APA531Hu61 100μg
Active Plasminogen Activator Inhibitor 2 (PAI2)
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: Met1~Pro415
Tags: N-terminal His-tag
Purity: >80%
Endotoxin Level: <1.0EU per 1μg (determined by the LAL method).
Buffer Formulation: PBS, pH7.4, containing 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: 48.2kDa
Accurate Molecular Mass: 60kDa 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 (pH7.4) 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]

MEDLCVANTLFALNLFKHLAKASPTQNLFLSPWSISSTMAMVYMGSRGSTEDQMAKVLQFNEVGANAVT
PMTPENFTSCGFMQQIQKGSYPDAILQAQAADKIHSSFRSLSSAINASTGNYLLESVNKLFGEKSASFR
EEYIRLCQKYYSSEPQAVDFLECAEEARKKINSWVKTQTKGKIPNLLPEGSVDGDTRMVLVNAVYFKGK
WKTPFEKKLNGLYPFRVNSAQRTPVQMMYLREKLNIGYIEDLKAQILELPYAGDVSMFLLLPDEIADVS
TGLELLESEITYDKLNKWTSKDKMAEDEVEVYIPQFKLEEHYELRSILRSMGMEDAFNKGRANFSGMSE
RNDLFLSEVFHQAMVDVNEEGTEAAAGTGGVMTGRTGHGGPQFVADHPFLFLIMHKITNCILFFGRFSS
P

[ACTIVITY]

Serpin B2, also known as PAI-2, is an approximately 60 kDa serine protease inhibitor. It is primarily secreted by macrophages and monocytes and can form disulfide-linked multimers. Serpin B2 inhibits both the urokinase-type and tissue-type plasminogen activators (uPA and tPA). Serpin B2 also promotes the clearance of uPA by enhancing its binding and uptake by LRP. It limits fibril formation by Huntington protein (HTT) and beta-Amyloid peptides. It promotes Th2 biased immune responses and is important for intestinal CCL2 production, monocyte recruitment, and nematode clearance. A non-glycosylated form of Serpin B2 is retained intracellularly where it interferes with TNF-a induced apoptosis by protecting the Retinoblastoma protein (RB1) from calpain digestion. It

also inhibits proteasome activity in activated endothelial cells. The activity of recombinant human PAI-2 was measured by its ability to inhibit uPA cleavage of a peptide substrate, N-carbobenzyloxy-Gly-Gly-Arg-7-amido-4-methylcoumarin (Z-GGR-AMC) in the assay buffer 50 mM Tris, 0.01% Tween 20, pH 8.5. The 50 ul different concentrations of rhPAI-2 (MW: 48.2 KD) was incubated with 50ul 2ug/ml rhuPA (EPA140Mu61) at room temperature for 15 minutes. Loading 50 μL of the incubated mixtures into empty wells of a plate, and start the reaction by adding 50 μL of 200 μM substrate (Z-GGR-AMC). Include a substrate blank containing 50 μL of assay buffer and 50 μL of 200 μM substrate. Then read at excitiation and emission wavelengths of 380 nm and 460 nm, respectively, in kinetic mode for 5 minutes. The result was shown in Figure 1 and it was obvious that recombinant human PAI2 significantly decreased uPA activity. The inhibition IC50 was <28 nM.

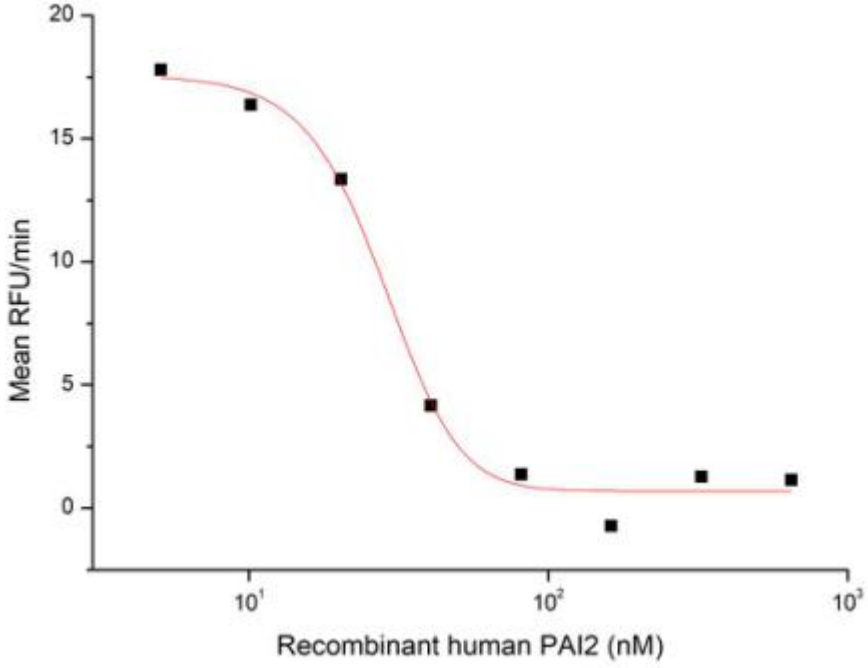

Figure 1. Inhibition of uPA activity by recombinant human PAI2

[IDENTIFICATION]

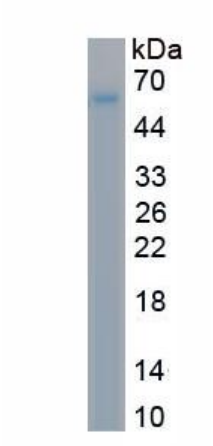

Figure 2. SDS-PAGE

Sample: Active recombinant PAI2, 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.