 30 µg
Lane 8 : U937 (human histiocytic lymphoma cell line) whole cell lysate at 30 µg
Lane 9 : MOLT-4 (human lymphoblastic leukemia cell line) whole cell lysate at 20 µg
Lane 10 : THP-1 (human monocytic leukemia cell line) whole cell lysate at 30 µg
Lane 11 : THP-1 whole cell lysate, PMA treated at 30 µg
Lane 12 : RAW 264.7 (mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate at 30 µg
Lane 13 : C6 (rat glial tumor cell line) whole cell lysate at 30 µg
Lane 14 : NR8383 whole cell lysate at 30 µg

Developed using the ECL technique.

Performed under reducing conditions.

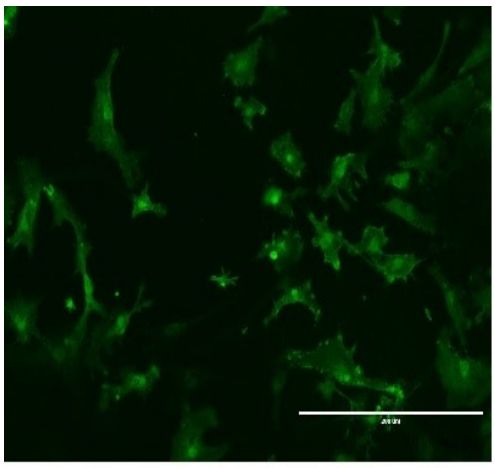
Predicted band size: 17 kDa

Exposure time: 1 minute

Abcam recommends blocking in milk for cleaner blots with reduced background, in comparison to BSA.

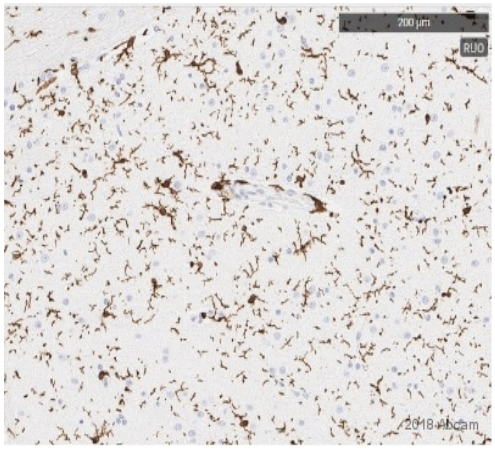
This blot was produced using a 4-12% Bis-Tris gel under the MOPS buffer system. The gel was run at 200V for 60 minutes before being transferred onto a nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour before being

incubated with ab178846 (anti-Iba1 antibody; 1/500 dilution) for 18 hours at 4°C. Antibody binding was detected using ab97040 (HRP-labelled goat anti-mouse IgG) at 1:50,000 dilution for 1 hour at room temperature and visualised using ECL development solution ab133406.



Immunocytochemistry/ Immunofluorescence - Anti-Iba1 antibody [EPR16588] (ab178846)

0.1% Triton-X 100 permeabilized paraformaldehyde-fixed Mouse cell Microglia cells labeling Iba1 (green) using ab178846 at 1/500 dilution in ICC/IF analysis.

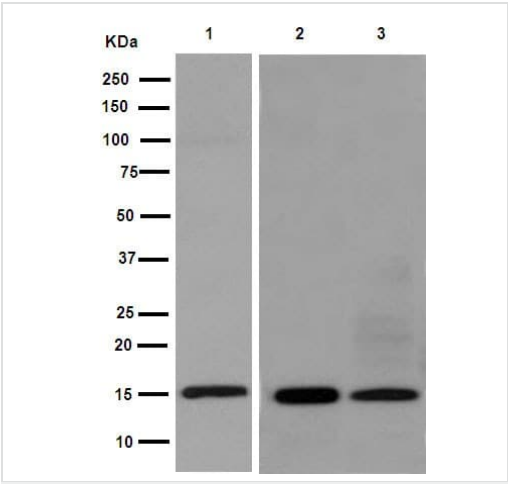


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody [EPR16588] (ab178846)

This image was courtesy of an annonymous Abreview

Formaldehyde-fixed, paraffin-embedded cynomolgus monkey brain tissue stained for Iba1 using ab178846 at 1/6000 dilution in immunohistochemical analysis.

Antigen Retrieval: Heat mediated - Buffer/Enzyme Used: pH 9.0 EDTA



Western blot - Anti-Iba1 antibody [EPR16588] (ab178846)

All lanes : Anti-Iba1 antibody [EPR16588] (ab178846) at 1/10000 dilution

Lane 1 : THP-1 (human monocytic leukemia cell line) whole cell lysate
Lane 2 : U937 (human histiocytic lymphoma cell line) whole cell lysate
Lane 3 : Human spleen whole cell lysate

Lysates/proteins at 10 µg per lane.

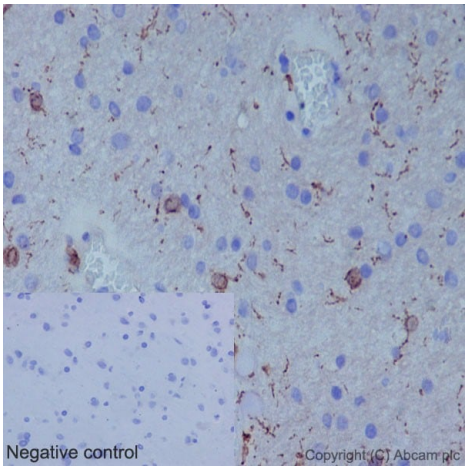
Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Developed using the ECL technique.

Predicted band size: 17 kDa
Observed band size: 15 kDa

Blocking buffer and concentration: 5% NFDM/TBST

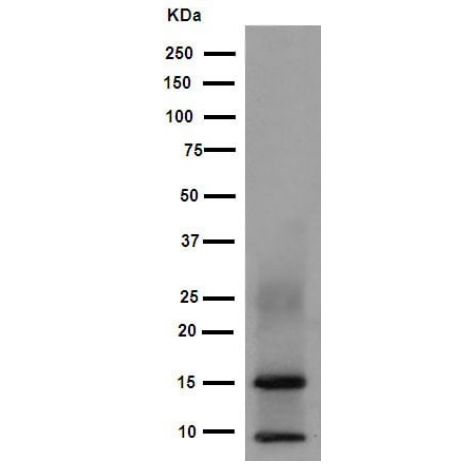
Diluting buffer and concentration: 5% NFDM /TBST



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Iba1 antibody [EPR16588] (ab178846)

Immunohistochemical analysis of paraffin-embedded Human cerebral cortex tissue labeling Iba1 with ab178846 at a 1/2000 dilution showing cytoplasm and nuclear staining on Glial cells. Counter stained with hematoxylin. Prediluted HRP Polymer for Rabbit/Mouse IgG was used as the secondary aantibody. Negative control also shown.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Western blot - Anti-Iba1 antibody [EPR16588] (ab178846)

Anti-Iba1 antibody [EPR16588] (ab178846) at 1/2000 dilution + HL-60 (human promyelocytic leukemia cell line) whole cell lysate at 10 µg

Secondary

Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

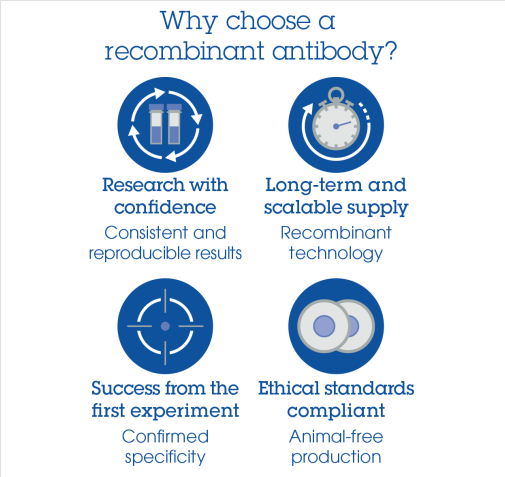
Predicted band size: 17 kDa
Observed band size: 10, 15 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration:

5% NFDM /TBST.

Based on sequence analysis, ab178846 recognizes 2 isoforms with the predicted MWs of 17KDa and 11KDa, respectively.



Anti-Iba1 antibody [EPR16588] (ab178846)

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Revision 3

Store at -20C

#3670

GFAP (GA5) Mouse mAb

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Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
WB, IHC-P, IF-F, IF-IC, F	H M R	Endogenous	50	Mouse IgG1	P14136	2670

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunohistochemistry (Paraffin)	1:50
Immunofluorescence (Frozen)	1:300
Immunofluorescence (Immunocytochemistry)	1:300
Flow Cytometry	1:400 - 1:1600

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.

Specificity / Sensitivity

GFAP (GA5) Mouse mAb detects endogenous levels of total GFAP protein.
Species Reactivity:
Human, Mouse, Rat

Source / Purification

Monoclonal antibody is produced by immunizing animals with native GFAP purified from pig spinal cord.

Background

The cytoskeleton consists of three types of cytosolic fibers: microfilaments (actin filaments), intermediate filaments, and microtubules. Major types of intermediate filaments are specifically expressed in particular cell types: cytokeratins in epithelial cells, glial fibrillary acidic protein (GFAP) in glial cells, desmin in skeletal, visceral, and certain vascular smooth muscle cells, vimentin in cells of mesenchymal origin, and neurofilaments in neurons. GFAP and vimentin form intermediate filaments in astroglial cells and modulate their motility and shape (1). In particular, vimentin filaments are present at early developmental stages, while GFAP filaments are characteristic of differentiated and mature brain astrocytes. Thus, GFAP is commonly used as a marker for intracranial and intraspinal tumors arising from astrocytes (2). In addition, GFAP intermediate filaments are also present in nonmyelin-forming Schwann cells in the peripheral nervous system (3).

1. Eng, L.F. et al. (2000) *Neurochem. Res.* 25, 1439-51.
2. Goebel, H.H. et al. (1987) *Acta. Histochem. Suppl.* 34, 81-93.
3. Jessen, K.R. et al. (1990) *Development* 109, 91-103.

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

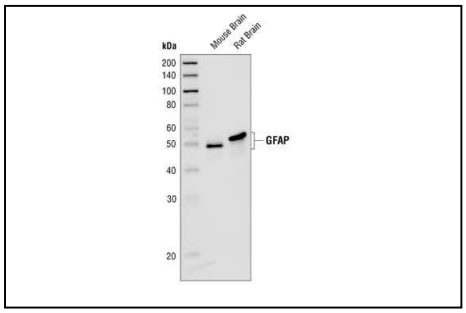
APPLICATIONS KEY **WB:** Western Blot **IP:** Immunoprecipitation **IHC:** Immunohistochemistry **ChIP:** Chromatin Immunoprecipitation **IF:** Immunofluorescence **F:** Flow Cytometry **E-P:** ELISA-Peptide

CROSS-REACTIVITY KEY **H:** human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **Mi:** mink **C:** chicken **Dm:** D. melanogaster **X:** Xenopus **Z:** zebrafish **B:** bovine **Dg:** dog **Pg:** pig **Sc:** S. cerevisiae **Ce:** C. elegans **Hr:** horse **All:** all species expected

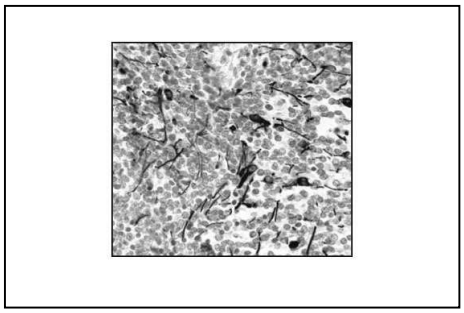
Revision 3

#3670

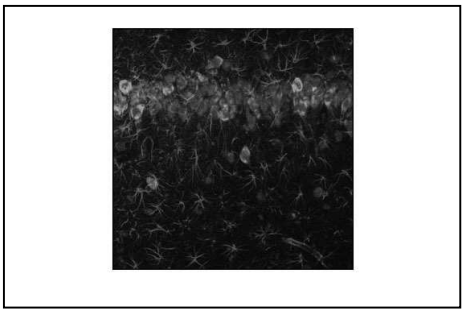
GFAP (GA5) Mouse mAb



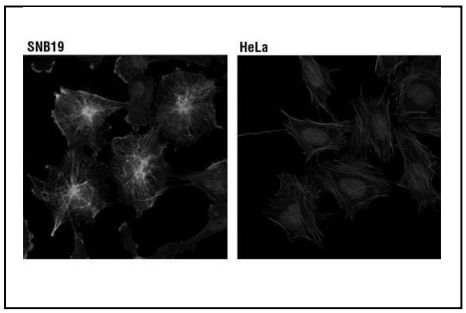
Western blot analysis of extracts from mouse and rat brain, using GFAP (GA5) Mouse mAb.



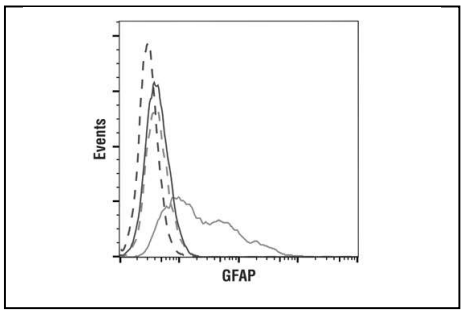
Immunohistochemical analysis of paraffin-embedded human medulloblastoma, using GFAP (GA5) Mouse mAb.



Confocal immunofluorescence image of rat hippocampus labeled with GFAP (GA5) Mouse mAb (red), Phospho-S6 Ribosomal Protein (Ser235/236) (2F9) Rabbit mAb (Alexa Fluor® 488 Conjugate) #4854 (green), and CREB (48H2) Rabbit mAb #9197 (blue).



Confocal immunofluorescent analysis of SNB19 (positive, left) and HeLa (negative, right) cells, using GFAP (GA5) Mouse mAb (green). Actin filaments were labeled with DyLight® 554 Phalloidin #13054 (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).



Flow cytometric analysis of HeLa cells (blue) and SNB-19 cells (green) using GFAP (GA5) Mouse mAb (solid lines) or a concentration-matched Mouse (G3A1) mAb IgG1 Isotype Control #5415 (dashed lines). Anti-mouse IgG (H+L), F(ab')$_2$ Fragment (Alexa Fluor® 488 Conjugate) #4408 was used as a secondary antibody.

#3670

GFAP (GA5) Mouse mAb

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Revision 3

Store at -20C

#9664

Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb

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Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
WB, IP, IHC-P, IF-IC, F	H M R Mk	Endogenous	17, 19	Rabbit IgG	P42574	836

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:50
Immunohistochemistry (Paraffin)	1:2000
Immunofluorescence (Immunocytochemistry)	1:400 - 1:1600
Flow Cytometry	1:6400

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.

For a carrier free (BSA and azide free) version of this product see product #94530.

Specificity / Sensitivity

Cleaved Caspase-3 (Asp175) (5A1) Rabbit mAb detects endogenous levels of the large fragment (17/19 kDa) of activated caspase-3 resulting from cleavage adjacent to Asp175. This antibody does not recognize full length caspase-3 or other cleaved caspases. Non-specific labeling may be observed by immunofluorescence in specific sub-types of healthy cells in fixed-frozen tissues (e.g. pancreatic alpha-cells). Cytoplasmic background may be observed in human and monkey samples.
Species Reactivity:
Human, Mouse, Rat, Monkey

Species predicted to react based on 100% sequence homology:
Bovine, Dog, Pig

Source / Purification

Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to amino-terminal residues adjacent to Asp175 of human caspase-3.

Background

Caspase-3 (CPP-32, Apoptain, Yama, SCA-1) is a critical executioner of apoptosis, as it is either partially or totally responsible for the proteolytic cleavage of many key proteins, such as the nuclear enzyme poly (ADP-ribose) polymerase (PARP) (1). Activation of caspase-3 requires proteolytic processing of its inactive zymogen into activated p17 and p12 fragments. Cleavage of caspase-3 requires the aspartic acid residue at the P1 position (2).

1. Fernandes-Alnemri, T. et al. (1994) *J Biol Chem* 269, 30761-4.
2. Nicholson, D.W. et al. (1995) *Nature* 376, 37-43.

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v nonfat dry milk, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

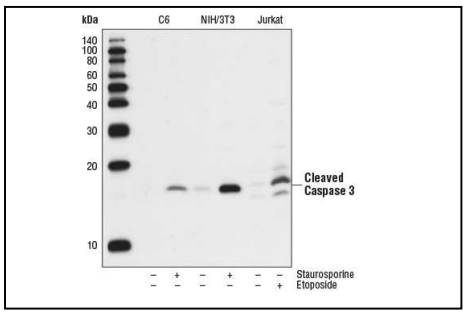
APPLICATIONS KEY **WB:** Western Blot **IP:** Immunoprecipitation **IHC:** Immunohistochemistry **ChIP:** Chromatin Immunoprecipitation **IF:** Immunofluorescence **F:** Flow Cytometry **E-P:** ELISA-Peptide

CROSS-REACTIVITY KEY **H:** human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **Mi:** mink **C:** chicken **Dm:** D. melanogaster **X:** Xenopus **Z:** zebrafish **B:** bovine **Dg:** dog **Pg:** pig **Sc:** S. cerevisiae **Ce:** C. elegans **Hr:** horse **All:** all species expected

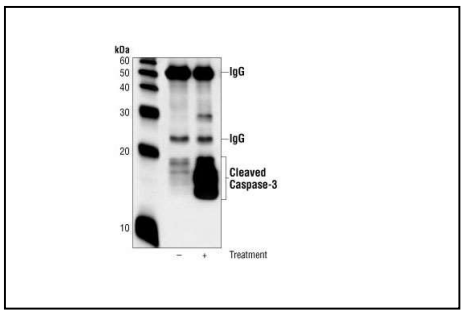
Revision 3

#9664

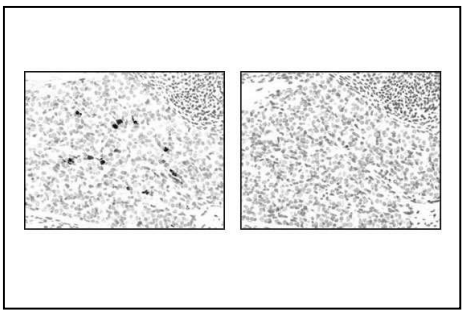
Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb



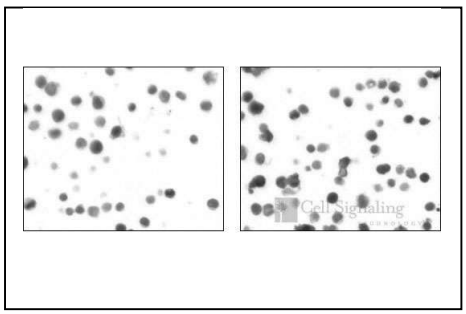
Western blot analysis of extracts from C6 (rat), NIH/3T3 (mouse), and Jurkat (human) cells, untreated or treated with staurosporine #9953 (1uM, 3hrs) or etoposide #2200 (25uM, 5hrs) as indicated, using Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb.



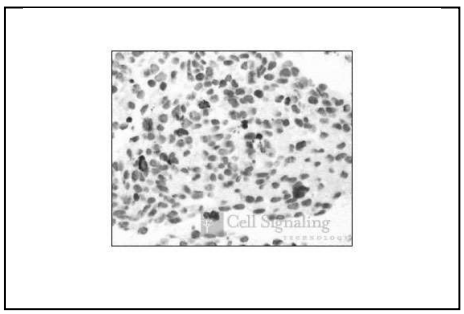
Immunoprecipitation of extracts from Jurkat cells, untreated or etoposide-treated (25uM, 5hrs), using Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb. Western blot was performed using the same antibody.



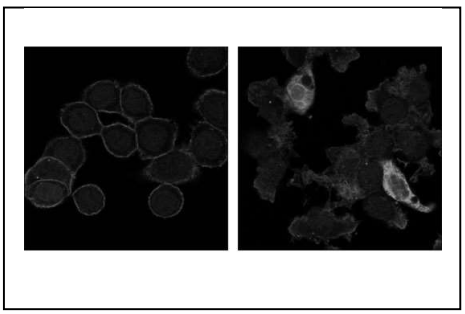
Immunohistochemical analysis of paraffin-embedded mouse embryo, using Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb in the presence of control peptide (left) or Cleaved Caspase-3 (Asp175) Blocking Peptide (#1050) (right).



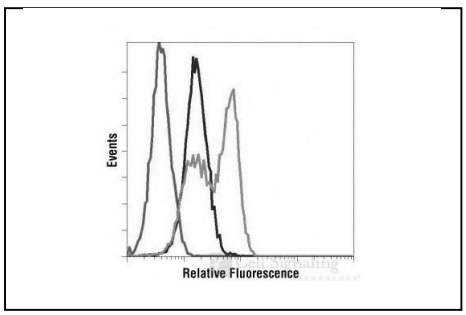
Immunohistochemical analysis using Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb on SignalSlide® Cleaved Caspase-3 IHC Controls #8104 (paraffin-embedded Jurkat cells, untreated (left) or etoposide-treated (right)).



Immunohistochemical staining of paraffin-embedded mouse embryo, showing cytoplasmic localization in apoptotic cells, using Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb.



Confocal immunofluorescent images of HT-29 cells, untreated (left) or Staurosporine #9953 treated (right) labeled with Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb (green). Actin filaments have been labeled with Alexa Fluor® 555 phalloidin #8953 (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).



Flow cytometric analysis of Jurkat cells, untreated (blue) or treated with etoposide #2200 (green), using Cleaved Caspase-3(Asp175) (5A1E) Rabbit mAb compared to a nonspecific negative control antibody (red).

#9664

Cleaved Caspase-3 (Asp175) (5A1E) Rabbit mAb

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abcam

Product datasheet

Anti-NeuN antibody [EPR12763] - Neuronal Marker ab177487

重组 RabMAb

★★★★★ 71 Abreviews 494 References 26 图像

概述

产品名称	Anti-NeuN抗体[EPR12763] - Neuronal Marker
描述	兔单克隆抗体[EPR12763] to NeuN - Neuronal Marker
宿主	Rabbit
经测试应用	**适用于:** Flow Cyt (Intra), IHC (PFA fixed), mIHC, IHC-P, WB, ICC/IF, IHC-Fr
种属反应性	**与反应:** Mouse, Rat, Sheep, Goat, Cat, Dog, Human, Zebrafish, Common marmoset **预测可用于:** Pig, Cynomolgus monkey
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: Mouse brain, mouse cerebellum, rat cerebellum and human fetal brain tissue lysates. ICC/IF: SH-SY-5Y and Mouse primary neuron cells. IHC-P: Human cerebellum, human gliocytoma tissue. mIHC: Human cerebellum tissue IHC-Fr: Mouse dentate gyrus tissue. Flow Cyt (intra): U-87 MG cells.
常规说明	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. **We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.**

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.

存储溶液	Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
纯度	Protein A purified
克隆	单克隆
克隆编号	EPR12763
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab177487于以下的经测试应用

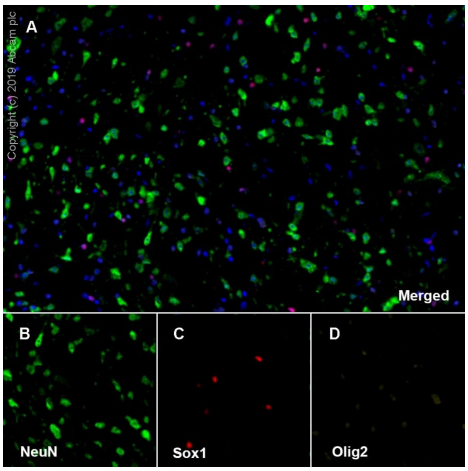
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		1/100. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC (PFA fixed)		Use at an assay dependent concentration.
mIHC		1/1000. Perform Sodium citrate antigen retrieval (pH 6.0) in between rounds of tyramide signal amplification to remove the antibody from the previous round, to avoid any cross-reactivity.
IHC-P	★★★★★ (16)	1/3000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols. **For unpurified use at 1/800.**
WB	★★★★★ (6)	1/1000 - 1/10000. Detects a band of approximately 48,50 kDa (predicted molecular weight: 34 kDa). **For unpurified use at 1/1000 - 1/2000.**
ICC/IF	★★★★☆ (14)	1/100 - 1/300. **For unpurified use at 1/80.**
IHC-Fr	★★★★★ (16)	Use at an assay dependent concentration.

靶标

功能	RNA-binding protein that regulates alternative splicing events.
序列相似性	Contains 1 RRM (RNA recognition motif) domain.
细胞定位	Nucleus. Cytoplasm.

图片



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse cerebrum tissue labelling NeuN with ab177487 at 1/100 dilution (B), SOX1 with ab242125 at 1/100 dilution (C) and Olig2 with ab109186 at 1/100 dilution (D). Anti-Rabbit and Mouse Polymer HRP was used as a secondary antibody, and DAPI was used for a nuclear counter stain. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20mins. Heat mediated antigen retrieval (Leica ER2, PH9.0, 20 minutes) was used in between rounds of tyramide signal amplification to remove the antibodies from the previous round, to avoid any cross-reactivity.

Panel A: merged staining of anti- NeuN (green, Opal™520), anti-SOX1 (red, Opal™570) and anti- Olig2 (yellow, Opal™690).

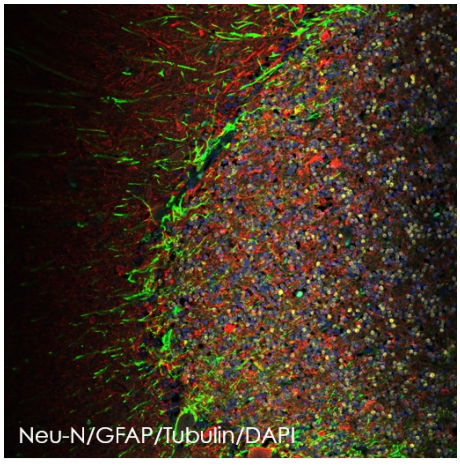
Panel B: anti-NeuN stands for neurons.

Panel C: anti-SOX1 stained on neural progenitors.

Panel D: anti-Olig2 stained on oligodendrocyte.

The section was incubated in three rounds of staining: in the order of ab177487, ab242125 and ab109186 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system.

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope.



Multiplex immunohistochemistry - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

Fluorescence multiplex immunohistochemical analysis of human cerebellum tissue (formalin-fixed paraffin-embedded section).

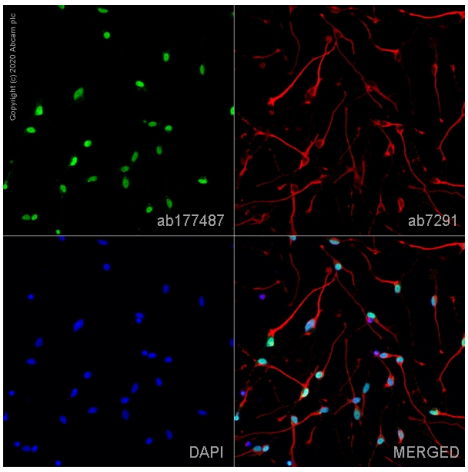
Merged staining of Neu-N (ab177487; yellow; Opal™570), anti-beta III Tubulin (ab52623; red; Opal™690) and anti-GFAP (ab68428; green; Opal™520).

The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal™ kit.

The section was incubated in three rounds of staining with ab177487 (1/1000 dilution), ab52623 (1/200 dilution) and ab68428 (1/250 dilution); each using a separate fluorescent tyramide signal amplification system.

Sodium citrate antigen retrieval (pH 6.0) was used in between rounds of tyramide signal amplification to remove the antibody from the previous round, to avoid any cross-reactivity.

DAPI (blue) was used as a nuclear counter stain.

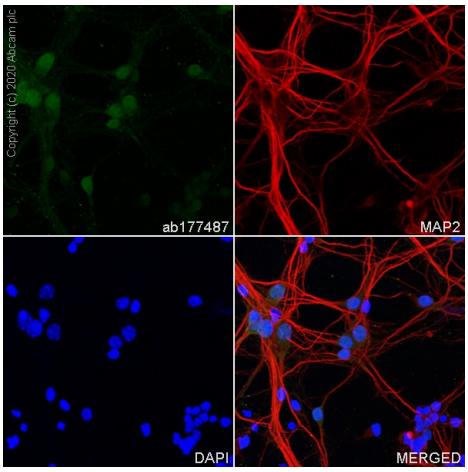


Immunocytochemistry/ Immunofluorescence - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

Immunofluorescence staining of NeuN using ab177487 in ioGlutamatergic Neurons (Human iPSC-Derived Glutamatergic Neurons, ab259259), which were differentiated for 1 day post induction.

The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% PBS-Tween for 5 mins 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 ab177487 at 1 µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin, at 1/1000 dilution. Cells were then incubated with ab150081, Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 dilution (shown in green) and ab150120, Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

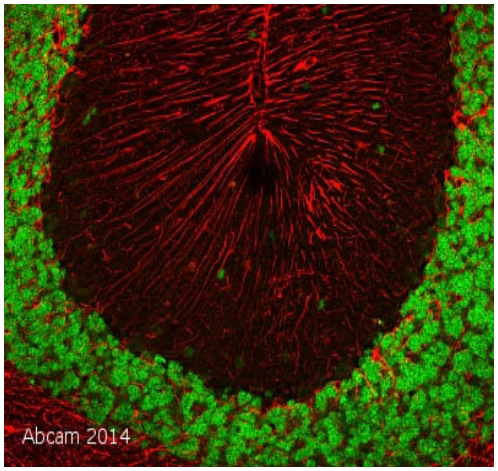
Images were acquired with the Perkin Elmer Operetta HCA and a maximum intensity projection of confocal sections is shown.



Immunocytochemistry/ Immunofluorescence - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

Immunocytochemistry/immunofluorescence analysis of Mouse primary neuron cells labelling NeuN with ab177487 at 1/100. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) at 1/1000 was used as the secondary antibody (green). Cells were counterstained with Anti-MAP2 mouse monoclonal antibody (ab11267) at 1/200 dilution and visualised using Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) at 1/1000 dilution (red). Nuclear DNA was labelled with DAPI (blue).

Confocal image showing mainly nuclear staining in mouse primary neuron cells. Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection.

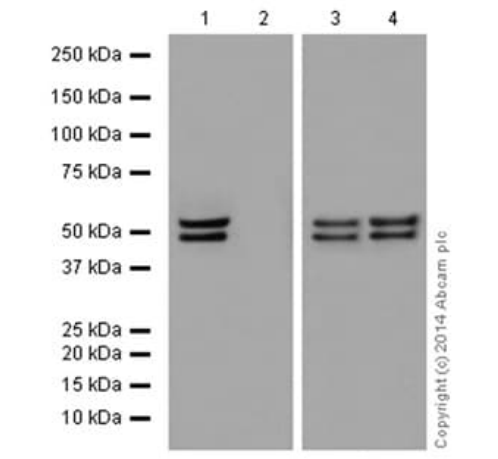


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

IHC-P image of NeuN (green) and GFAP (red) double staining on mouse cerebellum sections using ab177487 (1/5000) and ab4674 (1/1500) respectively.

The sections were deparaffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were then incubated with Rabbit Monoclonal to NeuN (ab177487) diluted at 1/5000 and Chicken Polyclonal to GFAP (ab4674) diluted at 1/1500. The primary antibody was detected using ab150097 Goat anti-rabbit IgG conjugated to Alexa Fluor® 488 (1/500) and ab150176 Goat anti-chicken IgY conjugated to Alexa Fluor® 594 (1/500)



Western blot - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

All lanes : Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487) at 1/10000 dilution (purified)

Lane 1 : Human fetal brain tissue lysate
Lane 2 : HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate
Lane 3 : Mouse brain tissue lysate
Lane 4 : Rat brain tissue lysate

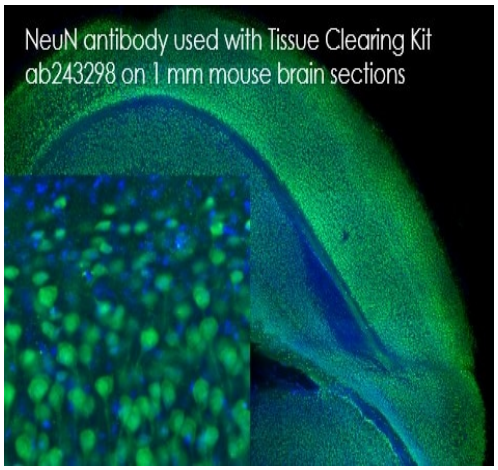
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Peroxidase conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 34 kDa
Observed band size: 46 kDa

Exposure time -
Lane 1-2: 3 minutes.
Lane 3-4: 1 minute.

Blocking and dilution buffer: 5% NFDM/TBST.

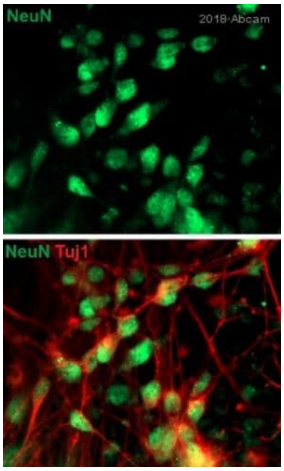


Immunohistochemistry (PFA fixed) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

NeuN antibody ab177487 was used with Tissue Clearing Kit ab243298 to penetrate, stain and clear a 1 mm coronal section of mouse brain. Blue: DAPI, Green: NeuN.

Learn more about tissue clearing kits, reagents, and protocols designed to make it easier to stain thick tissue sections and get more data from each valuable tissue section.

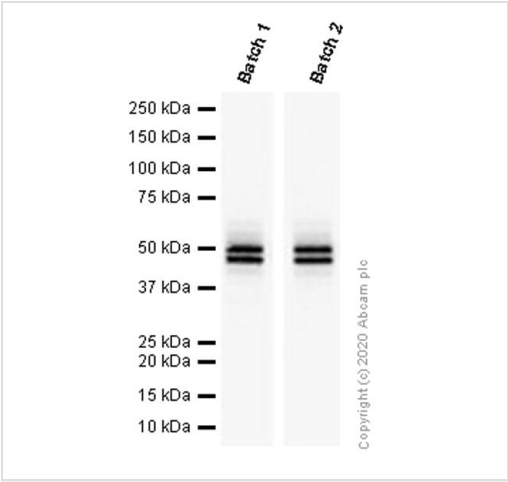
For 1 mm brain sections, we recommend a starting dilution of 1:200, and also using Goat Anti-Rabbit IgG H&L AlexaFluor488 (ab150077) at a dilution of 1:400.



Immunocytochemistry/ Immunofluorescence - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

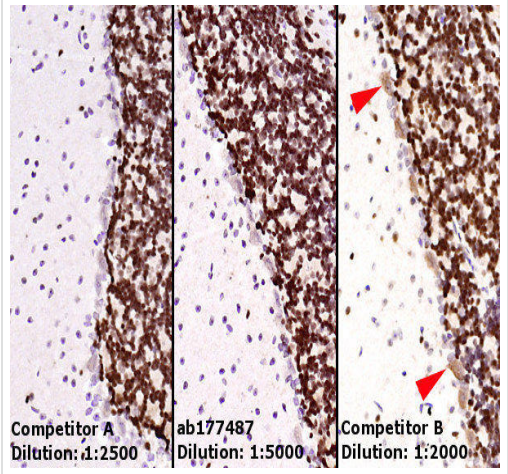
This image is courtesy of an Abreview submitted by Vladimir Milenkovic

Immunocytochemistry/immunofluorescence analysis of human neurons differentiated from iPSCs labelling NeuN (green) with ab177487 at 1/500 in 0.1% TritonX-100, 1% goat serum, 1X PBS for 16 hours at 4°C. Cells were fixed with paraformaldehyde and permeabilized with 0.5% Triton X-100. Then, cells were blocked with 5% serum for 20 minutes at 23°C. Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) at 1/1000 was used as the secondary antibody. Tuj1 antibody was used to stain neuronal dendrites and axons (red).



Western blot - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

Different batches of ab177487 were tested on mouse brain lysate at 2.0 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 46,48 kDa.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

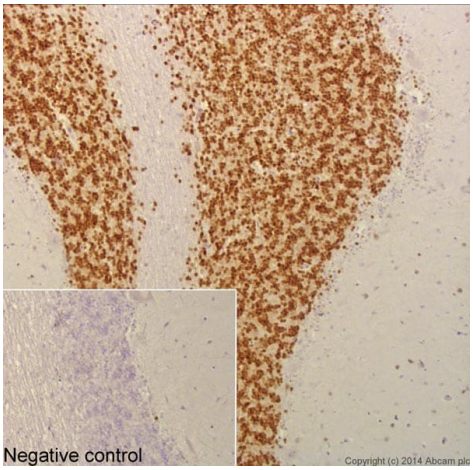
An independent comparison of commercially available NeuN clones in IHC-P.

Competitor A: Leading mouse monoclonal.

Competitor B: Non-Abcam rabbit monoclonal.

Sodium citrate was used for antigen retrieval in all 3 samples.

ab177487 produces specific staining, equivalent to the leading mouse monoclonal at half the dilution. The non-Abcam mouse monoclonal was less specific as it stained Purkinje cells, which do not express NeuN.



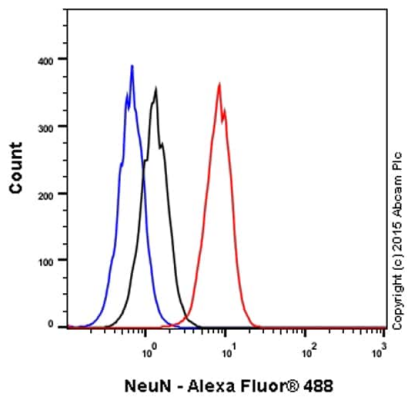
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

IHC image of NeuN (ab177487) with Anti-Rabbit IgG VHH Single Domain Antibody (HRP) (ab191866) staining in formalin fixed paraffin embedded normal human cerebellum tissue section.

The section was dewaxed and then pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6) in a Dako Pascal pressure cooker using the standard factory-set regime. Non-specific protein-protein interactions were then blocked using in TBS containing 0.025% (v/v) Triton X-100, 0.3M (w/v) glycine and 3% (w/v) BSA for 1 hour at room temperature. The section was then incubated with rabbit monoclonal antibody [EPR12763] to NeuN (ab177487, 0.1µg/ml) in TBS containing 0.025% (v/v) Triton X-100 and 3% (w/v) BSA overnight at +4°C. Endogenous peroxidases were quenched using 1.6% (v/v) hydrogen peroxide in TBS containing 0.025% (v/v) Triton X-100 for 30 minutes at room temperature, with agitation. The secondary antibody, Anti-Rabbit IgG VHH Single Domain Antibody (HRP) (ab191866, 1.0µg/ml) was then applied for 1 hour at room temperature in TBS containing 0.025% (v/v) Triton X-100 and 3% (w/v) BSA before being developed for 10 minutes at room temperature using Steady DAB/Plus (ab103723). The section was then counterstained with hematoxylin and mounted with DPX.

The negative control (secondary antibody only, no primary) inset shows no staining, demonstrating secondary antibody specificity.

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



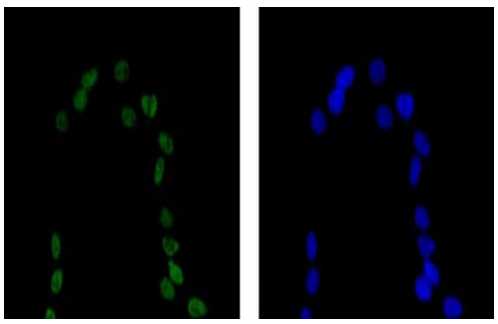
Flow Cytometry (Intracellular) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

Overlay histogram showing U-87 MG (Human glioblastoma-astrocytoma epithelial cell line) cells stained with ab177487 (red line).

The cells were fixed with 80% methanol (5 minutes) and then permeabilized with 0.1% PBS-Tween for 20 minutes. 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 (ab177487, 1/100 dilution) for 30 minutes at 22°C. The secondary antibody used was Alexa Fluor®488 goat anti-rabbit IgG (H&L) (ab150081) at 1/2000 dilution for 30 minutes at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (ab172730, 1µg/1x10^6cells used under the same conditions. Unlabeled sample (blue line) was also used as a control.

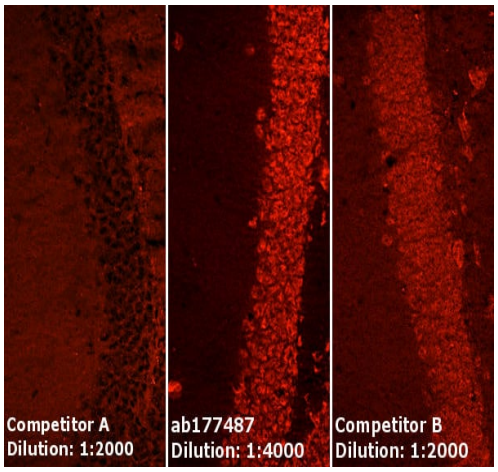
Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

Alexa Fluor® 488 (ab190195) and Alexa Fluor® 647 (ab190565) conjugated versions are available for this clone.



Immunocytochemistry/ Immunofluorescence - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

Immunocytochemsitry/Immunofluorescence analysis of SH-SY5Y (Human neuroblastoma cell line from bone marrow) cells labeling NeuN (green) with ab177487 at 1/300. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/200) was used as the secondary antibody. Counterstained with DAPI (blue).



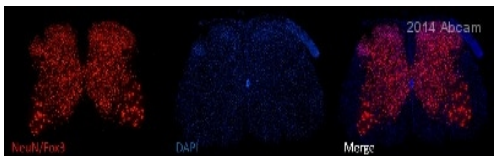
Immunohistochemistry (Frozen sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

An independent comparison of commercially available NeuN clones in IHC-Fr (acetone-fixed mouse dentate gyrus sections).

Competitor A: Leading mouse monoclonal.

Competitor B: Non-Abcam rabbit monoclonal.

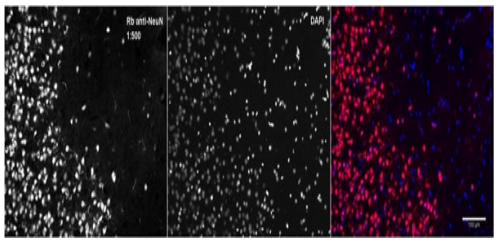
ab177487 produces intense, specific staining with minimal background, even at half the dilution of competing antibodies.



Immunohistochemistry (Frozen sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an Abreview submitted by Jianning Lu

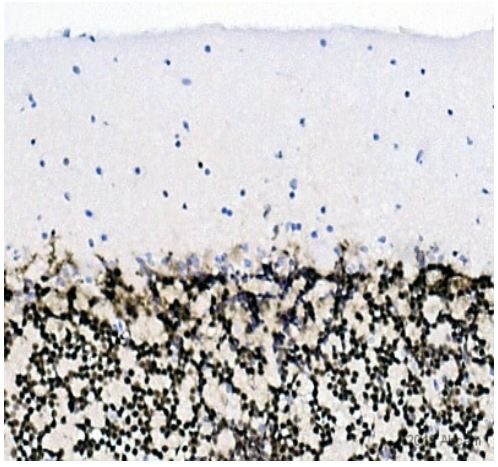
ab177487 staining NeuN in mouse free floating 50 micron lumbar spinal cord tissue sections by Immunohistochemistry (IHC-Fr - frozen sections). Tissue was fixed with formaldehyde, permeabilized with Triton X-100 and blocked with 10% serum for 2 hours at 25°C. Samples were incubated with primary antibody (1/500 in PBS + Triton) for 16 hours at 4°C. An Alexa Fluor® 594-conjugated donkey anti-rabbit IgG polyclonal (1/700) was used as the secondary antibody.



Immunohistochemistry (Frozen sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an Abreview submitted by Eva Borger

ab177487 staining NeuN in mouse brain tissue sections by Immunohistochemistry (IHC-Fr - frozen sections). Tissue was fixed with formaldehyde and blocked with Triton X-100 + 0.4% horse seurm for 30 minutes at 20°C. Samples were incubated with primary antibody (1/500 in blocking solution) for 16 hours at 4°C. An Alexa Fluor® 594-conjugated donkey anti-rabbit IgG polyclonal (1/200) was used as the secondary antibody.

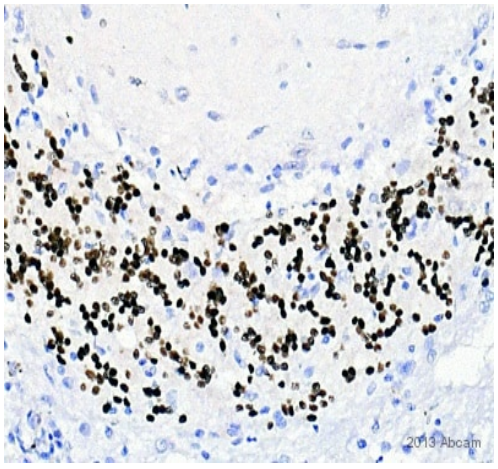


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

IHC-P image of FOX3/NeuN staining on cat cerebellum sections using ab177487 (1/1000).

Sections were de-paraffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 minutes at 21°C. ab177487 was diluted 1/1000 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was goat polyclonal to rabbit IgG conjugated to biotin (1/250).

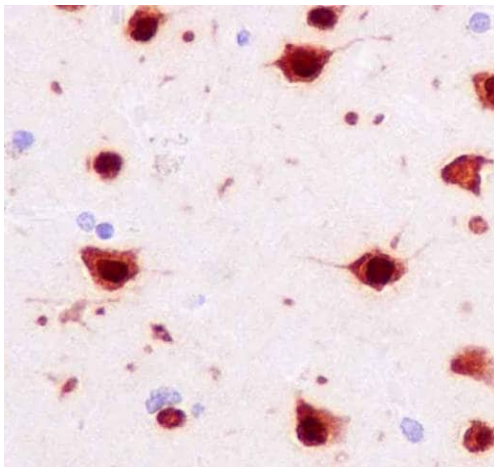


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

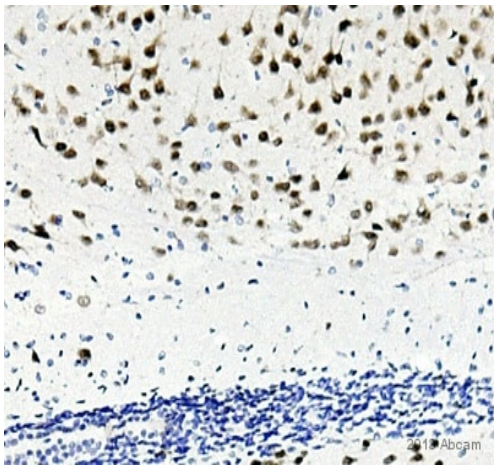
IHC-P image of FOX3/NeuN staining on dog cerebellum sections using ab177487 (1/500).

Sections were de-paraffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 minutes at 21°C. ab177487 was diluted 1/500 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was goat polyclonal to rabbit IgG conjugated to biotin (1/250).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

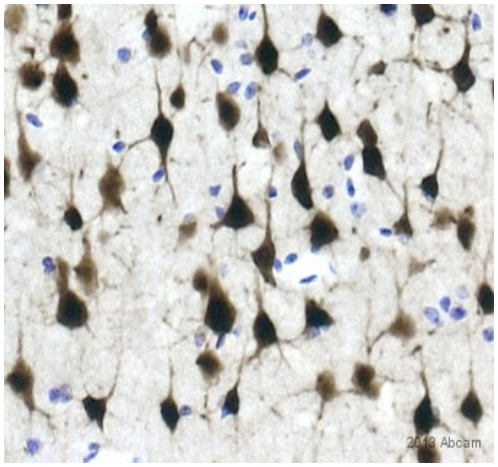
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human gliocytoma tissue labelling NeuN with ab177487 at 1/3000. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. A prediluted HRP-polymer conjugated anti-rabbit IgG was used as the secondary antibody. Counterstained with Hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

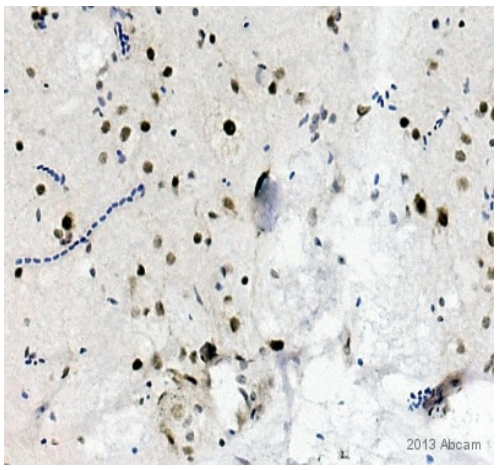
IHC-P image of FOX3/NeuN staining on rat brain (SVZ) sections using ab177487 (1/2000). Sections were de-paraffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 minutes at 21°C. ab177487 was diluted 1/2000 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was goat polyclonal to rabbit IgG conjugated to biotin (1/250).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

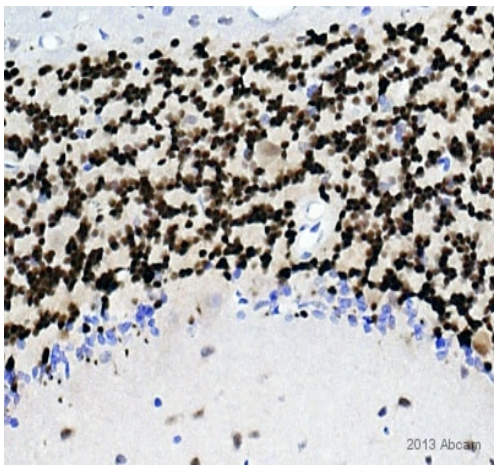
IHC-P image of FOX3/NeuN staining on mouse brain (frontal cortex) sections using ab177487 (1/800). Sections were de-paraffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 minutes at 21°C. ab177487 was diluted 1/800 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was goat polyclonal to rabbit IgG conjugated to biotin (1/250).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

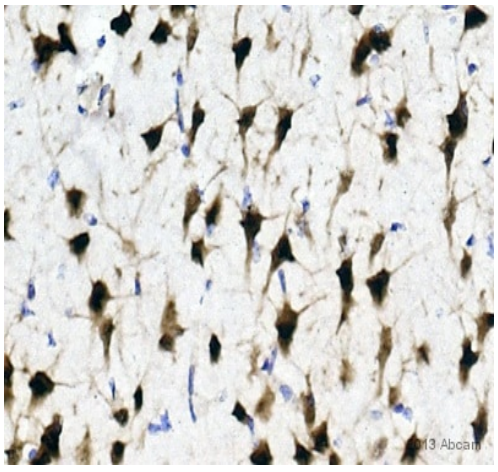
IHC-P image of FOX3/NeuN staining on zebrafish spinal cord sections using ab177487 (1/500). Sections were de-paraffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 minutes at 21°C. ab177487 was diluted 1/500 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was goat polyclonal to rabbit IgG conjugated to biotin (1/250).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

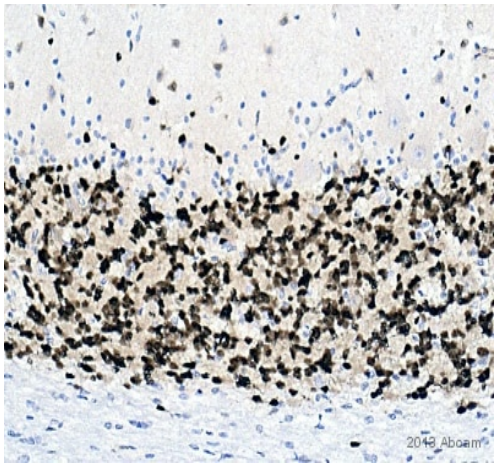
IHC-P image of FOX3/NeuN staining on marmoset cerebellum sections using ab177487 (1/2000). Sections were de-paraffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 minutes at 21°C. ab177487 was diluted 1/2000 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was goat polyclonal to rabbit IgG conjugated to biotin (1/250).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

IHC-P image of FOX3/NeuN staining on sheep brain (Frontal cortex) sections using ab177487 (1/1000). Sections were de-paraffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 minutes at 21°C. ab177487 was diluted 1/1000 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was goat polyclonal to rabbit IgG conjugated to biotin (1/250).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

This image is courtesy of an abreview submitted by Carl Hobbs, Kings's College London, United Kingdom.

IHC-P image of FOX3/NeuN staining on goat cerebellum sections using ab177487 (1/500). Sections were de-paraffinized and subjected to heat mediated antigen retrieval using citric acid. The sections were blocked using 1% BSA for 10 minutes at 21°C. ab177487 was diluted 1/500 and incubated with the sections for 2 hours at 21°C. The secondary antibody used was goat polyclonal to rabbit IgG conjugated to biotin (1/250).

Anti-NeuN antibody [EPR12763] - Neuronal Marker (ab177487)

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