

APA097Po01 100µg
Active Matrix Metalloproteinase 1 (MMP1)
Organism Species: Sus scrofa; Porcine (Pig)
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

13th Edition (Revised in Aug, 2023)

[PROPERTIES]

Source: Prokaryotic expression.

Host: E. coli

Residues: Phe100~Asn469
Tags: N-terminal His-tag

Purity: >90%

Endotoxin Level: <1.0EU per 1µg (determined by the LAL method). **Buffer Formulation:** PBS, pH7.4, containing 0.01% SKL, 5%Trehalose .

Original Concentration: 200µg/mL

Applications: Cell culture; Activity Assays.

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

Predicted isoelectric point: 5.3

Predicted Molecular Mass: 46.3kDa

Accurate Molecular Mass: 46kDa as determined by SDS-PAGE reducing conditions.

[USAGE]

Reconstitute in 10mM PBS (pH7.4) 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.

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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]

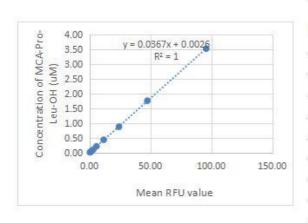
F VLTPGNPRWE NTHLTYRIEN YTPDLSREDV DRAIEKAFQL WSNVSPLTFT KVSEGQADIM ISFVRGDHRD NSPFDGPGGN LAHAFQPGPG IGGDAHFDED ERWTKNFRDY NLYRVAAHEL GHSLGLSHST DIGALMYPNY IYTGDVQLSQ DDIDGIQAIY GPSENPVQPS GPQTPQVCDS KLTFDAITTL RGELMFFKDR FYMRTNSFYP EVELNFISVF WPQVPNGLQA AYEIADRDEV RFFKGNKYWA VRGQDVLYGY PKDIHRSFGF PSTVKNIDAA VFEEDTGKTY FFVAHECWRY DEYKQSMDTG YPKMIAEEFP GIGNKVDAVF QKDGFLYFFH GTRQYQFDFK TKRILTLQKA NSWFNCRKN

[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 pig 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 rpMMP1 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 rpMMP-1 is diluted to 1 ug/mL in assay buffer. Loading into a black well plate 50 µL of 1 ug/mL rpMMP-1 and start the reaction by adding 50 µL of 20 µM substrate, with a substrate blank containing 50 µL assay

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buffer, 50 μ L substrate, and no rpMMP-1. Then read at excitiation and emission wavelengths of 320 nm and 405 nm, respectively, in kinetic mode for 5 minutes. The specific activity of recombinant pig MMP1 is > 10 pmol/min/ μ g.



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) =

Adjusted Vmax *(RFU/min) x Conversion Factor **(pmol/RFU)
amount of enzyme (ug)

[IDENTIFICATION]

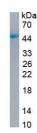


Figure 2. SDS-PAGE

Sample: Active recombinant MMP1, Pig

^{*}Adjusted for Substrate Blank

^{**}Derived using calibration standard MCA-Pro-Leu-OH



[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.