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# **Product Information**

Anti-Matrix Metalloproteinase-1, Hinge Region produced in rabbit, affinity isolated antibody

Catalog Number M4177

Synonym: Anti-MMP-1

## **Product Description**

Anti-Matrix Meltalloproteinase-1, Hinge Region is produced in rabbit using as immunogen a synthetic peptide corresponding to the hinge region of human MMP-1 (interstitial collagenase). Affinity isolated antibody is obtained from anti-MMP-1 by immunospecific purification which removes essentially all rabbit serum proteins, including immunoglobulins, which do not specifically bind to the peptide.

Anti-Matrix Meltalloproteinase-1, Hinge Region recognizes human MMP-1, hinge region (other species not tested). The antibody specifically binds to collagenase and does not cross-react with other MMP family members (MMP-2, MMP-3, MMP-9, etc). By immunoblotting, the antibody reacts with a band at ~52 kDa (unglycosylated proform of MMP-1) and ~57 kDa (glycosylated pro-form of MMP-1). Although the sequence homology for the hinge region of MMP-1 is well conserved, higher antibody concentrations may be necessary for non-human samples.

The antibody may be used for the detection and localization of MMP-1 by various immunochemical techniques such as immunoblotting and immunohistochemistry (not tested).

The matrix metalloproteinases (MMPs) are a family of at least eighteen secreted and membrane-bound zincendopeptidases. Collectively, these enzymes can degrade all the components of the extracellular matrix, including fibrillar and non-fibrillar collagens, fibronectin, laminin and basement membrane glycoproteins. In general, the structure of MMPs is characterized by a single peptide, a propeptide, and a catalytic domain containing the highly conserved zinc-binding site. In addition, fibronectin-like repeats, a hinge region, and a C-terminal hemopexin-like domain allow categorization of MMPs into the collagenase, gelatinase, stomelysin and membrane-type MMP subfamilies. MMPs contain the motif His-Glu-Xaa-His that binds zinc in the

catalytic site, as well as another zinc molecule and two calcium molecules structurally. They fall within the matrixin subfamily, and are EC designated 3.4.24.x. This group also contains astacin, reprolysin, and serralysin, as well as other more divergent metalloproteinases. All MMPs are synthesized as proenzymes, and most of them are secreted from the cells as proenzymes. Thus, the activation of these proenzymes is a critical step that leads to extracellular matrix breakdown.

MMPs are considered to play an important role in wound healing, apoptosis, bone elongation, embryo development, uterine involution, angiogenesis,<sup>4</sup> and tissue remodeling, and in diseases such as multiple sclerosis,<sup>2,5</sup> Alzheimer's,<sup>2</sup> malignant gliomas,<sup>2</sup> lupus, arthritis, periodontis, glumerulonephritis, atherosclerosis, tissue ulceration, and in cancer cell invasion and metastasis.<sup>6</sup> Numerous studies have shown that there is a close association between expression of various members of the MMP family by tumors and their proliferative and invasive behavior and metastaic potential.

MMPs and TIMPs can be divided into two groups with respect to gene expression: the majority exhibit inducible expression and a small number are produced constitutively or are expressed at very low levels and are not inducible. Among agents that induce MMP and TIMP production are the inflammatory cytokines TNF $\alpha$  and IL-1 $\beta$ . Marked cell type specificity is a hallmark of both MMP and TIMP gene expression (i.e., a limited number of cell types can be induced to make these proteins).

Matrix Metalloproteinase-1 (MMP-1) is also known as interstitial collagenase and collagenase-1. MMP-1 degrades fibrillar collagens types I, II, III, VII, VIII, X, aggrecan, serpins and  $\alpha_2$ -macroglobulin. All collagenases cleave fibrillar collagens at one specific site resulting in generation of N-terminal  $^3\!\!/_4$  and C-terminal  $^1\!\!/_4$  fragments, which then denature to gelatin at body temperature.

The substrate specificity of collagenases is variable: MMP-1 degrades type III collagen more efficiently than type I or type II collagen, whereas MMP-8 is more potent in degrading type I collagen than type III or type II collagen. MMP-13, in turn degrades type II collagen 6-fold more efficiently than type I and type II collagens and displays almost 50-fold stronger gelatinolytic activity than MMP-1 and MMP-8. Increased synthesis of MMP-1 is caused by a wide variety of reagents that include: TNF,  $^{11,12}_{}$  IL-1,  $^{13}_{}$  serum, EGF and TGF- $\beta$ ,  $^{14-16}_{}$  phorbol ester tumor promoter, PMA,  $^{17}_{}$  ECM components,  $^{18}_{}$  and polyoma and RSV infections. In contrast to these activators, several well-known antagonists, dexamethasone and all-transretinoic acid (RA) block the induced gene expression.

### Reagent

Supplied in phosphate buffered saline, pH 7.4, containing 0.05% sodium azide as preservative.

#### **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

# Storage/Stability

For continuous use, this antibody may be stored at 2-8 °C for up to one year. For extended use, the antibody may be aliquoted and stored at –20 °C. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

# **Product Profile**

Immunoblotting: a working antibody dilution of 1:500-1:1,000 (1-2µg/mL) is determined using recombinant human MMP-1.

Antibody concentration is ~200 µg/mL

**Note**: In order to obtain the best results and assay sensitivity in various techniques and preparations, we recommend determining optimum working dilutions by titration.

**Note**: Collagenase levels in quiescent cells and tissues are minimal, and stimulation of protein concentration is often needed to visualize the bands. In addition, cell types differ greatly in the quantity of collagenase produced.

#### References

- Borkakoti, N., *Prog. Biophy. Mol. Biol.*, **70**, 73 (1998).
- 2. Yong, V.W., et al., *Trends in Neuroscience*, **21**, 75 (1998).
- 3. Kahari, V.-M., and Saarialho-Kere, U., *Exp. Dermatol.*, **6**, 199 (1997).
- 4. Halbert, I., et al., *Proc. Natl. Acad. Sci.*, *USA*, **93**, 9748 (1996).
- 5. Chandler, S., et al., *J. Neuroimmunol.*, **72**, 155 (1997).
- Birkedal-Hansen, H., et al., Crit. Rev. Oral. Biol. Med., 4, 197 (1993).
- 7. Hasty, K.A., et al., *J. Exp. Med.*, **159**, 1455 (1984).
- 8. Mallya, S.K., et al., *Biochemistry*, **29**, 10628 (1990).
- 9. Knuper, V., et al., *J Biol. Chem.*, **271**, 1544 (1996).
- 10. Mitchell, P.G., et al., *J. Clin. Invest.*, **97**, 761 (1996).
- 11. Brenner, D.A., et al., Nature, 337, 661 (1989).
- 12. Sciavolina, P.J., et al., *J. Biol. Chem.*, **269**, 21627 (1994).
- 13. Dayer, J-M., et al., *J. Clin. Invest.*, **77**, 645 (1986).
- 14. Delany, A.M., and Brickerhoff, C.E., *J. Cell Biochem.*, **50**, 400 (1992).
- 15. Edwards, D.R., et al., *EMBO J.*, **6**, 1899 (1987).
- Partridge, N.C., et al., *Endocrinology*, **120**, 1956 (1987).
- 17. Brickerhoff, C.E., et al., *Biochemistry*, **21**, 2674 (1982).
- 18. Shapiro, S.D., et al., *J. Biol. Chem.*, **268**, 8170 (1993).
- 19. Brickerhoff, C.E., et al., *N. Engl. J. Med.*, **303**, 432 (1980).

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