

Anti-MMP-2 Antibody [PSH11-26]

HA723308



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 74 kDa
Clone number:	PSH11-26

Description: Proteins of the matrix metalloproteinase (MMP) family are involved in the breakdown of extracellular matrix (ECM) in normal physiological processes, such as embryonic development, reproduction, and tissue remodeling, as well as in disease processes, such as arthritis and metastasis. Most MMP's are secreted as inactive proproteins which are activated when cleaved by extracellular proteinases. This gene encodes an enzyme which degrades type IV collagen, the major structural component of basement membranes. The enzyme plays a role in endometrial menstrual breakdown, regulation of vascularization and the inflammatory response. Activation of MMP-2 requires proteolytic processing. A complex of membrane type 1 MMP (MT1-MMP/MMP14) and tissue inhibitor of metalloproteinase 2 recruits pro-MMP 2 from the extracellular milieu to the cell surface. Activation then requires an active molecule of MT1-MMP and auto catalytic cleavage. Clustering of integrin chains promotes activation of MMP-2. Another factor that will support the activation of MMP-2 is cell-cell clustering. A wild-type activated leukocyte cell adhesion molecule (ALCAM) is also required to activate MMP-2.

Immunogen: Recombinant protein within human MMP-2 aa 1-660.

Positive control: U-87 MG cell lysate, NIH/3T3 cell lysate, L6 cell lysate, Mouse skin tissue lysate, Rat skin tissue lysate, U-87 MG, NIH/3T3, L6, human colon tissue, human placenta tissue, human skin tissue, mouse colon tissue, mouse skin tissue, rat colon tissue, rat placenta tissue, rat skin tissue.

Subcellular location: Secreted, extracellular space, extracellular matrix Membrane, Nucleus; Cytoplasm, Mitochondrion.

Database links: SwissProt: P08253 Human | P33434 Mouse | P33436 Rat

Recommended Dilutions:

WB	1:2,000
IF-Cell	1:100
IHC-P	1:1,000
FC	1:1,000
IP	1-2µg/sample

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

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

Purity: Protein A affinity purified.

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Technical:0086-571-89986345

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Images

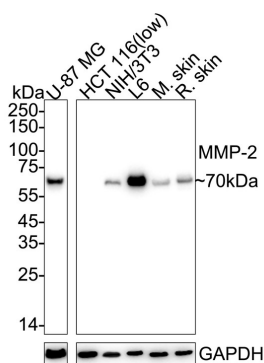


Fig1: Western blot analysis of MMP-2 on different lysates with Rabbit anti-MMP-2 antibody (HA723308) at 1/2,000 dilution.

Lane 1: U-87 MG cell lysate (15 µg/Lane)
 Lane 2: HCT 116 cell lysate (low expression) (15 µg/Lane)
 Lane 3: NIH/3T3 cell lysate (15 µg/Lane)
 Lane 4: L6 cell lysate (15 µg/Lane)
 Lane 5: Mouse skin tissue lysate (20 µg/Lane)
 Lane 6: Rat skin tissue lysate (20 µg/Lane)

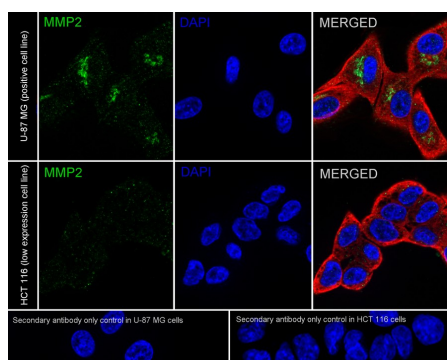
Predicted band size: 74 kDa
 Observed band size: 70 kDa

Exposure time: 30 seconds; ECL: K1802;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDN/TBST for 1 hour at room temperature. The primary antibody (HA723308) at 1/2,000 dilution was used in 5% NFDN/TBST at 4°C 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 U-87 MG (positive) and HCT 116 (low expression) labeling MMP-2 with Rabbit anti-MMP-2 antibody (HA723308) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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-MMP-2 antibody (HA723308) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. 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 (HA601187, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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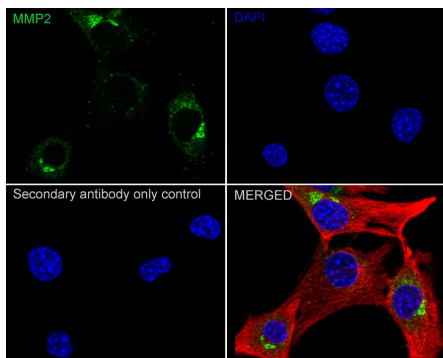
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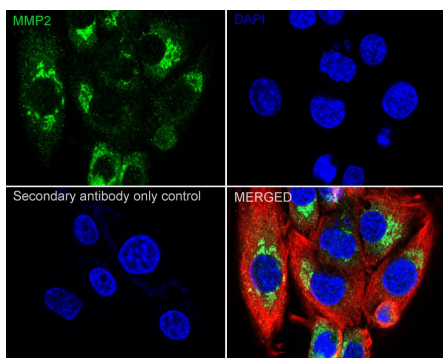
Fig3: Immunocytochemistry analysis of NIH/3T3 cells labeling MMP-2 with Rabbit anti-MMP-2 antibody (HA723308) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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-MMP-2 antibody (HA723308) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. 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 (HA601187, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

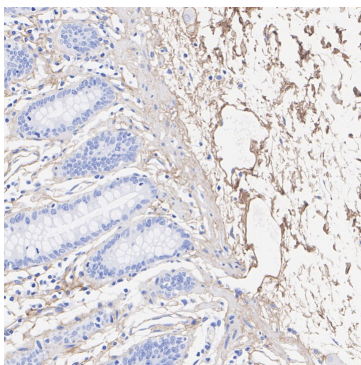
Fig4: Immunocytochemistry analysis of L6 cells labeling MMP-2 with Rabbit anti-MMP-2 antibody (HA723308) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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-MMP-2 antibody (HA723308) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. 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 (HA601187, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Fig5: Immunohistochemical analysis of paraffin-embedded human colon tissue with Rabbit anti-MMP-2 antibody (HA723308) 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 ddH₂O and PBS, and then probed with the primary antibody (HA723308) 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.

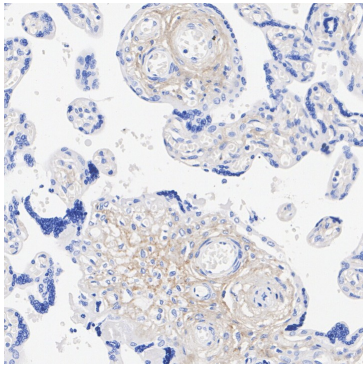


Fig6: Immunohistochemical analysis of paraffin-embedded human placenta tissue with Rabbit anti-MMP-2 antibody (HA723308) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA723308) 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.

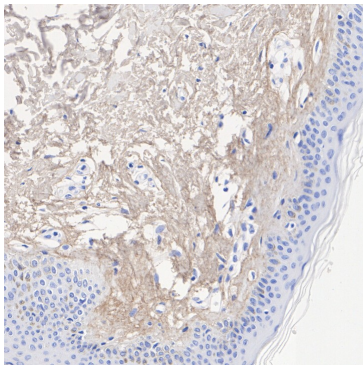


Fig7: Immunohistochemical analysis of paraffin-embedded human skin tissue with Rabbit anti-MMP-2 antibody (HA723308) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA723308) 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.

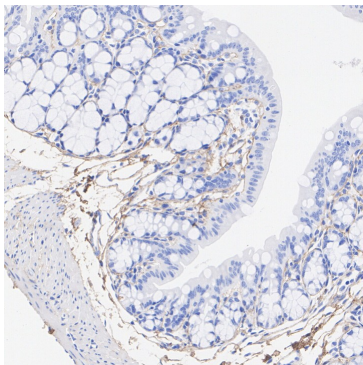


Fig8: Immunohistochemical analysis of paraffin-embedded mouse colon tissue with Rabbit anti-MMP-2 antibody (HA723308) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA723308) 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.

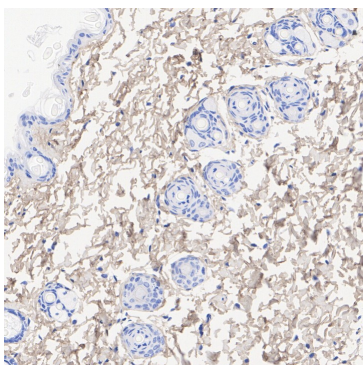


Fig9: Immunohistochemical analysis of paraffin-embedded mouse skin tissue with Rabbit anti-MMP-2 antibody (HA723308) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA723308) 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.

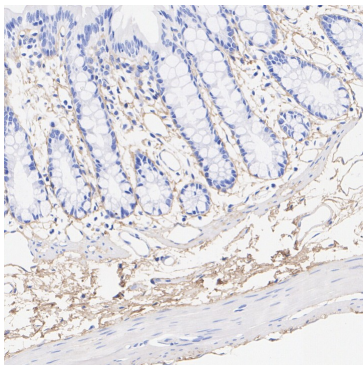


Fig10: Immunohistochemical analysis of paraffin-embedded rat colon tissue with Rabbit anti-MMP-2 antibody (HA723308) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA723308) 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.

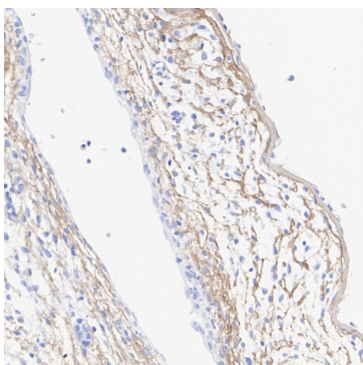


Fig11: Immunohistochemical analysis of paraffin-embedded rat placenta tissue with Rabbit anti-MMP-2 antibody (HA723308) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA723308) 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.

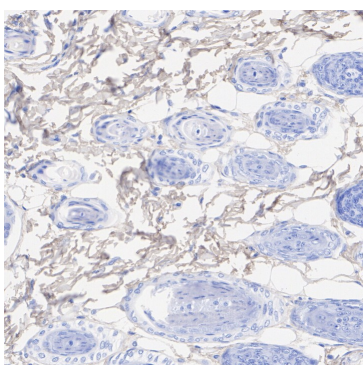


Fig12: Immunohistochemical analysis of paraffin-embedded rat skin tissue with Rabbit anti-MMP-2 antibody (HA723308) at 1/1,000 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA723308) 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.

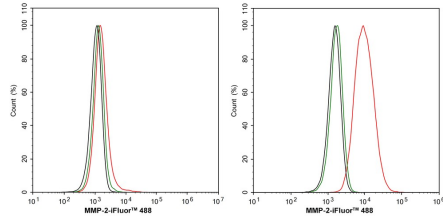


Fig13: Flow cytometric analysis of HCT 116 (left, low expression) and U-87 MG (right, positive) cells labeling MMP-2.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA723308, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C 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°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

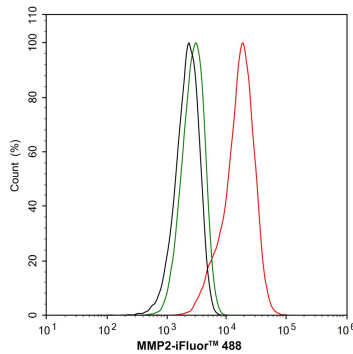


Fig14: Flow cytometric analysis of NIH/3T3 cells labeling MMP-2.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA723308, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C 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°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

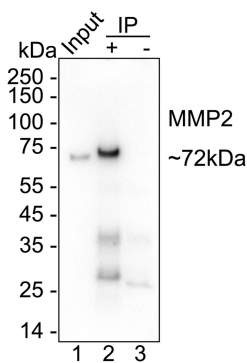


Fig15: MMP-2 was immunoprecipitated from 0.2 mg U-87 MG cell lysate with HA723308 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using HA723308 at 1/1,000 dilution. HRP Conjugated Anti-Rabbit IgG for IP Nano-secondary antibody at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: U-87 MG cell lysate (input)
Lane 2: HA723308 IP in U-87 MG cell lysate
Lane 3: Rabbit IgG instead of HA723308 in U-87 MG cell lysate

Blocking/Dilution buffer: 5% NFDm/TBST
Exposure time: 59 seconds; ECL: K1801

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Yan Y et al. Association of MMP2 and MMP9 gene polymorphisms with the recurrent spontaneous abortion: A meta-analysis. *Gene*. 2021 Jan
2. Kalev-Altman R et al. Mmp2 Deficiency Leads to Defective Parturition and High Dystocia Rates in Mice. *Int J Mol Sci*. 2023 Nov

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