abcam

Product datasheet

Anti-Tau (phospho T231) antibody [EPR2488] ab151559

Recombinant RabMAb

★★★★★ 6 Abreviews 44 References 15 Images

Overview

Product name Anti-Tau (phospho T231) antibody [EPR2488]

Description Rabbit monoclonal [EPR2488] to Tau (phospho T231)

Host species Rahhit

Specificity The specificity of this antibody refers to P10636-8.

Tested applications Suitable for: IHC-Fr, WB, IP, IHC-P Species reactivity Reacts with: Mouse, Rat, Human

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

(Peptide available as ab242015)

Positive control WB: Human hippocampus, C57 and SH-SY5Y treated with sorbitol whole cell lysates; Rat

> cerebral cortex lysate; IHC-P: Human Alzheimer hippocampus tissue; Human, rat and mouse cerebrum tissues; IP: Rat cerebral cortex lysate; IHC-Fr: Mouse cerebrum tissue, Hu Alzheimer

brain.

General notes This is the corresponding antibody for ab242015.

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**.

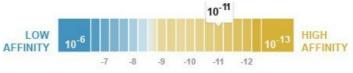
Properties

Form

Storage instructions 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.

 $K_D = 1.26 \times 10^{-11} M$ Dissociation constant (K_D)



Learn more about K_D

Storage buffer Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number EPR2488

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab151559 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
АР		Use at an assay dependent concentration. Antibody concentration range - 4.17, 2.08, 1.04, 0.52, 0.26, 0 nM/mL
IHC-Fr		1/50. Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20)
WB		1/1000 - 1/10000. Predicted molecular weight: 46 kDa.Can be blocked with Human Tau (phospho T231) peptide (ab242015).
IP		1/20. For unpurified use at 1/50 dilution.
IHC-P	★★★★★ (4)	1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. See IHC antigen retrieval protocols. For unpurified use at 1/200 dilution.

Target

Function

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.

Tissue specificity

Expressed in neurons. Isoform PNS-tau is expressed in the peripheral nervous system while the

Involvement in disease

others are expressed in the central nervous system.

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 presentle 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.

Sequence similarities

Developmental stage

Domain

Post-translational modifications

Contains 4 Tau/MAP repeats.

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.

The tau/MAP repeat binds to tubulin. Type I isoforms contain 3 repeats while type II isoforms contain 4 repeats.

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, sevenfold 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.

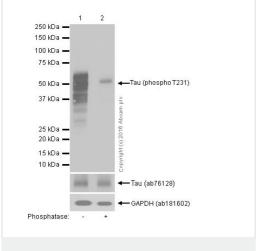
found in the axons of neurons, in the cytosol and in association with plasma membrane

components.

Form There are 9 isoforms produced by alternative splicing.

Images

Cellular localization



Western blot - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

All lanes : Anti-Tau (phospho T231) antibody [EPR2488] (ab151559) at 1/1000 dilution (purified)

Lane 1: Human hippocampus tissue lysate

Lane 2 : Human hippocampus tissue lysate incubated with phosphatase

Lysates/proteins at 15 µg per lane.

Secondary

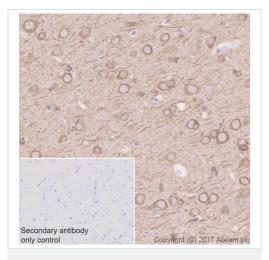
All lanes: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000

dilution

Predicted band size: 46 kDa **Observed band size:** 50-70 kDa

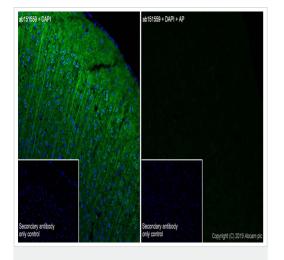
Exposure time: 1 second

Blocking and dilution buffer: 5% NFDM/TBST.



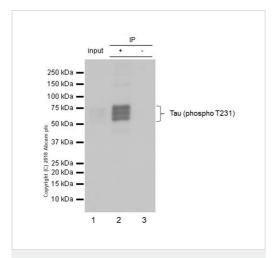
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Rat cerebrum tissue sections labeling Tau with Purified ab151559 at 1:500 dilution (0.24 µg/ml). Heat mediated antigen retrieval was performed using citrate (pH 6.0)



Immunohistochemistry (Frozen sections) - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

Immunohistochemistry (Frozen sections) analysis of mouse cerebrum tissue sections labeling Tau with Purified ab151559 at 1/50 (1.5 μ g/ml).Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20). Goat anti rabbit lgG (Alexa Fluor® 488, <u>ab150077</u>) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. DAPI was used as a counterstain.



Immunoprecipitation - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

ab151559 (purified) at 1:20 dilution (0.6μg) immunoprecipitating Tau in Rat cerebral cortex lysate.

Lane 1 (input): Rat cerebral cortex lysate 10µg

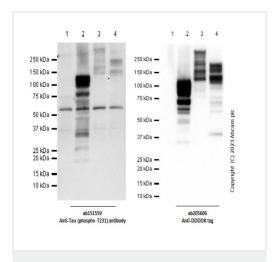
Lane 2 (+): ab151559 & Rat cerebral cortex lysate

Lane 3 (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab151559 inRat cerebral cortex lysate

For western blotting, VeriBlot for IP Detection Reagent (HRP)

(ab131366) was used for detection at 1:1000 dilution.

Blocking and diluting buffer: 5% NFDM/TBST.



Western blot - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

All lanes : Anti-Tau (phospho T231) antibody [EPR2488] (ab151559) at 1/1000 dilution

Lane 1 : 293T cells transfected with an empty vector containing a flag tag whole cell lysate

Lane 2: 293T cells transfected with a human Tau expression vector containing a flag whole cell lysate

Lane 3: 293T cells transfected with a human MAP2 expression vector containing a flag whole cell lysate

Lane 4: 293T cells transfected with a human MAP4 expression vector containing a flag whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

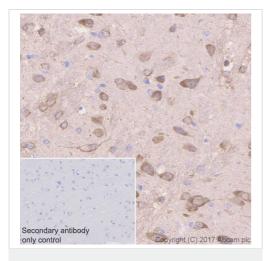
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 46 kDa **Observed band size:** 50-100 kDa

Exposure time: 1 second

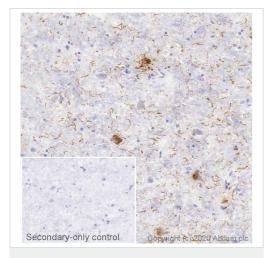
Blocking and dilution buffer: 5% NFDM/TBST.

Western blot for $\underline{ab205606}$ is positioned on the right-hand side and serves as a control for the detection of the flag tag.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

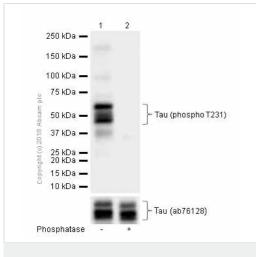
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse cerebrum tissue sections labeling Tau or Primary ab team with Purified ab151559 at 1:500 dilution (0.24 μ g/ml). Heat mediated antigen retrieval was performed using citrate (pH 6.0)



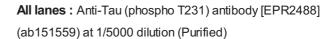
Immunohistochemistry (Frozen sections) - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

IHC image of Tau (phospho T231) staining in a section of frozen normal human Azheimer brain performed on a Leica BONDTM system using the standard protocol. The section was fixed in 10% paraformaldehyde (10 min) prior to staining. The section was incubated with ab151559, 1/1000 dilution, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset secondary-only control image is taken from an identical assay without primary antibody.

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



Western blot - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)



Lane 1: Rat cerebral cortex lysates with 5% NFDM/TBST

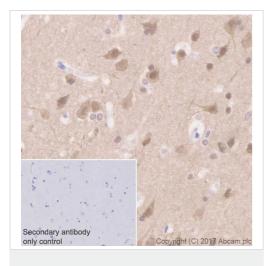
Lane 2: Rat cerebral cortex lysates. Then the membrane was incubated with phosphatase. with 5% NFDM/TBST

Lysates/proteins at 15 µg per lane.

Secondary

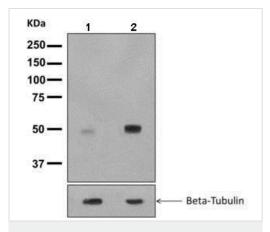
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution (Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated)

Predicted band size: 46 kDa **Observed band size:** 50-70 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human cerebrum tissue sections labeling Tau with Purified ab151559 at 1:500 dilution (0.24 μ g/ml). Heat mediated antigen retrieval was performed using citrate (pH 6.0)



Western blot - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

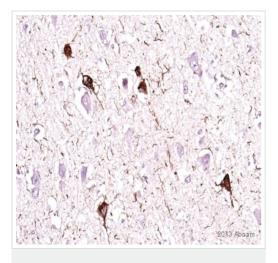
All lanes : Anti-Tau (phospho T231) antibody [EPR2488] (ab151559) at 1/1000 dilution (unpurified)

Lane 1: SH-SY5Y (human neuroblastoma cell line from bone marrow) cell lysate, untreated

Lane 2: SH-SY5Y cell lysate, treated with sorbitol

Lysates/proteins at 10 µg per lane.

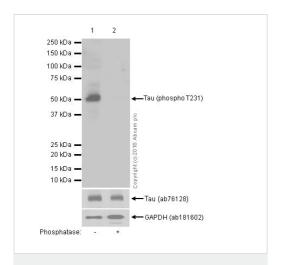
Predicted band size: 46 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

Image courtesy of Carl Hobbs, Kings College London.

IHC image of Tau (phospho T231) staining in human Alzheimer hippocampus formalin fixed paraffin embedded tissue section. The section was pre-treated using heat mediated antigen retrieval with citrate buffer. The section was then incubated with unpurified ab151559 at 1/2000 dilution for 2 hours at 21°C. A biotin conjugated goat-anti-rabbit antibody was used as a secondary at 1/250. The section shows clear neurofibrillary tangles in a subset of neurons.



Western blot - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

All lanes : Anti-cardiac Troponin I antibody (<u>ab1000</u>) at 1/1000 dilution (purified)

Lane 1: C57 (cerebral cortex) whole cell lysate

Lane 2: C57 (cerebral cortex) whole cell lysate incubated with phosphatase

Lysates/proteins at 15 µg per lane.

Secondary

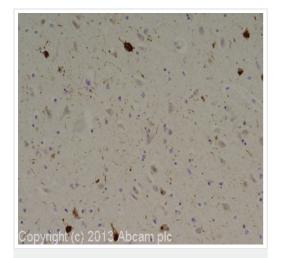
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 46 kDa

Observed band size: 50-70 kDa

Exposure time: 1 second

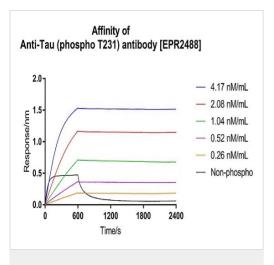
Blocking and dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

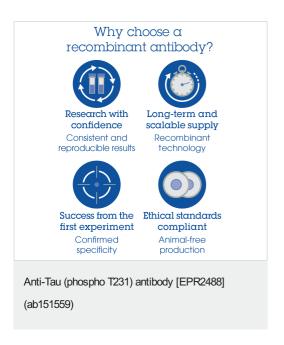
IHC image of Tau (phospho T231) staining in human Alzheimer hippocampus formalin fixed paraffin embedded tissue section, performed on a Leica Bond system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6, epitope retrieval solution 1) for 20 minutes. The section was then incubated with unpurified ab151559, 1/200 dilution, for 15 minutes 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.



Affinity Purification - Anti-Tau (phospho T231) antibody [EPR2488] (ab151559)

Biotinylated Human Tau (pT231) [0.3125 μg/ml] was loaded to SA biosensor on Fortebio RED96e Machine, then associate with recombinant Anti-Tau (phospho T231) antibody [EPR2488] in serial concentration points [4.17, 2.08, 1.04, 0.52, 0.26 nM/mL] by 2-fold dilution, next to dissociate in blank testing buffer [0.1% BSA in PBST (0.05%Tween-20)]. Calculated signals had already subtracted blank control, curve fitting using 1:1 binding model. Non-phospho Tau protein's association and dissociation were also showed in graph. KD(M) value of Anti-Tau (phospho T231) antibody [EPR2488] is 1.26E-11



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