Anti-Fibrillarin Antibody [JE04-54]

HA722842

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, FC, IP
Molecular Wt:	Predicted band size: 34 kDa
Clone number:	JE04-54

Description: rRNA 2'-O-methyltransferase fibrillarin is an enzyme that in humans is encoded by the FBL gene. This gene product is a component of a nucleolar small nuclear ribonucleoprotein (snRNP) particle thought to participate in the first step in processing pre-ribosomal (r)RNA. It is associated with the U3, U8, and U13 small nucleolar RNAs and is located in the dense fibrillar component (DFC) of the nucleolus. The encoded protein contains an N-terminal repetitive domain that is rich in glycine and arginine residues, like fibrillarins in other species. Its central region resembles an RNA-binding domain and contains an RNP consensus sequence. Antisera from approximately 8% of humans with the autoimmune disease scleroderma recognize fibrillarin.

Immunogen: Synthetic peptide within human Fibrillarin aa 272-321 / 321.

Positive control: HeLa cell lysate, HepG2 cell lysate, O-2 OS cell lysate, NIH/3T3 cell lysate, Neuro-2a cell lysate, C6 cell lysate, PC-12 cell lysate, HeLa, NIH/3T3, PC-12.

Subcellular location: Nucleus, nucleolus, Nucleus, nucleoplasm.

Database links: SwissProt: P22087 Human | P35550 Mouse | P22509 Rat

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:1,000
FC	1:1,000
IP	1-2μg/sample

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

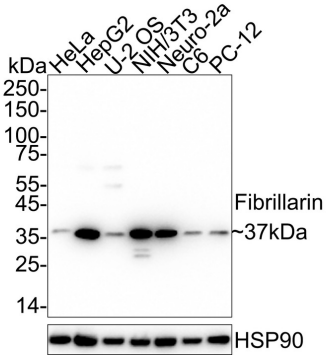

Fig1: Western blot analysis of Fibrillarin on different lysates with Rabbit anti-Fibrillarin antibody (HA722842) at 1/1,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HepG2 cell lysate
Lane 3: O-2 OS cell lysate
Lane 4: NIH/3T3 cell lysate
Lane 5: Neuro-2a cell lysate
Lane 6: C6 cell lysate
Lane 7: PC-12 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 34 kDa
Observed band size: 37 kDa

Exposure time: 20 seconds; ECL: K1801;

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 (HA722842) at 1/1,000 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.

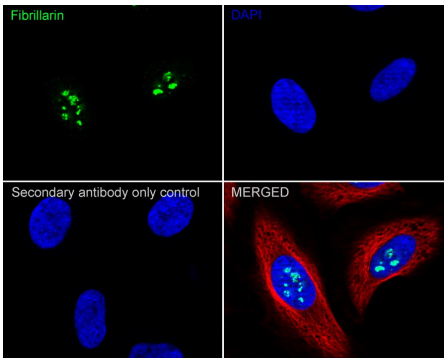

Fig2: Immunocytochemistry analysis of HeLa cells labeling Fibrillarin with Rabbit anti-Fibrillarin antibody (HA722842) at 1/1,000 dilution.

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-Fibrillarin antibody (HA722842) at 1/1,000 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.

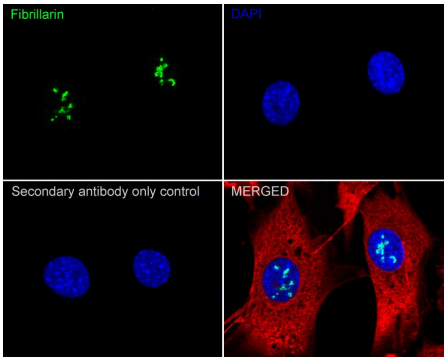

Fig3: Immunocytochemistry analysis of NIH/3T3 cells labeling Fibrillarin with Rabbit anti-Fibrillarin antibody (HA722842) at 1/1,000 dilution.

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-Fibrillarin antibody (HA722842) at 1/1,000 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.

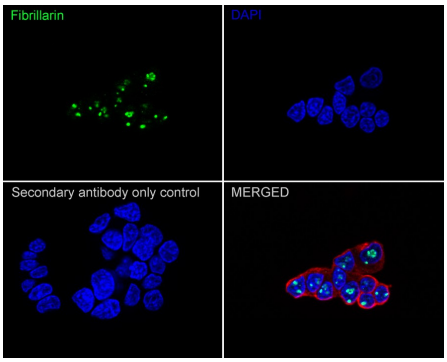

Fig4: Immunocytochemistry analysis of PC-12 cells labeling Fibrillarin with Rabbit anti-Fibrillarin antibody (HA722842) at 1/1,000 dilution.

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-Fibrillarin antibody (HA722842) at 1/1,000 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.

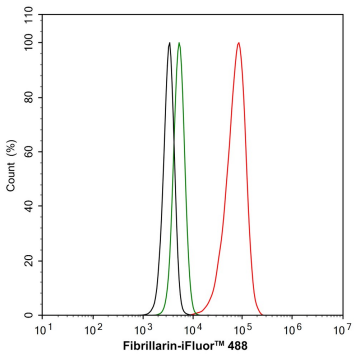

Fig5: Flow cytometric analysis of HeLa cells labeling Fibrillarin.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA722842, 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).

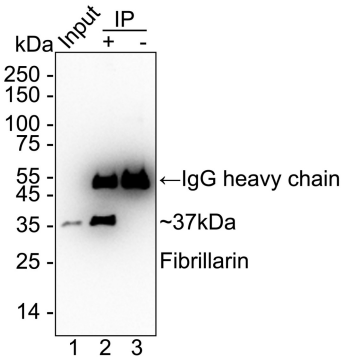

Fig6: Fibrillarin was immunoprecipitated from 0.2 mg HepG2 cell lysate with HA722842 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using HA722842 at 1/1,000 dilution. Mouse anti Rabbit IgG heavy chain (Fc) secondary antibody (M1003-7) at 1/100,000 dilution was used for 1 hour at room temperature.

Lane 1: HepG2 cell lysate (input)
Lane 2: HA722842 IP in HepG2 cell lysate
Lane 3: Rabbit IgG instead of HA722842 in HepG2 cell lysate

Blocking/Dilution buffer: 5% NFDM/TBST
Exposure time: 20 seconds; ECL: K1801

Background References

1. Decle-Carrasco S et al. Plant viral proteins and fibrillarin: the link to complete the infective cycle. Mol Biol Rep. 2021 May
2. Wang T et al. Fibrillarin-GFP Facilitates the Identification of Meiotic Competent Oocytes. Front Cell Dev Biol. 2021 Apr