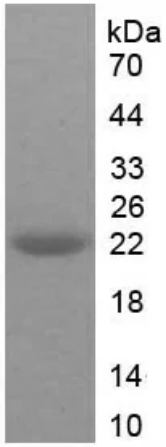
Active Tissue Inhibitors Of Metalloproteinase 3 (TIMP3)

Catalog#:PER12223

概述（Summary）

别称（Alternative Names）	SFD; TIMP Metallopeptidase Inhibitor 3; Metallopeptidase Inhibitor 3; Sorsby Fundus Dystrophy; Pseudoinflammatory
纯度（Purity）	Greater than 97% as determined by reducing SDS-PAGE.
内毒素（Endotoxin level）	Please contact with the lab for this information
蛋白构建（Construction）	【Ser27-Thr209】
Accession #	P48032
表达宿主（Host）	E.coli
种属（Species）	Rattus norvegicus (Rat)
预测分子量（Predicted MW）	22.4 kDa
制剂（Form）	Lyophilized from a 0.2 μm filtered solution of 20mM Tris, 150mM NaCl, pH8.0, containing 1mM EDTA, 1mM DTT, 0.01% SKL, 5% Trehalose and Proclin300.
运输方式（Shipping）	The product is shipped with ice bag. Upon receipt, store it immediately at the temperature listed below.
稳定性&储存（Stability &Storage）	12 months at -20°C to -80°C in lyophilized state.6 months at -20°C to -80°C under sterile conditions after reconstitution.7-10 days at 2°C to 8°C under sterile conditions after reconstitution.Please minimize freeze-thaw cycles.
复溶（Reconstitution）	Reconstitute in 20mM Tris, 150mM NaCl (pH8.0) to a concentration of 0.1-1.0 mg/mL. Do not vortex.
背景介绍（References）	Complexes with metalloproteinases (such as collagenases) and irreversibly inactivates them by binding to their catalytic zinc cofactor. May form part of a tissue-specific acute response to remodeling stimuli.

电泳图（SDS-PAGE image）



Greater than 97% as determined by reducing SDS-PAGE.

活性图（Bioactivity image）

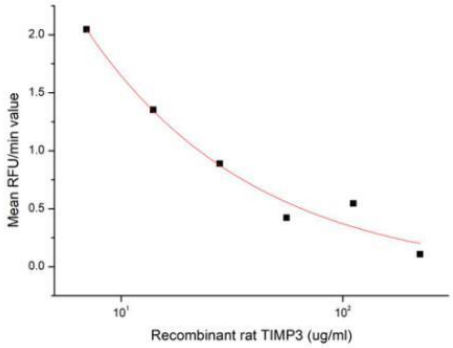
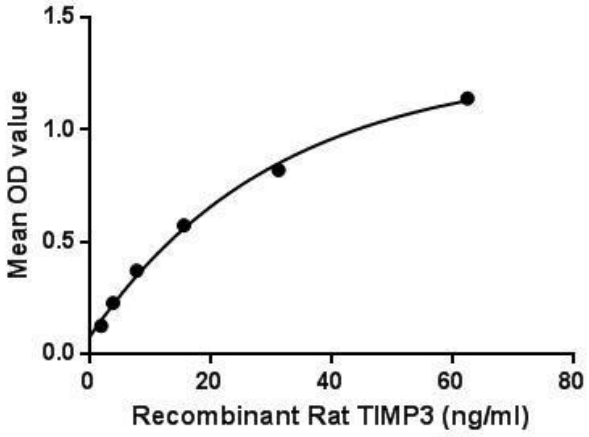


Figure 2. Inhibition of MMP2 activity by recombinant rat TIMP3

Figure. The binding activity of TIMP3 with MMP2. Tissue Inhibitors Of Metalloproteinase 3 (TIMP3) is an 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 rat TIMP3 and recombinant rat 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℃. 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℃. 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. The activity of recombinant rat TIMP3 was measured by its ability to inhibit rhMMP2 cleavage of a fluorogenic peptide substrate MCA-Pro-Leu-Gly-Leu-DPA-Ala-Arg-NH2 in the assay buffer 50 mM Tris, 10 mM CaCl2, 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 rrTIMP3 (MW: 23.25 KD) was diluted to different concentrations with the assay buffer. Mix 8 µl of rrTIMP3 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 µl of assay buffer and 50 µl of 20 µM substrate. Then read at excitiation 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 rat TIMP3 significantly decreased rhMMP2 activity. The inhibition IC50 was <1 nM.



注意事项（Note）

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