



MONOCLONAL MOUSE ANTI-HUMAN PHF-TAU, CLONE AT8

Presentation

This mouse monoclonal antibody to human PHF-

CODE BR-03

Immunoglobulin class	IgG1 κ
Clone	AT8
Mass/vial	100 μ g
Volume/vial	0.5 ml

tau is supplied in PBS, sterile filtered (0.22 μ m) and without addition of preservatives.

Source

Mouse myeloma SP2/0 cells were fused with spleen cells of a Balb/c mouse immunized intraperitoneally with partially purified human PHF-tau (1, 2). This antibody has been purified from serum-free culture supernatant by protein A affinity chromatography.

Purity

The final product is more than 95% pure as determined by SDS-PAGE.

Applications

This antibody can be used for immunohistochemical staining (3), Western blot and ELISA techniques.

Specificity

This antibody recognizes PHF-tau and does not cross-react with normal tau as determined by a sandwich ELISA. Furthermore, no signal was obtained using alkaline phosphatase-treated PHF-tau as antigen, indicating that this monoclonal is directed against a phosphatase-sensitive epitope (2).

The epitope has been shown to contain the phosphorylated Ser202* residue (4,5).

Instructions for use

1. For immunohistochemistry: use this antibody in a concentration range of 5-10 μ g/ml for the localization of PHF-tau in formalin-fixed, paraffin-embedded brain tissue.
2. For Western blot: a final concentration of 20-60 μ g/ml can detect 50 ng of SDS-denatured and β -mercaptoethanol-PHF-tau.
3. For ELISA: this antibody can be used at a concentration of 5-10 μ g/ml as a capturing reagent for PHF-tau in a sandwich ELISA.

Note: The recommended concentrations are approximate values. For each application, a dose-response assay should be performed to determine the optimal concentration for use.

Storage and stability

Monoclonal mouse anti-human PHF-tau, as shipped, is stable for at least six months when stored at -20°C. Avoid multiple freeze/thaw cycles by storage in appropriate aliquots.

This antibody should be diluted with PBS or medium containing a suitable carrier protein (e.g. 0.1 to 1% BSA). Failure to add carrier protein to diluted product will result in loss of activity.

FOR RESEARCH USE ONLY

BR 3-5

* numbering according to human tau40 (6).

References

- (1) Greenberg SG, Davies P. Proc Natl Acad Sci USA 1990; 87: 5827-31.
- (2) Mercken M, et al. Acta Neuropathol 1991; 84: 265-72.
- (3) Braak E, et al. Acta Neuropathol 1994; 87: 554-67.
- (4) Goedert M, et al. Proc Natl Acad Sci USA 1993; 90: 5066-70.
- (5) Biernat J, et al. EMBO J 1992; 11: 1593-7.
- (6) Goedert M, et al. Neuron 1989; 3: 519-26.

Specific references to applications of AT8 antibody

Western blotting: 1. Biernat J, Mandelkow EM, Schroter C, Lichtenberg-Kraag B, Steiner B, Berling B, Meyer H, Mercken M, Vandermeeren A, Goedert M, et al

The switch of tau protein to an Alzheimer-like state includes the phosphorylation of two serine-proline motifs upstream of the microtubule binding region.

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2. Goedert M, Jakes R, Crowther RA, Six J, Lubke U, Vandermeeren M, Cras P, Trojanowski JQ, Lee VM

The abnormal phosphorylation of tau protein at Ser-202 in Alzheimer disease recapitulates phosphorylation during development.

Proc Natl Acad Sci U S A 1993 Jun 1;90(11):5066-70

Specificity and peptide reactivity:

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Monoclonal antibody AT8 recognises tau protein phosphorylated at both serine 202 and threonine 205.

Neurosci Lett 1995 Apr 21;189(3):167-9

2. DeLeys R, Hendrickx G, Dekeyser F, Demol H, Raymackers J, Brasseur R, Borremans F, Vanmechelen E, Van de Voorde A

Mapping and sequence requirements of the phosphorylation-sensitive epitopes recognized by the monoclonal antibodies Tau1, BT2, and AT8.

Schneider CH 239-244. 1996. Chichester, New York, John Wiley & Sons, Ltd.
Peptides in Immunology

Neuropathology:

1. Braak E, Braak H, Mandelkow EM

A sequence of cytoskeleton changes related to the formation of neurofibrillary tangles and neuropil threads.

Acta Neuropathol (Berl) 1994;87(6):554-67

2. Spillantini MG, Goedert M, Crowther RA, Murrell JR, Farlow MR, Ghetti B

Familial multiple system tauopathy with presenile dementia: a disease with abundant neuronal and glial tau filaments.

Proc Natl Acad Sci U S A 1997 Apr 15;94(8):4113-8

Use in modelsystems:

1. Gotz J, Probst A, Spillantini MG, Schafer T, Jakes R, Burki K, Goedert M
Somatodendritic localization and hyperphosphorylation of tau protein in transgenic mice expressing the longest human brain tau isoform.

EMBO J 1995 Apr 3;14(7):1304-13

2. Sturchler-Pierrat C, Abramowski D, Duke M, Wiederhold KH, Mistl C, Rothacher S, Ledermann B, Burki K, Frey P, Paganetti PA, Waridel C, Calhoun ME, Jucker M, Probst A, Staufenbiel M, Sommer B

Two amyloid precursor protein transgenic mouse models with Alzheimer disease-like pathology.

Proc Natl Acad Sci U S A 1997 Nov 25;94(24):13287-92

ELISA

1. Vandermeeren M, Mercken M, Vanmechelen E, Six J, van de Voorde A, Martin JJ, Cras P

Dear Hiromi Shiokawa,

The antigen used for immunization is PHF-tau isolated from an Alzheimer brain

(Mercken et al, 1991; see datasheet). Upon dephosphorylation AT8 immunoreactivity is abolished and using mutated recombinant in vitro phosphorylated tau protein the site of AT8 has been shown to contain phosphorylated serine 202 and threonine 205 (Goedert et al, 1995; see new datasheet).

The antigen is PHF-tau and is full size.

You will also find a draft of an update of the datasheet.

Kind regards,

Vanmechelen Eugene

Innogenetics N.V.