abcam

Product datasheet

MMP1 Inhibitor Screening Assay Kit (Colorimetric) ab139443

2 References 4 Images

Overview

Product name	MMP1 Inhibitor Screening Assay Kit (Colorimetric)
Detection method	Colorimetric
Sample type	Inhibitor compounds
Assay type	Enzyme activity
Product overview	Abcam MMP1 Inhibitor Screening Assay Kit (Colorimetric) (ab139443) is a complete assay system designed to screen MMP1 inhibitors using a thiopeptide as a chromogenic substrate (Ac-PLG-[2-mercapto-4-methyl-pentanoyl]-LG-OC_2H_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 (ε=13,600 $M^{-1}cm^{-1}$ at pH 6.0 and above). The assays are performed in a convenient 96-well microplate format.
Notes	This kit is useful to screen inhibitors of MMP1, 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.
Platform	Microplate reader

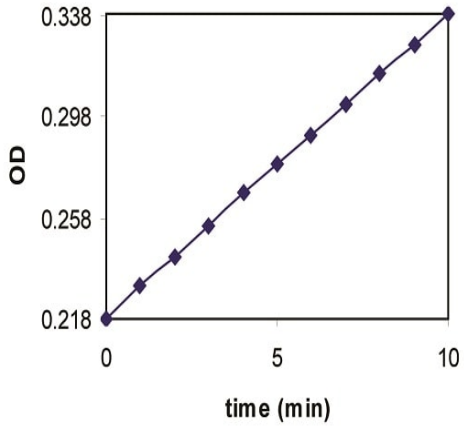
Properties

Storage instructions	Please refer to protocols.

Components	1 x 96 tests
96-well Clear Microplate (1/2 Volume)	1 unit
Colorimetric Assay Buffer	1 x 20ml
MMP Inhibitor	1 x 50µl
MMP Substrate	1 x 50µl
MMP1 Enzyme (Human, Recombinant)	1 x 66µl

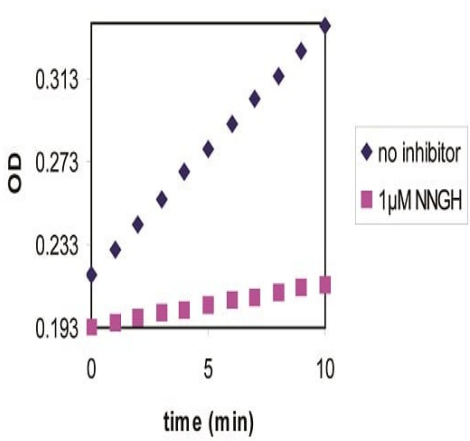
Function	Cleaves collagens of types I, II, and III at one site in the helical domain. Also cleaves collagens of types VII and X. In case of HIV infection, interacts and cleaves the secreted viral Tat protein, leading to a decrease in neuronal Tat's mediated neurotoxicity.
Sequence similarities	Belongs to the peptidase M10A family. Contains 4 hemopexin-like domains.
Domain	There are two distinct domains in this protein; the catalytic N-terminal, and the C-terminal which is involved in substrate specificity and in binding TIMP (tissue inhibitor of metalloproteinases). 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	Undergoes autolytic cleavage to two major forms (22 kDa and 27 kDa). A minor form (25 kDa) is the glycosylated form of the 22 kDa form. The 27 kDa form has no activity while the 22/25 kDa form can act as activator for collagenase.
Cellular localization	Secreted > extracellular space > extracellular matrix.

Images



Plot of OD vs time

Slope = V = 1.20E-02 OD/min

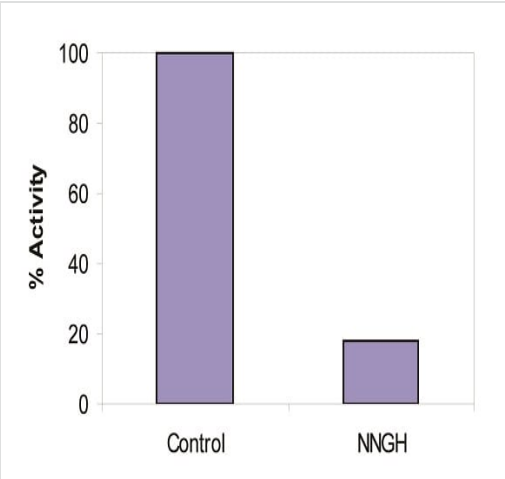


Inhibition of MMP1 by NNGH (1 μM)

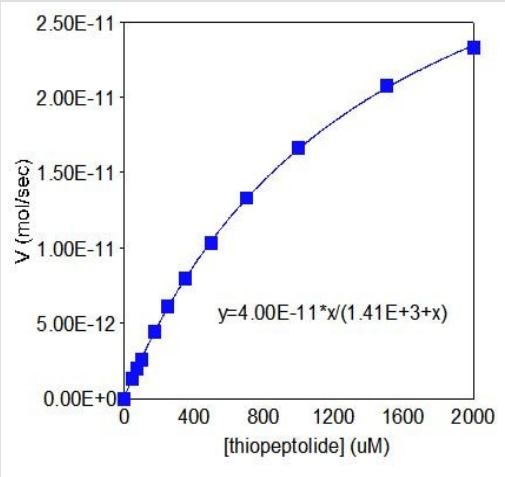
control slope = 1.20E-02 OD/min

inhibitor slope = 2.13E-03 OD/min

inhibitor % activity remaining = (2.13E-03/1.20E-02) x 100 = 17.7%



Inhibition of MMP1 by NNGH (1 µM)



Example graph for Km and Vmax determination

Km = 1410 µM

Vmax = 40.0 pmol/sec

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit https://www.abcam.com/abpromise or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors