

APA800Hu01 50µg
Active Pregnancy Associated Plasma Protein A (PAPPA)
Organism Species: *Homo sapiens* (Human)
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: Leu317~Pro532

Tags: N-terminal His-tag

Purity: >95%

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

Buffer Formulation: PBS, pH7.4, containing 0.01% SKL, 5% Trehalose.

Original Concentration: 350µg/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: 25.9kDa

Accurate Molecular Mass: 33kDa 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]

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LLDT SLEPPLCGQT LCDNTEVIAS YNQLSSFRQP  
KVVRYRVVNL YEDDHKNPTV TREQVDFQHH QLAEAFKQYN ISWELDVLEV  
SNSSLRRRLI LANCDISKIG DENCDPECNH TLTGHGGDC RHLRHPAFVK  
KQHNGVCDMD CNYERFNFDG GECCDPEITN VTQTCFDPDS PHRAYLDVNE  
LKNILKLDGS THLNIFFAKS SEEELAGVAT WP
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[ACTIVITY]

Pregnancy-associated plasma protein A (PAPPA), also known as pappalysin-1, is a secreted protease whose main substrate is insulin-like growth factor binding proteins. PAPPA's proteolytic function is activated upon collagen binding. It is thought to be involved in local proliferative processes such as wound healing and bone remodeling. Low plasma level of this protein has been suggested as a biochemical marker for pregnancies with aneuploid fetuses (fetuses with an abnormal number of chromosomes). Besides, Plasminogen (Plg) has been identified as an interactor of PAPPA, thus a binding ELISA assay was conducted to detect the interaction of recombinant human PAPPA and recombinant human Plg.

Briefly, PAPPAs were diluted serially in PBS with 0.01% BSA (pH 7.4). Duplicate samples of 100 μ L were then transferred to Plg-coated microtiter wells and incubated for 2h at 37 $^{\circ}$ C. Wells were washed with PBST and incubated for 1h with anti-PAPPA 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 $^{\circ}$ C. Finally, add 50 μ L stop solution to the wells and read at 450nm immediately. The binding activity of PAPPA and Plg was shown in Figure 1, and this effect was in a dose dependent manner.

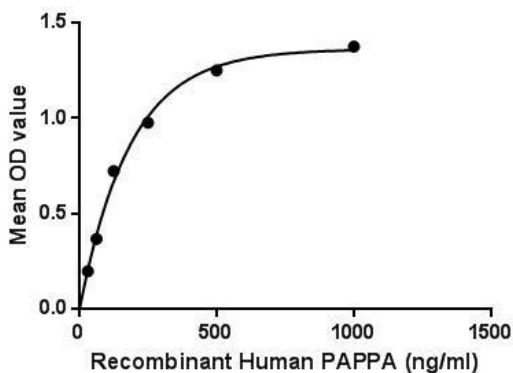


Figure 1. The binding activity of PAPPA with Plg.

[IDENTIFICATION]

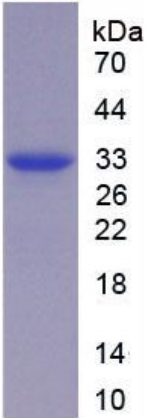


Figure 2. SDS-PAGE

Sample: Active recombinant PAPP A, Human

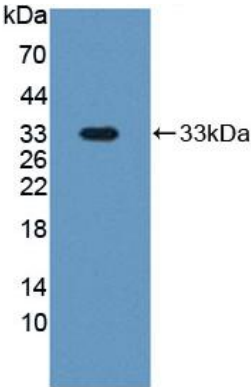


Figure 3. Western Blot

Sample: Recombinant PAPP A, Human;

Antibody: Rabbit Anti-Human PAPP A Ab (PAA800Hu01)

[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.