

APA097Hu61 100µg
Active Matrix Metalloproteinase 1 (MMP1)
Organism Species: *Homo sapiens (Human)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Phe20~Asn469

Tags: N-terminal His-tag

Purity: >95%

Endotoxin Level: <1.0EU per 1µg (determined by the LAL method).

Buffer Formulation: PBS, pH7.4, containing 5% trehalose.

Applications: Cell culture; Activity Assays.

(May be suitable for use in other assays to be determined by the end user.)

Predicted isoelectric point: 6.9

Predicted Molecular Mass: 53.5kDa

Accurate Molecular Mass: 58&62kDa as determined by SDS-PAGE reducing conditions.

Phenomenon explanation:

The possible reasons that the actual band size differs from the predicted are as follows:

1. Splice variants: Alternative splicing may create different sized proteins from the same gene.
2. Relative charge: The composition of amino acids may affects the charge of the protein.
3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.
4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.
5. Polymerization of the target protein: Dimerization, multimerization etc.

[USAGE]

Reconstitute in 10mM PBS (pH7.6) to a concentration of 0.1-1.0 mg/mL. Do not

vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.

Store at 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

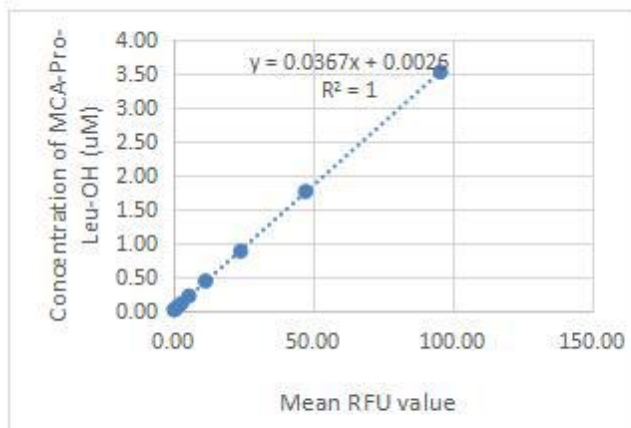
[SEQUENCE]

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F PATLETQEQD VDLVQKYLEK YYNLKNDGRQ
VEKRRNSGPV VEKCLKMQEF FGLKVTGKPD AETLKVMKQP RCGVPDVAQF
VLTEGNPRWE QTHLTYRIEN YTPDLPRADV DHAIEKAFQL WSNVTPLTFT
KVSEGOADIM ISFVRGDHRD NSPFDGPGGN LAHAFQPGPG IGGDAHFDDE
ERWTNNFREY NLHRVAAHEL GHSLGLSHST DIGALMYPSTY TFSGDVQLAQ
DDIDGIQAIY GRSQNPVQPI GPQTPKACDS KLTFDAITTI RGEVMFFKDR
FYMRTNPFYP EVELNFISVF WPQLPNGLEA AYEAFADRDEV RFFKGNKYWA
VQQQNVLHG YPKMIAHDFP GIGHKVDVAV MKDGGFFYFFH GTRQYKFDPK
TKRILTQKA NSWFNCRKN
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[ACTIVITY]

MMP1 is a zinc-dependent enzymes capable of cleaving components of the extracellular matrix, which belongs to the matrix metalloproteinase (MMP) family. MMP-1 (interstitial collagenase), can degrade a broad range of substrates including types I, II, III, VII, VIII, and X collagens as well as casein, gelatin and so on. MMP-1 is expressed by fibroblasts, keratinocytes, endothelial cells, monocytes and macrophages. 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. The activity of recombinant human MMP1 is measured by its ability to cleave a fluorogenic peptide substrate Mca-KPLGL-Dpa-AR-NH₂ in the assay buffer 50 mM Tris, 10 mM CaCl₂, 150 mM NaCl, 0.05% (w/v) Brij-35, pH 7.5. The rhMMP1 is diluted to 50 ug/ml in

assay buffer, then activated by p-aminophenylmercuric acetate (APMA) in a final concentration of 1 mM incubated at 37 ° C for 2 hours. The activated rhMMP-1 is diluted to 1 ug/mL in assay buffer. Loading into a black well plate 50 µL of 1 ug/mL rhMMP-1 and start the reaction by adding 50 µL of 20 µM substrate, with a substrate blank containing 50 µL assay buffer, 50 µL substrate, and no rhMMP-1. Then read at excitation and emission wavelengths of 320 nm and 405 nm, respectively, in kinetic mode for 5 minutes. The specific activity of recombinant human MMP1 is > 140 pmol/min/µg.



RFU (320/405)	MCA-Pro-Leu-OH (product) uM
95.78	3.52
47.46	1.76
24.20	0.88
11.63	0.44
5.71	0.22
3.05	0.11
1.52	0.05
0.77	0.03

Figure 1. The standard curve of MCA-Pro-Leu-OH

Specific Activity (pmol/min/µg) =

$$\frac{\text{Adjusted Vmax} * (\text{RFU/min}) \times \text{Conversion Factor} ** (\text{pmol/RFU})}{\text{amount of enzyme (ug)}}$$

*Adjusted for Substrate Blank

**Derived using calibration standard MCA-Pro-Leu-OH

[IDENTIFICATION]

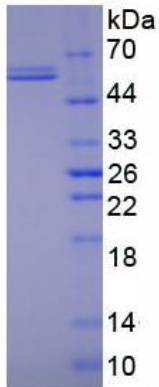


Figure 2. SDS-PAGE

Sample: Active recombinant MMP1, Human

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.