

**APA128Hu02 100µg**  
**Active Tissue Inhibitors Of Metalloproteinase 2 (TIMP2)**  
**Organism Species: *Homo sapiens* (Human)**  
***Instruction manual***

FOR RESEARCH USE ONLY  
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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13th Edition (Revised in Aug, 2023)

## **[ PROPERTIES ]**

**Source:** Prokaryotic expression.

**Host:** *E. coli*

**Residues:** Cys27~Pro220

**Tags:** N-terminal His and GST 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:** 6.5

**Predicted Molecular Mass:** 51.8kDa

**Accurate Molecular Mass:** 53kDa 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.

**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.

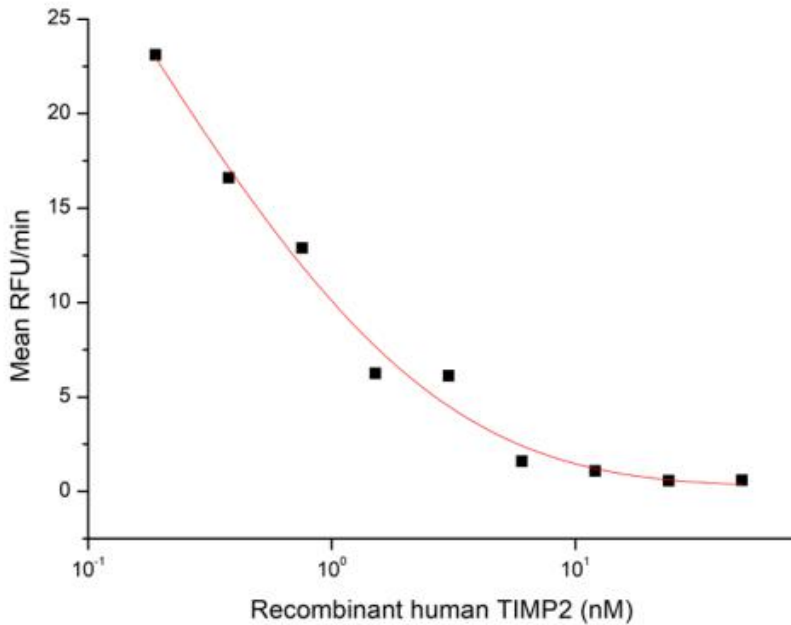
## **[ SEQUENCE ]**

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CSCS PVHPQQAFCN ADVVIRAKAV  
SEKEVDSGND IYGNPIKRIQ YEIKQIKMFK GPEKDIEFIY TAPSSAVCGV  
SLDVGGKKEY LIAGKAEGDG KMHITLCDFI VPWDTLSTTQ KKSLSNHRYQM  
GCECKITRCP MIPCYISSPD ECLWMDWVTE KNINGHQAKF FACIKRSDGS  
CAWYRGAAPP KQEFLDIEDP
```

## **[ ACTIVITY ]**

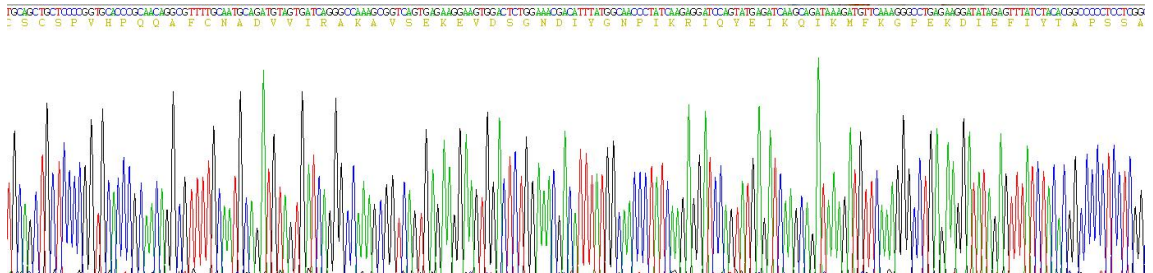
Tissue inhibitors of metalloproteinases or TIMPs are a family of proteins that regulate the activation and proteolytic activity of the zinc enzymes known as matrix metalloproteinases (MMPs). There are four members of the family, TIMP-1, TIMP-2, TIMP-3, and TIMP-4. TIMP-2 is a non N-glycosylated protein with a molecular mass of 22 kDa produced by a wide range of cell types, which inhibits MMPs non-covalently by the formation of binary complexes. TIMP-2 also has erythroid-potentiating and cell growth promoting activities. The activity of recombinant human TIMP2 was measured by its ability to inhibit rhMMP2 cleavage of a fluorogenic peptide substrate MCA-Pro-Leu-Gly-Leu-DPA-Ala-Arg-NH<sub>2</sub> in the assay buffer 50 mM Tris, 10 mM CaCl<sub>2</sub>, 150 mM NaCl, 0.05% (w/v) Brij-35, pH 7.5. rhMMP2 was diluted to 100 ug/ml and activated with 1 mM APMA at 37 ° C for 1 hour and rhTIMP2 (MW: 51.75 KD) was diluted to different concentrations with the assay buffer. Mix 8 µl of rhTIMP2 curve dilutions, 12.8 µl of activated rhMMP-2, and 59.2 µl of assay buffer, including a control containing assay buffer and the diluted rhMMP-2 and incubate the reactions for 2 hours at 37 ° C. Loading 50 µl of the incubated mixtures which were diluted five-fold in assay buffer into empty wells of a plate, and start the reaction by adding 50 µl of 20 µM substrate. Include a substrate blank containing

50  $\mu$ l of assay buffer and 50  $\mu$ l of 20  $\mu$ M substrate. Then read at excitation and emission wavelengths of 320 nm and 405 nm, respectively, in kinetic mode for 5 minutes. The result was shown in Figure 1 and it was obvious that recombinant human TIMP2 significantly decreased rhMMP2 activity. The inhibition IC<sub>50</sub> was <0.04 nM.

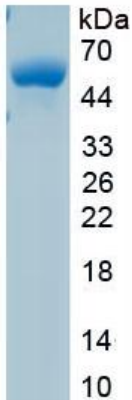


**Figure 1. Inhibition of MMP2 activity by recombinant human TIMP2**

**[ IDENTIFICATION ]**



**Figure 2. Gene Sequencing (extract)**



**Figure 3. SDS-PAGE**

**Sample: Active recombinant TIMP2, 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.