

APA525Hu62 100µg

Active Tissue Plasminogen Activator (tPA)

Organism Species: *Homo sapiens (Human)*

Instruction manual

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Ser36~Pro562

Tags: N-terminal His-tag

Purity: >95%

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

Buffer Formulation: PBS, pH7.4, containing 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: 7.5

Predicted Molecular Mass: 60.7kDa

Accurate Molecular Mass: 70kDa 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 (pH7.6) 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]

SYQVICRDEKTMIIYQQHQSWLRPVLRSNRVEYCWCNSGRAQCHSVPVKSCSEPRCFNGGTCQQALYFSDFVCQCEGFAGKCCCEIDT
RATCYEDQGGISYRGTWSTAESGAECTNWSSALAQKPYSGRRPDAIRLGLGNHNYCRNPDRDSKPWCYVFKAGKYSSEFCSTPACSEG
NSDCYFGNGSAYRGTHSLTESGASCLPWNSMILIGKVYTAQNPSAQUALGLGKHNHYCRNPDGDAKPWCHVLKNRRLTWEYCDVPSCSTC
GLRQYSQPQFRIKGLFADIASHPWQAIFAKHRRSPGERFLCGGILISSCWILSAAHCFQERFPPHHLTVILGRTYRVVPGEEEQKF
EVEKYIVHKEFDDDTYDNDIALLLQLKSDSSRCAQESSVVRTVCLPPADLQLPDWTECELSGYGKHEALSPFYSERLKEAHVRLYPSR
CTSQHLLNRTVTDNMLCAGDTRSGGPQANLHDACQGDSSGGLVCLNDGRMTLVGIIISWGLGCGQKDVPGVYTKVTNYLDWIRDNMRP

[ACTIVITY]

Plasminogen activators are serine proteases that catalyze the activation of plasmin via proteolytic cleavage of its zymogen form plasminogen. Plasmin is an important factor in fibrinolysis, the breakdown of fibrin polymers formed during blood clotting. There are two main plasminogen activators: urokinase (uPA) and tissue plasminogen activator (tPA). Tissue plasminogen activators (tPA) are used to treat medical conditions related to blood clotting including embolic or thrombotic stroke, myocardial infarction, and pulmonary embolism. Besides, Plasminogen Activator Inhibitor 1 (PAI1) has been identified as an interactor of tPA, thus a binding ELISA assay was conducted to detect the interaction of recombinant human tPA and recombinant human PAI1. Briefly, tPA were diluted serially in PBS, with 0.01% BSA (pH 7.4). Duplicate samples of 100ul were then transferred to PAI1-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-tPA 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 tPA and PAI1 was shown in Figure 1, and this effect was in a dose dependent manner.

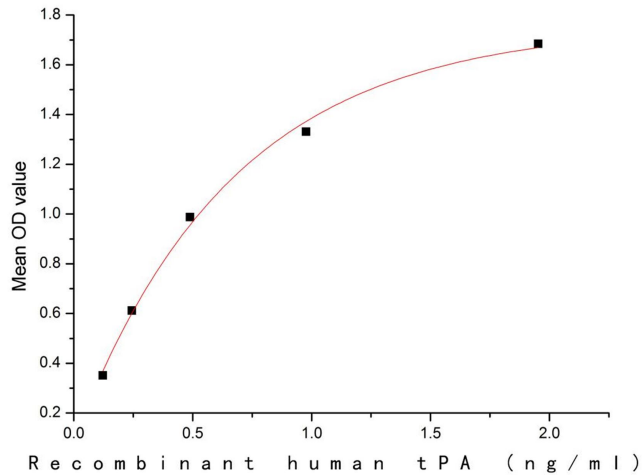


Figure 1. The binding activity of tPA with PAI1

[IDENTIFICATION]

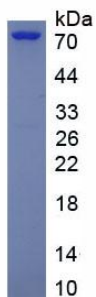


Figure 2. SDS-PAGE

Sample: Active recombinant tPA, Human

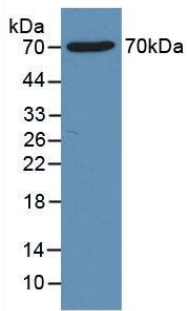


Figure 3. Western Blot

Sample: Recombinant tPA, Human;

Antibody: Rabbit Anti- Human tPA Ab (PAA525Hu06)

[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.