Anti-FUS/TLS Antibody [JJ09-31] - BSA and Azide free

HA750326

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Cynomolgus monkey, Pig
Applications:	WB, IHC-P, FC, IF-Cell
Molecular Wt:	Predicted band size: 53 kDa
Clone number:	JJ09-31

Description: EWS and FUS/TLS are nuclear RNA-binding proteins. As a result of chromosome translocation, the EWS gene is fused to a variety of transcription factors, including ATF-1, in human neoplasias. In the Ewing family of tumors, the N-terminal domain of EWS is fused to the DNA-binding domain of various Ets transcription factors, including Fli-1, ETV1 and FEV. The EWS/Fli-1 chimeric protein acts as a more potent transcriptional activator than Fli-1 and can promote cell transformation. In human myxoid liposarcomas and myeloid leukemias, chromosomal translocation results in the fusion of the N-terminal region of FUS/TLS with the open reading frame of CHOP. In normal cells, FUS/TLS binds to the DNA-binding domains of nuclear steroid receptors and is also present in subpopulations of TFIID complexes, indicating a potential role for FUS/TLS in the processing of primary transcripts that are generated in response to hormone-induced transcription.

Immunogen: Synthetic peptide within Human aa 1-46 / 526.

Positive control: K562 cell lysates, HepG2, NIH/3T3, human tonsil tissue, human colon carcinoma tissue, mouse brain tissue.

Subcellular location: Nucleus.

Database links: SwissProt: P35637 Human | P56959 Mouse

Recommended Dilutions:

WB	1:1,000-1:5,000
IHC-P	1:50-1:200
FC	1:1,000
IF-Cell	1:100

Storage Buffer: PBS (pH7.4).

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

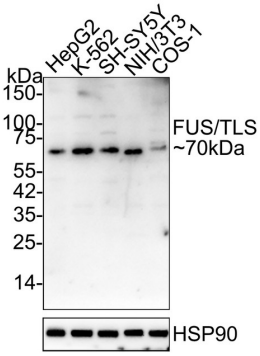

Fig1: Western blot analysis of FUS/TLS on different lysates with Rabbit anti-FUS/TLS antibody (HA750326) at 1/1,000 dilution.

Lane 1: HepG2 cell lysate
Lane 2: K-562 cell lysate
Lane 3: SH-SY5Y cell lysate
Lane 4: NIH/3T3 cell lysate
Lane 5: COS-1 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 50 kDa
Observed band size: 70 kDa

Exposure time: 1 minutes 20 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750326) at 1/5,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

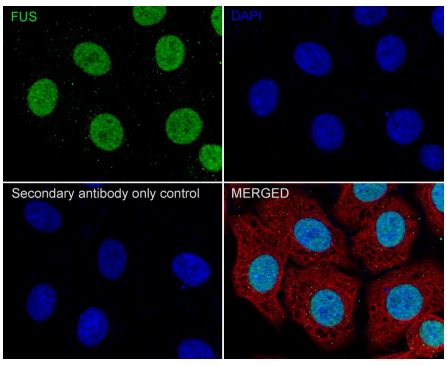

Fig2: Immunocytochemistry analysis of HepG2 cells labeling FUS/TLS with Rabbit anti-FUS/TLS antibody (HA750326) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-FUS/TLS antibody (HA750326) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

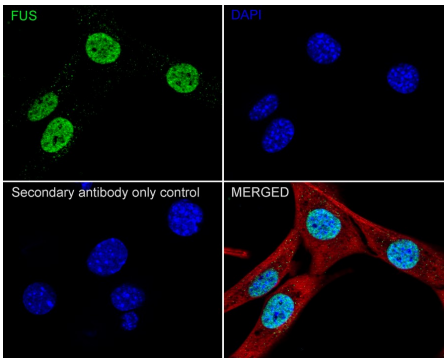

Fig3: Immunocytochemistry analysis of NIH/3T3 cells labeling FUS/TLS with Rabbit anti-FUS/TLS antibody (HA750326) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-FUS/TLS antibody (HA750326) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

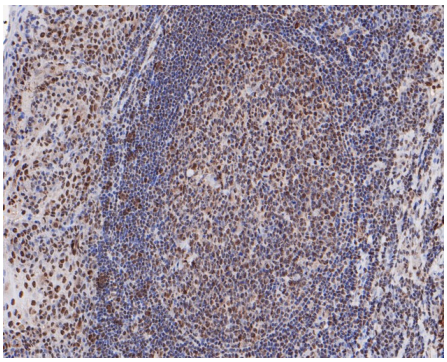

Fig4: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-FUS/TLS antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750326, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

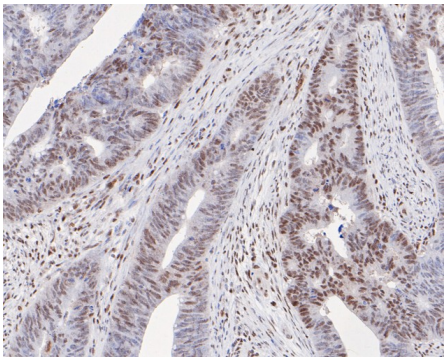

Fig5: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue using anti-FUS/TLS antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750326, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

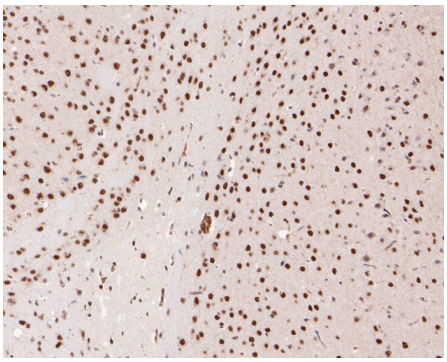

Fig6: Immunohistochemical analysis of paraffin-embedded mouse brain tissue using anti-FUS/TLS antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750326, 1/200) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

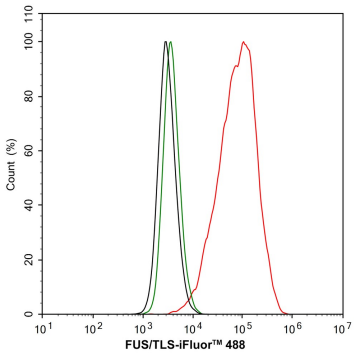

Fig7: Flow cytometric analysis of HepG2 cells labeling FUS/TLS.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750326, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

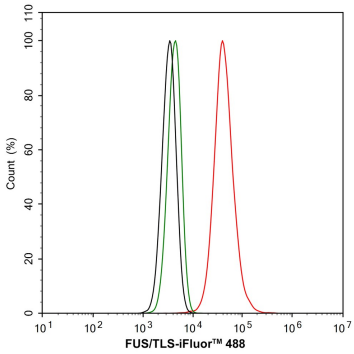

Fig8: Flow cytometric analysis of NIH/3T3 cells labeling FUS/TLS.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750326, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Joardar A et al. PPAR gamma activation is neuroprotective in a Drosophila model of ALS based on TDP-43. Hum Mol Genet 24:1741-54 (2015).
2. Shen W et al. 2'-Fluoro-modified phosphorothioate oligonucleotide can cause rapid degradation of P54nrb and PSF. Nucleic Acids Res 43:4569-78 (2015).