

Product datasheet

Anti-MMP9 antibody ab38898

★★★★☆ 44 Abreviews 308 References 9 Images

Overview

Product name	Anti-MMP9 antibody
Description	Rabbit polyclonal to MMP9
Host species	Rabbit
Specificity	The antibody binds to Gelatinase-B, but does not cross react with the other MMP family members (MMP-1, MMP-2, MMP-3). In our hands, we observe a weaker signal in WB in human samples compared to mouse samples (BLAST of full length mouse protein sequence showed 72% homology with the Human MMP9 sequence).
Tested applications	Suitable for: IHC-P, IHC-Fr, WB, IP, ELISA, ICC/IF, ICC, IHC-FoFr
Species reactivity	Reacts with: Mouse, Rat, Dog, Human
Immunogen	Full length protein corresponding to Mouse MMP9.
Positive control	IHC-Fr: Mouse liver and heart tissue. IHC-P: Mouse lung and pancreatic carcinoma tissue; Zebrafish pancreas tissue WB: Recombinant Human MMP9 protein; U937 and Raw 264.7 cell lysate. ICC/IF: Mouse neutrophils and monocytes.
General notes	Abcam recommended secondaries - Goat Anti-Rabbit HRP (ab205718) and Goat Anti-Rabbit Alexa Fluor® 488 (ab150077). See other anti-rabbit secondary antibodies that can be used with this antibody.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Store at -20°C.
Storage buffer	Preservative: 0.05% Sodium azide Constituent: 50% Glycerol
Purity	Immunogen affinity purified
Clonality	Polyclonal
Isotype	IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab38898** in the following tested applications.

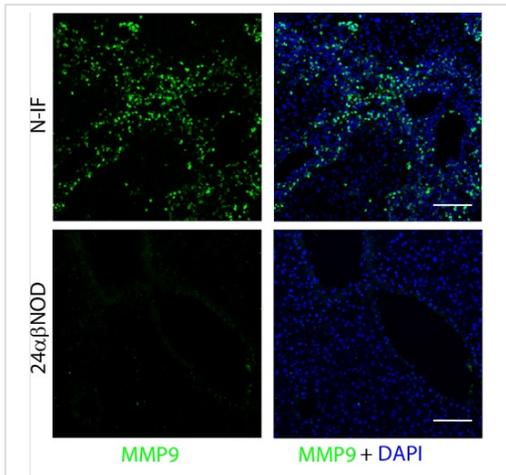
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P	★★★★☆	1/100 - 1/1000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
IHC-Fr	★★★★☆	1/1000. (see Abreview submitted by Greg Gibson) We recommend using Goat Anti-Rabbit IgG H&L (Cy3®) preadsorbed (ab6939) secondary antibody
WB	★★★★☆	1/1000. Detects a band of approximately 92 kDa. When using colorimetric substrates such as BCIP/NBT use at a 1/5000 dilution (for chemiluminescent substrates). Detects a band of approximately 92-95 kDa (pro-form) and 82kDa (active form) (Human samples). Mouse MMP9 is larger, and on SDS PAGE gels runs about 102-105 kDa. Dilution optimised using Chromogenic detection.
IP		Use at an assay dependent concentration.
ELISA	★★★★☆	Use at an assay dependent concentration.
ICC/IF	★★★★☆	1/500.
ICC		Use at an assay dependent concentration.
IHC-FoFr	★★★★★	Use at an assay dependent concentration. PubMed: 19295156

Target

Function	May play an essential role in local proteolysis of the extracellular matrix and in leukocyte migration. Could play a role in bone osteoclastic resorption. Cleaves KiSS1 at a Gly-Leu bond. Cleaves type IV and type V collagen into large C-terminal three quarter fragments and shorter N-terminal one quarter fragments. Degrades fibronectin but not laminin or Pz-peptide.
Tissue specificity	Produced by normal alveolar macrophages and granulocytes.
Involvement in disease	Intervertebral disc disease Metaphyseal anadysplasia 2
Sequence similarities	Belongs to the peptidase M10A family. Contains 3 fibronectin type-II domains. Contains 4 hemopexin repeats.
Domain	The conserved cysteine present in the cysteine-switch motif binds the catalytic zinc ion, thus inhibiting the enzyme. The dissociation of the cysteine from the zinc ion upon the activation-peptide release activates the enzyme.
Post-translational modifications	Processing of the precursor yields different active forms of 64, 67 and 82 kDa. Sequentially processing by MMP3 yields the 82 kDa matrix metalloproteinase-9. N- and O-glycosylated.
Cellular localization	Secreted, extracellular space, extracellular matrix.

Images

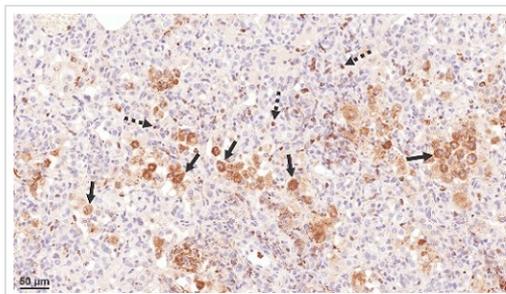


Immunohistochemistry (Frozen sections) - Anti-MMP9 antibody (ab38898)

Fransén-Pettersson N. et al PLoS One. 2016 Jul 21;11(7):e0159850. doi: 10.1371/journal.pone.0159850. eCollection 2016.

Immunofluorescence staining showing MMP9 (green) and DAPI (blue) in 12 weeks old N-IF and 24αβNOD control mouse livers. Scale bars are 100 μm.

Immunohistochemical staining was performed on liver biopsies fixed in 4% paraformaldehyde and embedded in OCT. The frozen tissues were cut in 5 μm thick sections and stained using primary antibody against matrix metalloproteinase 9 (MMP9) (1:300, Abcam, ab38898). The nuclei were visualized with DAPI. The sections were analyzed using confocal microscopy.

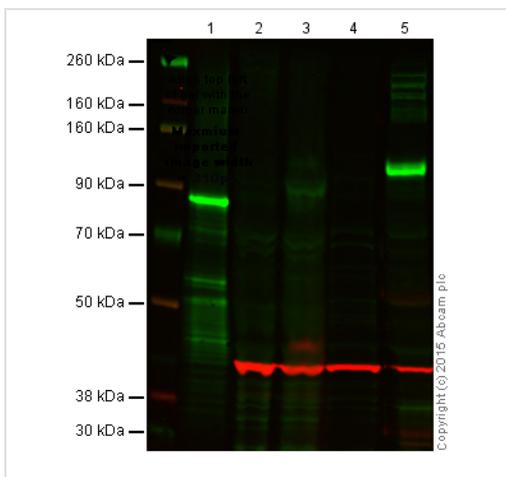


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MMP9 antibody (ab38898)

Ordonez AA et al PLoS One. 2018 May 14;13(5):e0197474. doi: 10.1371/journal.pone.0197474. eCollection 2018.

MMP-9 immunohistochemistry in the lung tissue of a C3HeB/FeJ mouse, 10 weeks after *M. tuberculosis* aerosol infection. MMP-9 was visualized in macrophages (black solid arrows) and neutrophils (black dotted arrows).

Paraffin embedded sections were rehydrated in graded alcohols, steamed in citrate buffer at pH 6 and probed at room temperature for 2 hours using the MMP-9 (rabbit polyclonal; 1:250; Abcam [AB38898]) and processed with a polymer-HRP kit (BioGenex) with diaminobenzidine development and Mayer hematoxylin counterstaining. Lungs from uninfected, and infected but untreated animals without primary antibody served as negative controls. Slides were scanned using the Apeiro digital scanner (Leica).



Western blot - Anti-MMP9 antibody (ab38898)

All lanes : Anti-MMP9 antibody (ab38898) at 5 μg/ml

Lane 1 : Recombinant Human MMP9, His tagged (ab82955) at 0.1 μg

Lane 2 : U937 whole cell lysate at 100 μg

Lane 3 : U937 whole cell lysate - treated with PMA and Brefeldin (24 hour treatment) at 100 μg

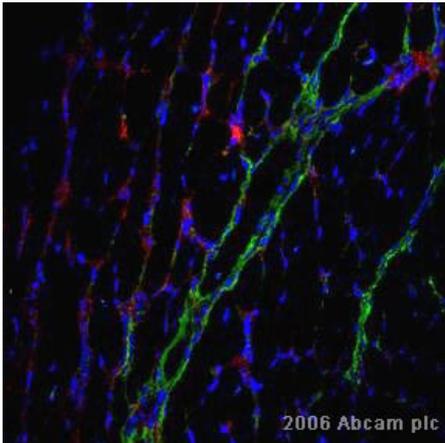
Lane 4 : Raw 264.7 (Mouse) whole cell lysate at 100 μg

Lane 5 : Raw 264.7 (Mouse) whole cell lysate - treated with LPS (6 hour treatment, 1 μg/mL) at 100 μg

Performed under reducing conditions.

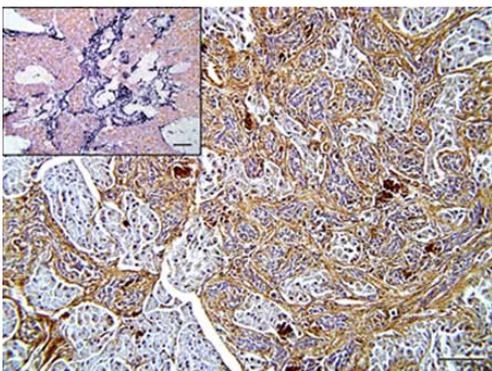
ab38898 detects recombinant Human MMP9 running at ~85 kDa, and endogenous full-length MMP9 in LPS-stimulated cells at ~100 kDa. This antibody also detects a band at 90 kDa in U937 PMA-treated cells.

ab38898 was incubated at 5 ug/mL and [ab8245](#) (loading control to GAPDH) was diluted at 0.1 ug/mL and both were incubated for 48 hours at 4°C. Blots were developed with goat anti-rabbit IgG (H + L) and goat anti-mouse IgG (H + L) secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.



Immunohistochemistry (Frozen sections) - Anti-MMP9 antibody (ab38898)

ab38898 at a 1/1000 dilution staining mouse heart tissue by Immunohistochemistry (Frozen sections). The tissue was removed from a mouse, rinsed in PBS and slowly frozen in supercooled isopentane. 14um sections were made using a cryostat. The sections were acetone fixed and blocked in 2% BSA prior to incubation with the MMP9 antibody. [Goat Anti-Rabbit IgG H&L \(Cy3 @\) preadsorbed \(ab6939\)](#) was used as the secondary antibody. In the image: red staining = MMP9, blue staining = nuclei, green = gelatinase activity.

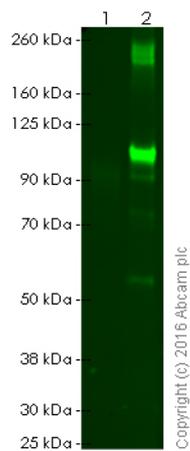


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MMP9 antibody (ab38898)

Image from Jung IH et al., PLoS One. 2011;6(12):e27941. Epub 2011 Dec 2. Fig 7.; doi:10.1371/journal.pone.0027941; December 2, 2011, PLoS ONE 6(12): e27941.

ab38898 staining MMP9 in 6 month-old transgenic zebrafish pancreas (Ihha overexpression) by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections).

Sections were incubated with primary antibody (1/500) and HRP-conjugated secondary antibody colored using DAB solution. Slides were counterstained with hematoxylin.



Western blot - Anti-MMP9 antibody (ab38898)

All lanes : Anti-MMP9 antibody (ab38898) at 2 µg/ml

Lane 1 : Native human MMP9 protein (Proenzyme, monomer) (ab157344)

Lane 2 : Recombinant Mouse MMP9 protein (ab39309)

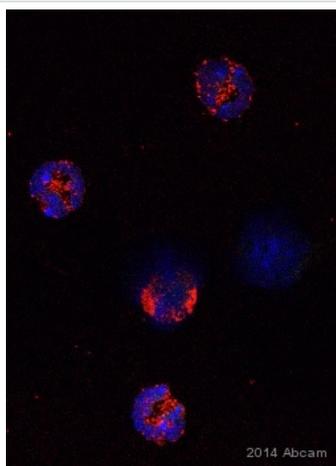
Lysates/proteins at 0.1 µg per lane.

Secondary

All lanes : Infrared labelled goat anti-rabbit (green) at 1/20000 dilution

Performed under reducing conditions.

This blot was produced using a 4-12% Bis-Tris gel under the MOPS buffer system. The gel was run at 200V for 60 minutes before being transferred onto a nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour before being incubated with anti-MMP9 antibody (ab38898; 2 microgram per mL) overnight at 4°C. Antibody binding was detected using infrared labelled goat anti-rabbit (green) antibody (diluted 1:20000) for 1 hour at room temperature before imaging.

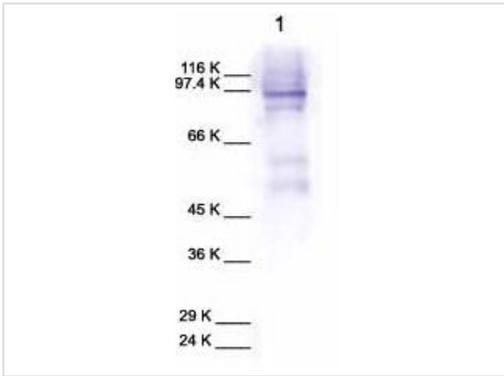


Immunocytochemistry/ Immunofluorescence - Anti-MMP9 antibody (ab38898)

Image courtesy of an anonymous Abreview

ab38898 staining MMP9 (red) in Mouse Neutrophils and Monocytes cells by ICC/IF

(Immunocytochemistry/immunofluorescence). Cells were fixed with paraformaldehyde and permeabilized with 2%BSA + 0.2% tritonX100 in PBS. Samples were incubated with primary antibody (1/200 in 2%BSA + 0.2% tritonX100 in PBS) for 25 minutes at 23°C. An Alexa Fluor® 568-conjugated Donkey anti-rabbit IgG polyclonal (1/1000) was used as the secondary antibody. DAPI is stained blue



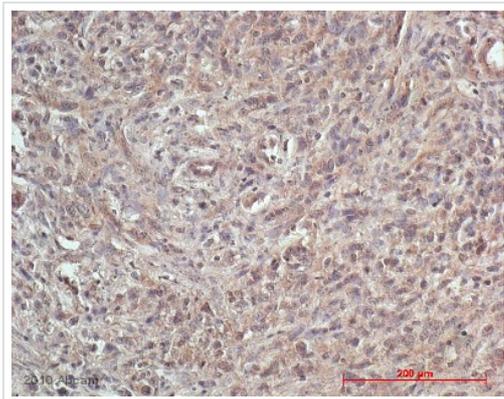
Western blot - Anti-MMP9 antibody (ab38898)

Anti-MMP9 antibody (ab38898) + Human MMP9

Observed band size: 88,92 kDa

[why is the actual band size different from the predicted?](#)

Ab38898 detects a band at 92 Kd (pro-form) and a band at 88 Kd (active form). Mouse MMP9 is slightly larger than human MMP9, and the antibody detects a band at about 105 Kd. It is recommended to concentrate samples by Gelatin-agarose affinity chromatography prior to Western blot usage. A recommended starting concentration for Western blots is 1:1000 when using colorimetric substrates such as BCIP/NBT, and 1:5000 for chemiluminescent substrates. Higher concentration of antibody may be needed for non-human samples.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MMP9 antibody (ab38898)

This image is courtesy of an anonymous Abreview

ab38898 staining MMP9 in Mouse Pancreatic carcinoma tissue sections by IHC-P (formaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with formaldehyde and blocked with 1% BSA for 1 hour at room temperature. Antigen retrieval was by heat mediation in citric acid (pH6). Samples were incubated with primary antibody (1/100) in 1% Aurion BSA for 12 hours. An HRP-conjugated Donkey polyclonal to rabbit IgG (1/100) was used as secondary antibody.

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