

APA129Mu01 100µg
Active Tissue Inhibitors Of Metalloproteinase 3 (TIMP3)
Organism Species: *Mus musculus (Mouse)*
Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Prokaryotic expression.

Host: *E. coli*

Residues: Gly20~Pro211

Tags: N-terminal His-tag

Purity: >98%

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

Buffer Formulation: 20mM Tris, 150mM NaCl, pH8.0, containing 0.05% sarcosyl and 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: 9.0

Predicted Molecular Mass: 23.3kDa

Accurate Molecular Mass: 28/22kDa 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 20mM Tris, 150mM NaCl (pH8.0) 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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          G AEACTCSPSH PQDAFCNSDI VIRAKVVGKK
LVKEGPFGLT VYTIKQMKMY RGFSKMPHVQ YIHTEASESL CGLKLEVNKY
QYLLTGRVYE GKMYTGLCNF VERWDHLTLS QRKGLNYRYH LGCNCKIKSC
YYLPCFVTSK NECLWTDMLS NFGYPGYQSK HYACIRQKGG YCSWYRGWAP
PDKSISNATD P
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[ACTIVITY]

Tissue Inhibitors Of Metalloproteinase 3 (TIMP3) is a protein belongs to the tissue inhibitor of metalloproteinases family. They are inhibitors of the matrix metalloproteinases. TIMP-3 is the only member of the TIMP family which is found exclusively in the extracellular matrix (ECM). It is regulated in a cell cycle-dependent fashion in certain cell types and may serve as a marker for terminal differentiation. Besides, Matrix Metalloproteinase 2 (MMP2) has been identified as an interactor of TIMP3, thus a binding ELISA assay was conducted to detect the interaction of recombinant mouse TIMP3 and recombinant mouse

MMP2. Briefly, TIMP3 were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100µl were then transferred to MMP2-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-TIMP3 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37°C. Finally, add 50µL stop solution to the wells and read at 450nm immediately. The binding activity of TIMP3 and MMP2 was shown in Figure 1, and this effect was in a dose dependent manner.

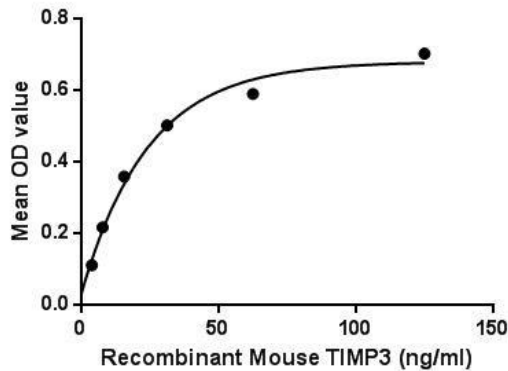


Figure 1. The binding activity of TIMP3 with MMP2.

[IDENTIFICATION]

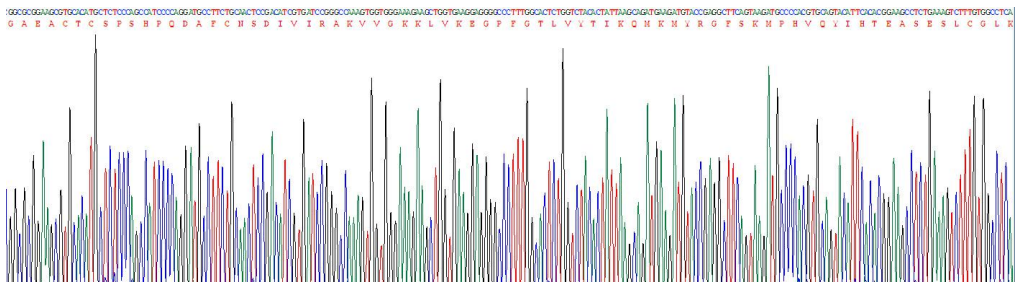


Figure 2. Gene Sequencing (extract)

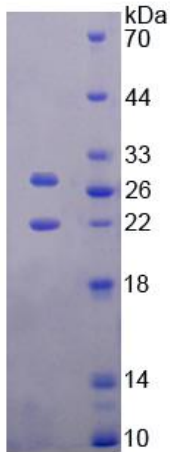


Figure 3. SDS-PAGE

Sample: Active recombinant TIMP3, Mouse

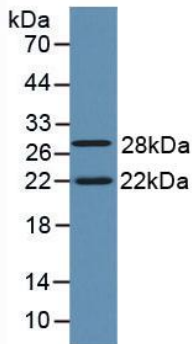


Figure 4. Western Blot

Sample: Recombinant TIMP3, Mouse;

Antibody: Rabbit Anti-Mouse TIMP3 Ab (PAA129Mu01)

[IMPORTANT NOTE]

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