APA889Hu61 100μg
Active Renin (REN)
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: CHO Cell
Residues: Pro25~Arg406
Tags: N-terminal His-tag
Purity: >97%
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: 6.1
Predicted Molecular Mass: 43.9kDa
Accurate Molecular Mass: 44kDa as determined by SDS-PAGE reducing conditions.

[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 37ºC 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]

PTDTTT FKRIFLKRMP SIRESLKERG
VDMARLGPEW SQPMKRLTLG NTTSSVILTN YMDTQYYGEI GIGTPPQTFK
VVFDTGSSNV WVPSSKCSRL YTACVYHKLF DASDSSSYKH NGTELTLRYS
TGTVSGFLSQ DIITVGGITV TQMFGEVTEM PALPFMLAEF DGVVGMGFIE
QAIGRVTPIF DNIISQGVLK EDVFSFYYNR DSENSQSLGG QIVLGGSDPQ
HYEGNFHYIN LIKTGVWQIQ MKGVSVGSST LLCEDGCLAL VDTGASYISG
STSSIEKLME ALGAKKRLFD YVVKCNEGPT LPDISFHLGG KEYTLTSADY
VFQESYSSKK LCTLAIHAMD IPPPTGPTWA LGATFIRKFY TEFDRRNNRI
GFALAR

[ACTIVITY]

Renin is an enzyme that plays a crucial role in the regulation of blood pressure and electrolyte balance by activating the renin-angiotensin-aldosterone system (RAAS). It is secreted by the kidneys in response to low blood pressure or low sodium levels.The activity of recombinant human REN is measured by its ability to cleave a fluorogenic peptide substrate Arg-Glu(EDANS)-Ile-His-Pro-Phe-His-Pro-Phe-His-Leu-Val-Ile-His-Thr-Lys(dabcyl)-Arg
in the assay buffer 50 mM Sodium Acetate, 150 mM NaCl, 2 mM EDTA, pH 5.0 .Recombinant human REN was diluted to 200ug/ml in the activation buffer 5 mM Tris, 15 mM NaCl, 1 mM CaCl2, 0.005% Brij-35, pH 7.5, then activated by Trypsin in a final concentration of 4ug/ml incubated at 37 °C for 1 hours. Stop activation with PMSF at 1 mM and incubate at room temperature for 30 min.The activated rhREN is diluted toto different concentrations in assay buffer. Loading into a black well plate 50 µL rhREN and start the reaction by adding 50 µL of 20 µM substrate, with a substrate blank containing 50 µL assay buffer, 50 µL substrate, and no rhREN.Then read at excitiation and emission wavelengths

of350 nm and 490 nm, respectively, in kinetic mode for 5 minutes. The specific activity of rhREN is >13 pmol/min/μg

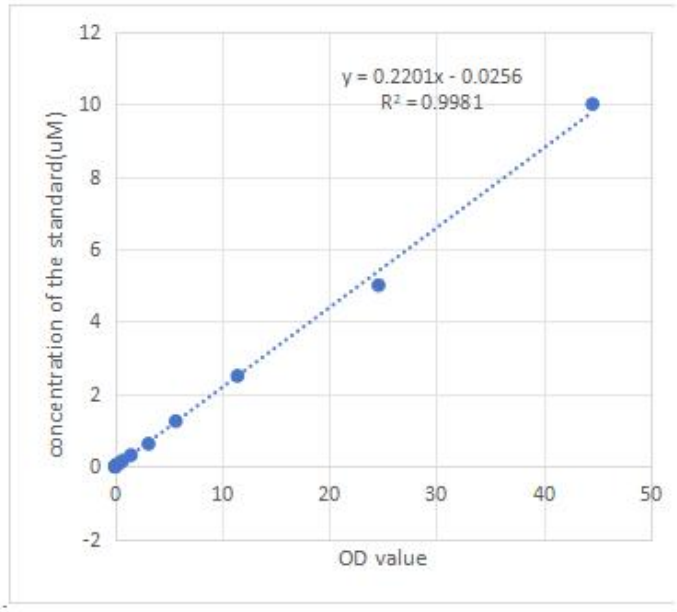

Figure 1. The standard curve of Fmoc-Glu(EDANS)-OH

OD value	Concentration of Fmoc-Glu(EDANS)-OH(uM)
44.5669	10
24.6069	5
11.4569	2.5
5.6999	1.25
3.1569	0.625
1.5169	0.3125
0.7300	0.15625
0.3860	0.078125
0.1616	0.0390625
0.0960	0.01953125
0.0584	0.009765625
0.0363	0.004882813
0.0557	0.002441406
0.0473	0.001220703
0.0177	0.000610352

$$\text{Specific Activity (pmol/min/μg)} = \frac{\text{Adjusted Vmax* (RFU/min) x Conversion Factor** (pmol/RFU)}}{\text{amount of enzyme (μg)}}$$

*Adjusted for Substrate Blank

**Derived using calibration standard Fmoc-Glu(EDANS)-OH (Bachem, Catalog # B3620).

[IDENTIFICATION]

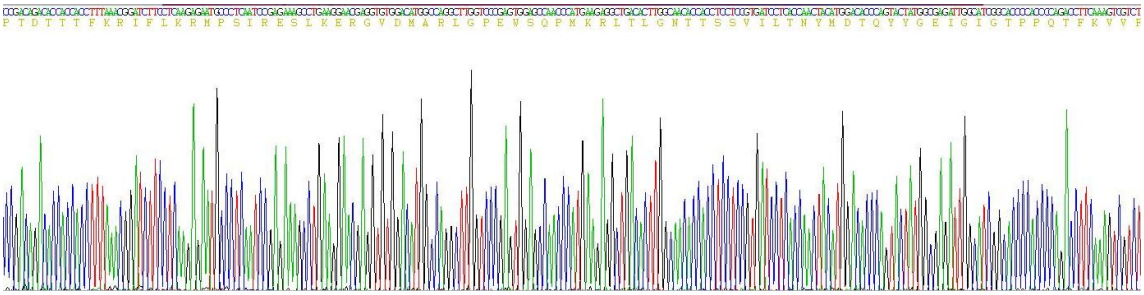

Figure 2. Gene Sequencing (extract)

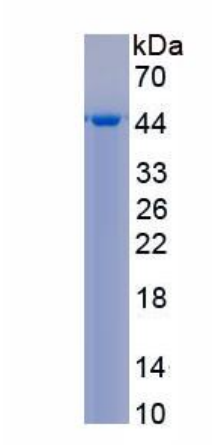

Figure 3. SDS-PAGE

Sample: Active recombinant REN, 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.