

Beta III Tubulin Recombinant Antibody [PSH15-32] - Rat IgG1 (Chimeric)

HA601655



Product Type:	Recombinant Chimeric Antibody, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	IHC-P, IF-Tissue, IHC-Fr, WB, IF-Cell
Molecular Wt:	Predicted band size: 50 kDa
Clone number:	PSH15-32

Description: Tubulin beta-3 chain, Class III β -tubulin, β III-tubulin (β 3-tubulin) or β -tubulin III, is a microtubule element of the tubulin family found almost exclusively in neurons, and in testis cells. In humans, it is encoded by the TUBB3 gene. Forebrain neuronal culture after 40 days of differentiation from induced human pluripotent stem cells. Class III β -tubulin is one of the seven β -tubulin isotypes identified in the human genome, predominantly in neurons and the testis. It is conditionally expressed in a number of other tissues after exposure to a toxic microenvironment featured by hypoxia and poor nutrient supply. Posttranslational changes including phosphorylation and glycosylation are required for functional activity. Class III β -tubulin's role in neural development has warranted its use as an early biomarker of neural cell differentiation from multi potent progenitors. TUBB3 inactivation impairs neural progenitor proliferation. Rescue experiments demonstrate the non-interchangeability of TUBB3 with other classes of β -tubulins which cannot restore the phenotype resulting from TUBB3 inactivation. Congenital neurologic syndromes associated with TUBB3 missense mutations demonstrate the critical importance of class III β -tubulin for normal neural development.

Immunogen:	Synthetic peptide within mouse Beta III Tubulin aa 401-450.
Positive control:	Neuro-2a, Human Cerebellum, Mouse Cerebellum, Rat Cerebellum.
Subcellular location:	Cytoplasm, cytoskeleton, growth cone, lamellipodium, filopodium.
Database links:	SwissProt: Q13509 Human Q9ERD7 Mouse Q4QRB4 Rat

Recommended Dilutions:

WB	1:5,000
IF-Cell	1:50
IHC-P	1:2,000
IF-Tissue	1:1,000
IHC-Fr	1:200-1:500

Storage Buffer:	1*PBS (pH7.4), 0.1% BSA, 40% Glycerol, 0.2% Proclean 950.
Storage Instruction:	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Store at -20°C long term.
Purity:	Protein A affinity purified.

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

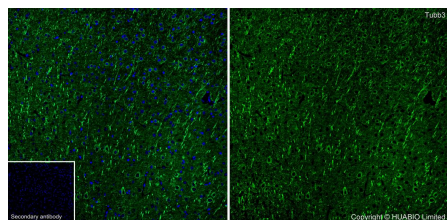
Images

Fig1: Application: Immunofluorescence (IHC-Fr)

Species: Mouse

Tissue: Cerebral cortex

Sample: Frozen section



Antigen retrieval: Recommend. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven.

Wash buffer: 1× TBST

Blocking: 10% normal goat serum + 1% Triton X-100 + 0.3 M Glycine in TBST, 30 minutes at room temperature.

Primary antibody: HA601655, 1/250, overnight at 4℃.

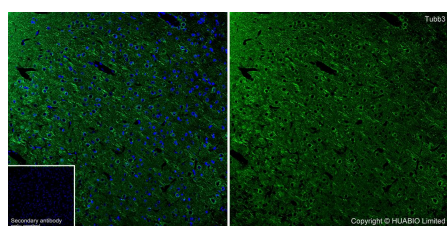
Secondary antibody: Goat Anti-Rat IgG (iFluor™ 488, HA1133), 1.5 hours at room temperature.

Fig2: Application: Immunofluorescence (IHC-Fr)

Species: Rat

Tissue: Cerebral cortex

Sample: Frozen section



Antigen retrieval: Recommend. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven.

Wash buffer: 1× TBST

Blocking: 10% normal goat serum + 1% Triton X-100 + 0.3 M Glycine in TBST, 30 minutes at room temperature.

Primary antibody: HA601655, 1/250, overnight at 4℃.

Secondary antibody: Goat Anti-Rat IgG (iFluor™ 488, HA1133), 1.5 hours at room temperature.

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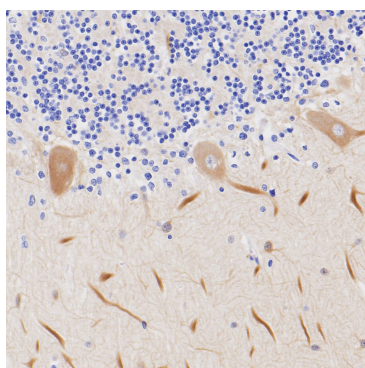


Fig3: Application: Immunohistochemistry (IHC-P)

Species: Human

Tissue: Cerebellum

Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST

Endogenous peroxidase blocking: 3% H₂O₂, 10 minutes at room temperature.

Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.

Primary antibody: HA601655, 1/2,000, 1 hour at room temperature.

Secondary antibody: HA1023, 30 minutes at room temperature.

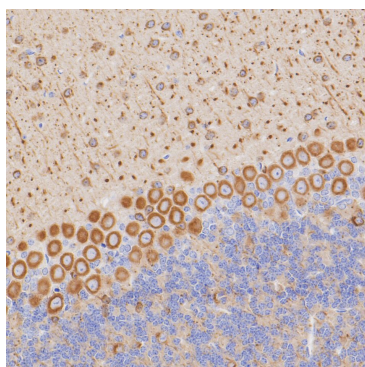


Fig4: Application: Immunohistochemistry (IHC-P)

Species: Mouse

Tissue: Cerebellum

Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST

Endogenous peroxidase blocking: 3% H₂O₂, 10 minutes at room temperature.

Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.

Primary antibody: HA601655, 1/2,000, 1 hour at room temperature.

Secondary antibody: HA1023, 30 minutes at room temperature.

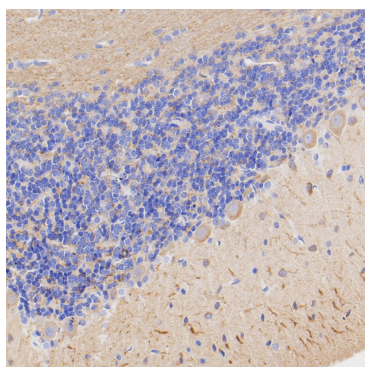


Fig5: Application: Immunohistochemistry (IHC-P)

Species: Rat

Tissue: Cerebellum

Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST

Endogenous peroxidase blocking: 3% H₂O₂, 10 minutes at room temperature.

Blocking: 1% BSA + 10% normal goat serum, 10 minutes at room temperature.

Primary antibody: HA601655, 1/2,000, 1 hour at room temperature.

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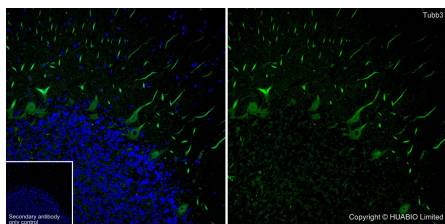


Fig6: Application: Immunofluorescence (IF-tissue)

Species: Human

Tissue: Cerebellum

Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST

Blocking: 10% normal goat serum + 1% Triton X-100 + 0.3 M Glycine in TBST, 30 minutes at room temperature.

Primary antibody: HA601655, 1/1,000, overnight at 4°C.

Secondary antibody: Goat Anti-Rat IgG (iFluor™ 488, HA1133), 1.5 hours at room temperature.

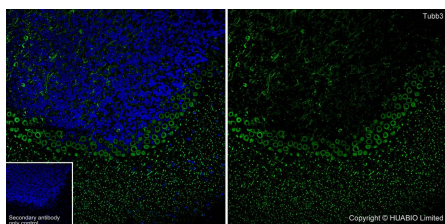


Fig7: Application: Immunofluorescence (IF-tissue)

Species: Mouse

Tissue: Cerebellum

Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST

Blocking: 10% normal goat serum + 1% Triton X-100 + 0.3 M Glycine in TBST, 30 minutes at room temperature.

Primary antibody: HA601655, 1/1,000, overnight at 4°C.

Secondary antibody: Goat Anti-Rat IgG (iFluor™ 488, HA1133), 1.5 hours at room temperature.

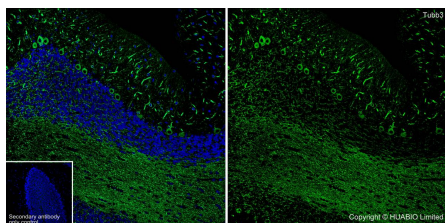


Fig8: Application: Immunofluorescence (IF-tissue)

Species: Rat

Tissue: Cerebellum

Sample: Paraffin-embedded section

Antigen retrieval: Heat-mediated, Tris-EDTA buffer (pH 9.0), 20 minutes at 95°C.

Wash buffer: 1× TBST

Blocking: 10% normal goat serum + 1% Triton X-100 + 0.3 M Glycine in TBST, 30 minutes at room temperature.

Primary antibody: HA601655, 1/1,000, overnight at 4°C.

Secondary antibody: Goat Anti-Rat IgG (iFluor™ 488, HA1133), 1.5 hours at room temperature.

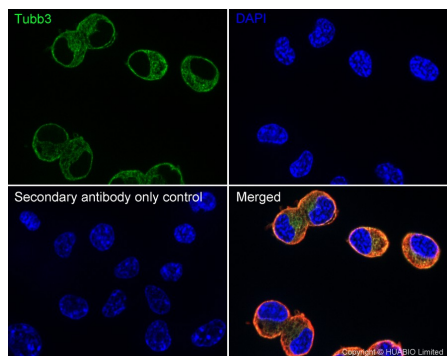


Fig9: Application: Immunocytochemistry (IF-cell)

Species: Mouse

Sample: Neuro-2a (Mouse brain neuroblastoma cell)

Fixation: 4% Paraformaldehyde, 15 minutes at room temperature.

Permeabilization: 0.1% Triton X-100, 15 minutes at room temperature.

Blocking: 1% BSA + 10% normal goat serum, 1 hour at room temperature.

Antibody dilution buffer: 1% BSA in PBST.

Primary antibody: HA601655, 1/50, overnight at 4℃.

Secondary antibody: Goat Anti-Rat IgG (iFluor™ 488, HA1133), 45 minutes at room temperature.

Counterstain: Beta tubulin (ET1602-4, Red), 1/500, overnight at 4℃. The nuclear counterstain was DAPI (Blue).

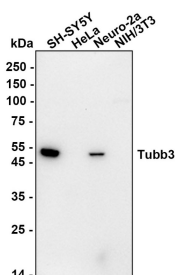
Fig10: Western blot analysis of Beta III Tubulin on different lysates with Rat anti-Beta III Tubulin antibody (HA601655) at 1/10,000 dilution.

Lane 1: SH-SY5Y (Human neuroblastoma cell)

Lane 2: HeLa (Human cervical adenocarcinoma cell)(Low expression)

Lane 3: Neuro-2a (Mouse brain neuroblastoma cell)

Lane 4: NIH/3T3 (Mouse fibroblast)(negative)



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Lysates/proteins at 10 µg/Lane.

Exposure time: 2 minutes; ECL: K1801

HeLa is a relative low expression control (PMID: 35573698)

and NIH/3T3 is a negative control (PMID: 25839941).

Blocking: 5% NFDM/TBST, 1 hour at room temperature

Primary antibody: HA601655, 1/10,000 in primary antibody dilution buffer (K1803), overnight at 4℃

Secondary antibody: Goat anti-Rat IgG-HRP (HA1023), 1/50,000 in 5% NFDM/TBST, 1 hour at room temperature

Predicted band size: 50 kDa

Observed band size: 50 kDa

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Background References

1. Puri D et al. TUBB3 and KIF21A in neurodevelopment and disease. Front Neurosci. 2023 Aug
2. Jin S et al. TUBB3 M323V Syndrome Presents with Infantile Nystagmus. Genes (Basel). 2021 Apr

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