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Product Information

**Anti-Matrix Metalloproteinase-16,
Cytoplasmic Domain**
Developed in Rabbit
Affinity Isolated Antibody

Product Number **M 4567**

Product Description

Anti-Matrix Metalloproteinase-16 (MMP-16), Cytoplasmic Domain is developed in rabbit using a synthetic peptide corresponding to the cytoplasmic domain (tail) of human matrix metalloproteinase-16 (MMP-16), also known as membrane-type matrix metalloproteinase-3 (MT3-MMP) as immunogen. Affinity isolated antigen specific antibody is obtained from rabbit anti-MMP-16 antiserum by immuno-specific purification which removes essentially all rabbit serum proteins, including immunoglobulins, which do not specifically bind to the peptide.

Anti-Matrix Metalloproteinase-16, Cytoplasmic Domain may be used for the detection and localization of human matrix metalloproteinase-16 (MMP-16, MT3-MMP). The antibody specifically binds to MMP-16 and does not cross react with the other MTMMP or MMP family members (MMP-14, MMP-15, MMP-17, MMP-1, MMP-2, MMP-3, MMP-9, etc.). The antibody can recognize native or reduced MT3-MMP, and has application for immunoblotting. The cytoplasmic tail is accessible to the cell surface thus the cells or tissue must be permeabilized for the antibody to gain access to the epitope. By immunoblotting against the reduced protein, the antibody identifies bands at 65 kDa and 63 kDa (the pro-form and active form), as well as activation/breakdown products.

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, reprotin, and serralytin, 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, periodontitis, 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-1 β . 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).

Membrane-type Matrix Metalloproteinase-3 (MT3-MMP, MMP-16) was identified as a membrane-bound MMP in normal and tumor cell lines. MT3-MMP is similar to other MT-MMPs. It contains a furin cleavage site, is membrane bound, and contains a cytoplasmic tail (MT4-MMP lacks the tail, and may not be intercalated into the membrane). MT3-MMP is also "shed" from the membrane in a soluble form. MT1-MMP is known to function in activating a number of MMPs, chiefly MMP-2, but the role for MT3-MMP is not clearly understood. MT3-MMP (MMP-16) is elevated in several tumor cell lines, and is constitutively produced by some normal cell lines.

Reagent

Anti-Matrix Metalloproteinase-16, Cytoplasmic Domain is supplied in 0.01 M phosphate buffered saline, pH 7.4, containing 50% glycerol and 0.05% sodium azide. The protein concentration is approximately 1 mg/ml.

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

For continuous use, store at 2-8 °C for up to six months. For extended storage, the solution may be stored -20 °C. Do not store below -22 °C. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

Product Profile

By immunoblotting, a minimum working dilution of 1:2,000 is recommended using a cell lysate, an alkaline phosphatase conjugated secondary antibody, and BCIP/NBT as the substrate. A starting dilution of 1:5,000 of anti-MMP-16 is recommended for chemiluminescent substrates.

Treatment of cell with Concanavalin-A or the phorbol ester TPA stimulates production of MT3-MMP in some cell types, and the enzyme can be recovered in cell lysates.

Note: Higher antibody dilutions may be necessary for non-human samples.

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

References

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