APA151Hu61 100μg
Active Prostate Specific Antigen (PSA)
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: Ala18~Pro261
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 .
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: 5.1
Predicted Molecular Mass: 28.5kDa
Accurate Molecular Mass: 35kDa 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.

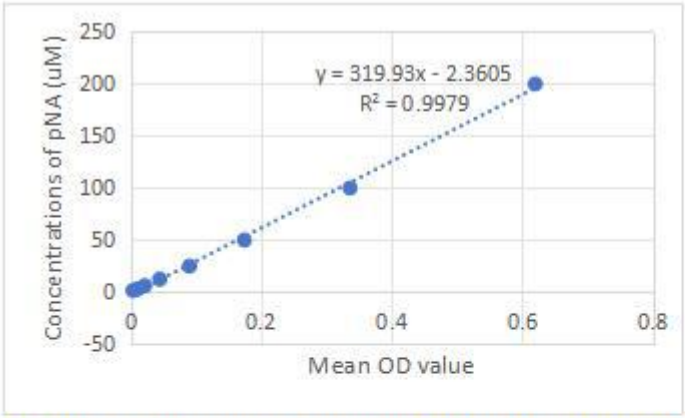
[SEQUENCE]

APL ILSRIVGGWE CEKHSQPWQV LVASRGRAVC
GGVLVHPQWV LTAAHCIRNK SVILLGRHSL FHPEDTGQVF QVSHSFPHPL
YDMSLLKNRF LRPGDDSSHD LMLLRLSEPA ELTDAVKVMD LPTQEPALGT
TCYASGWGSI EPEEFLTPKK LQCVDLHVIS NDVCAQVHPQ KVTKFMLCAG
RWTGGKSTCS GDSGGPLVCN GVLQGITSWG SEPCALPERP SLYTKVVHYR
KWIKDTIVAN P

[ACTIVITY]

Kallikrein 3, commonly known as prostate specific antigen (PSA), is a serine protease of the human tissue Kallikrein gene family. PSA is synthesized in the ductal and acinar epithelium of the prostate gland and secreted into the seminal plasma in high concentrations (0.5 - 2 g/L). A small portion of PSA “leaks” into the systemic circulation, the levels of which increase significantly (30-fold) from prostate cancer tissue than normal prostate tissue. PSA has become a well established tumor marker that aids the diagnosis, staging, and follow up of prostate cancer. The activity of recombinant human KLK3 is measured by its ability to cleave a colorimetric peptide substrate Suc-Arg-Pro-Tyr-pNa in the assay buffer 50 mM Tris, 1 M NaCl, pH 8.0. The rhKLK3 is diluted to 200 ug/ml in activation

buffer 50 mM Tris, 10 mM CaCl2, 150 mM NaCl, 0.05% Brij-35, pH 7.5, then activated with a final concentration of 1 ug/ml Thermolysin at 37 ℃ for 5min. Adding a final concentration of 10 mM 1,10 Phenanthroline to stop the activation. The activated rhKLK3 is diluted to 50 ug/mL in assay buffer. Loading into a clear well plate 50 µL of 50 ug/mL rhKLK3 and start the reaction by adding 50 µL of 2 mM substrate, with a substrate blank containing 50 µL assay buffer, 50 µL substrate, and no rhKLK3. Then read at 405 nm in kinetic mode for 5 minutes. The specific activity of recombinant human KLK3 is > 50 pmol/min/µg.



OD (405 nm)	pNA (uM)
0.6202	200
0.3364	100
0.1746	50
0.0906	25
0.0449	12.5
0.0221	6.25
0.0116	3.125
0.004	1.5625

Figure 1. The standard curve of P-nitroaniline (pNA)

Specific Activity (pmol/min/µg) =

$$\frac{\text{Adjusted Vmax * (OD/min) x Conversion Factor ** (pmol/OD)}}{\text{amount of enzyme (ug)}}$$

*Adjusted for Substrate Blank

**Derived using calibration standard P-nitroaniline

[IDENTIFICATION]

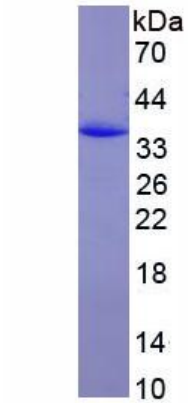


Figure 2. SDS-PAGE

Sample: Active recombinant PSA, 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.