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ProductInformation

Matrix Metalloproteinase-1, Stable
from human fibroblasts

Product Number **M 3691**

Synonyms: MMP-1, Collagenase-1, Interstitial Collagenase; EC 3.4.24.7

Product Description
Matrix Metalloproteinase-1 (MMP-1) is a matrix metalloproteinase that has been substrate-affinity purified from cell culture media of stimulated human fibroblasts. MMP-1 is free of endogenous inhibitors, TIMPs, and other matrix metalloproteinases.

Matrix Metalloproteinase-1, Stable may be used in various immunochemical techniques such as Immunoblotting, ELISA, enzyme kinetics assays, and substrate assays. This enzyme consists as a mixture of the zymogen and active enzyme. By immunoblotting against the reduced protein, bands may be detected at approximately 53 kDa and 51 kDa.

The matrix metalloproteinases (MMPs) are a family of at least eighteen secreted and membrane-bound zinc-endopeptidases. 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, a signal peptide, a propeptide, and a catalytic domain containing the highly conserved zinc-binding site characterizes the structure of the MMPs. 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.[1-3] MMPs contain the motif His-Glu-X-X-His (X represents any amino acid) 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,[4] and tissue remodeling, and in diseases such as multiple sclerosis,[2, 5] Alzheimer's,[2] malignant gliomas,[2] lupus, arthritis, periodontis, glomerulonephritis, atherosclerosis, tissue ulceration, and in cancer cell invasion and metastasis.[6] 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 metastatic potential.

The tissue inhibitors of metalloproteinases (TIMPs) are naturally occurring proteins that specifically inhibit matrix metalloproteinases and regulate extracellular matrix turnover and tissue remodeling by forming tight-binding inhibitory complexes with the MMPs. Thus, TIMPs maintain the balance between matrix destruction and formation. An imbalance between MMPs and the associated TIMPs may play a significant role in the invasive phenotype of malignant tumors. 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-α and IL-1β. A 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 a true collagenase and along with MMP-8 and MMP-13 cleaves all three strands of intact native collagen. The substrate specificity of the 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.[7, 8] 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.[9, 10]

MMP-1 degrades fibrillar collagens types I, II, III, VII, VIII, X, aggrecan, serpins and α_2-macroglobulin. All collagenases cleave fibrillar collagens at one specific site resulting in the generation of N-terminal ¾ and C-terminal ¼ fragments, which then denature to gelatin at body temperature.

Structurally, MMP-1 may be divided into several distinct domains: a pro-domain, which is cleaved upon activation, a catalytic domain containing the zinc binding site, a short hinge region, and a carboxyl terminal (hemopexin-like) domain.

Increased synthesis of MMP-1 is caused by a wide variety of agents that include: TNF, [11, 12] IL-1, [13] serum, EGF and TGF-β, [14-16] phorbol ester tumor promoter, PMA, [17] ECM (extracellular matrix) components, [18] and polyoma and RSV infections. In contrast to these activators, several well-known antagonists, dexamethasone and all-trans-retinoic acid (RA) block the induced gene expression.[19] MMP-1 is expressed by fibroblasts, keratinocytes, endothelial cells, monocytes, and macrophages.

Collagenase levels in quiescent cells and tissues are minimal, so stimulation or protein concentration is often needed to visualize the bands. When MMP-1 production is stimulated, the native MMP inhibitors (TIMPs) will usually follow to quench them. In addition, cell types differ greatly in the quantity of collagenase produced.

The human MMP-1 gene, about 17 kb, maps to chromosome 11q22.2-22.3.

Reagent
Matrix Metalloproteinase-1, Stable is supplied as a mixture of the zymogen and active enzyme in a buffer solution of 10 mM MES, pH 5.5, containing 0.175 M sodium chloride, 5 mM calcium chloride, 5 mM EDTA, 0.025 % Brij-35, 50% (v/v) glycerol, and 0.01 % sodium azide.

Precautions and Disclaimer
Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.

Storage/Stability
Store at –20 °C in aliquots. Do not store below –22 °C. Repeated freezing and thawing is not recommended. Storage in “frost-free” freezers is not recommended.

Product Profile
Purity: >95 % as determined by SDS-PAGE, visualized by silver stain.

The EDTA in the preparation stabilizes MMP-1 and slows autoactivation and degradation. For enzymatic uses, the EDTA in the preparation should be removed before use by dialysis or spin-column, or the calcium chloride concentration should be increased to compete out the EDTA. After removal of the EDTA, the product may not regain enzymatic activity. For applications where active MMP-1 is required, the companion product, Sigma No. M1802, is recommended.

Because sodium azide is toxic, dialysis or spin-column should be done to remove it before using this product on cells.

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