

MMP2 Inhibitor Screening Assay Kit (Colorimetric) ab139446

画像数 4

製品の概要

製品名	MMP2 Inhibitor Screening Assay Kit (Colorimetric)
検出方法	Colorimetric
サンプルの種類	Inhibitor compounds
アッセイタイプ	Enzyme activity
製品の概要	MMP2 Inhibitor Screening Assay Kit (Colorimetric) (ab139446) is a complete assay system designed to screen MMP2 inhibitors using a thiopeptide as a chromogenic substrate (Ac-PLG-[2-mercapto-4-methyl-pentanoyl]-LG-OC ₂ H ₅). The MMP cleavage site peptide bond is replaced by a thioester bond in the thiopeptide. Hydrolysis of this bond by an MMP produces a sulfhydryl group, which reacts with DTNB [5,5'-dithiobis(2-nitrobenzoic acid), Ellman's reagent] to form 2-nitro-5-thiobenzoic acid, which can be detected by its absorbance at 412 nm ($\epsilon=13,600 \text{ M}^{-1}\text{cm}^{-1}$ at pH 6.0 and above). The assays are performed in a convenient 96-well microplate format.
特記事項	This kit is useful to screen inhibitors of MMP2, a potential therapeutic target. The MMP inhibitor NNGH is also included as a prototypic control inhibitor. Thiol inhibitors should not be used with this kit, as they may interfere with the colorimetric assay.
試験プラットフォーム	Microplate reader

製品の特性

保存方法 Please refer to protocols.

内容	1 x 96 tests
96-well Clear Microplate (1/2 Volume)	1 unit
Colorimetric Assay Buffer	1 x 20ml
MMP Inhibitor	1 x 50 μ l
MMP Substrate	1 x 50 μ l
MMP2 Enzyme (Human, Recombinant)	1 x 45.7 μ l

機能 Ubiquitous metalloproteinase that is involved in diverse functions such as remodeling of the

vasculature, angiogenesis, tissue repair, tumor invasion, inflammation, and atherosclerotic plaque rupture. As well as degrading extracellular matrix proteins, can also act on several nonmatrix proteins such as big endothelial 1 and beta-type CGRP promoting vasoconstriction. Also cleaves KISS at a Gly-

-Leu bond. Appears to have a role in myocardial cell death pathways. Contributes to myocardial oxidative stress by regulating the activity of GSK3beta. Cleaves GSK3beta in vitro.

PEX, the C-terminal non-catalytic fragment of MMP2, possesses anti-angiogenic and anti-tumor properties and inhibits cell migration and cell adhesion to FGF2 and vitronectin. Ligand for integrin/beta3 on the surface of blood vessels.

組織特異性

Produced by normal skin fibroblasts. PEX is expressed in a number of tumors including gliomas, breast and prostate.

関連疾患

Defects in MMP2 are the cause of Torg-Winchester syndrome (TWS) [MIM:259600]; also known as multicentric osteolysis nodulosis and arthropathy (MONA). TWS is an autosomal recessive osteolysis syndrome. It is severe with generalized osteolysis and osteopenia. Subcutaneous nodules are usually absent. Torg-Winchester syndrome has been associated with a number of additional features including coarse face, corneal opacities, patches of thickened, hyperpigmented skin, hypertrichosis and gum hypertrophy. However, these features are not always present and have occasionally been observed in other osteolysis syndromes.

配列類似性

Belongs to the peptidase M10A family.

Contains 3 fibronectin type-II domains.

Contains 4 hemopexin-like domains.

ドメイン

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.

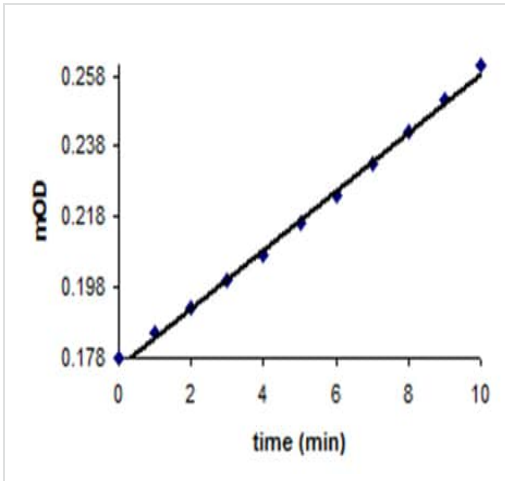
翻訳後修飾

Phosphorylation on multiple sites modulates enzymatic activity. Phosphorylated by PKC in vitro. The propeptide is processed by MMP14 (MT-MMP1) and MMP16 (MT-MMP3). Autocatalytic cleavage in the C-terminal produces the anti-angiogenic peptide, PEX. This processing appears to be facilitated by binding integrin/beta3.

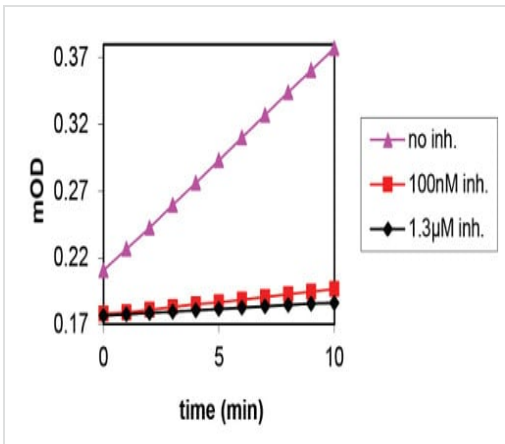
細胞内局在

Secreted > extracellular space > extracellular matrix. Membrane. Nucleus. Colocalizes with integrin alphaV/beta3 at the membrane surface in angiogenic blood vessels and melanomas. Found in mitochondria, along microfilaments, and in nuclei of cardiomyocytes.

画像



Plot of OD vs time

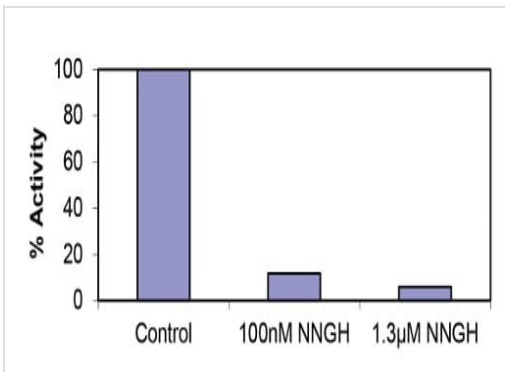


Inhibition of MMP3 by NNGH

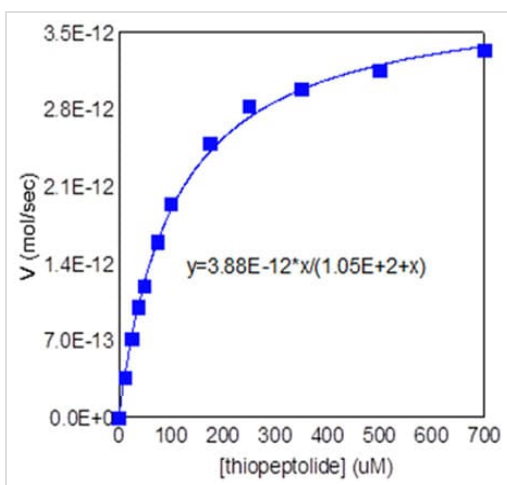
Control slope = $1.65E-02$ OD/min

Inhibitor slope = $9.80E-04$ OD/min

inhibitor % activity remaining = $(9.80E-04/1.65E-02) \times 100 = 5.94\%$



Inhibition of MMP3 by NNGH



$K_m = 105 \mu\text{M}$

$V_{\text{max}} = 3.9 \text{ pmol/sec}$

Example Graph for K_m and V_{max} Determination

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