Anti-MMP-12 Antibody [SR03-23]

ET1602-42

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP, FC
Molecular Wt:	Predicted band size: 54 kDa
Clone number:	SR03-23

Description: The matrix metalloproteinases (MMP) are a family of peptidase enzymes responsible for the degradation of extracellular matrix components, including collagen, gelatin, fibronectin, laminin and proteoglycan. Transcription of MMP genes is differentially activated by phorbol ester, lipopolysaccharide (LPS) or staphylococcal enterotoxin B (SEB). MMP catalysis requires both calcium and zinc. MMP-12 (also designated macrophage metalloelastase) is produced in alveolar macrophages and degrades elastin. MMP-12 may contribute to elastin degradation occurring in granulomatous skin diseases and may also participate in macrophage migration through the epidermal and vascular basement membranes in inflammatory disorders.

Immunogen: Synthetic peptide within Human MMP12 aa 421-470 / 470.

Positive control: A549 cell lysate, Hela cell lysate, MCF-7 cell lysate, THP-1 cell lysate, human breast carcinoma tissue lysate, mouse lung tissue lysate, rat lung tissue lysate.

Subcellular location: Secreted.

Database links: SwissProt: P39900 Human | P34960 Mouse | Q63341 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:20-1:50
IF-Tissue	1:20-1:50
IHC-P	1:50-1:200
FC	1:50-1:100
IP	Use at an assay dependent concentration.

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

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

Purity: Protein A affinity purified.

Images

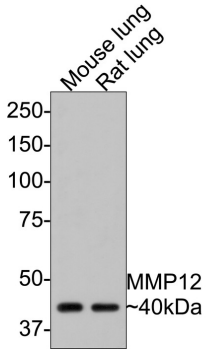

Fig1: Western blot analysis of MMP-12 on different lysates with Rabbit anti-MMP-12 antibody (ET1602-42) at 1/500 dilution.

Lane 1: Mouse lung tissue lysate
Lane 2: Rat lung tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 54 kDa
Observed band size: 40 kDa

Exposure time: 1 minute;

8% 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 (ET1602-42) at 1/500 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

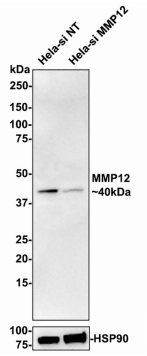

Fig2: Western blot analysis of MMP12 on different lysates with Rabbit anti-MMP12 antibody (ET1602-42) at 1/1,000 dilution.

Lane 1: Hela-si NT cell lysate
Lane 2: Hela-si MMP12 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 54 kDa
Observed band size: 40 kDa

Exposure time: 6 minutes;

4-20% SDS-PAGE gel.

ET1602-42 was shown to specifically react with MMP12 in Hela-si NT cells. Weakened band was observed when Hela-si MMP12 sample was tested. Hela-si NT and Hela-si MMP12 samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ET1602-42, 1/1,000) and Loading control antibody (Rabbit anti-HSP90, ET1605-56, 1/10,000) were used in 5% BSA at room temperature for 2 hours. Goat Anti-rabbit IgG-HRP Secondary Antibody (HA1001) at 1:100,000 dilution was used for 1 hour at room temperature.

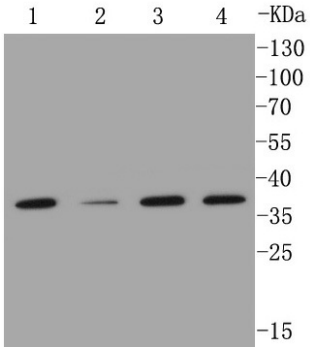

Fig3: Western blot analysis of MMP-12 on different lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET1602-42, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:5,000 dilution was used for 1 hour at room temperature.
Positive control:
Lane 1: A549 cell lysate
Lane 2: Hela cell lysate
Lane 3: MCF-7 cell lysate
Lane 4: THP-1 cell lysate

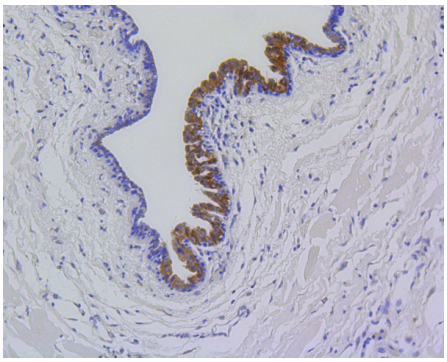

Fig4: Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue using anti-MMP-12 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1602-42, 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.

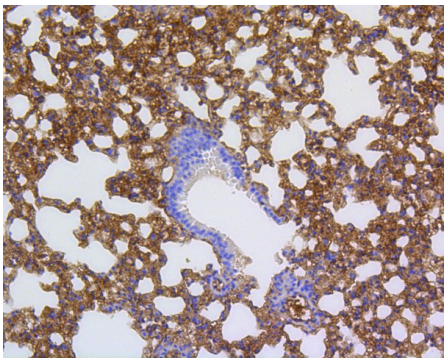

Fig5: Immunohistochemical analysis of paraffin-embedded mouse lung tissue using anti-MMP-12 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1602-42, 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.

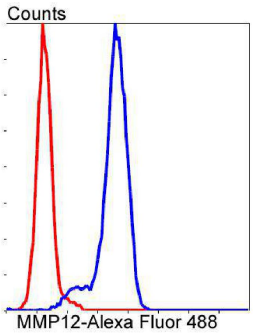

Fig6: Flow cytometric analysis of MMP-12 was done on A549 cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1602-42, 1/50) (blue). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes.Unlabelled sample was used as a control (cells without incubation with primary antibody; red).

Background References

1. John-Schuster G et al. Cigarette smoke-induced iBALT mediates macrophage activation in a B cell-dependent manner in COPD. Am J Physiol Lung Cell Mol Physiol 307:L692-706 (2014).
2. Stawski L et al. MMP-12 deficiency attenuates angiotensin II-induced vascular injury, M2 macrophage accumulation, and skin and heart fibrosis. PLoS One 9:e109763 (2014).