Anti-COL1A1 Antibody [PSH06-20]

HA722517

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IF-Tissue, IF-Cell, FC, IP, mIHC, IHC-Fr
Molecular Wt:	Predicted band size: 139 kDa
Clone number:	PSH06-20

Description: This gene encodes the pro-alpha1 chains of type I collagen whose triple helix comprises two alpha1 chains and one alpha2 chain. Type I is a fibril-forming collagen found in most connective tissues and is abundant in bone, cornea, dermis and tendon. Mutations in this gene are associated with osteogenesis imperfecta types I-IV, Ehlers-Danlos syndrome type VIIA, Ehlers-Danlos syndrome Classical type, Caffey Disease and idiopathic osteoporosis. Reciprocal translocations between chromosomes 17 and 22, where this gene and the gene for platelet-derived growth factor beta are located, are associated with a particular type of skin tumor called dermatofibrosarcoma protuberans, resulting from unregulated expression of the growth factor. Two transcripts, resulting from the use of alternate polyadenylation signals, have been identified for this gene.

Immunogen: Synthetic peptide within Human COL1A1 aa 1197-1208.

Positive control: HFF-1 cell lysate, NIH/3T3 cell lysate, Human lung tissue lysate, Mouse skin tissue lysate, Rat skin tissue lysate, HFF-1, human colon cancer tissue, human kidney tissue, human lung tissue, mouse kidney tissue, mouse lung tissue, rat kidney tissue, rat lung tissue.

Subcellular location: Secreted.

Database links: SwissProt: P02452 Human | P11087 Mouse | P02454 Rat

Recommended Dilutions:

WB	1:1,000
IHC-P	1:1,000
IF-Tissue	1:200
IF-Cell	1:500
FC	1:1,000
IP	1-2μg/sample
mIHC	1:1,000
IHC-Fr	1:1,000

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

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

Purity: Protein A affinity purified.

Images

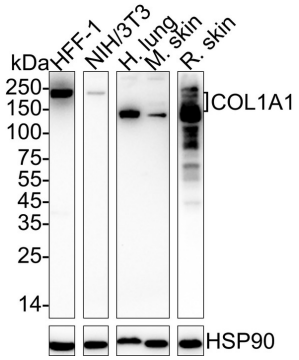

Fig1: Western blot analysis of COL1A1 on different lysates with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

Lane 1: HFF-1 cell lysate (20 µg/Lane)
Lane 2: NIH/3T3 cell lysate (20 µg/Lane)
Lane 3: Human lung tissue lysate (40 µg/Lane)
Lane 4: Mouse skin tissue lysate (40 µg/Lane)
Lane 5: Rat skin tissue lysate (40 µg/Lane)

Predicted band size: 139 kDa
Observed band size: 200/139 kDa

Exposure time: Lane 1-4: 1 minute; Lane 5: 3 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 (HA722517) 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.

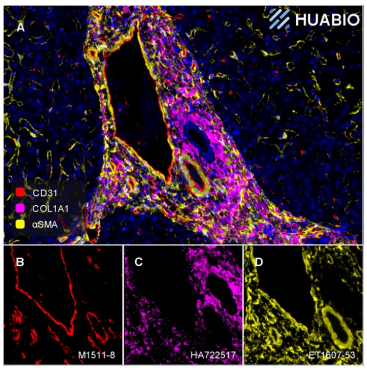

Fig2: Fluorescence multiplex immunohistochemical analysis of human liver (Formalin/PFA-fixed paraffin-embedded sections). Panel A: the merged image of anti-CD31(M1511-8, Red), anti-COL1A1(HA722517, Magenta) and anti-αSMA (ET1607-53, Yellow) on human liver. HRP Conjugated UltraPolymer Goat Polyclonal Antibody HA1119/HA1120 was used as a secondary antibody. The immunostaining was performed with the Sequential Immuno-staining Kit (IRISKit™MH010101, www.luminiris.cn). The section was incubated in three rounds of staining: in the order of M1511-8 (1/1000 dilution), HA722517 (1/10000 dilution) and ET1607-53 (1/5000 dilution) for 20 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 30 mins at 95C. DAPI (blue) was used as a nuclear counter stain. Image acquisition was performed with Olympus VS200 Slide Scanner.

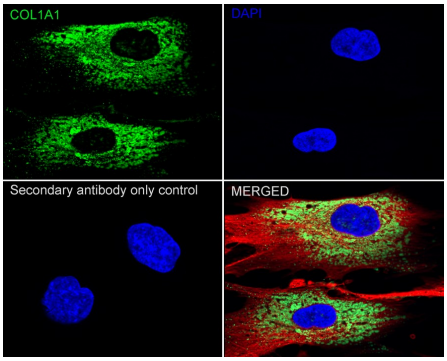

Fig3: Immunocytochemistry analysis of HFF-1 cells labeling COL1A1 with Rabbit anti-COL1A1 antibody (HA722517) at 1/500 dilution.

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-COL1A1 antibody (HA722517) at 1/500 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.

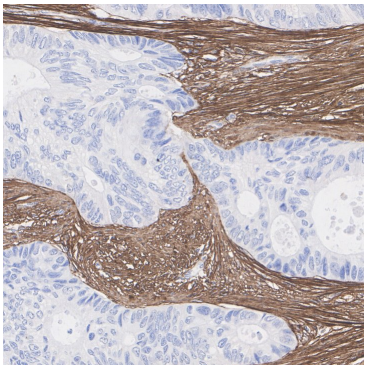

Fig4: Immunohistochemical analysis of paraffin-embedded human colon cancer tissue with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722517) at 1/1,000 dilution for 1 hour 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.

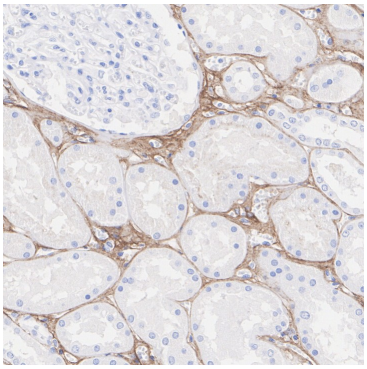

Fig5: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722517) at 1/1,000 dilution for 1 hour 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.

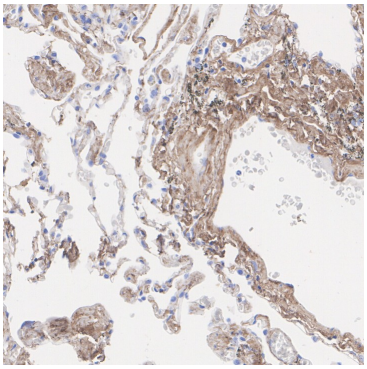

Fig6: Immunohistochemical analysis of paraffin-embedded human lung tissue with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722517) at 1/1,000 dilution for 1 hour 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.

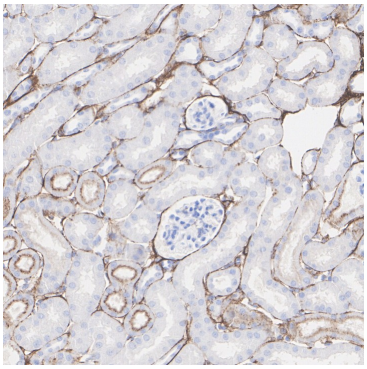

Fig7: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722517) at 1/1,000 dilution for 1 hour 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.

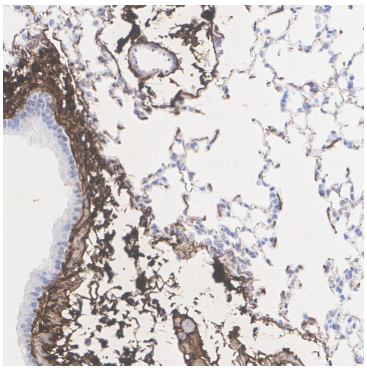

Fig8: Immunohistochemical analysis of paraffin-embedded mouse lung tissue with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722517) at 1/1,000 dilution for 1 hour 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.

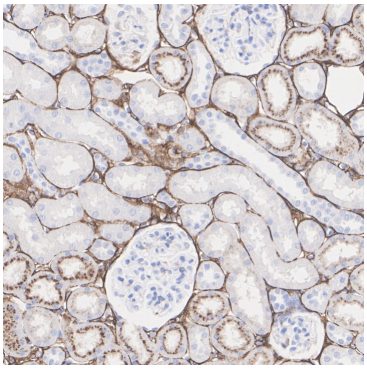

Fig9: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722517) at 1/1,000 dilution for 1 hour 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.

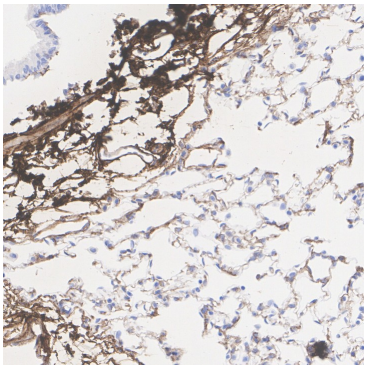

Fig10: Immunohistochemical analysis of paraffin-embedded rat lung tissue with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA722517) at 1/1,000 dilution for 1 hour 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.

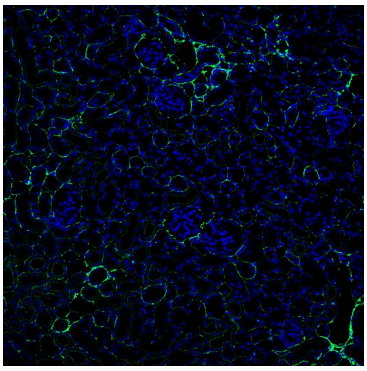

Fig11: Immunofluorescence analysis of paraffin-embedded mouse kidney tissue labeling COL1A1 with Rabbit anti-COL1A1 antibody (HA722517) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA722517, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

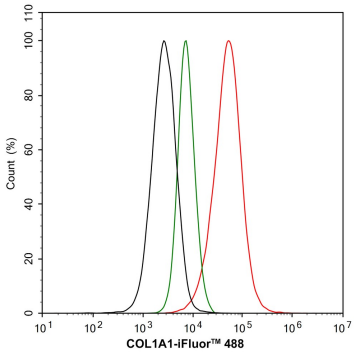

Fig12: Flow cytometric analysis of HFF-1 cells labeling COL1A1.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA722517, 1μg/mL) (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).

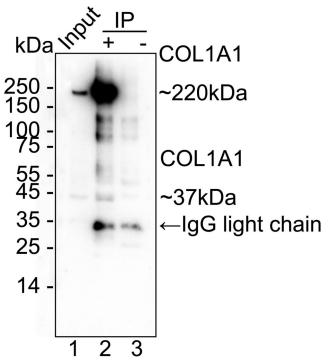

Fig13: COL1A1 was immunoprecipitated from 0.2 mg HFF-1 cell lysate with HA722517 at 2 µg/25 µl agarose. Western blot was performed from the immunoprecipitate using HA722517 at 1/1,000 dilution. Anti-Rabbit IgG for IP Nano-secondary antibody (NBI01H) at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: HFF-1 cell lysate (input)
Lane 2: HA722517 IP in HFF-1 cell lysate
Lane 3: Rabbit IgG instead of HA722517 in HFF-1 cell lysate

Blocking/Dilution buffer: 5% NFDM/TBST
Exposure time: 8 seconds; ECL: K1802

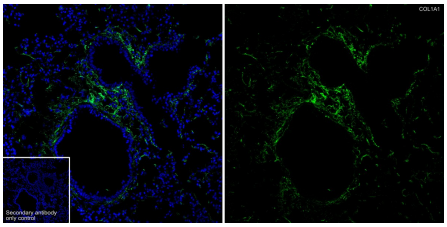

Fig14: Immunofluorescence analysis of frozen mouse lung tissue with Rabbit anti-COL1A1 antibody (HA722517) at 1/1,000 dilution.

The section was not undergone antigen retrieval. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA722517, green) at 1/1,000 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

Background References

1. Dang PN et al. Controlled Dual Growth Factor Delivery From Microparticles Incorporated Within Human Bone Marrow-Derived Mesenchymal Stem Cell Aggregates for Enhanced Bone Tissue Engineering via Endochondral Ossification. Stem Cells Transl Med 5:206-17 (2016).
2. Huang W et al. Astragalus and Paeoniae radix rubra extract inhibits liver fibrosis by modulating the transforming growth factor- /Smad pathway in rats. Mol Med Rep 11:805-14 (2015).